Indian Society of Human Genetics

ISHG 2025

49th Annual Meeting of Indian Society of Human Genetics and International Conclave on Neurogenetics

JANUARY 20–22, 2025

NIMHANS Convention Centre, Bengaluru

www.ishg2025.org

ABSTRACT BOOK

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CONTENTS

WELCOME MESSAGE

Dear all,

Greetings from the Organisers of ISHG 2025, Bengaluru!

We are happy and deeply honoured to host the **49th Annual Meeting of the Indian Society of Human Genetics (ISHG 2025) and International Conclave on Neurogenetics at the Convention Centre, NIMHANS, Bengaluru, from January 20 - 22, 2025.** NIMHANS, an Institute of National Importance, embodies the vision and mission 'To be a world leader in the area of Mental Health and Neurosciences and evolve state-of-the-art approaches to patient care through translational research'. With its foundation built on the three pillars of patient care, teaching/training, and research, NIMHANS stands out as a beacon of excellence.

The **three days** conference will cover a wide range of topics, including **new technologies and their applications and other research breakthroughs, highlighting the latest and most thrilling advancements in genetics and genomics for the current year.** It will provide an ideal platform for sharing pioneering research findings, acquiring fresh insights, and staying abreast of the progress in the field.

We are delighted to extend a warm invitation to this premier academic event dedicated to expanding our knowledge and understanding of health and disease, fostering collaboration, and inspiring innovation in Human Genetics. ISHG 2025 features stalwarts in the field as keynote speakers and a diverse array of didactic lectures by national and international experts, in addition to a compelling panel discussion on 'Genetic Research in India: quo vadis?', young scientist presentations, oral and interactive poster presentations by budding researchers. ISHG 2025 promises to be one of the biggest academic feasts highly relevant to fledgling scholars and experienced academicians seeking to delve deeper into the latest advancements and collaborations in Human Genetics.

Bengaluru, often referred to as the Silicon Valley of India, is the hub for scientific and technological advancements and is home to a multitude of prestigious organisations fostering a culture of innovation. Beyond the academic realm, the city with its rich cultural heritage and natural beauty showcases it's lush green parks and serene gardens, museums, historical monuments, and religious getaways. It is also complemented by the theatre and entertainment industry, silk weaving and wood craftsmanship, multicuisine eateries, and cosmopolitan populace. Notwithstanding the extreme and intense traffic, Bengaluru is a treasure trove waiting to be explored. As the capital of Karnataka, it exudes energy, seamlessly blending tradition with modernity. It's warm hospitality and vibrant atmosphere make it an appealing destination that leaves visitors spellbound.

We heartily welcome distinguished researchers, clinicians, scholars, Life Science/biotech companies as well as diagnostic and pharmaceutical firms, and genome artists to join us at the largest scientific gathering of Human Genetics in India. Let us together explore the current advances in genomic sciences, expand the frontiers of research in human diseases and celebrate the exchange of knowledge as well as the spirit of academic excellence at NIMHANS, Bengaluru.

Prof. Monojit Debnath
Organising Chairperson - ISHG 2025
Department of Human Genetics,
NIMHANS, Bengaluru

Dr. Mathivanan Jothi
Organising Secretary & Treasurer - ISHG 2025
Department of Human Genetics,
NIMHANS, Bengaluru

DIRECTOR'S MESSAGE

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It is an immense pleasure to welcome you all to the **49th Annual Meeting of the Indian Society of Human Genetics (ISHG 2025) and the International Conclave on Neurogenetics** being held at the **National Institute of Mental Health and Neuro Sciences (NIMHANS), Bengaluru,** from **January 20-22, 2025.**

This conference represents a unique platform where the leading experts in human genetics, in particular neurogenetics, can exchange their groundbreaking ideas, fostering collaborations and exploring the future frontiers in genetics. With the rapid advancements in human genetic research and the generation of vast amounts of genetic data, I hope this gathering will promise to address critical challenges, develop innovative approaches, and advance translational research for the benefit of the deserving. The conference theme reflects the significance of neurogenetics in unraveling complex neuropsychiatric disorders and its potential to transform diagnostics, therapeutics, and healthcare delivery. Over three days, this event will host lectures by distinguished speakers, insightful panel discussions, oral and poster presentations, and awards to encourage young researchers, which will definitely deepen our genetic understanding.

As one of the India's premier Institutes for Mental Health and Neurosciences and an Institute of National Importance, it is a perfect venue to host this meeting. I encourage all delegates to engage wholeheartedly, share their expertise, and explore opportunities to collaborate beyond this event. I hope together, we can push the boundaries of human genetic research to new horizons.

On behalf of the organizing committee, I extend my warmest wishes for a groundbreaking conference. Let us make ISHG 2025 an event to remember, driving impactful research and meaningful progress in the field of genetics.

I am looking forward to seeing you all in NIMHANS, Bengaluru!

Prof. Pratima Murthy
Director, NIMHANS, Bengaluru

MESSAGE FROM THE ISHG PRESIDENT

Greetings for a very happy, healthy and fulfilling new year!

It is my privilege to pen this Presidential message on the occasion of the 49th Annual meeting of Indian Society of Human Genetics (ISHG) and International Conclave on Neurogenetics, being hosted at the National Institute of Mental Health and Neurosciences (NIMHANS), Bengaluru. NIMHANS, mapped as an Institute of national importance, is also renowned worldwide for excellent clinical services, empowering teaching/training programs and high-quality research. All these activities are dedicated to Brain health and disease, Brain biology, and overall Mental wellbeing. Therefore, it is only appropriate that ISHG 2025 encompasses substantial coverage of all aspects of neurosciences under the associated umbrella of the international conclave on Neurogenetics.

My warm greetings to all the participants from India and overseas for ISHG 2025 conference which I am confident will turn out to be truly informative, thought provoking and scientifically enriching. ISHG always strives for excellence and novelty in all its activities year after year. The Society as all the members would know has also initiated summer training fellowships and Outreach activities in the last year. In addition to the ongoing Young Scientist Award (YSA) presentations which are research scholars' and post docs' dream, Emerging Scientist Award (ESA) is a new attraction for the early/mid-career faculty during ISHG 2025, which gives them an opportunity to showcase their original research findings.

"For last year's words belong to last year's language. And next year's words await another voice" - T.S. Eliot.

You would all witness, I hope, that this one single line conveys the tone, the theme and the essence of this year's meeting which is focused on current findings and updates in topics of contemporary significance to the human genetics and genomics community.

My congratulations to the Organisers of this conference for their painstaking efforts in structuring the meeting to represent/reflect a complete range of domains from Discovery genomics to disease biology, to therapeutics and big data analyses.

Secretariat: Centre for DNA Fingerprinting and Diagnostics (CDFD), Inner Ring Road, Uppal, Hyderabad – 500 039, Telangana, INDIA
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MESSAGE FROM THE ISHG PRESIDENT

The Indian Society of Human Genetics

(Registered under S. R. Act, 1860)

With leaders from the field as keynote and invited speakers to share the latest trends and findings in this fast-changing face of Human Genetics and Biomedical Genomics arenas, this meeting is indeed one to be remembered for a long time to come.

A notable balance between high quality basic research in neurosciences as well as other areas of rare disorders, complex traits including malignancies, equally important clinical domains, fascinating cellular models of disease offering insights into biology and new therapeutics, and finally, the early success stories of the ultimate merger of disease related genetic findings with genome (somatic) editing technology and subsequent gene therapy for a couple of hematological disorders in the country are promising highlights of the meeting. Of note, the emerging area of Regulatory genomics is another fascinating area being covered in this meeting.

The Panel discussion on 'Whither to' in Human Genetics research in India, needs a special mention. Aimed at covering the advancements, lacunae/limitations and thoughts/action plans for the future in both academic research and industry/translation settings offers an exciting possibility of a strong message or a white paper emerging from ISHG 2025 to be shared with the policy makers. This is particularly relevant given the laudable achievements that the country has witnessed over the last year. These include the successful completion and data release of the landmark Genome India project; 1st-in-Human lentiviral gene therapy trial for hemophilia A; Numerous animal models and patient derived organoids for functional genomics to establish genotype-phenotype correlation; for screening and development of new therapeutics; multi-OMICS approaches for biomarker discovery and of course Artificial Intelligence in diagnostics and more.

With technologies available and all this work becoming possible, the time to achieve Good Health and Well-being for all, SDG#3 is NOW. Therefore, I would take this opportunity to encourage all the participants to actively interact with each other and explore the possibility of establishing mutually beneficial collaborative efforts among the clinical, basic research and industry community gathered at ISHG 2025. This would help leap frog country's research efforts to the desired and highly likely next and essential level of translation to clinical settings for the myriad of genetic disorders which continue to pose a challenge.

"The future belongs to those who believe in the beauty of their dreams." Eleanor Roosevelt.

Thelma B K
President, ISHG

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MESSAGE FROM THE ISHG SECRETARY

The Indian Society of Human Genetics

(Registered under S. R. Act, 1860)

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Message from the Secretary:

The last two decades have witnessed major breakthroughs in genetic research leading to major innovative solutions to some of the rare and complex human diseases, offering a path to a healthy life. Nobel prizes in physiology, medicine, and chemistry for recent years, including discoveries of microRNA, nucleotide base modification, and genomes of extinct hominins and human evolution are all testimony to these great advances in genetic research. It is my privilege and honor to extend a warm welcome to all the researchers working on human genetics. across India for the 49th Annual Meeting of the Indian Society of Human Genetics (ISHG 2025) and International Conclave on Neurogenetics at the Convention Centre, NIMHANS, Bengaluru, from January 20-22, 2025.

I am sure this meeting spanning over three days would provide a platform for stimulating interactions and sharing of knowledge pertaining to genetic research being done in India, particularly by the young researchers, forging newer connections and collaborations. I extend my heartfelt gratitude to each participant, speaker, and sponsor who are contributing to making this conference a great success. I hereby extend my best wishes to the organizers and distinguished guests from abroad and India for a great and productive meeting.

Dr. Inderjeet Kaur

L V Prasad Eye Institute, Hyderabad

CONFERENCE PROGRAM

20th January, 2025 (First Day)				
8.45 AM – 10.00 AM	**Inauguration and high tea** **Chief Guest: Prof. Vinod K. Paul, Member, NITI Aayog, Government of India**			
SESSION 1 **Genome editing/ Gene Therapy**				
	No.	**Name of the Speaker**	**Venue**	**Chairpersons**
10.00 AM – 10.50 AM	OP1	**Dr. Debojyoti Chakraborty**, IGIB, New Delhi Navigating the translational route of therapeutic gene editing in resource limited settings.	Hall A	Prof. Alok Srivastava, CMC, Vellore
	OP2	**Prof. Ramachandran Shaji,** CMC, Vellore Lentiviral gene therapy vectors for haemoglobinopathies.		
SESSION 2 **Panel Discussion on "Genetics Research in India: Quo vadis?**				
10.50 AM – 12.05 PM		**Panellists:** **Prof. RNK Bamezai,** Former VC, Shri Mata Vaishno Devi University **Topic:** Current challenges for genetic research in India and way forward. **Dr. Giriraj Chandak,** Sir JC Bose Fellow, CCMB, Hyderabad **Topic:** Phenomic complexities of diseases transcend genomic complexity: newer strategies for fishing in troubled waters! **Dr. K. Thangaraj,** Former Director, CDFD, Hyderabad **Topic:** The Hype, The Hope, The Promise and The Reality of Genome India initiative: Where India stands now? **Prof. Alok Srivastava,** St. John's National Academy of Health Sciences, Bengaluru **Topic:** Developing technologies for gene therapy in India and their translation to products. **Dr. Vinod Scaria,** Chief Data Officer, Karkinos Healthcare, Bengaluru **Topic:** Mushrooming genome testing Industry in India: urgent need for constituting a regulatory body and policies. **Dr. Anuradha Acharya,** CEO, Mapmygenome, Hyderabad **Topic:** Pharmacogenomics vis-à-vis Precision Medicine: How to surmount the bumpy roads?	Hall A	**Moderators:** Prof. Binay Panda, JNU, New Delhi Dr. Moinak Banerjee, RGCB, Thiruvanathapuram
		Prof. B.K. Thelma, National Science Chair, University of Delhi, Delhi **Topic:** Biobanking and ethical issues in research: Can India replicate the UK biobank facility?	Hall A	

CONFERENCE PROGRAM

CONCURRENT SESSIONS 3 Theme: Neurogenetics, genetics of ageing and neurodegeneration				
12.10 PM - 1.15 PM	OP3	**Prof. Divya Mehta,** Queensland University of Technology, Australia Biopsychosocial drivers of the stress response in different populations.	Hall A	Prof. Prabha Chandra, Dean, Behavioural Sciences, NIMHANS, Bengaluru Prof. Pramod. K. Pal, NIMHANS, Bengaluru
	OP4	**Dr. Manu Sharma,** University of Tubingen, Germany Genomic peaks in Parkinson's Disease: Paving the way for precision medicine.		
	OP5	**Dr. Mohammed Faruq,** IGIB, New Delhi Conventional screening and Whole Exome Sequencing in Progressive Ataxias: Unravelling Genetic Complexity.		
12.10 PM - 1.15 PM	OP6	**Dr. Bratati Kahali,** Centre for Brain Research,Indian Institute of Science, Bengaluru Deciphering genetic architecture for complex disease traits: Insights from the Indian and other world populations.	Hall B	Prof. Anuranjan Anand, JNCASR, Bengaluru, Prof. K. Satyamoorthy, Research Director, SDM University, Karnataka.
	OP7	**Dr. Santhosh Girirajan,** Pennsylvania State University, USA Untangling genetic interactions in complex disease.		
	OP8	**Prof. Udai Bhan Pandey,** Pittsburgh School of Medicine, USA Geminopathies: mining for GEMs in rare genetic disorders.		
FREE COMMUNICATION SESSION 1 (10 mins oral presentations for young researchers) Theme: Genetics of Brain Disorders				
12.10 PM - 1.15 PM	ST1	**Dr. Bhagyalakshmi Shankarappa,** NIMHANS, Bengaluru Spatiotemporal Epigenetic variation in postmortem brain tissue samples from an Indian Population.	Hall C	
12.10 PM - 1.15 PM	ST2	**Ms. Kalyani Karunakaran,** NIMHANS, Bengaluru Temporal gene expression signatures across neurodevelopment: a transdiagnostic analysis of bipolar disorder, schizophrenia, autism, and epilepsy.	Hall C	Prof. G. Venkatasubramanian, NIMHANS, Bengaluru Dr. Moinak Banerjee, RGCB, Thiruvanathapuram
		Dr. Surya G. Krishnan, CHG, Bengaluru Plexiform Neurofibromatosis: Early experience from a Multispecialty Clinic at a single centre.		
	ST4	**Ms. Smriti Jha,** RIMS, Ranchi Association study of blood based and genetic markers predictive of cognitive decline post stroke.		
	ST5	**Dr. Pradip Paul, NIMHANS,** Bengaluru Deciphering cellular mechanisms of bipolar disorder and lithium response.		

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1.15 PM - 2.00 PM	**Lunch Break**			
2.00 PM - 3.00 PM		**Plenary Talk 1:** **Prof. Aravinda Chakravarti,** New York School of Medicine, USA Diversity of Mutations in Human Disease.	Hall A	Prof. Samir K. Brahmachari, Former Director General, Council of Scientific and Industrial Research (CSIR), Government of India. Dr. K. Thangaraj K., Former Director, CDFD, Hyderabad
SESSION 4 **Theme: Translational Genomics and Precision Medicine**				
3.10 PM - 4.15 PM	OP9	**Prof. Aurora Pujol,** Bellvitge Biomedical Research Institute, Spain Improving genomic diagnosis and therapies for brain metabolic disorders.	Hall A	Prof. Radha Rama Devi, Rainbow Children's Hospital, Hyderabad. Prof. Udai Bhan Pandey, USA
	OP10	**Dr. Prerana Jha,** Director-Lab Operations, Bencos Healthcare Solutions Pvt. Ltd., Mumbai, Maharashtra. Exome or Genome? Decoding the way forward in Genomic Medicine.		
	OP11	**Prof. Manisha Madkaikar,** Director, ICMR-NIIH, Mumbai Resolving the Unknown: Advancing Precision Diagnostics and Therapeutics in Inborn Errors of Immunity.		
SESSION 5 **Theme: Metagenomics and Human Health**				

CONFERENCE PROGRAM

3.10 PM - 4.15 PM	OP12	**Prof. Niranjan Nagarajan,** Genome Institute of Singapore, Singapore Deconstructing and Reconstructing Microbiomes and their role in AMR Transmission.	Hall B	Prof. Yogesh Shouche, Director, SKAN Research Foundation, Bengaluru Prof. Arindam Maitra, NIBMG, Kalyani
	OP13	**Mr. Mainak Chakraborty,** Senior Solutions Architect, Amazon Web Services. Revolutionize Genomics Research with AI on AWS cloud.		
	OP14	**Dr. Varun Aggarwala,** Jio Institute, Mumbai Antimicrobial Resistance Reservoir in the Gut: Baseline, Dynamics, and Reset through microbiota engineering.		
FREE COMMUNICATION SESSION 2 (10 mins oral presentations for young researchers) Theme: Genetics of complex disorders				
3.10 PM - 4.15 PM	ST6	**Dr.Sushil Selvarajan,** CMC, Vellore Impact of cancer and therapy on epigenetic landscapes of normal cells: Describing a novel endpoint in survivorship.	Hall C	Prof. Raghunath Chatterjee, ISI, Kolkata Prof. Biren Banerjee, Silicon University Bhubaneswar.
	ST7	**Ms. Khadija Sana Hafeez,** Turin, Italy. Multi-Omics Integration in Pleural Mesothelioma Reveals Key Molecular Signatures.		
	ST8	**Dr. Hafeeda Kunhabdulla,** Yenepoya University, Mangalore Molecular profiling of Oral Squamous Cell Carcinoma patients of south Indian population.		
	ST9	**Mrs. Megha Radhakrishnan,** BHU, Varanasi Molecular pathology of cervical carcinogenesis: Role of telomere and THOR in tumor initiation and progression.		
	ST10	**Ms. Uma Dharshini Karuppiah,** University of Madras, Chennai Transcriptome analysis reveals potential biomarkers in Head and Neck squamous cell Carcinoma.		
4.15 PM - 4.35 PM	**Tea Break**			
4.35 PM - 5.25 PM	**Dr. L.D. Sanghvi Oration of ISHG**		Hall A	Prof. B. K. Thelma Dr. K. Thangaraj
5.30 PM - 6.30 PM	**Plenary Talk-2:** **Prof. Eric Green,** Director, NIHGR, USA The Forefront of Genomics.		Hall A	Prof. Pratima Murthy, Director, NIMHANS, Bengaluru
6.30 PM - 7.30 PM	**Poster Presentation**			
6.30 PM - 8.00 PM	**ISHG EC MEETING**		Boardroom, Convention Centre	
7.30 PM - 8.30 PM	**QUIZ COMPETITION**		**Hall A**	
8.30 PM - 9.30 PM	**Dinner**			

CONFERENCE PROGRAM

21st January, 2025 (Second Day)				
8.00 AM-8.45AM	**Breakfast**			
8.45 AM-9.35 AM	**Plenary Talk-3:** **Prof. Henry Houlden,** University College of London, UK Using new technology to diagnose unsolved neurological disorders.		Hall A	**Chairpersons:** Prof. Yasha TC, Dean, Clinical Neuroscience, NIMHANS, Bengaluru Prof. Atchayaram Nalini, NIMHANS, Bengaluru
CONCURRENT SESSIONS 6 **Theme: Genetics of Complex Disorders/Epigenetics/Gene-environment Interactions**				
9.40 AM - 10.45 AM	OP15	**Prof. Amit Dutt,** University of Delhi, New Delhi Genetic insights into lung cancer resistance to targeted therapy.	Hall A	Prof. Sagar Sengupta, Director, NIBMG, Kalyani Prof. Annapoorni Rangarajan, Indian Institute of Science, Bengaluru
	OP16	**Prof. Sanjeev Shukla,** IISER, Bhopal Epigenetic reprogramming under hypoxia: The role of CTCF and PRMT5 in breast cancer progression.		
	OP17	**Prof. Dil-Afroze,** Sher-i-Kashmir Institute of Medical Sciences, J & K Navigating the *Helicobacter pylori* driven immune landscape from Inflammation to gastric carcinogenesis.		
9.40 AM - 10.45 AM	OP18	**Prof. Chittaranjan Yajnik,** KEM Hospital, Pune Life course evolution of diabetes in Indians: developmental origins of health and disease.	Hall B	Dr. Subhabrata Chakrabarti, LV Prasad Eye Institute, Hyderabad Prof. Dwaipayan Bharadwaj, JNU, New Delhi
	OP19	**Prof. Raghunath Chatterjee,** ISI, Kolkata Compensatory role of interleukin-17A in regulating the keratinocyte cell proliferation.		
	OP20	**Dr. Kiran Polavarapu,** CHEO Research Institute, Canada. Solving the unsolved in neuromuscular diseases: Genomic re-analysis through international collaborations using RDConnect GPAP.		
10.45 AM - 11.05 AM	**Tea break and visit to corporate stalls**			

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SESSION 6 ISHG Young Scientist Presentation				
11.05 PM - 1.20 PM	YSA1	**Mr. Sohail Rafik Mansuri,** CCMB, Hyderabad Epigenome-wide association study of childhood cognitive function identifies inflammation as a key differentially regulated pathway.	Hall A	Prof. B.K. Thelma, President, ISHG Dr. Inderjeet Kaur, Secretary, ISHG
	YSA2	**Ms. Debakreeta Ghosh**, ISI, Kolkata Unraveling the Role of the lncRNA RP11-215G15.5 in Psoriasis Pathogenesis.		
	YSA3	**Mr. Bipin Raj Shekhar,** NIRRCH, Mumbai CNTNAP2 gene deletion confers risk to schizophrenia through dendritic spine abnormalities evaluated using hiPSCs derived neuron.		
	YSA4	**Mr. Saikat Dey,** NIMHANS, Bengaluru Genetic modulation of systemic and neuro-inflammation in progressive Supranuclear Palsy.		
	YSA5	**Ms. Debolina Saha,** NIRRCH, Mumbai Developing a patient-friendly diagnostic method for mitochondrial disorders: correlation of genetic variants with multi-complex deficiencies.		
	YSA6	**Ms. Kavya V.P.,** NIMHANS, Bengaluru Transgenerational effect of maternal stress on the behavioral outcomes of children: the role of maternal immune activation and epigenetic modulation.		
	YSA7	**Ms. Smita Saha,** CDFD, Hyderabad Mutations in HYPK cause a novel neurodevelopmental disorder by impairing neuronal proteostasis and autophagy.		
	YSA8	**Ms. Pratibha Banerjee,** CUP, Bathinda. Gliadin-induced pathogenic differential transcriptome signature of Gut mucosa in celiac disease and its mitigation through a gluten-free dietary component using a patient-derived organoid model.		
	YSA9	**Ms. Swati Singh,** Kasturba Medical College, Manipal Biallelic variants in CCN2 underlie an autosomal recessive kyphomelic dysplasia.		
1.20 PM - 2.10 PM	**Lunch Break**			
2.10 PM - 3.00 PM	**Plenary talk-4:** **Prof. Jian Jun Liu,** Director, Genome Institute of Singapore, Singapore Genetics of leprosy susceptibility and treatment outcome.		Hall A	**Chairpersons:** Prof. Kunal Ray, Ramakrishna Mission Vivekananda Educational & Research Institute, Kolkata
SESSION 7 Emerging Scientist Award				

CONFERENCE PROGRAM

3.00 PM - 4.20 PM		**Dr. Aditi Chandra,** Saha Institute of Nuclear Physics, Kolkata A multi-enhancer hub controls dosage of Ets1 gene expression and protects from allergic inflammation.	Hall A	Dr. G.R. Chandak, CCMB, Hyderabad Prof. S. Ganesh, IIT, Kanpur
		Dr. Ashitha S.N.M., NIMHANS, Bengaluru Neurodevelopmental dysregulation underpins pathogenesis of severe mental illnesses.		
		Dr. Jeru Manoj Manuel, uMaster, Canada Decoding the transcriptome usage & Alternative Splicing signature (AS) during neuronal differentiation: Deep gene regulatory approach.		
		Dr. Priyanka Upadhyai, Kasturba Medical College, Mangalore A tale of two cellular processes: cilia-autophagy crosstalk in bone development and the pathogenesis of skeletal dysplasias.		
		Dr. Sabyasachi Senapati, Central University of Punjab, Bhatinda A cryptic tale besides HLA- Associations in Celiac Disease		
4.20 PM - 4.45 PM	**Tea Break and Visit to Corporate Stalls**			
SESSION 8 Theme: Genetic landscape and treatment avenues for rare genetic disorders				
4.45 PM - 5.45 PM	OP21	**Dr. Ashwin Dalal,** CDFD, Hyderabad Indian Genomics Landscape: Achievements and opportunities.	Hall A	Prof. Reena Gulati, JIPMER, Pondicherry Prof. Sumita Danda, CMC, Vellore
	OP22	**Prof. Radha Rama Devi,** Rainbow Children's Hospital, Hyderabad. MPS I – calibrating treatment outcome & India experience.		
	OP23	**Prof. Anju Shukla,** Kasturba Medical College, Manipal, Karnataka Initiating multi-center efforts for addressing rare disease challenges in India.		
SESSION 9 Theme: Genetics of malignancies				
4.45 PM - 5.45 PM	OP24	**Prof. Ellora Sen,** NBRC, Manesar Metabolic-Epigenetic shifts in Gliomas: Role of IDH1 mutation.	Hall B	Prof. Gopeshwar Narayan, BHU, Varanasi Prof. Sunita Singh, BHU, Varanasi.
	OP25	**Prof. Hirofumi Nakaoka,** Sasaki Institute, Japan Clonal selection and diversification of somatic mutations in normal human endometrial epithelium.		
	OP26	**Prof. Pranay Tanwar,** AIIMS, New Delhi Decoding the hedgehog signaling pathway in acute myeloid leukemia: Biomarkers and therapeutic perspectives.		

CONFERENCE PROGRAM

SESSION 10 Theme: Advances in Genomic Technologies and Clinical Awareness			
5.45 PM - 6.45 PM	**Dr. Jim Zhang,** Senior Field Application Scientist at Twist Biosciences. Maximizing Sensitivity in Liquid Biopsy: Advances in twist cfDNA library preparation and custom panel solutions.	Hall A	Prof. K. Premkumar, Bharathidasan University, Tiruchirappalli Dr. Kamlesh Guleria, Guru Nanak Dev University, Amritsar
	Dr. Ravi Kumar Chilukoti, Head, India & SA Technical Support & PM, MGI Tech co., Ltd. MGI's SEQ ALL Landscape: Full spectrum of sequencing technologies from short-read to Long-read sequencing.		
	Mr. Amit Bhat, Business Development Manager, Molecular Devices. Exploring Neurobiology in 3D.		
6.25 PM - 6.45 PM	**Poster Presentation**		
7.00 PM - 8.00 PM	**ISHG GB MEETING**	**Hall A**	
8.00 PM - 9.30 PM	**CULTURAL PROGRAMME AND GALA DINNER**		

CONFERENCE PROGRAM

22nd January, 2025 (Third Day)				
8.00 AM- 8.45 AM	**Breakfast**			
8.45 AM - 9.35 AM	**Plenary talk 5:** **Prof. Vorasuk Shotelersuk,** Chulalongkorn University, Thailand Genomics Thailand and Rare Diseases: Advancing precision public health and uncovering novel disease genes.		Hall A	**Chairpersons:** Prof. Ashutosh Halder, AIIMS, New Delhi Dr. Ashwin Dalal, CDFD, Hyderabad
CONCURRENT SESSIONS 11 **Theme: Chromosome biology, genome architecture and regulation**				
9.35 AM - 10.40 AM	OP27	**Prof. Sagar Sengupta,** Director, NIBMG Small molecules reverting chemoresistance via chromatin remodelling are viable candidates for adjunct therapy in colon cancer.	Hall A	Prof. S. Ganesh, IIT, Kanpur Prof. RNK Bamezai, Former Vice Chancellor, Shri Mata Vaishno Devi University, J&K
	OP28	**Prof. Sathees Raghavan** Indian Institute of Science, Bengaluru Mechanism of chromosomal translocations in lymphoid neoplasia: Focus on non-B DNA and RAGs.		
	OP29	**Prof. Chandrima Das,** SINP, Kolkata Epigenetic regulator ZMYND8 drives tumor heterogeneity through metabolic reprogramming in breast cancer.		
9.35 AM - 10.40 AM	OP30	**Dr. Padavattan Sivaraman,** NIMHANS, Bengaluru Unveiling the Parkinson's disease-associated a-Synuclein role in chromatin regulation.	Hall B	Prof. AJS Bhanwer, SGRDIMSR, Amritsar Dr. Bibhas Kar, Kokilaben Dhirubhai Ambani Hospitals, Mumbai
	OP31	**Dr. Hemachander Subramanian,** NIT, Durgapur Structure-function relationship in DNA: The importance of kinetics.		
	OP32	**Dr. Divya Tej Sowpati,** Centre for Cellular and Molecular Biology, Hyderabad Optical Genome Mapping: Next generation cytogenetics		

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FREE COMMUNICATION SESSION 3 (10 mins oral presentations for young researchers) Theme: Disease Genetics				
9.35 AM - 10.40 AM	ST11	**Mrs. Syed Ali Fathima Afrin J,** SASTRA Deemed University, Tamil Nadu Exploring the DNA methylation profile associated with Bardet-Biedl syndrome: An EPIC array-based approach.	Hall C	Prof. Inusha Panigrahi, PGIMER, Chandigarh Dr. Renu Saxena, Sir Ganga Ram Hospital, New Delhi
	ST12	**Dr. Shantanab Das,** ISI, Kolkata HLA-C and HLA-B Shape Psoriasis risk and protection profiles.		
	ST13	**Dr. Amol N Patil,** PGIMER, Chandigarh Development and validation wise assessment of genotype guided warfarin dosing algorithm in Indian population.		
	ST14	**Mr. Kondyarpu Abhishek,** ILS, Bhubaneswar Otitis Media, an underrated complex disease: From candidate gene to next generation sequencing and from confusion to clarity, in the end pragmatically a covalent compromise?		
	ST15	**Dr Shalini S Nayak,** Kasturba Medical College, Manipal Investigation of clinical and genetic heterogeneity in human fetuses with arthrogryposis multiplex congenita.		
10.40 AM - 11.00AM	**Tea break and visit to corporate stalls**			
SESSION 12 Theme: Genomic Technologies, big data, and precision medicine				
11.00 AM - 12.05 PM	OP33	**Prof. Analabha Basu,** NIBMG, Kalyani Why India needed a genome sequencing project of its own?	Hall A	Prof. B. K. Thelma, University of Delhi, Delhi Prof. Manisha Madkaikar, Director, ICMR-NIIH, Mumbai
	OP34	**Prof. Binay Panda,** JNU, New Delhi *X-plat*: a cross-platform transformation tool based on the nearest neighbor joining framework.		
	OP35	**Dr. Arunabha Majumdar,** IIT, Hyderabad A unified Bayesian approach to transcriptome-wide association study.		

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SESSION 13 Theme: Genome to Phenome				
11.00 AM - 12.05 PM	OP36	**Prof. Arindam Maitra,** NIBMG, Kalyani Born Too Soon: The molecular underpinnings of preterm birth in Indian women.	Hall B	Prof. Mitali Mukherjee, IIT, Jodhpur Prof. Govindasamy Kumaramanickavel, Director of Research, Narayana Netralaya, Bengaluru
	OP37	**Prof. Uppala Radhakrishna,** The University of Pittsburgh School of Medicine, USA Placental epigenetic signatures can predict long-term health outcomes in infants prenatally exposed to opioids and neonatal opioid withdrawal syndrome.		
	OP38	**Prof. Ashutosh Halder,** AIIMS, New Delhi Genetic landscape of Polycystic Ovary Syndrome (PCOS).		
FREE COMMUNICATION SESSION 4 (10 mins oral presentations for young researchers) Theme: Functional Genomics				
11.00 AM - 12.05 PM	ST16	**Mr. Bishnu Prasad Parida,** BHU, Varanasi Choline kinase isoform regulation in Cervical cancer: Implications in tumor progression and therapeutic targeting.	Hall C	Dr. Indrani Datta, NIMHANS, Bengaluru Dr. Dhanjit Das, National Institute for Research in Reproductive Health, Mumbai
	ST17	**Ms. Vibhaa Kumar,** TIGS, Bengaluru Gene editing reveals mutation specific disease manifestations in a human pluripotent stem cell derived Pompe disease model.		
	ST18	**Ms. Aishwarya Murali,** IIT, Madras Cellular specificity of ribosomal protein gene expression in human tissues.		
	ST19	**Dr. Priyanka Srivastava,** PGIMER, Chandigarh Cytogenetic landscape of products of conceptions from recurrent pregnancy loss in north Indian population.		
	ST20	**Ms. Merin George,** NIIH, Mumbai Comprehensive genomic and global DNA methylation profiling in Indian Fanconi Anaemia subjects.		

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SESSION 14 Theme: Innovations in genomic tools and treatment of genetic disorders				
12.05 PM - 1.20 PM	OP39	**Dr. Kriti Kaushik,** Product Manager- Multiomics at Premas Life Sciences. Advancing human healthcare through multiomics' insights.	Hall A	Dr. Vinod Scaria, Karnikos Healthcare, Bengaluru Dr. Prabhakar Kedar, NIIH, Mumbai
	OP40	**Prof. Sumita Danda,** Christian Medical College, Vellore Uncovering ASMD: Recent updates and management.		
	OP41	**Dr. Dan Fordham,** Director, Strategic Product Management EMEA, Oxford Nanopore Technologies. The path to a true genome - Reimagining a new standard using all read sequencing.		
	OP42	**Dr. Rahul Solanki,** Application Scientist, Agilent Technologies, India. Elevating Precision Oncology: Tumor genomic profiling solutions advancing precision oncology.		
1.20 PM - 2.00 PM	**Lunch Break and Poster Presentation**			
SESSION 15 Theme: Modelling of Human Diseases/Regenerative Medicine				
2.00 PM - 3.05 PM	OP43	**Prof. Jens Christian Schwamborn** University of Luxembourg, Germany Modeling parkinson's disease with human midbrain organoids.	Hall A	Prof. Krishanu Ray, Director, NBRC, Gurgaon Dr. Sridhar Sivasubbu, Former Chief Scientist, IGIB, New Delhi
	OP44	**Dr. Aarti Sevilimedu,** Dr. Reddy's Institute of Life Sciences, Hyderabad Modelling rare genetic disorders in Zebrafish: insights into disease mechanisms.		
	OP45	**Prof. Anindya Ghosh Roy,** NBRC, Manesar Regenerating and reconnecting Neural circuits.		
SESSION 16 Theme: Regulatory Genomics				
2.00 PM - 3.05 PM	OP46	**Prof. Swapan K. Nath,** OMRF, USA Decoding the genetic architecture of SLE: insights from Post-GWAS analyses and functional characterization of an Asian-Specific regulatory variant.	Hall B	Dr. V. Babu Rao, NIIH, Mumbai & Prof. A.K. Munirajan, University of Madras, Chennai
	OP47	**Dr. Rachel A.J.,** Formerly Associated with CCMB, Hyderabad Regulation of autosomal genes by noncoding RNAs from Y-chromosomal heterochromatin and implications in male fertility.		
	OP48	**Dr. Dimple Notani,** NCBS, Bengaluru Roles of transcriptional enhancers in disease susceptibility.		

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FREE COMMUNICATION SESSION 5 (10 mins oral presentations for young researchers) Theme: Population genetics				
2.00 PM - 3.05 PM	ST21	**Dr. Amit Gourav Ghosh,** Nanyang Technological University, Singapore Genomic Insights into North Eurasian and South American indigenous populations shaped by diverse environments.	Hall C	Dr. Swarkar Sharma, Central University of Jammu, J&K Dr. Anamika Singh, National University of Singapore, Singapore
	ST22	**Dr. Ranajit Das,** Yenepoya University, Karnataka Individuals' unique genetic makeup influences their susceptibility towards infectious diseases: COVID-19 as a case study.		
	ST23	**Mr. Sachin Rathod,** IIT, Jodhpur Genetic basis of climatic adaptation in Thar desert populations.		
	ST24	**Ms. Haya Afreen,** NIBMG, Kalyani Understanding South Asia's population history through archaic Introgression.		
	ST25	**Dr. M.S. Mustak,** Mangalore, University, Mangalore. Maternal genetic landscape of Konikanī population.		
SESSION 17 Theme: Genetics of brain disorders				
3.05 PM - 4.30 PM	OP49	**Prof. Quasar S Padiath,** University of Pittsburgh, USA Mechanisms underlying cell type specificity in CNS disorders: Lessons from autosomal dominant leukodystrophy.	Hall A	Prof. B.S. Sankarnarayana Rao, Registrar, NIMHANS, Bengaluru Prof. Anju Shukla, Kasturba Medical College, Manipal
	OP50	**Prof. Subrata Sinha,** AIIMS, New Delhi Behaviour, genetics & genes: connecting some dots in familial dyslexia.		
	OP51	**Dr. Ganesh Chauhan,** RIMS, Ranchi MRI markers of cerebral small vessel diseases and their genetic determinants.		
	OP52	**Prof. Santasree Banerjee,** Jilin University, China Application of genomic sequencing technologies: Genetic molecular diagnostics of developmental delay and intellectual disability.		

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SESSION 18 Theme: Gene discovery and management of genetic diseases				
3.05 PM - 4.30 PM	OP53	**Prof. Virginia Kimonis,** University of California, USA The use of antisense oligonucleotides for treating rare genetic diseases.	Hall B	Dr. P.V. Ramchander, ILS, Bhubaneswar Dr. Inderjeet Kaur, LV Prasad Eye Institute, Hyderabad
	OP54	**Prof. Arijit Mukhopadhyay,** University of Salford, UK Beyond the Lab: How storytelling with genetics is revolutionizing glaucoma care.		
	OP55	**Prof. Nachimuthu Senthil Kumar,** Mizoram University, Mizoram Environmental and genetic studies reveal potential risks factors for substance Use disorder in the high incidence mizo population of North-East India.		
	OP56	**Dr. Swarkar Sharma,** Central University of Jammu, Jammu Pharmacogenomics in populations of Jammu and Kashmir.		
4.30 PM - 5.30 PM	**High Tea & Valedictory Session**			

ABSTRACTS

Plenary Sessions

The Diversity of Mutation in Human Disease

Prof. Aravinda Chakravarti, *Director, Center for Human Genetics and Genomics, New York School of Medicine, United States. Email: Aravinda.Chakravarti@nyulangone.org*

Much of what we now know of human genetic disease has been learnt from monogenic disorders of high penetrance and high expressivity. However, most of human genetic disease has properties unlike these and has genetic lessons quite unlike what we have learnt. Most human diseases are polygenic, susceptible to environmental influences, have low penetrance and highly variable expressivity. I will discuss work from my laboratory, and those of others, to propose a new functional model for such so called 'complex' disorders and how monogenic disorders can emerge from this architecture.

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The Forefront of Genomics

Prof. Eric Green, *M.D., Ph.D., Director, National Human Genome Research Institute National Institutes of Health, United States.*

The coming decade offers great promise for human genomics and genomic medicine. Since the completion of the Human Genome Project over two decades ago, genomics has become progressively entrenched within the bedrock of the biomedical research enterprise. Capitalizing on the momentum of the project's successful completion, the field of genomics has increasingly expanded and matured, such that genomics is now central and catalytic in basic and translational research, and studies now regularly demonstrate the vital role that genomic information can play in clinical care. Looking ahead, the anticipated advances in technologies, biological insights, and clinical applications (among others) will lead to more widespread dissemination of genomics throughout biomedical research, a growing adoption of genomics into medical and public-health practices, and an increasing relevance of genomics in everyday life. To capitalize on these opportunities, the U.S. National Human Genome Research Institute continually develops and leads programs and initiatives that address the most pressing challenges at *The Forefront of Genomics*, with a particular emphasis on understanding the biological complexities of the human genome, on untangling the complex roles that genomic variants play in health and disease, and making genomics broadly and equitably integrated into medicine.

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Using new technology to diagnose unsolved neurological disorders

Prof. Henry Houlden, *University College London Institute of Neurology, Queen Square, London*
Email: h.houlden@ucl.ac.uk

There are many neurological conditions and families that clearly have a inherited aetiology, and the disease segregates in families but we have yet to find the genetic cause. This is even after using exome and genome sequencing in patients from the UK and countries around the world. We are therefore, looking at using new technology to define these mystery disorders and their often difficult to define causes with a particular focus on bioinformatics and long-read sequencing that can sequence very difficult and repetitive regions of the genome. Repeat expansion disorders are increasing common in neurological disorders and many new repeats have been found over the last few years in children and adults. Repeat expansion disorders can cause disorders such as Friedreich's ataxia, juvenile Huntington's disease, intellectual disability in Fragile X, childhood parkinsonism and epilepsy. Repeat expansions can be missed and be difficult to diagnose in undefined families and present clinically as cerebral palsy and developmental delay. Genetic diagnosis is key in these conditions as the repeat expands in subsequent family generations, and is clinically more severe, known as anticipation, making family planning essential.

Examples are given of how we use advanced bioinformatics and long-read sequencing, combined with genetic and clinical expertise to investigate families. With examples and video's from patients, recent difficult to find and newly published genes and novel repeat expansion disorders. I discuss how to identify and genetically diagnose patients and the use of long-read genetic sequencing to do this. I also speculate on common undiagnosed conditions and discuss how mystery genetic causes could be a key factor in their causation and offer collaboration to teams interested in these conditions.

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Genetics of Leprosy Susceptibility and Treatment Outcome

Prof. Jianjun Liu, *Executive Director, Genome Institute of Singapore, Singapore*
Email: liuj3@gis.a-star.edu.sg

Leprosy is a chronic infectious disease caused by the bacteria *Mycobacterium leprae*. It primarily affects the skin, peripheral nerves, mucous membranes of the upper respiratory tract, and eyes, and if untreated, can lead to disfigurement, sensory loss, and disabilities. Leprosy remains to be a public health challenge, with more than 180,000 new cases of leprosy reported globally in 2023, most of that were from South and

Southeast Asia, South America and Africa. Multidrug therapy (MDT) can treat the disease effectively, but may cause adverse drug reaction (ADR) in certain individuals.

Biological understanding of leprosy is limited. Microbiological research of *M. leprae* has been challenging, because this bacteria cannot be cultured *in vitro*. And, human is the primary host for *M. leprae*, which made difficult to investigate the disease using animal models. By focusing on Chinese population, we have carried out a series of genetic association studies and discovered a good number of genetic susceptibility loci for leprosy. These discoveries have provided valuable biological insights into molecular mechanism of leprosy as well as genetic information for risk stratification and leprosy prevention. In addition, our studies have also revealed the genetic basis of leprosy treatment-induced ADR. This discovery has enabled the development of clinical assay and pre-emptive test that can help to eliminate this deadly ADR. Our efforts have demonstrated the power of human genetic study to advance the biological understanding of this infectious disease.

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Genomics Thailand and Rare Diseases: Advancing Precision Public Health and Uncovering Novel Disease Genes

Prof. Vorasuk Shotelersuk, *Chulalongkorn University, Faculty of Medicine, Bangkok, Thailand*

Email: vorasuk.s@chula.ac.th

Genomics Thailand (GeTh) is a groundbreaking national initiative driving human genome research and applications in Thailand. Launched in 2020, Phase I of GeTh aims to sequence the whole genomes of 50,000 Thai individuals by 2024, targeting key areas such as cancer, rare diseases, non-communicable diseases, pharmacogenomics, and infectious diseases. This effort, involving 36 medical centers nationwide, has successfully collected blood samples from all participants, with the majority already sequenced.

As GeTh transitions into Phase II, the program is poised to integrate genomic technologies into clinical practice while fostering new industrial developments. This next phase emphasizes workforce expansion through training clinical geneticists, genetic counselors, molecular scientists, and bioinformaticians. Additionally, it prioritizes enhancing genomic literacy, public engagement, and addressing ethical, legal, and social considerations.

In the rare disease research domain, GeTh leverages long-read sequencing to uncover novel disease genes, including *YEATS2*, linked to Benign Adult Familial Myoclonic Epilepsy 4 (BAFME4), and *RAI1* associated with BAFME8. This technology proves invaluable in diagnosing complex cases unresolved by exome or short-read genome sequencing, particularly in disorders involving challenging genes like *CYP21A2* in congenital adrenal hyperplasia and *D4Z4* in facioscapulohumeral muscular dystrophy. Furthermore, its utility in rapidly diagnosing critically ill patients underscores its transformative potential.

This presentation highlights the accomplishments of GeTh Phase I and offers insights into its role in advancing precision public health and rare disease diagnostics, paving the way for a genomics-driven healthcare future.

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ABSTRACTS

Invited Oral Presentations

OP1

Navigating the translational route of therapeutic gene editing in resource limited settings

Dr. Debojyoti Chakraborty, *Institute of Genomics and Integrative Biology, New Delhi*

Genome and transcriptome editing toolboxes are expanding at an extremely rapid pace but the road to transformative clinical translation is limited by multiple factors ranging from comprehensively profiling the safety of these advanced technologies to developing strategies for equitable accessibility options for patients from diverse backgrounds. Our work addresses both these aspects through protein engineering of CRISPR effectors and their derivatives to developing pipelines for taking lab level discoveries to the clinic through coordinated programs with multiple stakeholders in the patient communities, academia, government and industry. Through focused disease associated gene therapy strategies, we are gradually developing and improving first generation platform technologies to make them affordable and accessible to patients suffering from inherited disorders.

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OP2

Lentiviral gene therapy vectors for haemoglobinopathies

Prof. Shaji R.V., *Department of Haematology, Christian Medical College, Vellore*

Lentiviral gene therapy vectors are suitable for gene therapy for hemoglobinopathies by reactivating fetal hemoglobin (HbF). We have engineered erythroid lineage-specific shRNA lentiviral vectors (Ery-Lin-shRNA) designed for the targeted silencing of gamma globin gene repressors. These vectors were optimized for enhanced knockdown efficiency and increased HbF expression. Our in vitro and in vivo evaluations confirm that Ery-Lin-shRNA vectors achieve specific and effective knockdown in erythroid cells, leading to substantial HbF induction (over 40%) while preserving the functionality of hematopoietic stem and progenitor cells (HSPCs) and maintaining specificity without affecting non-erythroid lineages. Furthermore, the vectors support efficient cloning of shRNAs, facilitating high-throughput RNAi screens to discover novel regulatory genes in erythropoiesis. The results highlight the potential of Ery-Lin-shRNA vectors as a powerful approach for hemoglobinopathy gene therapy.

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OP3

Biopsychosocial drivers of the stress response in different populations

Prof. Divya Mehta, *Queensland University of Technology, Australia*

Why some people develop mental health disorders after facing trauma and others do not is a key question in research. Our research at the Stress Genomics Lab, Queensland University of Technology, aims to investigate the biological, psychological, and social drivers of health and well-being after exposure to trauma or stress across a range of different populations including combat veterans, first year university students, emergency responders and elite athletes. Methodologies include a longitudinal and prospective study design and an integrative approach of omics and novel statistical pipelines to uncover health trajectories. Results of risk and protective factors of health as well as genes and pathways that drive the stress response will be presented.

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OP4

Genomic peaks in Parkinson's Disease: paving the way for precision medicine

Dr. Manu Sharma, *Centre for Genetic Epidemiology, Institute for Clinical Epidemiology and Applied Biometry, University of Tuebingen, Germany*

Preceding the discovery of the alpha-synuclein (SNCA) gene, the role of genetics in the etiopathogenesis of Parkinson's disease (PD) was contested. Since the discovery of the SNCA gene, the number of genetic loci implicated in familial forms of Parkinson's disease has increased to 26. In parallel, the economic affordability of array-based approaches has revolutionized genomic research by allowing the processing of hundreds of thousands of subjects to perform genome-wide association studies (GWAS) to understand the role of common genetic variants in myriad diseases. During the last 15 years, the number of genetic loci in PD has increased from 2 to 78. Though this has provided a set of novel genetic readouts to understand the disease mechanisms, it also highlighted pertinent questions: (i) which genetic loci should be prioritized for downstream analyses; (ii) what strategies should be implemented to refine these putative regions; (iii) do these genetic loci have a transferability in other populations? What is the road ahead for PD genomic research in the underrepresented populations?

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OP5

Conventional screening and Whole Exome Sequencing in Progressive Ataxias: Unraveling Genetic Complexity

Dr. Mohd Faruq, *Institute of Genomics and Integrative Biology, New Delhi*

Purpose: To improve diagnosis and understanding of genetically complex cerebellar ataxias.

Methods: n=4600, Whole exome sequencing of 613 cases, gene burden analysis, and case-based analysis.

Results: Identified novel and known ataxia-associated genes, including ATM, SETX, ANO10, and SACS. Uncovered potential genetic links with other neurological disorders. Achieved 60% diagnostic yield, combining SNV and tandem repeat analysis.

Conclusion: Integrative genetic approach enhances diagnostic accuracy and provides insights into the molecular basis of cerebellar ataxias.

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OP6

Deciphering genetic architecture for complex disease traits: Insights from the Indian and other world populations

Dr. Bratati Kahali, *Centre for Brain Research, Indian Institute of Science, Bengaluru*

The focus in human genetics has shifted in the past couple of decades to conducting large-scale biomedical projects with deep phenotyping and genome sequencing to gather population-specific knowledge for the benefit of human health in several countries. The GenomeIndia project is one such national flagship project aimed at identifying genetic variations from 10,000 individuals hailing from 80+ well-defined ethnolinguistic groups from more than 100 distinct geographical locales. The successful completion of this project has successfully identified >135 million high confidence SNVs and INDELs, including ultra rare variants, singletons, and doubletons. A substantial proportion of these variations are population-specific and rare. We have also created a haplotype reference imputation panel with this dataset, which shows improved imputation accuracy and allelic concordance for Indian population genotypes compared to the well-known TOPMed and Haplotype Reference Consortium panels. We have thereafter, used these discoveries to decipher the genetic architecture of complex traits manifesting phenotypic variations for assessing dyslipidemia, glycemic imbalance, and cognitive measures in the Indian population. These results sometimes overlap with known disease associations in the other world populations, nevertheless, some of them are novel in Indians, with implications for understanding and ameliorating health disparities. We have identified several genetic loci susceptible for cognitive domains, with a few overlaps and intriguing diXerential

haplotype specificities across Indian and other ancestry individuals. A subset of significant genetic hits for cognition indicates mediating eXects via metabolic traits, example, lipid and glycemic factors. Some of the

genes and variants identified have known function in neurodevelopmental, neurodegenerative and synaptic pathways. Therefore, these ongoing studies are helping us to understand the population level genetic diversity of India, emphasizing the medical relevance of discovered genetic variants, thereby facilitating the contribution of genomics in public health for the nation.

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OP7

Untangling genetic interactions in complex disease

Prof. Santhosh Girirajan, *Pennsylvania State University, United States*

Recent studies have suggested that genetic variants of different classes, frequency, and effect sizes individually or collectively contribute to complex disorders, including autism, obesity, and intellectual disability. Several themes have emerged from these studies that present challenges rendering our understanding of these disorders incomplete. Despite initial claims, most single genes or variants are now known to be associated with incomplete penetrance and variable expressivity, and this has complicated our understanding the molecular basis of these disorders and contributed to challenges in genetic diagnosis and counseling of affected families. My research uses a combination of human genetics, model systems, and computational approaches to dissect the genetic architecture of complex disorders. I will present examples of our work on family-based genetic studies, animal models, and population-scale datasets to provide evidence for how genetic interactions can explain the missing heritability of complex disorders.

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OP8

Geminopathies: mining for GEMs in rare genetic disorders

Prof. Udai Bhan Pandey, *Director, Children's Neuroscience Institute, Children's Hospital of Pittsburgh University of Pittsburgh Medical Center, Pittsburgh, United States.*

GEMIN5 is essential for core assembly of small nuclear Ribonucleoproteins (snRNPs), the building blocks of spliceosome formation. We identified novel autosomal recessive mutations in GEMIN5 gene among patients presenting with developmental delay, motor dysfunction and cerebellar atrophy. We found that these GEMIN5 variants perturb snRNP complex protein expression and assembly. While doing an in vivo genetic screen, we identified SMN as a genetic suppressor of GEMIN5-mediated neurotoxicity in Drosophila. We discovered that an increase in SMN expression by either genetically or the antisense oligonucleotide (ASO) Nusinsersen, significantly upregulated the expression of GEMIN5 in mammalian cells and mutant GEMIN5

derived iPSC neurons. Furthermore, we identified a strong functional association between the expression patterns of SMN and GEMIN5 in patient Spinal Muscular Atrophy (SMA) derived motor neurons harboring loss of function mutations in the SMN gene.

Interestingly, SMN binds to the C-terminus of GEMIN5 and regulates GEMIN5 expression through the Tudor domain. Lastly, we observed that SMN upregulation ameliorates defective snRNP biogenesis and alternative splicing defects caused by loss of GEMIN5 in iPSC neurons and in vivo. Collectively, our work indicates that SMN is a potent regulator of GEMIN5 expression and neuropathologies.

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OP9

Advancing Diagnosis and Treatment for Brain Metabolic Disorders

Prof. Aurora Pujol, Director, *Neurometabolic Diseases Laboratory, IDIBELL, Barcelona, Spain*

Diagnosing rare disorders through genomics remains a significant challenge. Current methods solve fewer than 50% of undiagnosed cases, often following years of challenging diagnostic odysseys. To address this, we developed **ClinPrior**, a computational tool that analyzes whole-exome and genome data based on clinical phenotypes and expanded associated interactome networks.

We applied ClinPrior to more than 300 undiagnosed patients presenting brain white matter abnormalities, spastic paraplegia, and/or ataxia phenotypes. Functional validation of variants of uncertain significance followed, achieving a diagnostic success rate of 70%. This yield doubles previous studies using next-generation sequencing (NGS) panels and clinical exomes for these entities. Our approach integrated multi-omics profiling of patient samples, *in vitro* studies, and *in vivo* disease modeling using *Drosophila melanogaster* and *Danio rerio*, demonstrating the value of combining clinical and functional genomics within the same lab.

Through this work, we discovered 12 novel ultra-rare brain disorders, many involving dysfunctions in critical genes controlling lipid metabolism. Examples include genes involved in ceramide biosynthesis (**DEGS1**), phosphatidylinositol metabolism (**PIK4A**), complex phospholipid synthesis (**PCYT2**, **SLC35B2**), and lipid droplet formation (**RINT1**). These findings highlight the essential role of lipid homeostasis in maintaining brain health.

Notably, some of the metabolic pathways identified are actionable, offering therapeutic potential. For **DEGS1 leukodystrophy**, we repurposed fingolimod, a multiple sclerosis treatment, which improved ceramide metabolism and locomotor abnormalities in a zebrafish model. This strategy earned Orphan Drug Designation from the European Medicines Agency (EMA) (EU/3/23/2776) and facilitated an international compassionate use trial. In patients, fingolimod corrected biochemical and neurological disease biomarkers, including NfL and GFAP, paving the way for clinical trials.

Beyond rare genetic diseases, ClinPrior has also been pivotal in uncovering host genetic factors underlying severe COVID-19 in adults and multisystem inflammatory syndrome in children (MIS-C). In collaboration with the COVID-Human Genetics Effort (CHGE) consortium, we identified mutations in Interferon-responsive genes, showcasing the broader societal benefits of clinical genomics.

This project reflects a collaboration between the Spanish Network for Rare Diseases (CIBERER), the IMPaCT Genómica Program, and international reference centers for leukodystrophies and spastic paraplegias, Robert Debré Hospital (Paris), Childrens Hospital of Philadelphia, Childrens Hospital of Pittsburg, UDNI (Undiagnosed Disease Network International, NIH-USA), and CHGE (Rockefeller University). We are deeply grateful to the families, clinicians, and funding agencies who made this work possible.

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OP10

Exome or Genome? Decoding the Way Forward in Genomic Medicine

Dr. Prerana Jha, *Ph.D. Director-Lab-Ops Bencos Healthcare Solutions Pvt Ltd.*

Genomic medicine is transforming healthcare by providing insights into various disease mechanisms, risk detection, rare disease, diagnosis, and personalized cancer treatments. Among genomic technologies, whole exome sequencing (WES) and whole genome sequencing (WGS) stand out for their broader diagnostic potential compared to single-gene assays, arrays, and targeted sequencing. However, the choice between WES and WGS remains crucial in advancing clinical applications, especially in resource-constrained settings. Exome sequencing, which focuses on protein-coding regions, offers a cost-effective solution with high diagnostic accuracy for many Mendelian disorders. On the other hand, WGS provides a more comprehensive analysis by capturing regulatory regions, structural variants, and non-coding elements—offering critical insights for rare and complex disorders but at higher costs and data complexity. In India, affordability challenges and limited access to advanced sequencing and analytical options hinders the routine adoption of these technologies. To bridge these gaps, we are introducing an innovative, cost-effective, end-to-end exome sequencing solution to enhance accessibility without compromising sensitivity or accuracy. Furthermore, we advocate for the technology that integrates the strengths of WES and WGS to optimize cost, diagnostic yield, and clinical utility. With robust bioinformatics, data science, and evidence-based clinical reporting, we aim to make affordable genomic technologies an integral part of precision medicine. Our goal is to ensure that genomic advancements benefit diverse populations, addressing affordability and technological challenges in both clinical and research settings.

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OP11

Resolving the Unknown: Advancing Precision Diagnostics and Therapeutics in Inborn Errors of Immunity

Dr. Manisha Madkaikar, *Director, ICMR-National Institute of Immunohaematology, Mumbai.*

Inborn Errors of Immunity (IEI) are inherited disorders of immune system manifesting with susceptibility to infections, autoimmunity, autoinflammation and malignancy. While NGS offers a very powerful tool for identifying pathogenic variants resulting in IEIs, we often identify variants of unknown significance (VUS), posing challenges for accurate diagnosis and management. Often either these VUS are ignored or considered as pathogenic while making key management decisions. This talk will highlight the importance of an integrated approach with comprehensive clinical, immunological and functional evaluation in resolving VUS. We will also discuss how these approaches enhance our understanding of immune dysregulation, enable precise variant classification, and inform personalized treatment strategies. By bridging the gap between genotype and phenotype offers new insights into IEI, driving advancements in precision medicine for immune disorders.

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OP12

Deconstructing and Reconstructing Microbiomes and their role in AMR Transmission

Prof. Niranjan Nagarajan, *Genome Institute of Singapore, Singapore*

We live in a microbial world estimated to contain >1 million species with vast genetic diversity arising from 4 billion years of evolution. Microorganisms are adapted to most habitats as complex communities shaped by ecological interactions and yet little is known about how this unseen world is organized. Humanity's perception of microbes is shaped by a small fraction of pathogenic species leading to an adversarial relationship defined by the pervasive use of antimicrobial agents. Efforts to eradicate microbes have met with limited success. Disinfected environments are rapidly recolonized while antibiotic treatments increasingly select for resistant pathogens.

The global rise in antimicrobial resistance (AMR) rates for common pathogens (e.g. ESKAPE) is recognized as a pre-eminent threat to healthcare systems. As the range of effective antibiotics shrinks we approach a tipping point where no antibiotic works for a pathogen, putting at risk the lives of millions of vulnerable patients in hospitals worldwide. Already >1 million deaths/year are attributed to AMR, surpassing estimates for COVID-19, HIV/AIDS and malaria. By 2050 the UN projects that AMR will be responsible for more deaths every year than all cancers (>10 million deaths/year), with >$3 trillion in economic impact/year by

2030. Parts of Asia are particularly vulnerable as AMR hubs, fuelled by several factors including higher infectious disease burden and inappropriate antibiotic usage.

We need radically new approaches to understand the microbial world we live in and change the rules of interaction. We propose that this involves (i) systematically *deconstructing* microbial communities by combining genome-resolved metagenomics and high-throughput culturing, (ii) *modelling* principles of community organization by studying defined complex communities *in vitro* and *in vivo*, and (iii) *reconstructing* ecological functions of natural microbial communities based on predictive models to suppress the burden of pathogenic species. In this talk, I will present some of the tools we have been developing towards these goals including for *genome-resolved metagenomics* and *generalized-Lotka-Volterra modelling* from metagenomic data. These have served to provide new insights into the impact of antibiotics on the gut microbiome and how colonization resistance might emerge, as well as the spread of multi-drug resistant organisms through hospital environments. I will highlight some of these applications in my talk and how we envision the development of *improved guidelines* and *new classes of interventions* to tackle the global challenge of antimicrobial resistance.

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OP13

Revolutionize Genomics Research with AI on AWS cloud.

Mr. Mainak Chakraborty, *Senior Solutions Architect, Amazon Web Services.*

The success rate for Phase I oncology clinical trials is significantly low. According to a study published in Nature Reviews Drug Discovery, the overall success rate for oncology drugs from Phase I to approval is around 5%, indicating a high failure rate of approximately 95%. According to the National Cancer Institute, cancer biomarkers, also known as tumor markers, are biological molecules found in blood, other body fluids, or tissues that indicate the presence of cancer. Biomarkers for patient stratification can improve the probability of success in clinical development; the average is between double and triple, but it can be as high as five-fold. In this session, we will demonstrate how agentic workflows with LLMs leverage planning, tool-use, and self-reflection to transform complex oncology research queries into actionable insights using an example analysis pipeline, specifically for lung cancer survival with clinical, genomics and radiology modalities of biomarkers. We will also showcase a variety of tools including database retrieval with Text2SQL, statistical models and visual charts with scientific libraries, biomedical literature search with public APIs and internal evidence, and medical image processing. We will discuss advanced capabilities of agents for self-review and planning that helps build trust with end users by breaking down complex tasks into a series of question and answers and showing the chain of thought to generate the final answer.

AWS is at the forefront of accelerating generative AI innovation, enabling organizations to harness the power of large language models (LLMs) and foundation models (FMs). By providing easy access to the

broadest set of high-performing generative AI models, AWS simplifies the process of building and scaling generative AI applications in life sciences from small scale process automation to fundamentally changing research and discovery. For example, AstraZeneca is accelerating the transformation of drug discovery and precision medicine using genomics, so that researchers can turn insights into science faster. Gilead is generating insights from key datasets that accelerate the analysis of large quantities of unstructured information from a variety of sources across their enterprise. Pfizer has deployed AI solutions to create medical/scientific content and patent applications, enabling breakthroughs to reach patients faster. Do join me in this session to understand how you can also harness AI to revolutionize your life sciences research using AWS.

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OP14

Antimicrobial Resistance Reservoir in the Gut: Baseline, Dynamics, and Reset through microbiota engineering.

Dr. Varun Aggarwala, *Jio Institute, Mumbai*

Antimicrobial resistance (AMR) is a global health threat, with the human gut microbiome serving as a critical reservoir for resistance genes. In India, AMR prevalence is exacerbated by antibiotic misuse, poor sanitation, and environmental contamination, yet comprehensive data on the Indian gut resistome remain scarce. Current methodologies for AMR profiling, culturing and deep metagenomics, are low-throughput and cost-prohibitive, making it impractical for large scale studies for preventive health. Here, we address these gaps by establishing a baseline of gut AMR reservoirs across demographics, health conditions, and antibiotic exposures in the Indian population and benchmark to global datasets. We also develop AMRGuard, a novel computational tool to identify and quantify AMR genes from low-depth sequencing data. We characterize the dynamics of the gut resistome, examining how it evolves with age, antibiotic use, infections, and early-life factors like breastfeeding. Finally, we show that Fecal microbiota transplantation (FMT), probiotics, and dietary modifications can reduce AMR genes and restore microbiome balance.

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OP15

Genetic insights into lung cancer resistance to targeted therapy

Prof. Amit Dutt, *Department of Genetics, University of Delhi South Campus, New Delhi*

Our efforts in lung cancer have led to describing the first large-scale systematic efforts in India, to define the landscape of all known actionable mutations among Indian lung cancer patients across subtypes - one of the most crucial pieces of information known to be ethnic specific. We identified a novel mutation in FGFR3 and presented an alternate dosing strategy of EGFR tyrosine kinase inhibitor Osimertinib in a lung cancer patient that could potentially lower the cost of treatment, and thus, could transform lung cancer care and treatment. While EGFR tyrosine kinase inhibitors initially improve outcomes, most patients relapse, often driven by concurrent genomic alterations. Here, I would present the impact of co-occurring tumor suppressor gene mutations among EGFR-mutant (mEGFR) lung adenocarcinoma patients who relapse in response to EGFR TKIs. Whole exome analysis of mEGFR patients at baseline and following relapse along with robust survival analysis will be presented. The presentation would also emphasize on clonal evolution across 200 odd longitudinally collected liquid biopsy samples using comprehensive gene panel for 500 odd genes. Our findings underscore the potential of co-occurring 17 TSG mutations as biomarkers for early relapse in mEGFR population and emphasize the importance of longitudinal monitoring to uncover the clonal dynamics. Early detection of co- occurring mEGFR and TSG mutations could guide more effective treatment strategies and improve patient outcomes.

OP16

Epigenetic Reprogramming Under Hypoxia: The Role of CTCF and PRMT5 in Breast Cancer Progression

Prof. Sanjeev Shukla, *Indian Institute of Science Education and Research, Bhopal, Bhopal, India*

Hypoxia triggers epithelial-mesenchymal transition (EMT) in cancer cells by altering chromatin landscapes, epigenetic modifications, and alternative splicing, thus promoting metastasis. Our study demonstrates that hypoxia upregulates CTCF expression through TET2-mediated DNA demethylation, which is crucial for its induction by HIF1α. This hypoxia-induced CTCF regulates the alternative splicing of genes involved in EMT, thereby advancing cancer progression. We elucidate a mechanism involving the EMT-associated gene COL5A1, where CTCF-mediated looping between the promoter and a distal exon influences DNA demethylation and RNA polymerase II pausing at exon 64, promoting alternative splicing toward EMT under hypoxic conditions. Furthermore, hypoxia-induced CTCF occupancy across the genome drives changes in gene expression and splicing related to cancer progression. Targeting the HIF1α-CTCF-COL5A1 axis using a dCas9-DNMT3A system reduces EMT potential, offering a promising therapeutic strategy for breast cancer. We also identify PRMT5, a type II protein arginine methyltransferase, as a key player under hypoxia.

Hypoxia induces PRMT5 via CTCF, which catalyzes symmetric arginine dimethylation of histones H4R3 and H3R8. This modification regulates the alternative splicing of Transcription Factor 3 (TCF3) by recruiting DNA methyltransferase 3A (DNMT3A), leading to DNA methylation at conserved intronic regions between exons 18a and 18b. This epigenetic change promotes the exclusion of exon 18a, leading to the expression of the E47 isoform, which enhances EMT and invasion in breast cancer cells under hypoxia. In summary, this work reveals the essential role of CTCF in regulating alternative splicing under hypoxia, driving EMT and contributing to cancer progression. These findings suggest potential therapeutic avenues by targeting hypoxia-induced pathways in breast cancer.

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OP17

Navigating the Helicobacter pylori driven immune landscape from inflammation to gastric carcinogenesis

Prof. Dil Afroze, *Advanced Center for Human Genetics, SKIMS-Srinagar (J&K) INDIA*

Helicobacter pylori infection has for long been known to be associated with onset of gastric mucosal inflammation and disbalance between T-helper (Th) cells and regulatory T cells (Tregs) which plays a critical role in determining the immune response, pathology, and persistence of infection. A dominant Th1/Th17 response leads to inflammation and tissue damage, which can result in symptomatic conditions like ulcers or gastritis while an enhanced Treg response limits inflammation but allows *H. pylori* to evade the immune system and establish chronic infection. This disbalance in Th/Treg activity contribute to various outcomes including gastric cancer. *H. pylori* infection augments the production of IL10, IL6, and TGF- β in the gastric milieu, in addition to atypical DNA methylation in gastric mucosa. Further, IL6/IL10 mediate the hyperactivation of the JAK/STAT pathway. On the other hand, IL17 is also known to up-regulate the expression of hTERT (human telomerase reverse transcriptase) which is also regulated by STAT3. Likewise, H. pylori infection is also known to stimulate the atypical DNA methylation in gastric mucosa. However, the precise role of cytokine signaling in induction of epigenetic modifications, that perturb signalling cascade during gastric carcinogenesis is vaguely understood. This interplay of Th/Treg imbalance and hTERT in gastric cancer forms a vicious cycle of inflammation, immune suppression, and tumor progression. Therefore, we seek to understand these dynamics in patients vital for developing immune-based therapies

for *H. pylori*-associated diseases for targeting these pathways aimed at improving therapeutic outcomes and patient survival.

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OP18

Life course evolution of diabetes in Indians: Developmental Origins of Health and Disease

Prof. C. S. Yajnik, *Diabetes Unit, KEM Hospital & Research Centre, Pune, India*

Conventionally, diabetes and other non-communicable diseases are thought to have genetic susceptibility and be precipitated by lifestyle factors. Over the last 50 years, research has highlighted the role of intrauterine epigenetic programming which is influenced by maternal nutrition, diabetes, stress, and endocrine disruptors. The association of birth weight with the risk of subsequent diabetes is U-shaped. Rapid post-natal catch-up growth in low birthweight babies is an additional risk factor.

India is the world's double capital of low-birth-weight babies and type 2 diabetes. The small and thin Indian babies are more adipose compared to the European babies and show higher levels of biomarkers for diabetes risk in the cord blood. Improving fetal growth and body composition by improving maternal nutrition and glycemia is expected to curtail the rising epidemic of type 2 diabetes. Primordial prevention will be more of a community action than in the clinics.

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OP19

Interleukin-17A in Regulating the Keratinocyte cell Proliferation

Prof. Raghunath Chatterjee, *Human Genetics Unit, Indian Statistical Institute, 203 B T Road, Kolkata.*

Integrating genomic and histopathological analysis can aid in comprehending the molecular mechanisms behind disease pathogenesis. Both psoriasis and cancer exhibit cellular hyper-proliferation, even though they have unique histological characteristics. Our study identified upregulation of tumor-suppressor miR-7-5p in psoriasis that positively correlated with rete-peg elongation, an indicator of keratinocyte proliferation in psoriasis. We confirmed miR-7-5p's tumor-suppressor role in various cancer cells. Mir-7-5p overexpression inhibits the proliferation of epidermal keratinocytes in both normal and psoriatic conditions by targeting EGFR and LIF. Conversely, miR-7-5p knockdown or inhibition resulted in a significant enhancement of keratinocyte proliferation, further demonstrating its anti-proliferative role in psoriasis. Our study showed that IL-17a has a dual role in psoriasis: while it elevates miR-7-5p to reduce keratinocyte proliferation, it

concurrently activates pro-inflammatory cytokines, including IL-6, to induce inflammation and keratinocyte proliferation by augmenting LIF expression. Therefore, in psoriasis, the upregulation of the tumor-suppressor miR-7-5p by IL-17a balances the keratinocyte proliferation induced by IL-17a. Meanwhile, miR-7 is often downregulated in various cancer cells, making miR-7-5p a promising therapeutic candidate for diseases characterized by hyperproliferation, such as cancer.

OP20

Solving the unsolved in neuromuscular diseases: Genomic re-analysis through international collaborations using RDConnect GPAP

Dr. Kiran Polavarapu*, CHEO Research Institute, Canada.*

Neuromuscular diseases are an important group among rare diseases and predominantly inherited as monogenic with a prevalence of 10-60/100,000 worldwide. With >600 known causative genes, about ~40-60% of patients still remain undiagnosed after initial genetic testing with exome or genome sequencing. Given the rapidly expanding knowledge and resources in genomics and rare disease research, re-analysis of existing genomic datasets can help in solving unsolved NMD patients' diagnosis by improving the variant identification and interpretation along with identification of novel genes and extended phenotypes.

The RD-Connect Genome Phenome Analysis Platform (GPAP) is a clinician-friendly genomic research platform which enables rare disease clinicians and researchers from across the world to upload and analyse unsolved next-generation sequencing datasets in order to identify disease causing variants in rare disease patients through both individual and collaborative efforts.

Utilizing functionalities of GPAP, a systematic re-analysis through international collaboration under pan-European project Solve-RD resulted in 96 (6.3%) new diagnoses among 1517 unsolved NMD families analysed. Further, Individual re-analysis of 163 unsolved NMD patients with exomes or genomes available led to 35 (21.5%) new diagnoses. Majority of the new diagnoses can be attributed to re-classification of variants of uncertain significance, missed variants (non-canonical splicing variants, mitochondrial variants, copy number variants etc.) and extended phenotypes in known NMD / non-NMD genes. Through individual re-analysis and matching patients with similar phenotypes in GPAP, we were also able to identify novel genes like *TEFM*, *ATP2A2* which were further studied in animal and tissue models to establish as new neuromuscular disease-causing genes.

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OP21

Indian Genomics Landscape: Achievements and opportunities

Dr. Ashwin Dalal, *Centre for DNA Fingerprinting and Diagnostics, Hyderabad*

Indian genomic landscape is rapidly evolving, driven by advancements in education, diagnostics, and research. Education in genomics has seen significant growth, with numerous institutions offering specialized programs and courses. The Indian government and various organizations have invested in capacity building, creating a skilled workforce adept at handling complex genomic data. In diagnostics, India has made remarkable strides. The introduction of next-generation sequencing (NGS) technologies and genomic databases has enhanced the accuracy of genetic testing. This progress is crucial for diagnosing genetic

disorders and personalizing treatment plans. Companies and hospitals are increasingly integrating genomic data into clinical practice, improving patient outcomes and enabling early disease detection. Research in genomics is vibrant, with India contributing to global discoveries. Several institutions are leading innovative projects in gene editing, genomics of rare diseases, and functional genetics. Collaborative efforts with international researchers further likely to boost India's role in global genomics. Overall, India's genomic landscape reflects a dynamic interplay of education, diagnostics, and research, positioning the country as a key player in the global genomics arena and paving the way for personalized medicine and advanced genetic therapies. The talk will focus on major achievements in Indian genomics field and the way forward.

OP23

Initiating multi-center efforts for addressing rare disease challenges in India

Prof. Anju Shukla, *Department of Medical Genetics, Kasturba Medical College and Hospital Manipal, Karnataka*

Rare genetic diseases pose unique challenges in terms of lack of awareness in the general as well as the medical community, difficulties in definitive diagnoses, lack of expertise and resources for optimal management, burden on healthcare due to protractive course of the illness and scarce avenues for treatment. India faces an uphill task to provide appropriate services and care to families with rare genetic diseases. Also, there is a need to develop large scale collaborative efforts for rare disease diagnosis, research and training. I will be highlighting one such multicentric effort to address these challenges which may represent a working model for investigators and centers catering to these families.

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OP24

Metabolic-Epigenetic Shifts in Gliomas: Role of IDH1 Mutation

Prof. Ellora Sen, *National Brain Research Centre, Manesar, Haryana*

Background Glioblastoma multiforme (GBM) - the most malignant of brain cancers characterized by aberrant metabolic profile, is largely refractory to current therapeutic regimens. Mutation in the metabolic gene isocitrate dehydrogenase (IDH1) is a key event in gliomagenesis, as these mutations alter epigenetic landscape to subsequently affect tumor progression.

Objectives We investigated how rewiring of metabolic reprogramming with epigenetic landscape in gliomas bearing IDH1 mutation can differentially influence genes associated with chemoresistance.

Materials and Methods Immunohistochemistry and Western blot analysis on IDH1 wild-type and mutant glioblastoma patient derived tumors. Whole transcriptome sequencing (RNA-seq) data from tumor tissues of low grade and high-grade glioma patients (TCGA-LGG and GBM dataset). In vitro genetic and pharmacological manipulations in IDH1MT and IDH1WT stable glioma cell lines.

Results and Conclusions

We obse0rve in IDH1 mutant gliomas (i) altered mitochondrial/ telomere function and redox status that have clinical relevance pertaining to their sensitivity to oxidative-stress inducers (ii) dependence on inflammatory molecule Pentraxin 3 (PTX3) for targeting towards ferritinophagy mediated cell death as a treatment regime (iii) involvement of anti-phagocytic CD47 gene in the regulation of immune-surveillance (iv) differential expression of senescence-associated secretory phenotype (SASP) linked with persistent DNA damage. Taken together, our findings suggest how distinct genetic/epigenetic landscape rewires energy metabolism and inflammation in the context of distinct molecular signatures associated with predictive and prognostic values in glioma patients.

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OP25

Clonal selection and diversification of somatic mutations in normal human endometrial epithelium

Prof. Hirofumi Nakaoka, *Department of Cancer Genome Research, Sasaki Institute, Sasaki Foundation, Chiyoda-ku, Japan.*

It has become evident that somatic mutations in cancer-associated genes accumulate in the normal endometrium, but spatiotemporal understanding of the evolution and expansion of mutant clones is limited. To elucidate the timing and mechanism of the clonal expansion of somatic mutations in cancer-associated genes in the normal endometrium, we sequenced 1,311 endometrial glands from 37 women. We showed that the burdens of somatic mutations with single base substitution signatures (SBS1, SBS5, and SBS18) increased with age and the mutations in cancer-associated genes were under strong positive selection in the normal human endometrium. By collecting endometrial glands from different parts of the endometrium, we showed that multiple glands with the same somatic mutations occupied substantial areas of the endometrium. By using three-dimensional imaging analysis, we demonstrated that “rhizome structures”, in which the basal glands ran horizontally along the muscular layer and multiple vertical glands rose from the basal gland, originated from the same ancestral clone. Moreover, mutant clones detected in the vertical glands diversified by acquiring additional mutations. These results suggest that clonal expansions through the rhizome structures are involved in the mechanism by which mutant clones extend their territories. Furthermore, we showed clonal expansions and copy neutral loss-of-heterozygosity events occurred early in life, suggesting such events can be tolerated many years in the normal endometrium. Our results of the evolutionary dynamics of mutant clones in the human endometrium will lead to a better understanding of the mechanisms of endometrial regeneration during the menstrual cycle and the development of therapies for the prevention and treatment of endometrium-related diseases.

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OP26

Decoding the Hedgehog Signaling Pathway in Acute Myeloid Leukemia: Biomarkers and Therapeutic Perspectives

Prof. Pranay Tanwar, *Dr.B.R.A. Institute Rotary Cancer, All India Institute of Medical Sciences, New Delhi*

Background/ Introduction: The Hedgehog (Hh) signaling pathway plays a critical role in embryonic development, stem cell maintenance, and tissue homeostasis. Dysregulation of this pathway has been implicated in various cancers, including acute myeloid leukemia (AML). However, the roles of specific Hh pathway genes, such as WNT6, BMP8B, HHIP, SUFU, CSNK1G3, and CSNK1A1, in AML remain unclear.

Objectives: This study investigates the expression, methylation status, survival correlation, and functional enrichment of these genes to elucidate their roles in AML pathogenesis and prognosis.

Materials and Methods: We analyzed 220 AML cases for differential expression and promoter methylation of the selected genes compared to normal controls. Methylation-associated gene silencing was evaluated, and survival analysis assessed the prognostic impact of gene expression levels. Functional enrichment and correlation analyses identified pathways and interactions involving these genes.

Results and Conclusions: Expression analysis revealed significant dysregulation of the Hh pathway genes in AML. WNT6, BMP8B, HHIP, and SUFU were upregulated, while CSNK1G3 and CSNK1A1 were downregulated. Hypermethylation of promoter regions contributed to the silencing of CSNK1G3 and CSNK1A1. Altered expression of these genes was associated with poorer survival outcomes. Functional enrichment analysis demonstrated that these genes are involved in key cellular processes, including cell cycle regulation, apoptosis, and stem cell maintenance, and interact with other pathways implicated in AML.This study highlights the dysregulation of Hh pathway genes in AML and their roles in leukemogenesis. The association of CSNK1G3 and CSNK1A1 downregulation with hypermethylation emphasizes the impact of epigenetic modifications on the Hh pathway. These genes show promise as potential biomarkers and therapeutic targets in AML. Future research should explore strategies to modulate the Hh signaling pathway for improved treatment outcomes.

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OP27

Small molecules reverting chemoresistance via chromatin remodelling are viable candidates for adjunct therapy in colon cancer

Prof. Sagar Sengupta, Director, *National Institute of Biomedical Genomics, Kalyani, West Bengal*

The human genome itself is extremely compact and needs to be remodeled so that every cell in the body have the full and complete repertoire of transcripts at the correct time and location. When the above process is aberrant - it can lead to multiple abnormalities - one of which is neoplastic transformation leading to

carcinogenesis. Hence maintenance of genome integrity by modulating the compaction and decompaction of the human genome is one of the hallmarks which can prevent neoplastic transformation.

Chemoresistance is one of the features of recurrent carcinogenesis. When chemoresistance occur - the frontline drugs used for cancer treatment either partially or completely lose their efficacy, leading to the recurrence of cancer at the same or distal sites. Using colon cancer as the model system, we have tried to understand whether chemoresistance can be linked to chromatin remodeling and if so - whether there are ways to revert the process. Some of these results will be presented.

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OP28

Mechanism of chromosomal translocations in lymphoid neoplasia: Focus on non-B DNA and RAGs

Prof. Sathees C. Raghavan, *Department of Biochemistry, Indian Institute of Science, Bangalore.*

Hematopoietic neoplasia like leukemia and lymphomas constitute 8-10% of all cancers reported worldwide. These are characterized by the presence of specific chromosomal translocations, which act as genetic markers for the diagnosis and prognosis of the disease. Chromosomal translocations result from aberrant DNA double-strand breaks (DSBs) on the heterologous chromosomes, followed by their anomalous joining by DNA repair pathways. The mechanism underlying such atypical DNA breaks is largely unknown. We have shown recently that many translocations fragile regions adopt non-B DNA conformation, such as G-quadruplex DNA structures, Cruciform DNA, Triplex DNA, and R-loops. Further, we observed that RAGs and AID, which are normally responsible for immunoglobulin diversity, could play a role in generating such chromosomal translocations. Our recent studies also showed that a nonamer near non-B DNA structures could enhance RAG cleavage on such structures, thereby providing a novel mechanism of RAG-mediated chromosomal translocations in lymphoid cancers. I will highlight some specific examples during my presentation, highlighting the role of non-B DNA structures and RAGs. Regulation of RAGs by miRNAs will also be discussed.

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OP29

Epigenetic Regulator ZMYND8 Drives Tumor Heterogeneity Through Metabolic Reprogramming in Breast Cancer

Prof. Chandrima Das, *Biophysics and Structural Genomics Division, Saha Institute of Nuclear Physics, Kolkata, India and Homi Bhabha National Institute, Mumbai, India*

Breast tumors exhibit remarkable heterogeneity, necessitating a comprehensive understanding of the underlying molecular mechanisms driving their transcription programs in a spatio-temporal manner. The

availability of the oxygen and nutrients within the solid tumors creates distinct regions, with the tumor core experiencing deprivation and the periphery exhibiting abundance. Cancer cells undergo genetic and epigenetic changes to adapt to these spatially diverse conditions. Histone reader Zinc finger MYND-type containing 8 (ZMYND8) has been identified as a regulator of hypoxia target genes in breast cancer. In this

study, we aimed to investigate the role of ZMYND8 in regulating gene expression under conditions resembling the tumor core and periphery. Our findings demonstrated elevated expression of ZMYND8 in tumor core-like conditions, where it governs the activity of several metabolic enzymes. We further extended our studies to human breast carcinoma patients, encompassing clinically distinct tumor core and peripheral tissues. Our study provides valuable insights into the distinct molecular mechanisms employed by ZMYND8 to regulate metabolic enzymes in breast tumor core and periphery. These findings contribute to a better understanding of tumor heterogeneity and may hold significant implication

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OP30

Unveiling the Parkinson's disease-associated α-Synuclein role in chromatin regulation

Dr. Sivaraman Padavattan, *Department of Biophysics, National Institute of Mental Health and Neurosciences, Bangalore-560029, India*

Background: α-Synuclein (αSyn) plays a critical role in the pathogenesis of Parkinson's disease (PD) and other Synucleinopathies. Although many cellular functions have been proposed for αSyn, its precise physiological role remains elusive. Multiple lines of evidence indicate that under pathological conditions, the nuclear αSyn level increases, eliciting neurotoxicity in dopaminergic neurons and mouse models independent of its aggregation property. These findings raise a fundamental question regarding the mechanism of αSyn toxicity in PD: the underappreciated nuclear function versus its aggregation property. Therefore, determining αSyn's physiological role in the nucleus is of great importance.

Materials and Methods: In this study, we have employed a combination of biochemical, biophysical, structural (X-ray crystallography), and cellular techniques.

Results and Conclusion: To date, studies on nuclear αSyn have been limited in its interaction with individual histones and dsDNA, leaving a significant gap in our understanding of its nuclear functions and role in chromatin regulation. Our earlier study showed that αSyn's interaction with dsDNA is weak and non-specific, whereas its interaction with individual core histones H3 is specific. Based on this study, we proposed that the nuclear function of αSyn is possibly driven through histone interaction. In the current study, we provide molecular-level details and structural insights into αSyn nuclear physiological function and its role in chromatin regulation for the first time.

Funding: This work was supported by a SERB-ECR grant (ECR/2018/002219) and NIMHANS intramural support.

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OP31

Structure-function relationship in DNA: The importance of kinetics

Dr. Hemachander Subramanian, *Department of Physics, National Institute of Technology, Durgapur, West Bengal.*

DNA, a molecule of central importance in biological systems, appears to have been evolutionarily optimized for its function of replication, storage and expression of information. However, some of the central biophysical properties of DNA are intriguing from an evolutionary perspective, because they add considerable, apparently avoidable complexity to DNA's function. Why is DNA daughter strand construction unidirectional? What is the need for the complicated lagging-strand replicational mechanism of DNA, or equivalently, of anti-parallel DNA strand orientation, when parallel strand orientation could have made DNA self-replication much simpler? What is the evolutionary significance of asymmetric distribution of nucleotides around replication origins? Why are nucleotides and other biomolecules not achiral? Answers to these disparate questions may lie in a kinetic property, which we call *Asymmetric Cooperativity*. In this talk, I will explain how evolutionary optimization for faster replication may have led to DNA acquiring these counter-intuitive properties.

Keywords: kinetic asymmetry in DNA, asymmetric cooperativity, evolutionary optimization, unidirectional replication, anti-parallel strand orientation, replication origins

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OP32

Optical Genome Mapping: Next Generation Cytogenetics

Dr. Divya Tej Sowpati, *CSIR-CCMB, Hyderabad*

Large genomic aberrations such as structural variants and copy number variations play a pathognomonic role in a wide range of genetic diseases. Conventional cytogenetics methods such as karyotyping, fluorescence in situ hybridization (FISH) and chromosomal microarray (CMA) have been at the forefront of standard of care recommendations for detection of these variants. Optical genome mapping (OGM) is a next generation one-stop cytogenomic technique to comprehensively identify all classes of structural variants and copy number changes. In this talk, I explain the principle and technology behind this method, and our application of OGM in identifying and diagnosing complex structural variants in a clinical setting.

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OP33

Why India needed a Genome Sequencing Project of its own?

Prof. Analabha Basu, *National Institute of Biomedical Genomics, Kalyani, West Bengal.*

The underrepresentation of non-Europeans in human genetic studies has limited the diversity of individuals in genomic datasets and led to reduced medical relevance for a large proportion of the world's population. Population-specific reference genome datasets as well as genome-wide association studies in diverse populations are needed to address this issue. This includes a whole-genome sequencing reference dataset from individuals of multiple population groups. We catalogue genetic variation, population structure, disease associations and founder effects. We also explore the role of novel variants in different populations.

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OP34

***X-plat*: a cross-platform transformation tool based on the nearest neighbor joining framework.**

Prof. Binay Panda, *School of Biotechnology, Jawaharlal Nehru University, New Delhi*

The advent of high-throughput genomics technologies like microarrays and next-generation sequencing have provided biologists with the tools to measure genome-wide activities of individual transcripts and genes. Before the high-throughput sequencing methods became ubiquitous and affordable, despite their dependence on previous annotations, microarrays were the tools of choice due to their wider availability, ease of use, and relatively inexpensive nature. Over the years, data from millions of samples were deposited in various databases and public repositories. Answers to many biological questions can benefit from combining data from legacy microarray platforms and high-throughput sequencing platforms via the discovery and validation of results, especially in the clinical domains where retrieving samples from deceased individuals is impossible. However, incompatibility from different technologies due to design considerations, target preparation, and dependence on prior annotations makes legacy data unusable.

During my talk, I shall describe our efforts in making a cross-platform data transformation tool, X-plat, that works both for expression and methylation data and inter-convert data for comparison between platforms without compromising throughput. X-plat builds a per-gene predictive model by connecting the nearest neighboring data points using a nearest neighbor-joining framework followed by Root Mean Square Error minimization. In its subsequent iterations of the algorithm, X-plat builds transformation rules for clusters of genes with lower RMSE over individual genes, thus reducing the model size and compute time. X-plat used the cross-platform conversion rules discovered and validated using paired mixed source microarray: sequencing gold standard datasets across conditions, sources and diverse organisms. X-plat outperformed the published normalization and conversion tools, harmony, distran, dwd, gq, mrs, qn and tdm in 70% and

89%, 94% and 98%, 91% and 96%, 95% and 99%, 94% and 98%, 85% and 95%, and 94% and 97%, of the genes, for two of the tested datasets, respectively.

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OP35

A unified Bayesian approach to transcriptome-wide association study

Dr. Arunabha Majumdar*[1]*, *Arnab Kumar Khan*[2] *and Tanushree Haldar*[3]

1. *Indian Institute of Technology Hyderabad, 2. Indian Statistical Institute Kolkata, and 3. University of California San Francisco.*

Background/ Introduction: The transcriptome-wide association study (TWAS) has discovered novel gene-trait associations that offer essential biological interpretations. TWAS integrates reference panel transcriptome data and genome-wide association study (GWAS) data. In standard TWAS methods, a prediction model for gene expression is built utilizing the transcriptome data, which is employed to impute the gene expression in the GWAS data. The complex trait in GWAS is regressed on the predicted expression to identify gene-trait associations. These two-step approaches ignore the uncertainty of the imputed expression and can lead to reduced inference accuracy.

Objectives: We develop a unified Bayesian method for TWAS to avoid a two-step approach, modeling the two datasets simultaneously and performing a comprehensive Bayesian inference.

Materials and Methods: We consider the horseshoe prior, an established global-local shrinkage prior, to model the relationship between gene expression and local SNPs and the spike and slab prior while testing for an association between the genetic component of expression and the trait. We extend our approach to conducting a multi-ancestry TWAS, focusing on discovering genes that affect the trait in all ancestries.

Results and Conclusions: Using simulations for a single-ancestry TWAS, we demonstrate that our approach estimates the effect size more accurately than the popular two-step approach PrediXcan. For multi-ancestry TWAS, our method offers better estimation accuracy than METRO, a method for multi-ancestry TWAS. While classifying non-null genes, our approach performs better than the comparing methods. We perform a single-ancestry and multi-ancestry TWAS for body mass index (BMI), integrating the Geuvadis transcriptome and UK Biobank GWAS data. Our approach identified 111 genes associated with BMI for the single ancestry TWAS of Europeans, which were enriched in the interferon gamma signaling pathway at a nominal level. For a multi-ancestry TWAS of Africans and Europeans, the method discovered 19 genes that affect BMI in both ancestries, which were nominally enriched in the molybdenum cofactor biosynthesis pathway.

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OP36

Born Too Soon: The Molecular Underpinnings of Preterm Birth in Indian Women

Prof. Arindam Maitra, *National Institute of Biomedical Genomics, Kalyani, West Bengal.*

South Asia and India contributes the highest numbers of preterm birth (PTB), the single largest cause of neonatal mortality. PTB is also associated with other health adversities in childhood and in adult life. We conducted a genome-wide association study (GWAS) of spontaneous preterm birth (sPTB) on 6,211 Indian women in the GARBH-Ini cohort followed by cross-ancestry meta-analyses. We identified both population-specific and trans-ethnic genetic associations with sPTB in Indian women. We integrated genotype, long read sequencing, DNA methylation, transcriptome and clinical data to delineate the mechanisms by which the identified SNPs might increase the risk of sPTB. We expect two major public health impacts from our findings. The SNPs and the downstream mechanistic linkages can help us identify potential biological pathways that may be amenable to interventions to prevent sPTB. Further, the set of SNPs identified in our study, might help us stratify pregnant women at risk of preterm birth, thus enabling clinicians to prioritise their antenatal care to improve birth outcomes. Both the impacts have immense potential to reduce the neonatal and infant mortality and morbidity and accelerate the achievement of sustainable development goals.

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OP37

Placental Epigenetic Signatures Can Predict Long-Term Health Outcomes in Infants Prenatally Exposed to opioids and Neonatal opioid withdrawal syndrome.

Prof. Uppala Radhakrishna, *Department of Anesthesiology and Perioperative Medicine, University of Pittsburgh, Pittsburgh, PA, United States*

Background. Opioid misuse has caused a global epidemic, with 16 million people affected by opioid use disorder (OUD) and over 120,000 deaths annually. Opioid use during pregnancy can lead to Neonatal Opioid Withdrawal Syndrome (NOWS), a complex condition characterized by neurodevelopmental delays, behavioral dysregulation, and, in severe cases, death. Infants with severe NOWS face an elevated risk of long-term developmental challenges, including lower IQ, attention deficits, developmental defects, and disabilities that may only emerge later in life. Mothers of NOWS infants also experience higher mortality rates and complications like placental abruption. Current tests are inadequate for predicting, assessing, or monitoring NOWS severity and treatment outcomes.

Study Design: We conducted genome-wide methylation analyses on a large cohort of 96 placental tissue samples using Illumina Infinium MethylationEPIC (850k) BeadChips. The study included three groups: 32

prenatally opioid-exposed infants requiring pharmacologic treatment for NOWS (+Opioids/+NOWS), 32 prenatally opioid-exposed infants with NOWS who did not require treatment (+Opioids/-NOWS), and 32 prenatally unexposed controls (-Opioids/-NOWS). Comprehensive statistical analyses, bioinformatics approaches, Artificial Intelligence (AI) techniques—including Deep Learning (DL)—and Ingenuity Pathway Analyses (IPA) were employed to identify and interpret the epigenetic changes.

Results: The study identified 1,778 significantly differentially methylated CpGs across 1,789 genes, many of which are pleiotropic and contribute to diverse biological functions. These genes are involved in key processes such as glucose metabolism, autophagy, neurotransmission, oxidative stress, obesity, pain modulation, stroke, eye anomalies, neurological functions, ion channel regulation, cytochrome activity, cytokine signaling, telomere maintenance, and circadian and ultradian rhythms. Gene ontology and pathway enrichment analysis identified 43 pathways, including Calcium signaling, Cocaine addiction, and Nicotine addiction.

Conclusion. In summary, our cohort study revealed epigenetic dysregulation in key genes linked to NOWS, shedding light on the long-term clinical impacts of these methylation changes. These genes are associated with autism, Alzheimer's disease, non-alcoholic fatty liver disease, emotional-behavioral dysregulation in infants, and other developmental defects. The findings emphasize the importance of epigenetic regulation in personalized pain management and its potential to identify high-risk patients, advance preventative therapies, and improve maternal and neonatal outcomes.

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OP38

Genetic Landscape of Polycystic Ovary Syndrome (PCOS)

Prof. Ashutosh Halder, *All India Institute of Medical Sciences, New Delhi*

PCOS is a common endocrinopathy among women of reproductive age, with a worldwide prevalence of 8 to 13%, depending on the criteria used for diagnosis. It is characterized by a constellation of features, including oligo/anovulation, clinical and/or biochemical hyperandrogenism, and polycystic ovarian morphology. Despite extensive research, the etiology of PCOS remains largely unknown. Familial clustering of the cases of PCOS points to a genetic component linked with it. However, the typical form of inheritance of PCOS follows a non-Mendelian pattern and involves complex genetic mechanisms.

WES was performed in a cohort of 100 PCOS cases. The variants were predicted for their probable deleterious effect on the functionality of the proteins through in-silico prediction tools such as CADD, SIFT, LRT, etc. Protein modeling was employed to predict the impact of these variants further.

We detected 100 rare coding variants in 50 known PCOS-causative genes, accounting for 61 (63.5%) PCOS cases. Functional annotations of these genes indicated their involvement in steroid hormone biosynthesis and action (POR, CYP21A2, STAR, PAPSS2, H6PD), ovarian folliculogenesis (FOXO3, BMP2), reproductive

hormones and receptors (AMHR2, AR, PGR, FSHR, GNRHR, AMH, INHBA, ACVR2A), and metabolic syndromes such as obesity and insulin resistance (MC4R, FTO, ADIPOQ, INSR, IRS2, LMNA). Cumulatively, 26 likely pathogenic variants and 74 variants of uncertain significance contributed to the PCOS cases. In-silico prediction analysis showed that these variants had deleterious effects on the protein function and a decreasing effect on their stability. There were significant differences in the frequencies of the variants in the cohort when compared to the population databases ($p<0.05$).

This study expands our knowledge of the mutational landscape that contributes to PCOS. Discovering the genetic factors and pathways involved in the disorder will help us better comprehend the underlying mechanisms of the disorder.

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OP42

Elevating Precision Oncology: Tumor genomic profiling solutions advancing precision oncology

Dr. Rahul Solanki, *Application Scientist, Agilent Technologies, India.*

In this session, we will introduce the Agilent SureSelect Cancer CGP portfolio, a comprehensive suite designed to revolutionize precision oncology. Discover the pan-cancer SureSelect Cancer CGP assay, the focused SureSelect Cancer Tumor-Specific assays, and the customizable SureSelect Cancer Custom panels. We will delve into the design and performance of a new hybrid-capture target enrichment NGS assay, which offers a modular approach to accurately characterize tumors. Learn how this pan-cancer assay, targeting 679 cancer-specific genes and an 80-gene RNA panel, can detect key classes of somatic alterations in a single assay, including the assessment of new and emerging biomarkers, the immuno-oncology biomarkers TMB and MSI, and HRD. We will also explore the flexibility of customizing NGS cancer panels to meet specific laboratory needs, utilizing Agilent SureDesign's machine learning capabilities for precise biomarker assessment.

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OP43

Modeling Parkinson’s disease with human midbrain organoids

Prof. Jens C. Schwamborn, *University of Luxembourg, Luxembourgh*

In Parkinson’s disease (PD) patients the dopaminergic neurons of one region in the midbrain, the substantia nigra, are highly vulnerable for degeneration, while the dopaminergic neurons of a neighboring region, the ventral tegmental area, are not. The reasons for this selective vulnerability are largely unknown to date. This lack of knowledge, to a good extend is the consequence of the absence of human specific models for the midbrain.

Here, we demonstrate that three-dimensional (3D) differentiation of expandable human midbrain floor plate neural progenitor cells (mfNPCs) leads to organoids that resemble key features of the human midbrain. These organoids are composed of midbrain dopaminergic neurons (mDANs), which produce and secrete dopamine. Additionally, the midbrain organoids contain other neuronal subtypes, astrocytes and oligodendrocytes. They can be further enriched with induced pluripotent stem cell (iPSC) derived microglia. Patient and disease specific midbrain organoids can be generated through the usage of patient derived iPSCs. Importantly, in these disease specific organoids, key hallmarks of PD including reduced numbers of dopaminergic neurons and appearance of alpha-Synuclein positive protein aggregates are recapitulated. Thus, we provide a robust method to reproducibly generate 3D human midbrain organoids containing mDANs to investigate PD-relevant patho-mechanisms.

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OP44

Modelling Rare genetic disorders in Zebrafish: insights into disease mechanisms

Dr. Aarti Sevilimedu, *Dr. Reddy's Institute of Life Sciences, Hyderabad*

Rare genetic disorders are a group of diseases with relatively low prevalence in the general population, however together the 7000 or so rare disease affect around 400 million people worldwide. Very few of these have known treatment, and that is due to the complex nature of the disease, poor understanding of the causal biology and small patient pool that makes clinical development challenging. Therefore, innovative and cost-effective preclinical studies are required. The zebrafish has emerged as a powerful model system to study rare diseases, due to the conservation of over 80% of the disease-causing genes, conserved physiology and easy laboratory manipulation. In this talk, I will focus on two case studies of zebrafish models we have created, to study the underlying mechanistic basis of the disease as well as discuss potential use of these models for therapy development.

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OP45

Regenerating and reconnecting neural circuits

Prof. Anindya Ghosh Roy, *National Brain Research Centre, Gurgaon.*

Aging is an inevitable process through which our brain and body undergo progressive changes, some of which has highly unfavorable outcomes. Many of the neurodegenerative diseases such as Alzheimer's, Parkinson's and Frontotemporal dementia are caused due to loss of large number of neurons in the functional brain circuits. One approach to treat these conditions is to introduce new neurons, which can further integrate in to the functional circuitry. It has been a long-standing problem for clinicians in finding a cure for patients suffering from nervous system injuries or degeneration due to failure in neurite regeneration in central nervous system and re-establishing functional connections with target tissues.

Our group at the National Brain Research Centre have been investigating the molecular mechanism of axon regeneration. Axons are long cables through which nerve cells propagate electrical signals to their post-synaptic neurons. We have identified a conserved signaling pathway involving Dual Leucine Zipper Kinase (DLK) required for axon regeneration following an injury. We have further shown that axon regeneration leads to functional restoration in early stage of life and this ability is lost in old age. Manipulation of the conserved *let-7* miRNA or Insulin (IIS) signaling can overcome age related decline in axon regeneration and aid in functional recovery. Rehabilitation and pharmacological paradigms were established to alleviate the decline in regenerative capacity of aging neurons.

Recently we found that dendrites, (the information receiving units of neurons) also can regenerate following physical injury. However, the axon regeneration pathway DLK-1 is not required for dendrite regeneration. This suggested that molecular mechanism for dendrite regeneration is distinct from axonal regeneration. Implications of these findings for therapeutic strategies in humans will be discussed.

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OP46

Decoding the Genetic Architecture of SLE: Insights from Post-GWAS Analyses and Functional Characterization of an Asian-Specific Regulatory Variant

Prof. Swapan K. Nath, *Arthritis & Clinical Immunology Research Program, Oklahoma Medical Research Foundation, 825 NE 13th St. Oklahoma City, OK 73104, USA*

Systemic lupus erythematosus (SLE) is a complex autoimmune disease driven by genetic, epigenetic, and environmental factors. Genome-wide association studies (GWAS) have identified numerous susceptibility signals, primarily single nucleotide polymorphisms (SNPs), associated with increased SLE risk. However, the causal mechanisms underlying these associations and the full landscape of susceptibility loci remain incompletely defined.

In the first part of this talk, we present an extensive post-GWAS analysis aimed at delineating independent susceptibility loci and mapping them within SLE's regulatory framework. By integrating fine-mapping, epigenomic profiling, and gene expression data, we compiled a comprehensive catalog of SLE susceptibility loci across European, Asian, Hispanic, and African ancestries, identifying 182 non-HLA loci linked to disease risk.

The second part highlights a GWAS locus at 19p13 specific to Asian populations. Using systematic post-GWAS prioritization, we identified rs4808485 as a key regulatory variant within this locus. Functional assays, including luciferase reporter assays, chromatin immunoprecipitation-qPCR (ChIP-qPCR), and chromosome conformation capture (3C), demonstrated that this variant functions as an active enhancer, modulating *KLF2* expression via allele-specific interactions with active histone marks, Pol II, and

transcription factors such as PARP1. CRISPR-mediated perturbation of the rs4808485 enhancer in Jurkat and lymphoblastoid cells confirmed its regulatory role. Loss of enhancer activity disrupted cellular homeostasis by dysregulating the NLRP3 inflammasome, leading to elevated CASPASE1, IL-1β, and GSDMD levels, while also increasing cell proliferation and accelerating cell cycle progression. RNA-seq analyses further validated the connection between *KLF2* expression and inflammasome activity in healthy controls and SLE patients, including those with lupus nephritis (LN). These findings establish a functional mechanism linking rs4808485 to SLE susceptibility through its regulatory effects on *KLF2*. This work provides critical insights into the genetic architecture of SLE and identifies promising therapeutic targets for SLE.

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OP47

Regulation of autosomal genes by noncoding RNAs from Y-chromosomal heterochromatin and implications in male infertility.

Rachel A Jesudasan*[1,2]*, *Kankadeb Mishra*[1,3]*, Pranatharthi Annapurna*[1,4]*, Anurag Chaturvedi*[1,5]*, Shrish Tiwari*[1]*, Nissankararao M Praveena*[1,6]*, Jomini L Alex*[1]*, Arpan Mukhoti*[1]*, Arvind Kumar*[1]*, Sivarajan Karunanithi*[1,7] *and Hemakumar M Reddy*[1,8]*.*

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Y chromosome, which is set aside for male reproduction, is rich in species-specific repeats, transposon elements and is heterochromatic, with few protein coding genes and no recombination during male meiosis. It is one of the smallest chromosomes in any genome and maintains the same theme across evolutionary strata. Understanding this apparently invariant functional pattern therefore holds the key to cracking the secrets of the Y. it was thought that the Y chromosome is transcriptionally silent and the major fraction of the repeats were considered junk at some point of time. Studies of the species-specific repeats on Y chromosomes have elucidated that these repeats regulate the expression of genes localizing to autosomes and X chromosomes, in testis, via long and small noncoding RNAs. Three different mechanisms by which this is achieved has been identified in our laboratory with respect to humans and mice (i) via trans-splicing (ii) via piRNA mediated regulation and (iii) by acquisition of DNA segments from autosomes on to the Y and subsequent amplification over there. Deletions of the Y chromosomal repeats result in differing degrees of

male infertility depending on the extent of deletion. Thus, species-specific repeats on Y chromosomes regulate genes involved in male fertility, in a universe where cross-species fertilization is not entertained.

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OP48

Roles of transcriptional enhancers in disease susceptibility

Dr. Dimple Notani, *National Centre for Biological Sciences, Bengaluru*

Genes are regulated by distal regulatory elements known as enhancers that exert their function on target genes by establishing looping with the promoter. Although discovered over forty years ago, the molecular mechanisms underlying enhancer functions still remain poorly understood. Recently, another layer of complexity has been uncovered by the discovery that in addition to widespread transcription of long non-coding RNAs (lncRNAs) in mammalian cells, bidirectional ncRNAs are transcribed on enhancers, and are thus referred to as enhancer RNAs (eRNAs). However, it has remained unclear whether these eRNAs are functional or merely a reflection of enhancer activation. Different roles of eRNAs in gene regulation are just emerging.

Investigations using Hi-C/5C technologies has revealed highly organized topologically associated domains (TADs) in chromosomes. These megabase sized regions are characterized by increased frequency (~2 fold) of interaction between loci within the TADs when compared to their interaction to loci located outside the confines of boundary elements. However, the defining principle behind these ordered structure is unknown. I will talk about our recent work that shows the relationship between mutations in enhancers, genome organization and disease susceptibility.

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OP49

Mechanisms underlying cell type specificity in CNS disorders: Lessons from Autosomal Dominant Leukodystrophy

Prof. Quasar S Padiath, *Department of Human Genetics, Graduate School of Public Health, University of Pittsburgh, Pittsburgh, PA – 15261, United States*

The role of non-coding regulatory elements and how they might contribute to tissue type specificity of disease phenotypes is poorly understood. Autosomal Dominant Leukodystrophy (ADLD) is a fatal, adult-onset, neurological disorder that is characterized by extensive CNS demyelination. Most cases of ADLD are caused by tandem genomic duplications involving the lamin B1 gene (LMNB1) while a small subset are caused by genomic deletions upstream of the gene. Utilizing data from recently identified families that carry LMNB1 gene duplications but do not exhibit demyelination, ADLD patient tissues, CRISPR modified cell lines and mouse models, we have identified a novel silencer element that is lost in ADLD patients and that specifically targets overexpression to oligodendrocytes. This element consists of CTCF binding sites that mediate three-dimensional chromatin looping involving the LMNB1 and the recruitment of the PRC2 repressor complex.

Loss of the silencer element in ADLD identifies a previously unknown role for silencer elements in tissue specificity and disease causation.

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OP50

Behaviour, genetics and genes: connecting some dots in familial dyslexia.

Prof. Subrata Sinha, *All India Institute of Medical Sciences, New Delhi, India*

The neurodevelopmental disorder, dyslexia, is a specific learning disability that manifests as difficulty in reading in spite of adequate intelligence or opportunity. It is a complex disorder, which has a strong heritable component. While these are a number of genes that have been implicated in the pathogenesis of dyslexia, none of these account for a significant proportion of the disability across populations.

We have studied three extended multigenerational families from distinct endogamous groups who had a high prevalence of dyslexia within the family. All these families had distinct patterns of inheritance. One of these had an autosomal dominant pattern of inheritance. A set of 17 polymorphisms on Chromosome 5p31.3 that encompassed a 1.9 Mb region comprising mainly the PCDHG cluster, was strongly associated with dyslexia in this family. Seven of the risk associated variants are ancestral variants preponderant Neanderthal and Denisovan sequences, while the non-risk associated variants are preponderant in modern humans. Another family had an s autosomal recessive inheritance pattern with a dinucleotide insertion at promoter of the BASP1 gene at the 5p15.1 locus, which overlapped with the first exon of a divergent long non-coding RNA, BASP1 AS1. This helped us identify a process involved in the neural differentiation of stem cells. In the third family there was a co morbidity with Attention Deficit Hyperactive Disorder (ADHD), a known co-morbidity of dyslexia which indicated a predisposing locus on the FAM43A gene, involved with GABAergic signalling.

Endogamous groups are common in the developing world, and could be studied for identifying disease pathogenesis associations and the molecular sub-classification of disease phenotypes. Because of genetic similarities, especially during studies of multigenerational families. they provide a better opportunity to identify pre disposing mutations and pathways. While the exact pathways thus identified may not be generalizable to the population at large, they provide vital clues for endo-phenotyping and pathogenesis, as well as insights into biology.

This study has been in collaboration with the research laboratories of Nandini Chatterjee Singh, Pankaj Seth (NBRC), Mitali Mukerji, Md Farukh (IGIB) and Deepti Jain (RCB).

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OP51

MRI markers of cerebral small vessel diseases and their genetic determinants.

Dr. Ganesh Chauhan, *Department of Genetics & Genomics, Rajendra Institute of Medical Sciences (RIMS), Ranchi, INDIA*

MRI based markers of cerebral small vessel disease (SVD) like, white matter hyperintensities, brain infarcts, cerebral microbleeds, dilated perivascular spaces, etc., are commonly seen in older persons. Most of these MRI based SVD markers are covert, not being associated with overt, clinical stroke symptoms. However, they cannot be considered benign, as they are often associated with subtle neurologic symptoms and increased risk of future stroke, cognitive decline, and in some studies dementia. Mechanisms and predictors of MRI based SVD markers remain incompletely understood. In this direction multiple collaborative studies from around the world have tried to understand the genetic risk variants determining risk of these MRI based SVD markers. Multiple genome wide association studies and sequencing studies using next generation sequencing approaches have been performed in large sample studies and have led to identification of new pathways and target molecules for future research. This presentation will describe these genetic findings and focus on future road maps for better understanding and treatment of stroke and dementia.

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OP52

Application of genomic sequencing technologies: Genetic Molecular Diagnostics of Developmental delay and intellectual disability

Prof. Santasree Banerjee, *Department of Genetics, College of Basic Medical Sciences, Jilin University, Changchun, Jilin, 130021, China.*

Intellectual Disability (ID) is characterised by impaired intellectual and adaptive function that starts during the developmental period. It is a lifelong condition and is among the most common neurodevelopmental disorders. Developmental Delay (DD) is a broad term that applies when one or more areas of a child's development are delayed. A disease-causing variant is identified in about half of individuals with DD and ID. Both copy number variants (CNVs) and single nucleotide variants (SNVs) are implicated as major causes of DD and ID; more than 130 rare CNVs and SNVs in more than 600 genes have been reported to be associated with this disease spectrum. Previous reports have shown that DD and ID can follow autosomal dominant, autosomal recessive, or X-linked modes of inheritance, as well as making a substantial contribution from *de novo* variants to the molecular aetiology. Further research into the identification of candidate causal genes will create opportunities to study the molecular mechanisms underlying the disease phenotype and will enable clinicians to make timely and accurate clinical diagnoses. This will make it possible to group cohorts of cases with similar genetic aetiology and refine their phenotypes including the

prognosis and course of the disease. Genetic diagnosis will allow mutational screening in families and make prevention possible through genetic counselling, prenatal, or pre-implantation diagnosis. According to the American Association on Intellectual and Developmental Disabilities, affected individuals with global developmental delay are characterised by a significant delay in the development of motor skills, speech, and cognition, while ID involves deficits in both intellectual function (learning, reasoning, and problem-solving skills) and adaptive function (communication, conceptual, social and practical skills) among children aged 5 years or younger. The global prevalence of DD is 1–3% among children aged 5 years or younger. The prevalence of ID is 2.7% and 2.17% among children and adults respectively. In addition, DD patients with de novo mutations have a prevalence rate of 1 in 213 to 1 in 448 live-births. Advances in variant detection technology have evolved in recent years, leading to accelerated causal gene discovery and understanding of genomic lesions in ID/DD cohorts. After preliminary confirmation of the disease through clinical, laboratory and radiological examination, clinicians can order further diagnostic testing, which is influenced by initial findings and availability and access to resources. The range of potential lines of inquiry include but are not limited to: karyotyping to identify gross chromosomal abnormalities; chromosome microarray (CMA) to identify deletions, duplications, loss of heterozygosity, and aneuploidy; and genomic sequencing (target-based sequencing of a gene panel, clinical whole exome sequencing or whole genome sequencing) to identify disease-causing variants. In conclusion, we identified both CNVs and SNVs were associated with DD and ID. In future, more refined cellular and molecular studies are required to understand the disease mechanisms. We emphasise the significance of genomic sequencing technology for molecular genetic diagnostics in ID/DD cohorts, thus laying the groundwork for a future Research Topic focused on studies that will identify new candidate genes and disease-causing variants, which in turn will allow us to develop novel therapeutic interventions.

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OP53

The use of antisense oligonucleotides for treating rare genetic diseases

Prof. Virginia Kimonis, *Division of Genetics and Genomic Medicine, Department of Pediatrics, Neurology and Pathology, UC Irvine, Irvine, California, USA*

Antisense Oligonucleotide (ASO) therapeutics include short synthetic RNA or DNA strands that bind to their complementary target RNA molecules to alter protein expression. ASOs have been shown to be safe and effective in diseases such as spinal muscular atrophy. I will share my lab's experience with ASOs in two rare neuromuscular diseases: Pompe disease and VCP related multisystem proteinopathy (MSP-1).

Pompe disease is a progressive myopathy caused by the aberrant accumulation of glycogen in skeletal and cardiac muscle resulting from the deficiency of the enzyme acid alpha-glucosidase (GAA). Administration of recombinant human GAA as enzyme replacement therapy (ERT) works well in alleviating the cardiac manifestations of Pompe disease but loses sustained benefit in ameliorating the skeletal muscle pathology.

The limited efficacy of ERT in skeletal muscle is partially attributable to its inability to curb the accumulation of new glycogen produced by the muscle enzyme glycogen synthase 1 (GYS1). Substrate reduction therapies aimed at knocking down GYS1 expression represent a promising avenue to improve Pompe myopathy. However, finding specific inhibitors for GYS1 is challenging given the presence of the highly homologous GYS2 in the liver. ASO-mediated Gys1 knockdown in the $Gaa^{-/-}$ mouse model of PD led to a robust reduction in glycogen accumulation in skeletal and cardiac muscle. In addition, combining Gys1 ASO with ERT further reduced glycogen content in muscle, eliminated autophagic buildup and lysosomal dysfunction, and improved motor function in $Gaa^{-/-}$ mice. Our results provide a strong foundation for further validation of the use of Gys1 ASO, alone or in combination with ERT, as a therapy for PD in patients.

Valosin-containing protein (VCP) multisystem proteinopathy 1 (MSP1) disease is an autosomal dominant disease caused by gain-of-function pathogenic variants in the VCP gene. The disease is associated with inclusion body myopathy, early-onset Paget's disease of the bones, frontotemporal dementia, and familial amyotrophic lateral sclerosis. There is currently no treatment for this progressive disease associated with early demise resulting from proximal limb girdle and respiratory muscle weakness. We hypothesize that reduction of expression in VCP with the use of ASOs may reduce the typical disease pathology. We assessed the effect of ASOs specifically targeting the human VCP gene in the patient R155H iPSC-derived skeletal muscle progenitor cells (SMPCs). ASOs were well tolerated up to 5 µM concentration and significantly reduced VCP mRNA and protein expression in the SMPCs ~ 50%. We also treated the transgenic mouse model which overexpresses the humanized VCP gene with the severe A232E mutation with weekly subcutaneous injections starting from 6 months of age for 3 months. ASO2-treated mice showed improvements in functional studies compared to mice treated with control ASOs. ASO2 demonstrated tolerability in VCP transgenic mice and showed over 50% knockdown of VCP at the mRNA level and the protein level compared to control ASO. We found improvement in the autophagy markers and reduction in TDP-43 expression, hallmarks of VCP disease. Understanding the pathogenesis of disease is key to developing novel treatments for rare genetic diseases.

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OP54

Beyond the Lab: How Storytelling with Genetics is Revolutionizing Glaucoma Care

Prof. Arijit Mukhopadhyay, *University of Salford, United Kingdom*

Glaucoma is a complex neurodegenerative disease affecting 100 million people globally. Glaucoma is the 2nd largest cause of blindness after cataract, and the largest cause of irreversible vision loss. Glaucoma has multiple subtypes, where the primary subtypes have a major genetic component. Primary Open Angle Glaucoma (POAG) is the most common subtype accounting for over 50% of the total disease prevalence.

POAG is largely multifactorial, with rare cases of highly penetrant familial transmission showing autosomal dominant inheritance.

We have identified a mutation (p.Pro370Leu) in the *Myocilin* gene causing an aggressive form of POAG in a large family from rural West Bengal. This mutation is reported from multiple studies and is always associated with aggressive forms of glaucoma.

To appeal to clinicians and different stakeholders for early interventions in children carrying this mutation, before a fully manifested disease, we created a fictional narrative telling the true story of the family. This is produced as a film and can be seen at https://www.youtube.com/watch?v=MgWYvDEY3NA.

During 2023-24, this film has moved many different strata of the society and led to change of clinical management of POAG saving sight in children.

During this meeting, I will share with the community how the research and the storytelling via film is impacting life of the common people with glaucoma helping translation of research beyond the laboratory.

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OP55

Environmental and Genetic studies reveal potential risks factors for substance use disorder in the high incidence Mizo population of North-East India

Prof. Nachimuthu Senthil Kumar, *Department of Biotechnology, Mizoram University, Aizawl, Mizoram.*

Substance use and substance use disorder (SUD) are leading causes of preventable morbidity and mortality and are often associated with socioeconomic consequences contributing towards preventable morbidity and mortality. Mizoram, a state in northeastern India, inhibited by a tribal community and stands out from other parts of the country in its distinct culture, food and lifestyle habits, and its long history of endogamy in the community. Mizoram shares international borders with Bangladesh and Myanmar which serves as a key transit route for drug trafficking which originates from the Golden Triangle. This brings about an increased availability of inexpensive heroin making it more affordable for new users. Our study explores the relationship between adverse childhood experiences (ACEs), family history, and socio-demographic factors with substance use disorder (SUD). Data were collected using a comprehensive questionnaire encompassing socio-demographic details, ASSIST, AUDIT, DAST, FTND, ACE, and family history assessments, providing a multidimensional perspective on SUD risk factors. Our study also characterized the effect of substance use and environmental factors on the oral microbiome using the V3-V4 hypervariable region of 16S rRNA gene. Alpha diversity, beta diversity, and Linear Discriminant Analysis Effect Size (LEfSe) and associations with environmental factors were analysed. PICRUSt2 was utilised to predict the putative functional pathways of the observed microbes. The results revealed significant higher rates of adverse

childhood experiences (ACEs) and family substance use or mental illness, among individuals with SUD. Significant association was identified between SUD and a family history of substance use was also observed. In metagenome analysis, Alpha diversity indices suggested a significant decrease in diversity within the SUD group and beta diversity analysis showed distinct clustering patterns between the SUD and Healthy control groups. A higher relative abundance of Firmicutes_D and Actinobacteria was observed in SUD, while unclassified Bacteria, Bacteroidetes, and Proteobacteria were more abundant in HC. Prenatal smoking exposure correlated with elevated *Streptobacillus* in HC, while *Megasphaera* and *Limosilactobacillus* in SUD cases. Analysis of whole exome sequence data identified key associations between alcohol use and variants in genes such as ABAT, ALDH1B1, ALDH2, AUTS2, CAT, CYP2E1, GABRA6, GABRB2, GABRG2, HTR1B, INPP1, OXT, OXTR, and SLC6A7. For opioid use, significant variants were observed in genes including RGS17, OPRM1, OPRD1, GAL, and ABCB1. The identification of SNPs linked to alcohol, nicotine, and opiate- dependence provided evidence of genetic predispositions that influence addiction susceptibility. This research underscores the importance of studying underrepresented populations to uncover unique environmental and genetic contributions to complex conditions like SUD, so that new therapeutic targets can be identified to prevent and/or control substance use.

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OP56

Pharmacogenomics in Populations of Jammu and Kashmir.

Prof. Swarkar Sharma, *Human Genetics Research Group, Centre for Molecular Biology, Central University of Jammu.*

Jammu and Kashmir (J&K), situated in the northernmost part of India and nestled in the Himalayas, is geographically isolated and underrepresented in research. We have recently completed a genome-wide scan of 500 healthy individuals from various Indo-European linguistic sub-groups in Jammu and Kashmir. This study aimed to explore genetic variability with a focus on pharmacogenomics. The population of Jammu and Kashmir is ethnically diverse, including major groups like Kashmiris, Dogras, Gujjars, Ladakhis, and Paharis, each with unique cultural and genetic attributes and sub-population groups. This diversity, shaped by historical admixture, results in a range of genetic traits that can impact health outcomes. In the field of pharmacogenomics, examining genetic variability in this region is crucial for developing personalized medicine strategies.

We noticed specific alleles linked to drug metabolism are more common in certain ethnic groups, which can influence how they respond to medications. Customizing treatments based on these genetic differences can enhance drug efficacy and minimize adverse reactions, particularly for complex genetic conditions.

Current efforts aim to deepen the understanding of regional population structures with goals such as disease risk prediction and personalized medicine. Additionally, a comprehensive database of SNP frequency data

is available for research through the portal www.jkdna.in, offering valuable resources for population-based studies in Northwest India.

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ABSTRACTS

Free Communication Sessions

Abstract ID: 124

Spatiotemporal Epigenetic variation in postmortem brain tissue samples from an Indian Population

Bhagyalakshmi Shankarappa*, Ashim Paul Deb, Bharath Holla, Kalyani Bindu Karunakaran, Anita Mahadevan, Biju Viswanath, Sanjeev Jain, Meera Purushottam*

National Institute of Mental Health and Neurosciences, Bengaluru.

Background/ Introduction: The spatial and temporal regulation of gene expression is crucial for normal brain development and function. Alterations in epigenetic and transcriptional regulation are linked to brain development, aging, and disease.

Changes in DNA methylation patterns in the brain have been associated with disorders such as Alzheimer's disease, autism, schizophrenia, and other neurological conditions. Therefore, understanding the patterns of DNA methylation across different brain regions during aging is crucial for uncovering the molecular mechanisms that underlie brain development, function, and disease.

Objectives: To compare genome wide DNA methylation patterns across brain regions, and age groups in an Indian sample To identify DNA methylation signatures and their potential implications for neurodevelopment and disease

Materials and Methods: Tissue samples were acquired from the Human Brain Tissue Repository, National Institute for Mental Health and Neurosciences, Bengaluru, India. Post-consent, samples from 47 individuals (ages 1 to 88, with 15 females and 32 males) were dissected, frozen at −80°C, and processed. Genomic DNA was isolated from ~35 mg of cerebellum and frontal cortex and analyzed for DNA methylation on the Infinium MethylationEPIC array using relevant software for data processing.

Results and Conclusions: We identified 5,000 differentially methylated CpG sites across brain regions. We evaluated genes known to be dysregulated in neurodevelopmental and neurodegenerative disorders, in our cohort and found age and region specific changes in particular genes.

Our findings offer a comprehensive view of the methylation landscape across different brain regions and ages, providing valuable insights into the epigenetic mechanisms underlying brain function and disease.

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Abstract ID: 180

Temporal gene expression signatures across neurodevelopment: a transdiagnostic analysis of bipolar disorder, schizophrenia, autism, and epilepsy

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Background/ Introduction: Bipolar disorder (BPD), schizophrenia (SCZ), autism spectrum disorder (ASD), and epilepsy (EPI) share biological processes. However, spatiotemporal transcriptomic profiles of the associated genes have not been examined in a transdiagnostic manner. Genes harboring rare variants identified through whole exome-sequencing (WES) are particularly valuable for investigating disease pathobiology, as they impact gene and protein function.

Objectives: Our aim was to integrate the WES genetic data of the four syndromes with spatiotemporal transcriptomic data and characterize their temporal and regional specificities. Previous studies have examined the genetic correlations between the syndromes. However, a collective analysis of the rare variant harboring genes associated with the four syndromes, and more importantly, their transcriptomic profiles, will help understand the underlying functional themes.

Materials and Methods: We compiled gene-level exome-wide association data of BPD, SCZ, ASD, and EPI from the BipEx consortium, SCHEMA consortium, Autism Sequencing Consortium, and EPI25 consortium, respectively. Then, we examined this data in conjunction with the BrainSpan Atlas transcriptomic data from 26 brain regions, spanning fetal stages to adulthood, using clustering and enrichment analyses.

Results and Conclusions: Temporal clustering showed specific signatures with BPD genes expressed from early infancy to adulthood and ASD genes in early prenatal stages. Spatial clustering revealed enrichment of BPD genes in visual, somatosensory, and motor cortical regions, and ASD genes in fetal ganglionic eminence. EPI gene expression patterns were similar to BPD, and SCZ to ASD, suggesting overlaps of the syndromes. BPD and ASD clusters were enriched for trans-synaptic signaling and chromatin modification, respectively. This study clarifies the neurodevelopmental context of the syndromes from the perspective of rare variants. It bridges the gap between genes, neural circuitry, and developmental stages, and provides insights into the functional implications of the rare variants. It identifies developmental windows and neural substrates for therapy, and highlights gene networks that offer opportunities for experimental research in patient-derived cell lines and animal models.

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Abstract ID: 194

Association study of blood based and genetic markers predictive of cognitive decline post stroke

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Background/ Introduction: Stroke is the second leading cause of death worldwide and the primary cause of cognitive impairment and long-term disability. Identification of biomarkers which can predicting cognitive decline can lead to mitigation of early intervention in term of cognitive therapy. Thereby reducing risk of cognitive decline post stroke.

Objectives: To identify genetic and blood-based biomarkers associate with cognitive decline post-stroke, and composite analysis of these biomarkers in predicting cognitive decline post stroke.

Materials and Methods: The study involves stroke patients admitted within 10 days of Symptom onset to Rajendra Institute of Medical Sciences, Ranchi, a tertiary care center in Jharkhand. Patients with a history of brain tumors, psychiatric diseases, and neurodegenerative diseases, which affect cognitive measures, are excluded. The methodology includes collecting clinical and sociodemographic profiles, assessing stroke severity using the NIH Stroke Scale, mRS, and Barthel Index, and evaluating cognitive function through standardized tests like MoCA and HMSE. Blood samples are collected, and plasma and serum are separated to measure blood-based biomarkers, including levels of CRP, BDNF, and GFAP, using ELISA. Genetic markers, including APOE, IL6, NOS3, and PICALM, are studied using PCR and Snapshot. Follow-up is done three months after recruitment. Data analysis involves statistical methods to identify significant associations of the biomarkers with cognitive decline.

Results and Conclusions: This study will lead to Identification of blood-based marker and genetic factors influencing cognitive decline post-stroke and development of a composite risk score predictive of cognitive decline. The research holds potential to revolutionize clinical care, enhance research efforts, and improve the lives of individuals affected by post-stroke cognitive impairment by providing early intervention.

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Abstract ID: 902

Plexiform Neurofibromatosis: Early experience from a Multispecialty Clinic at a single centre.

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Background: Neurofibromatosis type-1 (NF1), OMIM# 162200, is an autosomal dominant disorder with an incidence of 1 in 3000. Plexiform Neurofibroma (PNF) is a benign peripheral nerve sheath tumour, seen in ~30-50% of individuals with NF1. PNFs originate from Schwann cell precursor population and are highly vascular, locally invasive, show rapid proliferation during 2nd decade. The risk of malignant transformation is about 8-13%.

The management of PNFs is complicated and debulking surgeries have been done for PNF though complete resection is not achieved. Recently, MEK (Mitogen activated protein kinase) inhibitor Selumetinib has been approved for inoperable PNFs in children older than 2 years. A Neurofibromatosis Multispecialty Clinic (NFMC) has been established at our centre offering multidisciplinary care to individuals with NF1 focusing on early identification of complications like hypertension or malignant transformation.

Objectives: 1. Early accurate diagnosis of mutations in NF1 with genotype-phenotype correlation, 2. Effective medical management of complications of NF1, and 3. Innovative therapies in reduction of tumour burden.

Materials and Methods: 31 individuals with clinically diagnosed NF1 (based on Revised diagnostic criteria for NF1, *E. Legius et al. 2021*) have been enrolled. Nine individuals with Plexiform Neurofibromas are described here. A specially designed proforma was used to capture the data including demographic details, family history, age of onset, site, extent and number of PNFs, progress, treatment history, psychosocial and quality of life assessment.

Results: The age group of the cohort varied from 4y9mo to 44y with male:female ratio of 6:3. Affected relatives were identified in 7 families. All individuals had café-au-lait macules since birth and PNFs were noticed from infancy in 4 and later childhood or adolescence in 5. The distribution of PNF was noted over head and neck in 4, trunk in 1, upper limbs in 3 and lower limb in 1. The size varied from more than 3cm (8/9), causing disfigurement (6/9), pain (2/9), functional impairment (4/9), progressive increase in size (6/9), psychosocial concerns (6/9) and associated learning disability (1/9). Debulking surgery of craniofacial lesion was attempted on three occasions in one affected, but complete resection was not achieved. 2 others have inoperable PNF.

Genetic analysis is available in 5 and a heterozygous pathogenic variant was identified in all (2 deletions, 1 duplication, 1 intronic splice-site variant and 1 nonsense variant).

Conclusion: Nearly 30% of individuals diagnosed with NF1 in our series had PNF. With an emerging therapy, to address this complication it is important that all individuals with NF1 are evaluated in multispecialty clinics such as ours.

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Abstract ID: 190

Deciphering cellular mechanisms of bipolar disorder and lithium response

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Background/ Introduction: The idea that psychiatric diseases have their roots in fetal brain development has been around for decades, but not explored much in bipolar disorder. Brain cells derived from human induced pluripotent stem cells (iPSC) offer an unprecedented opportunity to model specific aspects of BD pathophysiology, including response to lithium.

Objectives: To investigate the role of neurodevelopment in adult-onset bipolar disorder (BD).

Materials and Methods: To investigate cellular aberrations in BD, we generated lymphoblastoid cell lines (LCLs) and iPSC derived neural precursor cells (NPCs), building block of the fetal brain and performed cellular and functional assays. The cells were also exposed to lithium in vitro to investigate if disease-associated phenotypes are reversed. These were investigated using both 2D and 3D brain models.

Results and Conclusions:

LCLs revealed aberrations in cell proliferation, mitochondrial membrane potential; and the reversal with in-vitro lithium. Patient-derived LCLs and NPCs exhibited reduced viability, rescued in clinical lithium responders. Dysfunctions in cellular phenotypes central to NPCs were also observed: in their capacity to proliferate and migrate. Much like in LCLs, in patient-derived NPCs, we observed a reversal in cellular phenotypic abnormalities with in vitro lithium exposure. The human cortical organoids also detected abnormalities for organisational, proliferation and migration defects in BD.

Conclusion:

Aforementioned studies demonstrate subtle changes in fetal brain development associated with BD. Ours is one of the few labs that have shown neurodevelopmental anomalies in BD using the iPSC model system. The relevance of cellular phenotypes examined here to clinical phenotypes is strengthened by rescue of observed defects with lithium exposure.

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Abstract ID: 903

Impact of cancer and therapy on epigenetic landscapes of normal cells: Describing a novel endpoint in survivorship

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Context: With the advent of modern multi-drug chemotherapy regimens, several hematologic malignancies now have excellent outcomes. However, long-term follow-up studies show increased incidences of secondary malignancies & lifestyle diseases - all features of accelerated aging. As per the prevailing information theory of aging, these drugs may affect epigenetic landscapes, resulting in an aged phenotype.

Objective: To study epigenetic effects of cancer and chemotherapy in hematological cancer survivors.

Methods: Whole-genome methylation patterns (using Illumina Infinium MethylationEPIC v2.0 BeadChips) in survivors of Hodgkin's Lymphoma (HL), Acute Promyelocytic Leukemia (APL), Acute Myeloid Leukemia (AML) and age-matched normal controls (n=10 each) were studied.

Results: We demonstrated that levels of aberrant DNA methylation were higher among cancer survivors; observing an average of 2.34%, 6.93%, and 10.68% higher methylation in the lowest quantile of the 935K CpG sites on the BeadChip array in HL, APL, and AML survivors respectively.

Of the differentially methylated regions (DMRs) identified, the HL, APL, and AML cohorts had 211, 2085, and 4248 DMRs of significance ($p<0.0001$), respectively. PCA of the top DMRs showed a clear separation between cohorts.

To understand if these methylation changes were associated with epigenetic age acceleration (EAA), we used the PCGrimAge epigenetic clock tool. When normalized against healthy controls, HL, APL, and AML survivors showed an age-acceleration of 13.8, 23.8 and 28.8 months, respectively.

Conclusions: Our findings suggest that different hematological malignancies and their chemotherapies cause varying long-term epigenetic changes and EAA in normal hematopoietic progenitors. The implications underscore an unmet need for further research and potential changes in treatment algorithms on this basis.

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Abstract ID: 82

Multi-Omics Integration in Pleural Mesothelioma Reveals Key Molecular Signatures

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Background/ Introduction: Pleural mesothelioma (PM) is a rare, aggressive cancer linked to asbestos exposure, with poor prognosis and limited treatment options. Integrating multi-omics data can help unravel PM's complex biology, potentially enhancing early detection, risk assessment, and therapy discovery.

Objectives: This study employs Multi-Omics Factor Analysis (MOFA) to identify distinct molecular factors associated with PM by integrating genomics, DNA methylation, miRNA, and proteomics data. By reducing dimensionality, we aim to reveal independent biological insights across omics layers, shedding light on PM pathogenesis.

Materials and Methods: We analyzed a dataset from a nested case-control study within the Prospective EPIC cohort, comprising 142 preclinical PM cases and matched controls, integrating different omics profiles. MOFA enabled unsupervised integration of these datasets, focusing on variance-capturing factors. We examined correlations between factors and clinical variables (e.g., age, sex) and used logistic regression to assess associations with PM susceptibility. Functional enrichment analysis was performed on top-weighted features from each factor for biological interpretation.

Results and Conclusions: Several high-variance factors were identified, notably Factor 2, which correlated strongly with genomics data and PM differentiation, showing significant functional enrichment in the chromosomal region 6q14, previously linked to PM susceptibility. Key regulatory elements like CTCF binding sites were found, potentially influencing genes like MYC and BAG1. The miRNA analysis revealed nine significant miRNAs, including hsa-mir-21-5p and hsa-mir-126-5p, suggesting a miRNA-mediated regulatory mechanism. Proteomic data highlighted processes related to wound healing, immune response, and stress. Additionally, miRNA analysis emphasized pathways related to aging, innate immunity, and inflammation,

crucial given the role of immune dysregulation in asbestos-related carcinogenesis. Multi-omics results aligned with prior single-omics findings, reinforcing our approach.

Conclusion: This multi-omics integration identifies key molecular signatures in preclinical PM, uncovering factors and pathways related to immune response, transcriptional regulation, and cellular stress. This approach enhances potential for PM biomarker discovery, improving risk assessment and targeted interventions. Further research is necessary to validate these factors clinically and explore therapeutic potential.

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Abstract ID: 117

Molecular profiling of Oral Squamous Cell Carcinoma patients of south Indian population

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Background/ Introduction: Oral Squamous Cell Carcinoma (OSCC) is a heterogeneous group of cancer arising from the mucosal lining of the oral cavity. The incidence and prevalence of oral cancer show significant geographic disparities, with substantial variation across different regions. In India, it is the third most common cancer contributing to mortality. The primary causes of OSCC vary by geographic location and are influenced by context-specific factors such as dietary habits, lifestyle, tobacco chewing, smoking, and poor oral hygiene, which increase the OSCC burden. It is therefore important to screen and validate the association of mutation identified for the accurate diagnosis and therapeutic purpose. Further, the role of genetic variants and risk factors in the management of OSCC is not comprehensively analyzed in patients living in south India. Hence, present work aimed to answer how inter-individual variability can possibly influence the outcome of OSCC in a given patient cohort of South India.

Objectives: Molecular profiling of oral squamous cell carcinoma patients among south Indian population compared to the Caucasian cohorts using FFPE tissue blocks

Materials and Methods: DNA isolated from 14 FFPE blocks of OSCC patients of the south Indian origin and adjacent normal tissue of the same patients used for the molecular profiling with whole-exome sequencing (WES). Integrative Genomics Viewer (IGV) was used to validate somatic mutations. Mutation burden and mutant genes were correlated to clinico-pathological parameters. We are also working to identify hotspot

mutations in the protein domains which can be used as a drug target. TCGA datasets were downloaded from cBioportal for comparison

Results and Conclusions: A comprehensive genomic analysis revealed an average of 1077 mutations per individual, predominantly C>T and C>A single nucleotide polymorphisms, consistent with smoking-related mutagenesis. In total, 13 subjects harbored hotspot mutations in TP53 and CDKN2A, established oncogenic drivers in HNSCC. While TP53 and CDKN2A mutations were observed in both South Indian and European OSCC patients, the South Indian cohort exhibited a unique mutational landscape, with a higher frequency of somatic functional mutations in KMT2D. This profile has been reported by Indian project team of ICGC consortium in gingivo-buccal oral squamous cell carcinomas. However, patients with this molecular profile were too few (~5%) in this ICGC cohort. These findings have significant implications that could inform targeted therapies and diagnostics, such as personalized treatment plans and early detection biomarkers.

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Abstract ID: 103

Molecular pathology of cervical carcinogenesis: Role of telomere and THOR in tumor initiation and progression.

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Background/ Introduction: Telomere dysfunction and subsequent genomic instability is an important hallmark triggering cellular immortalization and tumor progression in cervical cancer. An epigenetically regulated region upstream of the core promoter region of hTERT that appears to be responsible for its activation in cancer is referred to as the TERT hypermethylated oncological region (THOR). However, correlation between THOR hypermethylation, hTERT upregulation and cervical cancer progression in North Indian population have not been addressed yet. Studies on THOR hypermethylation status can be useful in prognosis and diagnosis of cervical cancer tumor initiation and metastasis.

Objectives: 1) mRNA profiling of hTERT and hTERC and relative telomere length analysis in cervical cancer biopsies. 2) HPV typing, sub-typing and copy numUmaber evaluation in cervical cancer patient samples. 3) Examining the functional role of THOR in cervical cancer progression.

Materials and Methods: Relative mRNA expression of hTERT and hTERC and relative telomere length in clinical samples as well as azacytidine treated cells (HeLa and SiHa) were determined by qPCR. MS-PCR was

done to determine the methylation status. HPV L1 copy number was estimated by creating standard curves with genomic DNA. hTERT protein profiling following Azacytidine treatment was done by flow-cytometry.

Results and Conclusions: The relative mRNA expression of hTERT and hTERC and relative telomere length is significantly upregulated in patient samples compared to control. It was found that 75% of the samples were positive for HPV while 25% samples were negative. Methylation status analysis showed that 65% of total control samples were homozygous unmethylated whereas 81% of cervical cancer patient samples were heterozygous for THOR methylation. Hypomethylation of THOR by azacytidine treatment reversed the methylation status of THOR and downregulated the hTERT mRNA and protein expression and thereby telomere length in cervical cancer cells.

Re-activation of telomerase plays an important role in cervical cancer progression. In cervical cancer, we have observed that the up regulation of hTERT, hTERC as well as telomere length are significantly correlated with HPV status. Hypomethylation of THOR decreases telomerase activity thereby inhibiting cancer progression.

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Abstract ID: 118

Transcriptome analysis reveals potential biomarkers in Head and Neck squamous cell Carcinoma

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Background/ Introduction: Head and neck cancer (HNC) is the sixth most common cancer worldwide, primarily affecting the oral cavity, lip, oropharynx, hypopharynx, and larynx. Oral squamous cell carcinoma (OSCC) arises primarily from the oral mucosal epithelium and accounts for approximately 90% of oral cancer cases. Despite the advances in therapy, 5-year survival remains poor. The molecular mechanisms underlying carcinogenesis is essential to explore the key contributing factors at the transcriptome level and identification of prognostic biomarker.

Objectives: we aimed to study the expression of the differentially expressed genes the transcriptome sequencing and its significance in disease progression.

Materials and Methods: The oral tumor tissues and the adjacent normal tissue were collected from 50 untreated oral cancer patients during surgical resection and 10 OSCC samples were subjected to RNA

sequencing using the Illumina platform. The differentially expressed genes were validated by qRT-qPCR and confirmed by Immunohistochemistry (IHC). Online web interfaces such as Tumor Immune Dysfunction and Exclusion (TIDE) and Tumor IMmune Estimation Resource 2.0 (TIMER2.0) were used for the biomarker evaluation and immune cell infiltrations respectively.

Results and Conclusions: The RNA sequencing identified 225 differentially expressed genes, of which 175 were upregulated and 50 were downregulated. Gene ontology pathway analysis identified PTGS2, IL-1A, IL-1B, CCL3, and MMP3 as key genes involved in tumor-promoting inflammation and invasion. The genes showed a strong association with immune checkpoint blockade (ICB) with area under curve (AUC) scores greater than 0.5 in the TIDE analysis. The TIMER2.0 analysis revealed significant infiltration of CD8+ T cells and myeloid-derived suppressor cells (MDSCs) implying immune evasion and correlated with the prognosis. Our study revealed key deregulated genes that play a significant role in modulating the tumor microenvironment and CCL3 could serve as a biomarker and personalized immunotherapy for oral cancer.

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Abstract ID: 917

Exploring the DNA methylation profile associated with Bardet-Biedl syndrome: An EPIC array-based approach

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Background: Bardet-Biedl Syndrome (BBS) is a ciliopathy characterized by retinitis pigmentosa, hexadactyly, truncal obesity, renal failure, intellectual disability, and reproductive abnormalities. The disease exhibits autosomal recessive inheritance with 26 genes (*BBS1-BBS26*) mapped till date. The protein products of these genes play crucial roles in the biogenesis of non-motile cilia. Metabolic syndrome are identified with specific DNA methylation pattern; given that BBS is characterized with metabolic features like, hyperleptinemia, increased intra-abdominal fat accumulation, type 2 diabetes, a steady increase in insulin resistance, we hypothesize that specific DNA methylation patterns occur unique to BBS. Here we present our preliminary findings on the methylation patterns observed in molecularly confirmed BBS cases.

Objectives: To identify the differential methylation pattern specific to BBS and its associated metabolic features.

Methodology: EPIC array-based genome-wide methylation data were generated for molecularly confirmed BBS patients (N=6) and non-disease controls (N=7). In brief, DNA samples were subjected to bisulfite conversion, followed by whole genome methylation analysis in the EPIC Array platform. The pair of intensity data files (IDAT) generated for each sample were further analyzed using the ChAMP R package. Differentially methylated probes (DMPs) were identified with a Benjamini-Hochberg (BH) adjusted P-value < 0.05 and a minimum methylation difference of 20%. The identified DMPs were used to construct SVM based binary classification model and the accuracy of the model was assessed by leave-one-out cross-validation. Using the ChAMP Bumphunter algorithm, differentially methylated regions (DMRs) were also identified which were defined by 7 consecutive probes with adjusted p-val <0.05. These DMRs were prioritized in the VarElect tool followed by Gene Set Enrichment Analysis (GSEA).

Results: ChAMP analysis identified a total of 1,636 differentially methylated probes after normalization, of which 187 were unique to BBS. Binary classification by SVM separated these two groups into different sample clusters. The SVM model was used to validate these disease-specific probes in another cohort of clinically confirmed BBS cases (N=8) irrespective of their molecular status. Further, 246 DMRs with at least seven probes in a gene were identified, of which 84 DMRs were prioritized based on the disease phenotype

using VarElect software and were further narrowed down to 69 DMRs based on their positioning in the transcription start sites. Gene Set Enrichment analysis of these DMRs in the Molecular Signature Database identified significant (FDR<0.05) biological processes such as development of retinal cone cells and cell-type specific biological events such as the development of retinal fibroblasts, fetal kidney stromal cells, fibroblasts in the developing heart, among others. One among the 69 DMRs is the *MCHR1* gene, which is involved in adipogenesis.

Conclusion: This study has identified specific methylation patterns in those genes associated with the disease manifestation. Identification of specific epigenetic signatures is emerging as potential diagnostic markers in many monogenic diseases. This is the first study elucidating the possible role of methylation in BBS. The prioritized genes that exhibit significant methylation changes are being validated in an independent cohort.

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Abstract ID: 44

HLA-C and HLA-B Shape Psoriasis Risk and Protection Profiles.

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Background/ Introduction: Psoriasis is an immune-mediated hyperproliferative chronic disorder that mainly affects the skin and has a high genetic complexity. It is considered a T-cell-mediated autoimmune skin disease. HLA-C and HLA-B are highly polymorphic Class I alleles that serve to present endogenous antigens to cytotoxic T cells. Therefore, HLA-C and B alleles are important in susceptibility and protection against the disease. Apart from HLA-C*06, no other susceptible HLA-C and B alleles have been reported yet.

Objectives: Determination of the HLA-C and B alleles among the psoriasis patients and compare it with the healthy controls. Additionally, the co-occurring HLA-C and B alleles, and specific genotypes may classify further susceptibility to psoriasis. The understanding of such combinations is crucial to disclose disease-specific genetic interactions that lead to the heterogeneous nature of the disease.

Materials and Methods: This study recruited 400 psoriasis cases and healthy controls from eastern India. To determine the HLA-B and C alleles, exons 2 and 3 were PCR amplified and subjected to Sanger sequencing. All the sequences from respective samples were subjected to BLAST with the IMGT/HLA database to identify the HLA-B and C alleles. As HLA-C and B are highly polymorphic, we identified an ambiguity for proper identification of the alleles, and it was solved using a novel cloning-specific approach. Exons 2 and 3 of HLA-

B and C of the respective samples were TA-cloned and sanger-sequenced, which in turn helped to fortify the exact identification of an allele.

Results and Conclusions: Among the psoriasis patients, apart from C*06, C*18 allele was significantly associated with the disease. When the data was classified based on the presence or absence of C*06 allele, C*18 was significantly associated with C*06 negative patients, suggesting C*18 as a key player apart from C*06. In the case of protective alleles, C*04, C*08, and C*07 showed significant associations. For HLA-B, B*15, B*56, B*57, B*58, and B*39 showed significant associations with the disease, whereas B*40, B*35, B*07, and B*48 showed protective alleles. We observed that C*06-positive individuals harbored B*56 and B*15 alleles and C*04-positive individuals harbored protective B*40 and B*35 alleles. When classified according to the genotype, B*56-B*15 conferred the highest risk and B*40-B*35 conferred the lowest risk. But, interestingly, in the case of HLA-C, C*12-C*06 conferred the highest and C*12-C*04 conferred the lowest risk suggesting the exclusivity of C*06 and C*04 among psoriasis and healthy individuals. The instability index of C*06 and C*04 specific amino acid sequences classified C*06 as unstable and C*04 as stable, respectively. This study reported a new approach for identifying HLA-C and B alleles and explored the association of HLA-C and B alleles with psoriasis patients in India.

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Abstract ID: 143

Development and validation wise assessment of genotype guided warfarin dosing algorithm in Indian population

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Background/ Introduction: Warfarin Pharmacogenetic panel for dose optimization in patient care is not yet validated for Indian Asian ethnicity.

Objectives: A study was conducted to develop and validate the warfarin pharmacogenetic dose optimization algorithm considering the clinical pharmacogenetic implementation consortium (CPIC) recommendations for the Asian ethnicity population.

Materials and Methods: The present prospective observational study recruited warfarin-receiving patients. We collected a three ml blood sample for VKORC1, CYP2C9*2, CYP2C9*3, and CYP4F2 polymorphism

assessment during the follow-up visits. Clinical history, sociodemographic and warfarin dose details were noted.

Results and Conclusions: The study recruited 300 patients (250 in derivation and 50 in validation timed cohort) receiving warfarin therapy. The baseline characteristics were similar in both cohorts. BMI, presence of comorbidity, VKORC1, CYP2C9*2, and CYP2C9*3 were identified as covariates significantly affecting the warfarin weekly maintenance dose ($p<0.001$ for all) and the same were included in warfarin pharmacogenetic dose optimization algorithm building. The algorithm built-in the present study showed a good correlation with Gage ($r=0.57$, $p<0.0001$), and IWPC ($r=0.51$, $p<0.0001$) algorithms, widely accepted in western side of the globe. The receiver operating characteristic curve analysis showed a sensitivity of 73 %, a positive predictive value of 96 %, and a specificity of 89 %. The algorithm correctly identified the validation cohort's warfarin-sensitive, intermediate reacting, and resistant patient populations.

Conclusions: Validation and comparisons of the warfarin pharmacogenetic dose optimization algorithm have made it ready for the clinical trial assessment.

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Abstract ID: 141

Otitis Media, an underrated complex disease: From candidate gene to next generation sequencing and from confusion to clarity, in the end pragmatically a covalent compromise?

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Background/ Introduction: Otitis media (OM) is a prevalent yet often underestimated disease backed by genetic, epigenetic, environmental, and anatomical factors. Chronic suppurative otitis media (CSOM) is a leading cause of hearing impairment, secondary complications, and financial strain. Research on OM's genetic and epigenetic mechanisms is limited, but epigenetic factors like DNA methylation, miRNAs, and lncRNAs are crucial in disease progression. Understanding these factors is essential for developing targeted treatments.

Objectives: Identifying the genetic and epigenetic reciprocations of candidate genes associated with OM and comparing it with the transcriptome and methylome data from patient tissues for focusing on pragmatically mitigable molecular targets.

Materials and Methods: The candidate gene study includes basic genetic/statistical/epidemiological analysis of 300 OM patients and healthy controls. Insilico tools were used to predict the putative methylation status and TF binding of SNVs in the promoter region. Gene expression/methylation levels in the blood and middle ear mucosa of patients with CSOM were evaluated by expression/methylation analysis utilizing ELISA, qPCR, MS-PCR, flow cytometry, and IHC. Whole transcriptome sequencing (WTS), miRNA seq., and whole genome bisulfite seq. (WGBS), were all performed on the same tissue sample to identify the cumulative/reciprocal

effects of genetic and/or epigenetic regulations of OM susceptibility. The associated candidate genes and the genes identified from NGS are being compared for the rational check in the tissue samples.

Results and Conclusions: This study investigates the genetic and epigenetic factors associated with OM. The cohort was 53% male and 47% female, with an average age of 32.48 years, and 20% had multiple affected family members. Significant differences in immune-inflammatory markers were observed across OM subtypes, and genetic analysis linked the ISL1 gene to OM, with SNP variants potentially influencing eosinophil levels. A TLR3 variant showed a protective effect in males. Elevated TGFβ1 levels in plasma and tissues pointed to its role in chronic inflammation. Flow cytometry revealed increased immune cell populations in CSOM patients. Epigenetic analysis led to the creation of Gememiom (www.gememiom.org), a database mapping OM-related genes, SNPs, and methylation effects. NGS revealed novel miRNA and lncRNA targets, and over 1600 differentially methylated regions were identified, with many being hyperregulated. Understanding and treating OM will need integrating multiple interventions, as it ultimately appears to involve a pragmatic and covalent balance. The term "covalent compromise" alludes to the metaphorical joining of several scientific viewpoints to create a more comprehensive and useful knowledge or treatment plan, much like covalent bonds keep atoms together.

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Abstract ID: 904

Investigation of clinical and genetic heterogeneity in human fetuses with arthrogryposis multiplex congenita

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Introduction: Arthrogryposis multiplex congenita (AMC) is a condition with contractures in two or more joints and associated with approximately 400 conditions. AMC is not a distinct diagnosis but a phenotypic finding. Reduced or absent fetal movements in utero are the primary manifestation.

Objectives: The study aims to elucidate the clinical characterization of fetal or neonatal losses with arthrogryposis by perinatal and postnatal evaluation and identifying the genomic alterations underlying them.

Material and Methods: We performed perinatal evaluation of fetuses referring to autopsy following pregnancy loss and clinical evaluation in neonates after taking informed consent. Fetal tissues and blood samples were used to obtain genomic DNA. Chromosomal microarray (CMA), whole exome, and genome sequencing were performed as required. Spinal muscular atrophy (SMA) and common aneuploidies were ruled out whenever necessary.

Results and Conclusion: We have recruited 50 families (58 subjects; males-39; females-17) with AMC in the study. Seventeen consanguineous families (17/50, 34%) were noted. Gestation ranged from 12-38 weeks and there were eight neonatal losses (first day of life to one month of age). Apart from contractures, pulmonary hypoplasia, hydrops and facial dysmorphism are the most notable findings in the cohort. Six families (6/35 families) were diagnosed with trisomy 18. The study identified rare disease-gene associations in thirty families (60%) following exome sequencing (30/43 families) with singleton exome sequencing yielding the molecular diagnosis majorly. Overall, there were disease-causing variants in 23 genes among 30 families (76.6%), in which 8 disease-gene associations (11 families) have been reported by us. Among the 15 disease-gene associations in remaining nineteen families, the most common ones are *COASY* (3 families), *ECEL1* (2 families) and *MYH3* (2 families). A biallelic variant c.644-13_644-9delCTTTC in *UNC50*, a gene of uncertain significance was identified. This variant leads to aberrant splicing with an insertion of 17 nucleotides following reverse transcriptase-polymerase chain reaction and identified as cause of recurrent fetal akinesia deformation sequence in a family.

We achieved a molecular diagnosis in thirty-six families (72%). Most of the families had autosomal recessive (26 families) inheritance and were lethal. We also noted 30 novel variants and 25/36 (69.4%) pathogenic or likely pathogenic variants in the cohort. Detailed perinatal evaluation with integration of genomic tests in fetuses with arthrogryposis improves diagnosis.

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Abstract ID: 102

Choline kinase is differentially regulated in cervical cancer progression

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Background/ Introduction: Choline kinase is the precursor enzyme in Kennedy pathway producing phosphatidylcholine, which is an important constituent of mammalian membrane system. Cancer cells are aggressive in division, thereby requiring over expression of Choline kinase to produce phosphatidylcholine exponentially to be used in rapid membrane formation. Choline kinase is a dimer of two isoforms CHKA (11q13.2) and CHKB (22q13.33). In cervical cancer progression, the role of Choline kinase is poorly understood.

Objectives: In this study we have profiled CHKA and CHKB in cervical cancer patients from Gangetic plain of north India. Also, we have analyzed the molecular mechanisms associated with the regulation of Choline kinase in cervical cancer.

Materials and Methods: Expression profiling was done by q-RT-PCR and western blotting. MS-PCR and bisulfite sequencing was done for analysis of promoter methylation status. Selective knockdown of CHKA was done by shRNA. Intracellular ROS generation, cell cycle analysis and mitochondrial potential was measured by flow cytometry. ATP synthesis and quantification of phosphatidylcholine levels was done by assay kit.

Results and Conclusions: Our results demonstrate that Choline kinase is differentially regulated; mRNA profiling suggests CHKA is frequently up regulated (60%) and CHKB is frequently down regulated (81.73%) in cervical cancer patients than hysterectomy control samples. MS-PCR analysis followed by bisulfite sequencing suggests methylated CpG island 3 associated with CHKB promoter region is the major cause of CHKB down regulation. Differential regulation of Choline kinase is significantly correlated clinically with low age strata, nodal metastasis and positive HPV infection. shRNA mediated CHKA knockdown is associated with low mitochondrial membrane potential and high intracellular ROS. Further G2/M transition is stalled in CHKA knocked down cells. CHKA being the major isoform of Choline kinase in cervical cancer cells; its down regulation results in low phosphatidylcholine formation. CHKA knockdown is compensated by CHKB up regulation in cervical cancer cells; but not sustainable.

Abstract ID: 175

Gene editing reveals mutation specific disease manifestations in a human pluripotent stem cell derived Pompe disease model

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Background/ Introduction: Pompe's disease (PD) is an inherited lysosomal storage disorder (LSD) with a global prevalence of 1 in 40,000. PD is caused by mutations affecting the enzymatic function of acid α-glucosidase (GAA) encoding gene. GAA is a hydrolase that breaks down glycogen within lysosomes and in PD, its reduced or complete loss of function causes glycogen accumulation in cells. This causes a progressive dysregulation of functions associated with heart, skeletal and the respiratory system. Enzyme replacement therapy (ERT), which involves administrating recombinant functional GAA enzyme remains the primary treatment for PD patients. However, it is inefficient in rescuing skeletal muscle defects and is not cost effective. Thus establishing skeletal muscle models can enable identifying mechanisms that can improve treatment outcomes

Objectives: To establish human pluripotent stem cell-based disease model to study Pompe associated disease pathogenesis in skeletal muscles

Materials and Methods: A previously established human embryonic stem cell line (BJNhem20) derived from a healthy donor was used. To derive a cell line that can display pompe disease pathogenesis, specific mutations were introduced in GAA gene using genome editing. The control and the mutant lines were differentiated to skeletal muscles and the disease pathogenesis was evaluated.

Results and Conclusions: we have successfully utilized a CRISPR-Cas9 based gene editing approach to generate PD embryonic stem cell lines with mutations that are prevalent in India (Ex.14-c.1942G>A; Ex.6-c.1003G>A) and a founder mutation in the East Asia (Ex.14-c.1935C>A). In line with the previous reports using PD derived patient iPSCs, the reduction in GAA enzyme activity did not affect pluripotency. Differentiation to skeletal muscle cells revealed variability in disease manifestations, with mutations c.1942G>A and c.1935C>A exhibiting severe loss of myotubes. They also displayed hallmarks of PD such as accumulation of lysosomal markers and glycogen accumulation. Taken together, using genome editing we were able to develop pluripotent stem cell derived skeletal muscle model for PD. Importantly, the model's ability to recapitulate a spectrum of disease severities can enable evaluation of a therapeutic intervention to rescue the mutation dependent heterogenous disease manifestations observed in Pompe patients.

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Abstract ID: 120

Cellular Specificity of Ribosomal Protein Gene Expression in Human Tissues

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Background/ Introduction: Ribosomes comprise rRNA and ribosomal proteins (RP) and can change their composition to perform specific functions based on the environment or tissue types. These changes in RP composition have been shown to cause preferential translation of specific mRNAs, leading to specific functions. In cancers, it has been observed that RP stoichiometry changes affect the translation of specific mRNAs and could lead to differential cancer progression. Previous research using bulk RNA-Seq data has shown that various human tissues and tumours have distinct mRNA expression signatures for RP genes. The tissues comprise many cell types of distinct functionality, and hence, the RP gene expression patterns can vary within a tissue.

Objectives: To elucidate the heterogeneity in RP gene expression within different cell types of a particular tissue compared to other genes and study the tissue-specific expression of RP genes in cell types found in multiple tissues.

Materials and Methods: We used publicly available 10X genomics-based scRNA-Seq datasets for 16 healthy tissues from the Human Cell Atlas and processed them as per our pipeline. The RP gene expression patterns across different cell types of the tissues were analysed. Besides, RPs were compared with other ubiquitous and random genes regarding their expression variability across the tissues. Finally, the RP gene expression patterns in a few common cell types across different tissues were analysed to understand the tissue-specific expression patterns of RP genes.

Results and Conclusions: Analyses of the 16 tissues showed that RP genes exhibit a range of expression patterns across different cell types of the tissue, with some tissues showing more divergent profiles than others. RP genes demonstrated the highest degree of variability in gene expression across the different cell types in the majority of tissues examined compared to other genes. Analysing common cell types like endothelial cells and fibroblasts across tissues revealed that these cell types have tissue-specific RP gene expression patterns. Overall, we showed RP gene expression patterns can vary at the cellular level and depend on the tissue in which the cell is located. This analysis is being extended to cancer scRNA-Seq datasets, which will help reveal how these RP gene expression patterns change with the disease phenotypes at a cellular level. Further, understanding the unique patterns of RP gene expressions in different tumour cell types will help explore if RPs can serve as potential genetic markers.

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Abstract ID: 905

Cytogenetic Landscape of Products of Conceptions from Recurrent Pregnancy Loss in North Indian Population

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Aims & Objectives: Recurrent Pregnancy Loss (RPL) is one of the major complications affecting reproductive age couples. Due to variable etiologies diagnostic evaluation of RPL poses a major challenge to clinicians. This study aimed at genetic evaluation of product of conception (POC) for comprehensive understanding of RPL genetic etiology and study its co-relation with clinical parameters.

Materials/Methods: This prospective multi-centric study was conducted for 2 years, wherein 30 couples with history of RPL were enrolled. Detailed clinical history was recorded for each patient, followed by couple karyotype. QF-PCR was done in all POC samples to rule out maternal cell contamination (MCC) followed by Chromosomal microarray (CMA) to look for the copy number variations (CNVs).

Results: In 6 individuals out of 30 couples (10%), abnormality in karyotype was detected. Balanced translocation was present in 2 individuals and heteromorphic variants was present in 4 patients. Out of 30 POCs, 9 were excluded because of MCC. CMA in POCs identified pathogenic CNVs in 24% cases (5/21), Variant of Unknown Significance (VUS) in 19% cases (4/21). Likely pathogenic/pathogenic CNVs were: 1. Mosaic Chr19 monosomy (47.9Mb) along with 1Mb deletion on chr17q24.2; 2. Partial trisomy of chr 9 (90.3Mb) and a deletion on 15q11.2-q15.3 (21.3Mb); 3. deletion on 11q24.3q25 (6.8Mb) with a duplication (34.1Mb) on 14q24.2q32.33; 4. mosaic Klinefelter (155Mb) and 5. trisomy 11. Among VUS CNVs, few important genes which were deleted were found to have important role during embryo development. A small deletion in 2q34 was identified in a POC which encompasses a gene ERBB4 which is an important receptor in the control of fetal lung type II cell maturation. Another case revealed a deletion on 14q13.2 including SRP54 gene which is associated with autosomal dominant form of Neutropenia, severe congenital. No co-relation was found with the clinical parameters except the advanced maternal age.

Conclusion: Cytogenetic analysis of POCs is important to get a clue of fetal lethality/loss and to estimate the risks of recurrence in future pregnancies. Early recognition of a potential risk to pregnancy loss and systematic monitoring can have beneficial effect in increasing live birth rates in RPL couples.

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Abstract ID: 75

Comprehensive genomic and global DNA methylation profiling in Indian Fanconi Anaemia subjects

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Background/ Introduction: Fanconi Anaemia (FA) is a rare genetic, autosomal recessive disorder; characterized by chromosomal breakages, bone marrow failure, congenital malformation, developmental disorders, physical abnormalities and an increased predisposition to solid tumours and haematological malignancies such as myelodysplastic syndrome (MDS) and acute myeloid leukaemia (AML). FA is generally presented with variable clinical presentation including severity of bone marrow failure and cancer development. Although genetic basis of FA is well studied the exact mechanism of cancer predisposition is not known. In our study we have studied epigenetic modifications particularly DNA methylation and its association with cancer predisposition in FA

Objectives: The study investigates the role of epigenetic factors in Fanconi Anemia phenotype and to understand the cancer predisposition through global DNA methylation profiling

Materials and Methods: Chromosomal breakage investigations induced with MMC and mutational analysis using NGS, MLPA and Sanger sequencing was carried out in all FA subjects. Genome-wide methylation profiling was done using the Illumina Infinium Methylation EPIC 850k Bead Chip Array in 12 subjects and the gene expression of selected methylated genes was validated in 162 subjects by qPCR.

Results and Conclusions: The study was carried out in two sets; the discovery cohort (n=12, 7 FA samples and 5 controls) and the validation cohort (n=162, 62 patients and 100 controls (30 siblings and 70 age-matched healthy controls). Median age of the patients were 8.9 years (0.8 years to 32 years range) and the median age of onset of BMF was 9 years. Genomic analysis of FA subjects could identify 10 different complementation groups in the study cohort. Global DNA methylation profiling in FA patients revealed an overall hyper-methylation across several CpGs compared to the controls. A significant ($p>0.05$) hyper-methylation pattern was observed in FANCA complementation groups when compared to FANCG and FANCL complementation groups. Pathway enrichment analysis and gene ontology of the differentially methylated genes identified involvement of key regulatory pathways such as mTOR, NF-κB, WNT and apoptosis pathways. Further validation of top 15 significantly differentially methylated genes revealed a significant ($p>0.05$) downregulation of FAM65B, NKAPL, ITGAM, CDIP1 and CDKN1b in patients as compared to controls. We also observed a significant ($p>0.05$) overexpression of DNA methylase (DNMT1) and histone methyl transferases enzymes (SETMAR, PRMT1, PRDM8, SETD2).

In conclusion these unique hyper-methylated genes such as FAM65b, NKAPL, CDIP1, CDKN1b, ITGAM & HDACs can be a useful predictor in cancer progression in FA subjects. Hence regular monitoring and long term follow up studies are essential to better understand the role of these genes in cancer development and their correlation with the FA phenotype.

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Abstract ID: 134

Genomic Insights into North Eurasian and South American Indigenous Populations Shaped by Diverse Environments

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Background/ Introduction: By the late Pleistocene, human expansion into North Eurasia led to the settlement of the Americas, reaching Patagonia, the farthest point from the migration out of Africa. However, key aspects of the genetic diversity and demographic history of Indigenous North Eurasian and Native South American populations remain unclear, largely due to underrepresentation in genetic studies and reliance on low-resolution genotype data.

Objectives: This study aims to uncover the following genetic characteristics of Indigenous populations in North Eurasia and South America using whole-genome sequencing data: 1. Infer population structure across regions, 2. Uncover past population dynamics shaping genetic diversity and its potential health impacts, and 3. Examine environmental influences in shaping genetic diversity and adaptation.

Materials and Methods: We analysed 72,354,450 genetic variants from whole-genome sequences of 1,537 individuals across 139 populations in North Eurasia and South America, generated by the GenomeAsia 100K consortium. Population structure was inferred using allele frequency and haplotype-sharing methods. Past

Population dynamics were explored through haplotype genealogy and Markovian coalescence-based approaches. Adaptive signals were detected by examining population differentiation and haplotype homozygosity decay-based methods.

Results and Conclusions: Our findings reveal the diverse ancestry in indigenous populations across North Eurasia and South America. We find that the West Siberian ancestry, represented by the ethnic minority Kets, thrived in Siberia 10,000 years ago but has since experienced a fourfold reduction in population size. West Beringian populations (Koryaks, Inuits) are genetically distinct, with adaptation in lipid metabolism and thermogenesis genes for Arctic survival.

The first South Americans, upon entering the uninhabited continent, were instantly isolated by environmental boundaries, diverging into four major lineages— Amazonians, Andeans, Chaco Amerindians, and Patagonians—13,900 years ago. Rapid and long migration into South America and geographic isolation significantly reduced overall genetic and immunogenic diversity, increasing vulnerability to unencountered pathogens. Combined with reduced genetic diversity, loss of traditional lifestyles, and natural habitat encroachments, pushed populations like the Patagonian Kawésqar to the brink of extinction.

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Abstract ID: 129

Individuals' unique genetic makeup influences their susceptibility towards infectious diseases: COVID-19 as a case study

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Background/ Introduction: Each individual is born with a unique genetic blueprint. While humans across the world are genetically 99.9% similar, the remaining 0.1% accounting for differences in physical traits, disease susceptibility, and responses to the environment. This unique genetic makeup can significantly influence an individual's vulnerability to infectious diseases such as COVID-19. Specific genetic variations may heighten the severity of COVID-19 and experiencing post-acute sequelae of SARS-CoV-2 infection (PASC).

Objectives: In this study we aimed to identify genetic variants, associated genes, and underlying biological pathways linked to the severity of COVID-19 and PASC.

Materials and Methods: We examined genomic data from diverse Indian samples, including 60 individuals with COVID-19 history, genotyped on Illumina Infinium Global Screening Array v3.0 in our laboratory, as well as data from the Gujarat Department of Biotechnology, and GenomeAsia 100K. Our analysis involved two different case-control models. The severity cohort compared the genomes of individuals with mild or

asymptomatic COVID-19 (controls) to those who experienced severe COVID-19 (cases), while the PASC cohort compared individuals with post-COVID complications (PASC) (cases) to those without any noticeable Long COVID symptoms (controls).

Results and Conclusions: Our findings revealed significant genetic differences between individuals with severe COVID-19 compared to those with mild cases, as well as among those experiencing Long COVID versus those who are not. Notably, most of the genetic variants that showed significant frequency variation in both the severity and PASC cohort are associated with the nervous and cardiovascular systems. Our study that combined various genomic, transcriptomic and proteomic datasets from humans and rodents underscored the genetic predisposition of certain individuals to COVID-19 and PASC, suggesting the potential for the development of prognostic marker panels based on germline mutations to identify susceptibility to such infectious diseases.

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Abstract ID: 114

Genetic Basis of Climatic adaptation in Thar Desert populations

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Background/ Introduction: Deserts are among Earth's harshest ecosystems, marked by extreme stressors like high UV radiation, limited food, aridity, temperature fluctuations, and dust. Yet, desert inhabitants thrive through unique physiological and behavioral adaptations.

Thar is amongst the world's most populous deserts with a large number of diverse endogamous populations who are with native or have migrated from different parts of the world. Studies in desert biota have revealed signatures of selection and convergent evolution in pathways related to food metabolism, thermoregulation, salt and water homeostasis many of which are relevant in non-communicable diseases. Studies on human genetic adaptation to Thar are limited. Baseline information on signatures of selection to extreme desert environment could be important to address risk to diseases during transitions from non-native to Thar environment or vice versa.

Objectives: The aim of this study is to explore signatures of selection in genetically distinct and representative populations of Thar.

Materials and Methods: A comprehensive literature review of historical archives, census data, and pilot surveys across four major districts in the Thar region identified key communities for study. Genotyping was performed on 185 individuals, with 176 passing GenomeStudio and PLINK quality control for downstream

analysis. Whole-genome sequencing was conducted on a representative sample from eight communities, adhering to ethical guidelines. Population structure analysis using PCA identified the closest comparative populations from India and globally. Imputation was applied to enhance the density of population-specific SNPs. Genes previously reported as under selection in desert biota were curated from literature, and allele frequency-based analyses including Fst, Tajima's D, and Population Branch Statistics (PBS) were conducted on our dataset.

Results and Conclusions: PCA analysis indicates that Thar populations are closely related to Indo-European groups, selected as a comparative population for further allele frequency-based analysis. This analysis also potential selection signatures in the form of suggestive SNPs, which overlap with genes previously reported to be adaptive, which shows may provide advantages for survival in the desert. Additional selection-signature methods are being applied to validate these genetic hotspots.

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Abstract ID: 83

Understanding South Asia's Population History Through Archaic Introgression

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Background/ Introduction: Human evolution spans over 7 million years, marked by milestones like bipedalism, tool use, and social development. Fossil and genetic data suggest that anatomically modern humans (AMHs) emerged in Africa around 200,000 years ago, migrating out between 50,000 and 100,000 years ago. During this period, archaic humans like Neanderthals inhabited parts of Eurasia, with their presence noted in fossil records as recently as 40,000 years ago, dating back as early as 400,000 years ago. This temporal and geographic overlap raises questions about interactions between AMHs and archaic humans. Such interactions left genetic imprints in present-day populations, and advances in ancient DNA (aDNA) sequencing now enable comprehensive studies of these interactions, deepening insights into human ancestry. Additionally, aDNA sequencing has advanced our understanding of human evolution with the discovery of previously unknown hominin species, Denisovans, a sister group to Neanderthals.

Objectives: This study examines patterns of archaic introgression within South Asian populations, focusing on Neanderthal and Denisovan genetic segments and their variations across linguistic and ancestral groups in the region.

Materials and Methods: Genomic sequences of archaic individuals were obtained from the Ensembl Genome Browser. Whole Genome Sequence data of >9500 individuals representing 83 South Asian populations was obtained through the GenomeIndia Project. Using allele counts, we detected archaic gene flow by identifying

mutations shared between AMHs and archaic humans, using 10-primate ancestral alleles as a reference. SPrime was applied to identify introgressed segments and match them to Neanderthal and Denisovan genomes, using West and Central African genomes as outgroups.

Results and Conclusions: Differing levels of Neanderthal and Denisovan ancestry were observed across South Asian populations. Tibeto-Burman speakers showed higher Neanderthal admixture, while Austro-Asiatic and Dravidian speakers exhibited greater Denisovan ancestry. A population, with East African ancestry and residing in South Asia, exhibited minimal archaic admixture. This pattern aligns with expectations based on their ancestral lineage, indicating limited gene flow from archaic populations. Another Tibeto-Burman speaking western Himalayan population revealed two distinct waves of Denisovan introgression, suggesting multiple sources of archaic admixture in this region.

This study highlights the complex history of archaic gene flow in South Asia and its impact on population diversity. Variations in Neanderthal and Denisovan ancestry across linguistic groups underscore the role of geography and cultural factors in shaping early human migration and adaptation.

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Abstract ID: 906

Maternal genetic landscape of Koṅkaṇī population

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Around 2 million people speak Koṅkaṇī language in India. The Koṅkaṇī population resides along the Koṅkaṇ Malabar region on the west coast of India. This is a point where two of the world's largest language families meet - Indo European and Dravidian. To understand the maternal genetic landscape of Koṅkaṇī population, we have sequenced complete mitogenomes of 85 and the hypervariable region of 225 Koṅkaṇī individuals. Haplogroup frequency distribution suggests that the Sārasvata and non-Sārasvata Koṅkaṇī groups exhibit distinct lineages within the Koṅkaṇī population. This distinction corresponds with the oral history of the Sārasvata group being migrated from north India, presumably from the banks of river Sarasvati. The clustering pattern also corresponds with the traditional occupation placing Khārvi, a fishing community away from Kuḍubi, a traditionally involved in hunting. We also observe low diversity in groups within the Koṅkaṇī population suggesting bottleneck events with subclades dating back to the Late Pleistocene. Based on the distinction derived from the N macrohaplogroup lineages, we suggest that the maternal ancestry of the Koṅkaṇī population is shaped greatly by the demographic changes that happened during the recovery phase of the Last Glacial Maximum.

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ABSTRACTS

ISHG Young Scientist Session

YSA1

Epigenome-wide association study of childhood cognitive function identifies inflammation as a key differentially regulated pathway

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Introduction: Cognitive function refers to an individual's overall ability of perception, memory, processing, and reasoning. Children with higher cognitive ability often have greater educational and occupational attainment, better cognitive capabilities as adults, and a lower risk of cardiometabolic, psychiatric, and neurodegenerative diseases. Hence, identifying the early determinants of cognitive function can help reduce disease burden and raise human capital.

Cognitive function is a highly polygenic trait shaped by the environment. Despite identification of more than 200 cognition-associated genetic loci, the heritability is less than 30%. Early life environment, including nutrition, stress, and socio-economic status, also significantly influence a child's cognition. Environmental factors can exert their effects via modifiable epigenetic mechanisms like DNA methylation (DNAm). Earlier epigenome-wide association studies (EWAS) of cognitive function in European adults have identified 37 differentially methylated CpGs (DMCs) and shown that DNAm explained 35% of the variance, independent of genetics. However, adult cognition-associated DMCs were poorly replicated in a European cord blood EWAS at birth, and failed to identify any novel loci, suggesting a complex interplay of genetics and environment during childhood and adulthood. Unfortunately, these have never been studied in children and hence remain an area of interest. This is especially important for Indians as they have a different early-life phenotype, a diverse nutritional environment, and possible genetic differences than Europeans. We hypothesize that gene-environment interactions during childhood influence cognitive function via DNAm, and the effects persist into adulthood.

Methodology: We utilized two longitudinal birth cohorts, SARAS ['excellent'] KIDS and Mysore Parthenon Cohort (MPC). The discovery EWAS and replication analyses were conducted in SARAS KIDS Cohort (n=1103), which included children born to mothers with and without pre-and periconceptional food-based micronutrient intervention. The children's anthropometric, cardiovascular, and cognitive outcomes were measured at the age of 5-7 years. The longitudinal analysis was conducted in the Mysore Parthenon Cohort (MPC; n=509), established to investigate the long-term effects of gestational diabetes and micronutrient status during pregnancy on the cardiovascular and cognitive outcomes of the offspring. The children were followed up at the ages 5, 9.5, 13.5, and 21 years and above phenotype parameters were measured serially.

Cognitive function was measured using batteries of age-appropriate and culturally validated tests. In SARAS KIDS (at 5-7 years) and MPC (at 9.5 and 13.5 years), a modified Kauffman battery was administered, consisting of seven tests. In MPC, at 21 years, the Wechsler Adult Intelligence Scale (WAIS-IV India), containing eight tests, was used. A general intelligence score ('g') representing the overall cognitive ability was calculated using the first principal component derived from the cognitive batteries.

DNAm on genomic DNA isolated from blood was measured using the Illumina EPIC V.1.0 array in 1) SARAS KIDS at 5-7 years, 2) MPC at 5 years, and 3) MPC at 21 years. The raw data were pre-processed using stringent QC followed by functional normalization to generate three high-quality DNAm datasets. Discovery EWAS of 'g' was conducted in SARAS KIDS (n=681) using robust linear regression while adjusting for the child's age, sex, and methylation-derived cell types. Replication of the top signals was done in additional 422 children from SARAS KIDS using pyrosequencing.

Results: We identified seven DMCs (FDR < 0.05), including four positively (standardized beta (β) range = 0.19 – 0.23) and three negatively (β range = -0.23 – -0.18) associated with 'g'. The positively associated DMCs annotated to NLRC5, SOCS3, and IFITM1 genes. NLRC5 encodes a transactivator of MHC-I expression while SOCS3 expresses a master regulator of cytokine signalling, and IFITM1 encodes a surface protein that is involved in anti-viral response. The negatively associated DMCs mapped to SCN3B (encodes a subunit of Sodium Voltage-Gated Channel) and FGF23 (a hormone involved in phosphate metabolism). The top two DMCs, in NLRC5 and SOCS3, were replicated in independent samples from SARAS KIDS (β = 0.11 – 0.12, P = 6.2 x 10-3 – 8.4 x 10-3).

In the EWAS catalog, the DMCs were reportedly associated with inflammatory markers (like C- Reactive Protein) and autoimmune diseases (like Chron's disease). Enrichment analyses identified genesets related to immune signalling pathways like JAK-STAT, PI3K/AKT, and IL-4 and IL-13 signalling. Similar observations were made using a poly-epigenetic score of chronic inflammation in SARAS KIDS, which was negatively associated with 'g', adding to the evidence of a link between inflammation and childhood cognition.

On longitudinal analysis in the MPC, 'g' measured at 9.5, 13.5, and 21 years were strongly correlated (r = 0.76-0.82), suggesting that cognitive function is stable from childhood to young adulthood. Moderate to high correlations were also observed between DNAm measured at 5 and 21 years at the above DMCs (Pearson r = 0.45-0.65). DNAm in the NLRC5, SOCS3, and IFITM1 continued to be significantly associated with 'g' at all three ages (β = 0.13 – 0.28, P = 4.7 x 10-3 – 4.6 x 10-6) and cross-sectionally at 21 years (β = 0.13 – 0.21, P = 0.018 – 3.9 x 10-5), showing the stability of the DMC-'g' associations from childhood to adulthood.

Conclusion: We report the first EWAS of childhood cognitive function, and the identified genes and pathways were predominantly related to inflammatory signalling. We also demonstrate a novel association between a poly-epigenetic score of chronic inflammation and childhood cognition. Our findings highlight

the potential interaction between inflammation and cognitive function as early as childhood, with the effects extending into adulthood.

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YSA2

Unraveling the Role of the lncRNA RP11-215G15.5 in Psoriasis Pathogenesis

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Introduction: Psoriasis (OMIM: 177900) is a chronic inflammatory dermatosis and affects approximately 0.44% to 2.8% of the Indian Population. While the genetic component of the disease is well understood, various epigenetic aspects are not explored well. Long non-coding RNAs (lncRNAs) are translationally inactive transcripts having more than 200 nucleotides in size. The key regulatory function of lncRNAs encompasses the sponging of microRNAs (miRNAs), which in turn regulates the target gene expressions. Thus, disease-specific differentially expressed lncRNAs can disrupt the 'lncRNA-miRNA-target gene'network homeostasis. In this work, we investigated the differentially expressed lncRNAs in psoriasis and the functional attributes of any majorly contributing lncRNA(s) through the lncRNA-mediated competitive endogenous RNA (ceRNA) network.

Objectives: We have undertaken the following objectives for our research work: • To identify the differentially expressed lncRNAs in the lesional psoriatic skin compared to the adjacent normal skin. • To identify the epidermal keratinocyte-specific lncRNAs to investigate their functions in disease-mimicking keratinocyte cell model system. • To explore the 'lncRNA-miRNA-target gene' network and the regulatory role of lncRNA(s) in psoriasis pathogenesis.

Methodologies: Selection of patients and sample collection: Psoriasis patients from Eastern India were recruited with their informed written consent. The clinical diagnosis of the disease was independently corroborated by at least two dermatologists and verified by histopathological examinations. We recruited 24 psoriasis vulgaris patients, and the lesional, along with adjacent normal skin tissues, were collected using a 4mm punch-hole biopsy for histopathological confirmation and Total RNA isolation. Total RNA sequencing and small RNA sequencing: Skin tissues collected in RNA Later were snap-frozen in liquid nitrogen and ground using mortar and pestle followed by total RNA isolation using the mirVana™ miRNA Isolation Kit (Thermo Fisher, USA) following the manufacturer's protocol. Isolated RNA was quantified and checked for quality. Then the total RNA sequencing and small RNA sequencing libraries were prepared followed by sequencing using the Illumina HiSeq X platform. Sequencing reads were QC checked followed by adapter trimming. QC passed reads were aligned to the human reference genome (GRCh37/hg19). Transcript assembly of the mapped reads was carried out using StringTie (version v1.3.6) in reference-guided mode, using the genome annotation file (gtf) downloaded from GENCODE (version GRCh37/hg19). The raw counts for all annotated genes obtained at the gene level were used for differential expression analysis using edgeR.

Bioinformatical analysis: We used four online repositories ('LNCRNASNPv3', 'miRDB', 'miRTARbase', and 'TargetScan 8.0') to construct the subsequent lncRNA-miRNA-target gene' pairs. Gene ontology has been performed using EnrichR to identify the probable biological roles.

Cell culture and the disease model generation: Normal primary human epidermal keratinocytes (NHEKs) were isolated from the healthy individuals and cultured in Keratinocyte Growth Medium 2 (KGM2) with the necessary supplements. Ker-CT (h-TERT modified human primary keratinocyte cell line) was cultured in Epilife medium along with HKGS. Lenti-X, a human embryonic kidney cell line, was cultured in a DMEM high glucose medium with 10% FBS and an antibiotic cocktail. All the cells were maintained at 37°c and 5% CO2. A combination of IL1a and IL17a [10ng/µl] along with IL6 and TNFα [5ng/µl], was used for treatment in keratinocytes to generate a 'psoriasis-like' (disease-induced) condition.

RNA isolation and qRT-PCR analysis: Total RNA isolation from the cell lines was performed using mirVana™ miRNA Isolation Kit (Thermo Fisher, USA) following the manufacturer's protocol. After cDNA synthesis, the expressions of the desired lncRNAs, miRNAs, and the target genes were checked using quantitative real-time PCR (qRT-PCR).

Luciferase reporter assay: Predicted miRNA target regions of the lncRNA were cloned in the pmirGLO Dual-Luciferase miRNA target expression vector. Respective partner miRNAs were transiently overexpressed using the pRNAU6 siRNA expression vector. Overexpression of the respective miRNAs was confirmed with respect to the empty vector controls. Luciferase reporter assay was performed to validate the predicted 'lncRNA-miRNA' interactions.

Stable knockdown of lncRNA: The short hairpin RNA (shRNA) sequence specific to the lncRNA was cloned in to the PLKO1 shRNA expression vector. Following a lentiviralmediated shRNA targeting, the respective lncRNA was stably downregulated in the Ker-CT cell line.

Biochemical assay: To check the cell proliferation rate upon the lncRNA knockdown, the WST1 assay (a colorimetric assay) and the colony formation assay (stained cell colonies with crystal violet) were performed. To measure the rate of cell migration, the wound-healing assay was performed.

Statistical tests: All the statistical tests have been performed using the GraphPad Prism 7.0 software. A 5% level of significance was used for all the tests performed.

Results: For 24 paired tissue samples, we performed Genome-wide total RNA sequencing and small RNA sequencing and identified 120 differentially expressed lncRNAs in the lesional psoriatic skin. The interacting miRNAs, of these differentially expressed lncRNAs were identified and 78% of them were found to be significantly deregulated in psoriasis and inversely correlated with respect to the lncRNA expression. This observation indicates that a substantial portion of differentially expressed miRNAs may be deregulated through the lncRNA axis. Similarly, we also identified the differentially expressed and inversely correlated target genes for these miRNAs. Gene ontology analysis with those genes showed significantly enriched

biological processes like cell population proliferation, migration, apoptosis, inflammation and cell differentiation, which are the hallmark pathways for psoriasis pathogenesis. We identified the overrepresented lncRNAs considering all the significant biological processes and found lncRNA RP11-215G15.5 as one of the central nodes regulating majority of these biological processes. RP11-215G15.5 was downregulated in the lesional psoriatic skin tissue as well as in the disease mimicking keratinocyte cell model system. Knockdown of the lncRNA in the keratinocyte cell model system caused its hyperproliferation and migration.

Next, we sought to explore the 'lncRNA-miRNA-target gene' axis through the functional characterization of RP11-215G15.5. We identified the interacting miRNA partners and through qPCR validation, we found miR-18a-5p, miR-142-5p, and miR-7-5p were significantly overexpressed upon RP11-215G15.5 knockdown and in the disease-mimicking Ker-CT. The luciferase reporter assay corroborated the qPCR data and showed a significant interaction of these miRNAs with RP11-215G15.5. We identified the target genes associated with these three miRNAs and also modelled common target genes by using different combinations of these miRNAs. We found three such genes, were getting targeted by the three miRNAs altogether. Among the several genes three genes, PRDM8 (3'UTR targeted by miR-142-5p), CADM3 (3'UTR targeted by miR-7-5p) and SOX6 (3'UTR targeted by miR-18a-5p) were previously reported to be involved in cell proliferation and migration. Expectedly, these genes showed reduced expression upon RP11-215G15.5 knockdown and corresponding miRNA overexpression. This observation underscores the pivotal role of RP11-215G15.5 in psoriasis pathogenesis as it regulates the disease pathogenesis by affecting the 'lncRNA-miRNA-target gene' axis. In a previous study, we explored the synergistic effect of miRNAs on corresponding target genes and found optimal binding distance enhances the synergistic effect. As we previously modelled common target genes with combinations of miRNAs, we are aiming to explore the synergistic effect of these miRNAs on these target genes. It will enlighten the lncRNA-miRNA-target gene axis path as well as the lncRNA regulation module in psoriasis pathogenesis.

Conclusion(s): RP11-215G15.5 remains downregulated in psoriasis and it enhances keratinocyte proliferation and migration by affecting the underlying 'lncRNA-miRNA-target gene' axis.

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YSA3

CNTNAP2 gene deletion confers risk to schizophrenia through dendritic spine abnormalities evaluated using hiPSCs derived neurons

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Background: Schizophrenia is a chronic neuropsychiatric disorder characterised by positive symptoms of hallucination, delusion, disorganised thinking, asociality, lack of motivation etc. Onset of symptoms typically occur at late adolescent or early adulthood with ~1.1% of world population affected. There are multitude of factors contributing to the onset of symptoms which includes neurotransmitter imbalances, neurodevelopmental defects, genetic predisposition, substance abuse, mitochondrial impairment, environmental cues etc. Of all these factors the interplay between genetic and environmental factors plays crucial role in the onset of schizophrenia. With heritability as high as 80%, it is the most heritable disorder among all other mental illnesses. Multiple Genome Wide Studies have identified various copy number variations (CNV), Single nucleotide polymorphisms, associated with Schizophrenia, however, their pathophysiology is still elusive. Furthermore, the lack of suitable model systems makes it challenging to investigate how neuronal morphology and function are affected. One such CNVs associated with schizophrenia is CNTNAP2. While CNTNAP2's connection to autism spectrum disorder is well-established and extensively researched, its involvement in schizophrenia stays unexplored and lacks comprehensive investigation. Objective Our aim is to study morphological and functional changes in human induced Pluripotent Stem Cells (hiPSCs) derived neurons of schizophrenic patient with CNTNAP2 deletion.

Methods: Familial cases of schizophrenia diagnosed by experts according to the Diagnostic and Statistical Manual of Mental Disorders fifth edition (DSM 5), at the psychiatry OPD of KEM Hospital, Parel, Mumbai, India, visiting Genetic Clinic run by ICMR-National Institute for Research in Reproductive and Child Health, Parel , Mumbai , for genetic diagnosis and counselling services were recruited for this study. As a part of the larger effort to understand the genetic etiology of schizophrenia, whole exome sequencing (WES) and genomic microarray was conducted of the recruited patients, after taking detailed written informed consent. In the process, we found a family where two sisters affected with schizophrenia was having copy number loss in 7q35-36.1 region encompassing CNTNAP2 gene. We generated hiPSCs of these two sisters along with two age and sex matched controls from peripheral blood mononuclear cells (PBMCs) using Cytotune Sendai virus kit (Invitrogen). After characterisation,

cortical neurons were differentiated from hiPSCs. For maturation differentiated neurons maintained in culture for 120 days and was characterised for mature neurons marker. The matured neurons were then transfected with eGFP and morphometry analysis was performed. For functional analysis of matured neurons, we used Fluo-4 Calcium imaging kit. Live cell imaging was conducted with excitation wavelength of 485 nm. Images were taken at the interval of 0.5 sec for 2 min. Initially basal activity was recorded thereafter without disturbing the setup, stimuli (10mM KCl) was applied, and activity was recorded. Image J was used to generate profile of fluorescent intensity for each cell overtime.

Results: Microarray analysis shown CNVs at locus 7q35-36.1 in both the affected siblings. This locus comprises CNTNAP2 gene which is one of the largest gene (2.3mb) in the genome out of which 2.07 mb was deleted in both the sisters. CNTNAP2 protein participates in the architecture of myelinated axons, spine development, dendritic arborization, and axon guidance. It is essential for clustering potassium voltage-gated channels. Complete loss of Cntnap2 results in a decrease of GABAergic neurons, impaired neuronal synchronization, disruptions in cortical projection, and issues with neuron migration in mice. The deletion was confirmed through breakpoint analysis, for which we designed a set of four primers: two for the deleted exons (exon 5 and exon 10) and two sets for intact exons (exon 1and exon 21). hiPSCs of these two siblings were generated along with two age and sex matched control which was characterised for stemness (OCT4, SOX2, NANOG, SSEA4) using Immuno Fluorescence (IF). The characterised hiPSCs was subjected for differentiation using dual SMAD inhibition protocol. Initially developed neural stem cell was characterised for marker (NESTIN and SOX2) using IF, it was differentiated into neurons. The differentiated neurons were characterised for immature neuronal marker (DCX, NeuroD1, β III Tubulin), Glial cells (GFAP), mature neuronal marker (PSD95, NeuN, MAP2) using IF. Neuronal lineages were identified using immature neuronal markers (DCX, NeuroD1, and βIII tubulin). The presence of mature neuronal markers (PSD95, NeuN, and MAP2) in cortical neurons indicated that most neurons matured within 4 months of continuous culture. Morphometric analysis revealed decreased dendritic arborization, dendritic length, spine density, soma area, and volume in the schizophrenia (SCZ) sisters compared to controls. Conversely, axon length increased in the sisters. A significant reduction in overall spine numbers was observed in patients compared to controls. Analysis of different spine types showed no significant decrease in stubby spines, but a decrease in long thin and mushroom types. Interestingly, the percentage of filopodia increased in the patients' neurons. This increase might be due to a lack of transformation to long thin and mushroom types, which is typically normal in adult brains, although other molecular factors may be involved and require further investigation. The observed spine variability could explain disruptions in adaptive experiences leading to unsettling social behavior. Calculated fluorescence intensity obtained from live cell imaging was plotted using GraphPad prism and it was observed that there was delayed firing in patient neurons as compared to control (for control firing maxima was at 2 sec while for patient it was 4 sec). At the same

time, the neurons firing spike intensity was 1.2 in control while for patient neurons it was 2.5. This neuron's firing spike is directly proportional to the calculated fluorescent intensity.

Conclusions: We have identified hemizygous deletion in CNTNAP2 gene in two siblings affected with schizophrenia using microarray. CNTNAP2 engages in dendritic arborisation axon guidance and spine development. We generated iPSCs derived cortical neurons of these two siblings and upon analysis we found that dendritic branching, dendritic length and spine density were decreased in patient's neurons whereas neurite length was increased in patient's neuron. The detailed analysis of spines showed that the percentage contribution of filopodia spines were increased in patients' neurons. Also, through calcium imaging analysis we observed defects in neurotransmission in patient's neurons.

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YSA4

Genetic modulation of systemic and neuro-inflammation in Progressive Supranuclear Palsy

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Background: Progressive Supranuclear Palsy (PSP) is the second most common neurodegenerative Parkinsonian disorder with a prevalence rate of 5-6/100000. The cardinal features of PSP are vertical gaze palsy, recurrent backward falls and mild cognitive impairment. Pathologically, PSP is defined by abnormal aggregation of tau protein in the basal ganglia, brain-stem and diencephalon mainly in the form of globose-type neurofibrillary tangles. Genes play an important role in the risk of PSP, however, failed to dissect its precise etiopathological basis. Recent research highlights immune dysfunction and neuroinflammation in tauopathies and neurodegenerative diseases like Alzheimer's disease (AD) and Parkinson's Disease (PD). Microglia activation and neuroinflammation in PSP are linked to tau aggregation and disease progression, but the underlying mechanisms for microglial activation remain unclear.

Cytokines, the key mediators of inflammation, are predominantly produced by T lymphocytes, which include subsets like Th1, Th2, Th17, and Treg cells. Among these lineages, the encephalitogenic role and brain infiltrating potential of Th17 cells and their subsequent link with neuroinflammation have received wide recognition in diseases like multiple sclerosis and AD. Besides, Th17 cells have been shown to cause neuronal cell death in both the mouse model as well as iPSC-based models of PD.

However, the role of peripheral adaptive immunity mediated by Th17 cells in PSP and its impact on neuroinflammation remains unexplored.

Objectives: i) To understand the impact of the Th17-pathway on systemic inflammation and neuroinflammation in PSP by looking at its initiators, master regulator transcription factors and effectors in lymphocytes, plasma, and cerebrospinal fluid. ii) To identify the signatures of neuroinflammation and neurodegeneration in PSP.

Methodology: A prospective case-control study was carried out in the Human Genetics and Department of Neurology of NIMHANS. PSP patients (n=60) fulfilling the Movement Disorder Society criteria for PSP and age-gender-matched healthy subjects (n=60) were recruited from the community. Ethical approval was obtained from the Institutional Ethics Committee (No.NIMHANS/DO/16th Ethics sub-committee BS & NS meeting/2022 dated 17.01.2022) and written informed consent was obtained from each participant.

The lymphocyte expression of Th17 pathway-related genes (*Tgfb*, *Il6*, *Stat3*, *Rorc*, *Il17*, *Il22*, *Il1b*, *Il23a* and *Tnfα*) were quantified using TaqMan real-time PCR assays on a Quant Studio 6 PCR system (Applied Biosystems, CA, USA). Additionally, quantification of the inflammasome pathway genes (*Casp1*, *Nlrp3* and *Il18*) and Treg effector gene (*Il10*) were also performed using TaqMan probes on an automated droplet digital PCR in a QX200 AutoDG Droplet Digital PCR System (Bio-Rad, USA). Additionally, the Th17 pathway-related cytokines such as IL-6, IL-1β, IL-4, IL-10, IL-17A, IL-17F, IL-21, IL-22, IL-23, IL-25, IL-31, IL-33, IFN-γ, sCD40L, and TNF-α were measured using a multiplex suspension array on the Bio-Plex 200 platform (Bio-Rad, Hercules, CA, USA) with the Bio-Plex Pro™ Human Th17 Cytokine Panel 15-Plex (Bio-Rad, USA). The positron emission tomography (PET) scan, followed by the acquisition of a routine magnetic resonance imaging (MRI) sequence was obtained using [^{11}C] PBR28 ligand on SIEMENS Biograph mMR (Erlangen, Germany). The ligand specificity was checked by genotyping rs6971 SNP of the translocator protein (*TSPO*) gene using TaqMan biallelic discrimination assay. Only a high or mixed-affinity binder was considered for PET/MRI imaging.

Data analysis for the components of the Th17 pathway i.e., gene expression, plasma and cerebrospinal fluid (CSF) cytokine levels and PET/MRI data was performed using the SPSS-25.0. The linearity of the data was checked by the Kolmogorov-Smirnov test. As our data was not normally distributed, we opted for the nonparametric test. Mann-Whitney U test and Spearman's rank correlation test were used for analysis.

Results: The mean age of the patients was 60.95±7.18 years, while that of the healthy controls was 55.57±9.26 years. The mean age of onset of PSP was 58.33±6.83 years. The gene expression levels of *Il6* (case/control= 1.51 ±3.33/0.25±1.12, ***p*=0.026)**, *Il1b* (case/control= 1.73±5.19/0.29±12.02,

***p*=0.037)**, *Casp1* (case/control= 16.33±41.56/8.12±11.17, ***p*=0.038**) and *Il18* (case/control= 0.95±2.06/0.43±0.6, ***p*<0.001)** were upregulated in PSP as compared to healthy controls while those of *Tgfb*, *Stat3*, *Rorc*, *Il17*, *Il22*, *Il23a*, *Tnfα* and *Nlrp3* were comparable between the two groups. Interestingly, strong positive correlations in gene levels were observed between *Il6* and *Tnfα* (c=0.70, ***p*<0.001**); *Il6* and *Il22* (c=0.57, ***p*<0.001**); *Il1b* and *Tnfα* (c=0.56, ***p*<0.001**); *Il1b* and *Il22* (c=0.60, ***p*=0.001**). We observed a higher plasma level of IL-1B (case/control= 1.32±1.53/0.93±0.65, ***p*=0.041**), IL-6 (case/control= 7.9±9.64/5.4±10.52, ***p*=0.014**) and IL-17A (case/control= 4.34±6.63/0.9±6.23, ***p*=0.031**). Additionally, a decreased level of IL-25 (case/control= 1.68±3.33/2.73±5.27, ***p*=0.043**) and IL-33 (case/control= 62.22±81.67/104.43±103.09, ***p*=0.015**) were observed in the plasma of patients with PSP as compared to healthy controls. Only the significantly dysregulated genes and cytokines were included in the analysis to evaluate their correlation with CSF cytokine levels. Additionally, the plasma level of IL-1β was significantly correlated with the CSF level of IL-1β (c=0.60, ***p*=0.004**), Additionally, in the PSP-RS group, the UPDRS-III score showed a positive correlation with IL-17F (c=0.61, ***p*=0.002**) and IL-23 (c=0.60, ***p*=0.017**). Similarly, the PSPRS score was significantly correlated with plasma IL-23 levels (c=0.67, ***p*=0.007**).

We checked the rs6971 (TSPO) polymorphism in PSP patients (n=13) and found that all patients exhibited either high or low binding affinity to the [^{11}C] PBR28 ligand. Six different regions of the brain including the pons, midbrain, striatum, thalamus, pallidum and substantia nigra were selected for the analysis. Additionally, the volume of the pons and midbrain was measured. However, we did not observe any correlation between PET/MRI parameters and gene expression, or the plasma and CSF levels of cytokines.

Conclusion: Elevated expression of *Il6*, *Il1b*, *Casp1* and *Il18* genes in the peripheral blood suggests the involvement of the Th17 and inflammasome pathway in the pathogenesis of PSP. These findings were further supported by the higher levels of IL-1B, IL-6, and IL-17A, along with decreased levels of IL-25 and IL-33 in the plasma. Additionally, the strong correlations between gene expression and plasma cytokine levels with CSF cytokines highlight the interplay between systemic and neuroinflammation, which may provide important insights into the immunopathogenic basis of PSP.

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YSA5

Developing a patient-friendly diagnostic method for mitochondrial disorders: correlation of genetic variants with multicomplex deficiencies

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Background: Mitochondrial disorders (MDs) are a wide group of disorders with clinical variability and variable age of onset. The clinical presentations primarily affect those organs and tissues that have a high energy requirement, however in the majority of the cases, MD's are multisystemic. Although, wide spectrum of clinical manifestations and relatively poor genotypephenotype correlation, a combinatorial diagnostic approach is required based on laboratory and metabolite analysis, respiratory chain enzyme assays, histopathology and genetic analysis. The respiratory chain enzyme analysis on skeletal muscle biopsy has remained the gold standard for diagnosis. However, knowing the invasive nature and the expense of a muscle biopsy, it is practically difficult to obtain it from children and infants for routine diagnostic purpose. Therefore, there has been an inclination towards alternative approaches for the diagnosis. Apart from muscle biopsy, other cell-types such as lymphocytes and cultured skin fibroblasts have also been used for the diagnostic evaluation of patients, however they are not representative of actual muscle enzyme defects, depending on variable culture conditions.

Objectives: The objective of the study is to develop a patient friendly diagnostic tool for mitochondrial disorders using buccal swabs along with a comparative study between muscle and buccal samples. Detailed genetic analysis using whole exome sequencing (WES) and the analysis of the diagnostic potential of buccal swabs as a potential non-invasive diagnostic tool for MD's. Material & methods: Buccal swabs were collected in buccal extraction media from 43 pediatric patients suspected with MD. Age matched control participants (n=37) were recruited for the study with no history of neurological or metabolic disorders in family and the patient. Biochemical analysis of mitochondrial complex enzymes (Complex I, II, III, IV and CS) were carried out using spectrophotometry. Further, genetic analysis was carried out using long-range PCR for the detection of large mitochondrial deletions and whole exome sequencing for specific detection of missense variants. Sanger sequencing was carried out to validate the variants detected using WES. Receiver operating characteristic (ROC) curve were calculated to analyze the diagnostic potential of buccal swabs as a non-invasive diagnostic tool for mitochondrial disorders.

Result: In our cohort of pediatric patients, using buccal swab as a non-invasive diagnostic tool, a significant decrease in enzyme activity of Complex I (p-value 0.0270), Complex III (p-value 0.0081) and complex IV (p-value 0.0031) was observed. Buccal swab mitochondrial enzyme analysis had shown deficiency in 35 patients with 13 patients (30.2%) showing isolated complex deficiency and 22 patients (51.1%) harboring multi-complex deficiency. A comparative analysis between muscle biopsy and buccal swab samples (n=4) have indicated a correlation in mitochondrial enzyme deficiencies. The diagnostic

value of buccal swabs when analyzed was observed to be within the potentially acceptable range with an AUC above 0.7 for mitochondrial complex I, III & IV.

Besides, development of a cost-effective diagnostic tool, genetic analysis has revealed the presence of genetic variants in 25 mitochondrial genes some of which being MTND6, MTATP6, CoQ8A, ECHS1, SURF1 etc encoded either by mtDNA and nDNA. Presence of genetic variants in 13 nonmitochondrial genes causing secondary enzyme deficiencies in mitochondrial enzymes have also been identified. Although, these genes do not directly interact or affect mitochondrial structure or functions, they have been observed to affect mitochondrial oxidative phosphorylation (OXPHOS) complexes. Long-range PCR has shown multiple large deletions in mtDNA in 22 patients.

A correlation between the genetic variants and the mitochondrial complex deficiencies had shown that, out of 13 patients with isolated complex deficiency, 10 patients had shown concordance with the genetic variants they harbored. TTC19 encodes for protein that is associated with complex III, those patients with mutations in TTC19 had shown a deficiency in complex III. Likewise, ECHS1 and DGUOK are part of complex I and IV respectively, patients with mutations in these two genes were also found to be deficient with the corresponding complexes. Eleven patients out of 22 harboring multi-complex deficiencies had shown concordance with genetic variants (e.g cases of MRPS16 had shown complex I & IV deficiency, NDUFS1 & NFUFS8 had shown Complex I & III deficiency and MT-CYB had shown complex I, III & IV deficiency). Eleven patients had shown multi-complex as well as isolated complex deficiencies with genetic variants in non-mitochondrial genes (such as SQSTM1, PIGG, KCTD7, SCN3A, DOCK3 etc) indirectly affecting mitochondrial function.

Therefore, in order to assess the diagnostic potential of buccal swabs as a non-invasive tool, the sensitivity was analyzed by comparing it with genetic investigations, was observed to be 78% which makes it a considerable non-invasive diagnostic tool for pediatric mitochondrial cases.

Conclusion: Our study demonstrates the use of buccal swabs as a non-invasive and cost effective tool for the preliminary diagnosis of mitochondrial complex I, III and IV. Although, smaller sample size is one of the limitations of our study, a correlation was observed between muscle and buccal samples of the patients. The diagnostic value of buccal swabs has also been analyzed to be within the potentially acceptable range with an AUC above 0.7. A sensitivity of 78% has been observed, hence, buccal swab approach can be used as a preliminary diagnostic technique before prior planning of the invasive and expensive procedure of muscle biopsy and also making it suitable for an effective diagnosis in a resource-limited setting for those who have remained undiagnosed till now.

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YSA6

Transgenerational effect of maternal stress on the behavioral outcomes of children: the role of maternal immune activation and epigenetic modulation

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Introduction: Maternal stress during pregnancy has gained significant attention due to its potential long-term effects on offspring's health. The second trimester is a critical period for fetal development, making the fetus particularly vulnerable to prenatal stressors. Maternal psychological stress activates physiological stress responses, such as the hypothalamic-pituitary-adrenal (HPA) axis, which can affect fetal development and lead to brain and behavioural changes. These stress-induced alterations may become embedded at the epigenomic level. DNA methylation, an essential epigenetic mechanism, regulates gene expression and is sensitive to early-life stress. Changes in DNA methylation during critical developmental windows, such as infancy, may influence childhood behaviour, cognitive function, and increase susceptibility to mental health issues. This study investigates maternal stress during pregnancy and its impact on maternal immune activation (MIA) and DNA methylation in children at 1, 2, and 5 years old.

Objectives: 1. To investigate the impact of maternal psychological stress on MIA and maternal physiological stress markers., 2. To examine the associations between maternal stress during the second trimester and DNA methylation changes in the offspring at first, second and five years of age, and 3. To understand the moderating role of child DNA methylation in mediating maternal stress-induced behavioural abnormalities in the child.

Methods: A total of 157 mother-child dyads were recruited for this study, part of the longitudinal Bangalore Child Health and Development Study (BCHADs). Ethical clearance was obtained from NIMHANS (IEC/2015, 02.07.2015) and University of Liverpool (RETH001024, 01.03.2016).

Maternal fasting blood samples were collected in the second trimester to measure cortisol levels, and maternal depression was assessed using the Edinburgh Postnatal Depression Scale (EPDS). Follow-ups were conducted at 1, 2, and 5 years postpartum, with child salivary DNA samples collected at each visit. Child behavior was assessed using the Brief Infant-Toddler Social and Emotional Assessment (BITSEA) at 1 and 2 years.

Maternal plasma cortisol was measured commercially available ELISA kit, and pro-inflammatory cytokines (IL-1β, IL-6, IL-8, IL-17A, TNF-α) were quantified using a multiplex suspension array in a Bioplex 200 platform.

The promoter DNA methylation of *Il6, Il17A,* and *Nr3c1* genes in the child was assessed after bisulfite conversion and PCR amplification, followed by sequencing in the Pyromark Q48 autoprep platform.

Data was analyzed using IBM SPSS 21. Spearman's correlation and hierarchical multiple regression were used to examine the moderating role of child DNA methylation in the relationship between maternal stress and child behavior.

Results: Maternal depression at second trimester had a negative correlation with maternal IL-6 levels (ρ= -0.232, p=0.005).

At the 1-year time point: **Maternal cortisol level exhibited** significant positive correlations with *Il6* CpG1 (ρ= 0.240, p=0.044), *Il6* CpG2 (ρ=0.308, p=0.009), *Il17a* CpG4 (ρ=0.359, p=0.003), and *Nr3c1*CpG3 (ρ= 0.345, p=0.003). **Maternal IL-1β** had significant positive correlations with *Il6* CpG1 (ρ= 0.592, p= 0.026), *Il6* CpG2 (ρ==0.654, p= 0.011), and *Il6* CpG3 (ρ==0.628, p= 0.016). **Maternal IL-6** had a significant negative corelation with *Il17a* CpG3 (ρ=-0.303, p=0.011), *Nr3c1* CpG2 (ρ=0.225, p=0.049). **Maternal TNF-α** showed significant positive correlation with *Il17a* CpG4 (ρ=0.276, p=0.025).

At the two-year time point: **Maternal depression** scores showed significant positive correlation with *Il17a* CpG3 (ρ=0.173, p=0.037) and CpG4 (ρ=0.189, p=0.022). **Maternal cortisol** showed significant positive correlations with *Il6* CpG1 (ρ=0.165, p=0.044) and *Nr3c1* CpG1(ρ=0.174, p=0.033). **Maternal IL-6** showed significant negative correlations with *Il6* CpG1(ρ=-0.293, p=0.000) , *Il6* CpG 2 (ρ=-0.3740, p=0.000), *Il6* CpG 3 (ρ=-0.379, p=0.000), *Il17a* CpG2 (ρ=-0.177, p=0.027), *Il17a* CpG3 (;ρ=-0.211, p=0.008), *Il17a* CpG4 (ρ= -0.158, p=0.049), *Nr3c1* CpG1 (ρ=-0.239, p=0.003), *Nr3c1* CpG2 (ρ=-0.217, p=0.006), and *Nr3c1* CpG3 (ρ=-0.270, p=0.001). **Maternal TNF-α** showed significant positive correlation with *Il6*CpG1 (ρ=0.184, p=0.027), *Il6*CpG 2 (ρ=-0.169, p=0.042), *Il6*CpG 3 (ρ=0.213, p=0.010), *Il17a* CpG3 (ρ=0.257, p=0.002), *Nr3c1* CpG1 (ρ=0.257, p=0.002), *Nr3c1* CpG 2 (ρ= 0.209, p=0.011), and *Nr3c1* CpG3 (ρ=0.261, p=0.002). **Maternal IL-17a** showed significant negative correlation with *Il17a* CpG3 (ρ= -0.285, p=0.002) and *Il17a* CpG4 (ρ= -0.289, p=0.002).

At the 5- year time point: **Maternal depression** showed significant negative correlations with *IL17a* CpG3 (ρ= -0.273, p=0.001) and *IL17a* CpG 4 (ρ= -0.233, p=0.005) and *Nr3c1* CpG3 (ρ= -0.174, p=0.035). **Maternal cortisol** had a negative correlation with *Il6* CpG3 (ρ= -0.195, p=0.017). **Maternal IL-6** showed negative correlation with *Il6* CpG3 (ρ= -0.188, p=0.019) and positive correlation with *IL17a* CpG3 (ρ= 0.247, p=0.002), *IL17a* CpG4 (ρ=0.252, p=0.002), *NR3C1* CpG2 (ρ= 0.209, p= 0.009), *NR3C1* CpG3 (ρ=0.297, p=0.000). **Maternal TNF-a** showed significant negative correlation with child *IL17a* CpG1 (ρ= -0.304, p=0.000), *IL17a* CpG 2 (ρ= -0.208, p=0.001), *IL17a* CpG3 (ρ= -0.483, p=0.000), *IL17a* CpG 4 (ρ= -0.541, p=0.000) and *NR3C1* CpG 3 (ρ= -0.178, p=0.032).

A significant positive correlation was found between maternal depression scores and child externalizing scores at two years (ρ= 0.201, p=0.015) and maternal IL-8 showed a significant positive correlation with child internalizing behaviour at two years (ρ= 0.240, p= 0.022).

There were no significant findings indicating the role of child DNA methylation at 1 year and two years in moderating the effect of prenatal stress on child internalising and externalising behaviour. Existing literature on prenatal stress and child behaviour has yielded mixed results regarding the role of DNA methylation, with some studies suggesting a potential link while others find no significant moderation. This reflects the complex nature of gene-environment interaction and challenges in detecting such effects in early childhood.

Conclusion: At 1-year, maternal IL-6 negatively correlated with *Il17a* and *Nr3c1* CpG sites, suggesting maternal inflammation may dampen specific methylation patterns. At 2 years, IL-6 showed significant negative correlations with multiple CpG sites, including *Il6*, *Il17a*, and *Nr3c1*, indicating consistent suppression. By 5 years, maternal depression and IL-6 negatively correlated with child DNA methylation at *Il6* and *Il17a* CpG sites, highlighting a sustained effect. These findings emphasize the complex, time-dependent relationship between maternal inflammation and child DNA methylation. However, no consistent role of child DNA methylation in moderating prenatal stress effects on behaviour was found, reflecting the complexity of gene-environment interactions.

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YSA7

Mutations in HYPK cause a novel neurodevelopmental disorder by impairing neuronal proteostasis and autophagy

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Background: HYPK (Huntingtin yeast interacting Partner K) is a conserved proteostasis regulatory protein playing a pivotal role in non-canonical, NEDDylation-dependent autophagy of protein aggregates in neurons. Functioning as a crucial autophagy receptor, HYPK mediates the clearance of NEDDylated aggregates by interacting with the ubiquitin-like molecule NEDD8 via its ubiquitin-associated (UBA) domain. This interaction facilitates the recognition of poly-NEDDylated protein aggregates while the LC3-interacting region (LIR) of HYPK binds to LC3, triggering the autophagosome formation around these aggregates. Importantly, HYPK exhibits a molten globule-like conformation, allowing its flexible binding to NEDD8 and other associated proteins.

Besides its autophagy function, HYPK functions as a chaperone-like protein within the Nterminal acetyltransferase (NatA) complex, where it cooperates with NAA10 and NAA15 to regulate proper protein folding and post-translational modification. HYPK is essential for embryonic viability, as knockout mutations in mice lead to lethality. Predictive evidence suggests that mutations in HYPK could be associated with human diseases, potentially contributing to neurodegenerative processes. However, the precise mechanistic underpinnings of the HYPK mutations in the nervous system remain largely unknown.

Objectives: This study aims to investigate the effects of heterozygous missense mutations in HYPK on neurodevelopment and neurodegeneration. The specific objectives are: 1. Neurological phenotypic effects of HYPK mutations: to examine the spectrum of neurological abnormalities (e.g., CNS characteristics, motor deficits, cognitive impairments) and behavioral phenotypes in probands harboring heterozygous missense mutations in HYPK, with a focus on identifying novel neurodevelopmental syndromes. 2. Characterizing HYPK's role in neurons and neurodegeneration: to elucidate the cellular and molecular functions of HYPK within neurons, particularly in autophagy and proteostasis pathways, and to assess the potential link between HYPK mutations and neurodegenerative conditions. 3. Impact of HYPK mutations on cellular mechanisms: to investigate how specific HYPK mutations disrupt molecular processes such as autophagy, proteostasis, and stress responses, leading to cellular dysfunction and potentially contributing to neurodegeneration. 4. Developing a Drosophila model of HYPK-deficient neurodegeneration: to create transgenic Drosophila models with HYPK knockouts to study the physiological effects of HYPK loss in a model organism context and to uncover the disease mechanisms underlying HYPK-associated neurodegeneration.

Methods: To achieve the above objectives, we employed the following approaches: • Clinical diagnostics and genetic analysis: detailed neurological examinations and radiological scans (MRI), genetic counseling, and family history assessments, followed by exome sequencing of the probands and Sanger sequencing of HYPK of probands and their parents and siblings to identify mutation types in HYPK. • Cell and molecular biology techniques: neuronal cultures and SH-SY5Y cell lines were generated with stable expression of mutant HYPK variants (E8A, K24E, S97A) to analyze their aggregation propensity and interaction with wild-type HYPK, autophagy assays using immunofluorescence to detect LC3-positive autophagosomes and NEDD8-positive puncta, immunoblotting and advanced fluorescence microscopy to assess protein aggregation, autophagy markers, and senescence-related proteins (p16, p21), cell viability and senescence assays to measure cellular stress responses. • Biophysical and Bioinformatics analysis: protein-protein interactions through dynamic light scattering and co-immunoprecipitation assays, ab initio molecular dynamics simulations (MDS) to assess the stability of mutant HYPK proteins, focusing on changes in backbone root-mean-square deviation (RMSD) and residue fluctuation (RMSF) compared to wild-type HYPK, pathogenicity prediction tools and conservation analyses to evaluate the evolutionary conservation of the mutated residues and their functional relevance. • In vivo studies: A Drosophila model with HYPK knockout to study the protein's role in neurodevelopment. Analysis of the central nervous system of Drosophila larvae to evaluate the impact of HYPK knockout on neurodevelopmental processes (neurons and neuroblasts). • Statistical Analysis: Statistical tests, including t-tests and ANOVA, to determine the significance of observed phenotypic and molecular differences between wild-type and mutant HYPK.

Results: Clinical observations: hereditary or de novo heterozygous mutations in HYPK, specifically E8A, K24E, and S97A, were identified in three probands (aged 9 months, 1 year 6 months, and 13 years) presenting with a novel neurodevelopmental disorder. Radiological scans revealed significant thinning of the corpus callosum, along with diffuse cerebral atrophy, while other brain regions, including the cerebellum and hippocampi, appeared normal. Probands exhibit a spectrum of mildto-moderate clinical phenotypes that affect both motor and cognitive functions. The motor deficits include ataxia, unsteady gait, and difficulty with fine motor tasks. Additionally, probands experience spasticity, contributing to challenges in movement and posture. Cognitive impairments of probands are evident through developmental delays, including delayed speech, learning difficulties, and impaired social interactions. One proband also displays hypotonia and dysphagia.

Cellular, molecular, and model organism findings: Molecular dynamics simulations revealed that all HYPK mutants displayed reduced structural stability compared to the wild-type protein, as evidenced by higher backbone RMSD and increased residue fluctuations (RMSF) of residues near the mutant residues. This instability likely contributes to the propensity of these mutants to form aggregates. HYPK

mutants (E8A, K24E, S97A) formed aggregates in SH-SY5Y neuronal cells and sequestered wild-type HYPK. In primary neurons expressing these mutants, a marked reduction in autophagy was observed, characterized by decreased LC3-positive autophagosomes and impaired clearance of NEDD8-positive puncta. Additionally, mutant-expressing neurons exhibited signs of cellular senescence, as indicated by increased expression of p16 and p21. The Drosophila model with HYPK knockout exhibited severe developmental delays and larval lethality. Significant neurodevelopmental defects were also observed, including pronounced structural abnormalities in the central nervous system. The brain lobe size was substantially reduced in HYPK knockout larvae compared to the wild type, and the formation of neurons and neuroblasts was significantly reduced in HYPK knockout Drosophila.

Conclusion: Our findings suggest that heterozygous missense mutations in HYPK lead to novel neurodevelopmental disorders characterized by impaired autophagy and neuronal proteostasis. The aggregation-prone mutants of HYPK likely disrupt NEDDylation-dependent clearance of protein aggregates, leading to cellular dysfunction and neurodegeneration. The development of Drosophila models further elucidates the physiological consequences of HYPK deficiency and advances our understanding of its role in neurodegenerative diseases.

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YSA8

Gliadin-induced Pathogenic Differential Transcriptome Signature of Gut Mucosa in Celiac Disease and its Mitigation through a Gluten-free Dietary Component Using a Patient-derived Organoid Model

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Introduction: Celiac Disease (CD) is a complex autoimmune disease of small intestine triggered by the ingestion of gluten in genetically susceptible individuals. Genetics of CD could explain ~50% disease risk but rest of 50% missing heritability remains unanswered. The transcriptomics, proteomics, lipidomics and epigenenomics could be explored to address the missing heritability. The known antigen in CD is gluten which is present in wheat, rye and barley. Gluten is composed of gliadin and glutenin, where the gliadin peptides have been known to elicit immunogenicity in CD patients. Several immunogenic peptides already reported in CD are associated with severity of mucosal damage.

This alteration could modulate the transcriptomic signature of duodenal mucosa in CD patients which is still under investigation.

Currently, there is no treatment available for CD except intake of gluten-free diet (GFD) which is neither curative nor does it stop from relapsing CD and also adherence to GFD is often difficult. Frequent relapse and incidences of refractory CD necessitate to investigation of alternative precision medicine for CD.

Objectives: i) in *silico* evaluation of polymorphic sites of HLA-DQ and TCR (T-cell receptor) involved in differential binding affinity with gliadin antigen in CD, ii) Identification of small intestinal tissue specific gene expression pattern and differential expression in CD, and iii) Attempt to mitigate gliadin-induced tissue damage using enterolactone using a 3D patient-derived organoid model.

Rationale: Through extensive literature review, it was found that the *in vitro* cell line couldn't reproduce CD pathology completely while animal models pose challenges including ethics issues and altered genetic architecture. In the present work we investigated the molecular interaction of immunogenic gliadin with HLA DQ2/8 and T-cell receptor (TCR) and identified key immunodominant gliadin peptides. These immunodominant gliadin peptides through HLA-DQ2/8 could alter the downstream transcriptional regulation in duodenum. This led to the identification of differentially expressed genes in the small intestine that are majorly associated with mucosal biology. Thus, we investigated the mitigation of gliadin induced intestinal tissue damage through the effect of plant phyto-estrogen (lignan) derived enterolactone (ENL) in CD. CD associated gut microbes were found to convert the plant lignans to enterolignans. A patient-derived 3D organoid was established that replicates tissue micro-environment (epithelium, mesenchymal and immune cell) and disease pathology without changing the genetic architecture. The 3D patient-derived organoid model of duodenal mucosa was used to study the mitigation of gliadin-induced epithelial injury by the bioactive enterolactone (ENL).

Methodologies: We analyzed immunogenic gliadin peptides binding affinity with HLA-DQ2/8 and their specific TCR in CD through *in-silico* molecular docking. Immunodominant peptides could alter the downstream transcriptional regulation. The molecular interaction of HLA with gliadin and TCR leads to mucosal damage in CD. Duodenal biopsies of three CD patients and four non-CD controls were subjected to whole RNA sequencing and transcriptomic analysis identified differentially expressed genes (DEGs) and further validated them using qRT-PCR in an independent cohort of 23 CD and 26 non-CD controls. Finally, patient derived 3D organoid was established using duodenal biopsy and characterisation was done using microscopic and gene expression analysis. Effect of ENL with gliadin challenge was evaluated using total RNA expression (qRT-PCR), protein expression (immunocytochemistry and western blot), cytokine release by ELISA method and tissue permeability assays (transepithelial efflux resistance (TEER) and FITC dextran 4kDa flux).

Results: 33 mer gliadin peptide was found to be most immunogenic to elicit differential immunogenicity in CD patients with *HLA-DQ2.5* and T-cell receptor variable gene pair (TRAV4/TRBV20). Both transcriptome and replication study identified one novel gene CDH18 (log2FC=-4.65, *p*=1.18E-39) and one already reported immune gene *CXCL9* (log2FC=4.36, *p*=0.002) to be significantly differentially expressed in active CD patients as compared to non-CD controls. Transcriptome study on CD suggested that the differentially expressed genes (DEGs) such as *MKI67* (log2FC=1.9, 7.80E-07), *CCND1* (log2FC=1.03, 1.00E-03), *RSPO2* (log2FC=-2.2, 1.05E-07), *WNT3* (log2FC=-2.14, 4.12E-05) and *MYC* (log2FC=1.63, 1.62E-08), predominantly down-regulated, were enriched in tissue-specific functions such as membrane transport complexes (p=1.34x10^{-07}), and cell-cell adhesion via plasma membrane adhesion molecules (p=8.05x10^{-06}), all of which are crucial for mucosal function and barrier integrity. The up-regulated genes, including *TRAV12-3, CTD-2631K10.1, CTC-548H10.2, CCDC81, RP11-334A14.8,* and *CXCL9* were primarily involved in antigen processing and presentation (p=1.83x10^{-06}), beta-catenin degradation via the destruction complex (p=1.13x10^{-23}), Wnt signaling (p =1.13x10^{-23}), auto degradation of *CDH1* by the CDH1:APC/C complex (p=2.29x10^{-19}), and immune system processes (p=1.50x10^{-16}).

The 3D organoid demonstrated the gene expression pattern of stem cell marker (*LGR5),* transient amplifying cell *(EPHB2),* inflammation associated cytokines *(IL6/15),* cell proliferation marker *(MKI67)* and *Wnt signaling associated gene (BMPR1A* and *CCND1),* similar to the biopsy sample of active CD patients found in our previous transcriptome study. ENL was found to limit gliadin-induced epithelial damage in CD patients during gliadin challenge by upregulating *CDH1 and VIL1* and downregulating *LGR5, EPHB2, TNFa,* and *IL-15* (p<0.05). 0.5mg/ml of gliadin increased the membrane permeability by decreasing TEER and elevated FITC 4kDa, and 25µM of ENL was found to reduce the mucosal damage caused by gliadin. Reduced expression of *LGR5* and increased expression of *CDH1, EPHB2* along with decreased transcellular and paracellular membrane permeability and anti-inflammatory effect (downregulation of *IL-15 and TNFa)* was observed with the treatment of ENL.

Conclusion: Specific molecular interactions of HLADQ-gluten-TCR could be exploited for therapeutic purposes. *CDH18/CXCL9,* key pathways, and their PPI could be targeted for therapy. ENL is a metabolic product of specific gluten-free diet. It thus could be tested as a recommended disease management strategy to limit the gliadin-induced damage of duodenal mucosa of CD patients. Patient-derived 3D organoid could be a proof of principle to investigate autoimmune diseases such as CD.

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YSA9

Biallelic Variants in CCN2 Underlie an Autosomal Recessive Kyphomelic Dysplasia

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Introduction: Kyphomelic dysplasia is a rare heterogenous group of skeletal dysplasia, characterized by bowing of the limbs, severely affecting femora with distinct facial features. The term "kypho" is derived from ancient Greek word "kyphos," meaning "bent", while "melia" refers to "limb". The term 'kyphomelia' was first used to describe a skeletal dysplasia by MacLean in 1983. Despite its first description nearly four decades ago, the precise molecular basis of this condition remained elusive. Moreover, it was conventionally considered an autosomal recessive condition. Recently, heterozygous de novo variants in KIF5B have been found to be associated with kyphomelic dysplasia, while several bent bone dysplasias do not have a known genetic basis.

Cellular communication network factor 2 (CCN2) is a matricellular protein spanning 349 amino acids, with five exons. It belongs to cysteine rich CCN protein family crucial for proper skeletal growth and development. In vitro studies on CCN2 demonstrated that it promotes DNA synthesis in chondrocytes. Previous investigations on Ccn2 deficient mice showed broader vertebrae, shortened and kinked sterna, along with bending in the radius, ulna, tibia, and fibula. Additionally, they exhibit craniofacial abnormalities including a distorted ethmoid bone, a domed cranial vault, shortened mandibles and secondary cleft palate. Owing to the high conservation of fundamental signalling pathways and cellular processes involved in skeletal development from fish to humans, zebrafish serve as valuable models for studying skeletal disorders. The function of ccn2a in early skeletal development in zebrafish has not yet been studied.

Zebrafish serve as valuable models for studying skeletal disorders due to the high conservation of fundamental signaling pathways and cellular processes involved in skeletal development. However, the role of ccn2a in early skeletal development in zebrafish has not yet been studied. We studied three probands from two consanguineous families with bowed long bones, and identified biallelic CCN2 variants segregating recessively. Using zebrafish models, we examined the impact of CCN2 loss of function.

Objectives: The objective of the study was to evaluate multiple affected individuals with kyphomelic dysplasia and to investigate the consequences of CCN2 loss of function in vivo using zebrafish models.

Methods: Three individuals with kyphomelic dysplasia from two unrelated families of different ethnicities were recruited. Clinical, radiological, and medical history were documented, with written informed consent obtained from participants and their families. The study was approved by the institutional ethics committees.

Exome sequencing was performed for all the three affected individuals with kyphomelic dysplasia. Filtered variants were further analyzed using several in silico pathogenicity prediction tools to assess their potential impact. Conservation analysis was performed using the Clustal Omega tool. The allele frequency of the identified rare variant was estimated from gnomAD and our in-house data of 3188 exomes. Sanger sequencing was performed to validate and segregate identified candidate variants in the proband and their family members. The variants were described according to Human Genome Variation Society (HGVS) nomenclature, with NCBI reference sequences (NM_001901.4, NP_001892.2). Both variants were submitted to the Leiden Open Variation Database (LOVD) database (variant ID: 0000972076; 0000972075). Disulfide bridge between cysteine 148 and a nearby cysteine residue was visualized using PyMOL (protein modelling tool).

CRISPR/Cas9 mediated gene editing was used for generation of ccn2a F0 knockout in zebrafish. Alcian blue staining was performed. Benchling software was used to choose target region of ccn2a. Single guide RNAs (sgRNAs) were synthesized, and phenotypic characterization of the ccn2a F0 knockout zebrafish was performed. Quantitative real-time qPCR analysis was done for investigating gene expressions of ccn2a, rac1a, rhoAa, col2a1a, sp7, runx2a and gapdh using Ct method ($\Delta\Delta$Ct).

Results: We ascertained two unrelated consanguineous families with kyphomelic dysplasia. They had six affected offsprings and we performed a detailed clinical evaluation, skeletal survey, and exome sequencing in three probands. All the probands had short stature, facial dysmorphism, cleft palate, and micro-retrognathia. Radiographs revealed kyphomelic femora, bowing of long bones (tibia, fibula, radius and ulna), radial head dislocations, scoliosis, mild platyspondyly and irregularities of the knee epiphyses and metaphyses.

Their clinical and radiological manifestations were consistent with what were previously reported in kyphomelic dysplasia. However, the phenotypic manifestations in family 1 were milder compared to those in family 2.

In Family 1, a shared biallelic missense variant, c.443G>A; p.(Cys148Tyr), located in exon 3 of the CCN2, was identified through duo exome sequencing. The variants are present in heterozygous state in the parents and absent in the unaffected sibling. This variant is absent in gnomAD (V3.1.2) and our in-house data of 3188 exomes. In silico mutagenesis analysis revealed a disulfide bridge formed between cysteine residue at position 148 and the nearby cysteine at position 166, is disrupted due to the substitution. Multiple sequence analysis performed revealed conservation of the cysteine residue across several vertebrate species. In family 2, we identified a homozygous frameshift variant, c.779_786del; p.(Pro260LeufsTer7) that was absent from the public and in-house datasets, located in exon 5 of CCN2. Sanger sequencing confirmed the heterozygous status of the parents.

A significant decrease in ccn2a mRNA was observed in the crispants as compared to the NT and WT controls, thus confirming ccn2a editing. The ccn2a crispants showed abnormal body curvature and bent tail suggesting defects in early skeletal development. The ccn2a crispants showed substantial defects in cartilage formation in the craniofacial region as seen by Alcian blue staining at 6.5 days post-fertilization. They had underdeveloped ceratohyal arches, bent or missing ceratobranchial arches and misshapen Meckel cartilage. The levels of established skeletal marker genes such as col2a1a (chondrocyte marker), rac1a and rhoAa (palatogenesis markers), sp7 and runx2a (osteoblast markers) in ccn2a crispants showed a significant decline, as is expected from the phenotype. The crispants also showed poor survival beyond seven days post-fertilization and few survived to adulthood. These adult crispants (F0 KO) showed defects in mineralization and bone structure in specific locations with known endochondral ossification such as missing structures in the tail region and abnormal trunk curvature. The level of knockdown of ccn2a in these adults was confirmed by measuring the mRNA level from trunk tissue and was found to be significantly reduced.

Conclusion: Our observations in humans and zebrafish combined with previously described skeletal phenotype of Ccn2 knock out mice, confirm that biallelic loss of function variants in CCN2 result in an autosomal recessive kyphomelic dysplasia. However, investigation of additional patients and cellular studies are necessary to establish the gene-disease relationship.

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ABSTRACTS

Emerging Scientist Session

A multi-enhancer hub controls dosage of Ets1 gene expression and protects from allergic inflammation

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Introduction: Allergic diseases affect around 20% of adult population in India. This leads to an estimated 200 million people suffering from allergies, including asthma, allergic rhinitis, eczema, food allergies, etc. Several studies have shown that genetic variants can predispose individuals towards allergic responses. However, the precise underlying mechanisms remain poorly understood.

Objectives: One particular genetic variant associated with allergic diseases overlaps with a super-enhancer associated with ETS1 gene locus. Previous studies from our lab have shown that enhancers tend to cluster together in the three-dimensional space of the nucleus forming hyperconnected hubs in T cells, of which the Ets1 locus has been a top candidate. We aimed to understand the functionality of genetic variation at the hyperconnected Ets1- super enhancer locus.

Methods: We deleted the Ets1- super enhancer element in mice using CRISPR-Cas9, and wanted to learn its effect on the mice. We performed in-vitro T cell polarization assays, chromatin conformation analysis, gene expression, CUT&RUN, and oligopaint FISH assays for this study.

Results and Conclusion: We observed that this region was dispensable for T cell development but important for T-helper (Th)-1 differentiation. Mice lacking this region were unable to develop Th1-mediated colitis, but showed a severe Th2-mediated allergic response to house-dust mite exposure. This indicated a loss of balance between Th1 versus Th2, and predisposition for Th2 – mediated immune-response in these mice. To understand the mechanism behind this phenotype, we performed single-cell oligopaint FISH studies and observed loss of chromatin connectivity at this locus after the deletion. This also led to reduced Ets1 gene expression, which impaired Th1-differentiation possibly through defective CTCF recruitment.

Thus, we showed that genetic variation at the Ets1-super enhancer locus reduced dosage of Ets1 gene expression, causing imbalance of Th1/Th2 response, leading to allergic predisposition. Our study thus demonstrates the importance of understanding genetic variants at hyperconnected enhancer regions, due to their crucial role in precise gene regulation as well as their potential to predispose to disease responses.

A tale of two cellular processes: cilia-autophagy crosstalk in bone development and the pathogenesis of skeletal dysplasias

Priyanka Upadhyai, *Kasturba Medical College, Manipal*

Background: Primary cilia (PC) are conserved immotile sensory antennae that transduce a wide array of cues and are required for embryonic and postnatal development. They are a hub of signalling and can interplay with cell processes, e.g., autophagy, involving the degradation and recycling of cellular proteins and organelles in cell-type specific manner. Individually PC and autophagy play vital roles in endochondral ossification, a major mechanism of bone development. Earlier studies by our lab found PC and autophagy deregulation in fibroblasts from achondrogenesis type 1A (ACG1A; OMIM 200600) patient carrying biallelic mutations in TRIP11/GMAP210. Few existing studies also reported abnormal autophagy and PC in chondrodysplasias, e.g., achondroplasia (ACH; OMIM 100800), a non-lethal form of dwarfism caused by gain-of-function mutations in FGFR3 receptor.

Objective: we sought to examine the nature and molecular mechanism of PC and autophagy crosstalk in chondrocyte differentiation using ATDC5 prechondrocytes. We also interrogated the PC-autophagy axis in chondrocytes from ACH patients.

Methods: Autophagy was chemically modified. PC ablation was by knockdown of Ift52 an essential subunit of the intraflagellar transport machinery required for ciliogenesis. Primary cultures of chondrocytes were established from ACH patient.

Results and conclusion: Manipulating autophagy and PC ablation revealed a mutual positive regulatory role between PC and autophagy during chondrocyte differentiation. Studies on ACH chondrocytes showed significantly impaired autophagy, PC, and differentiation. Augmenting autophagy rescued ciliation in mutant ACH chondrocytes. Currently we are studying how FGFR3 overactivation modifies PC-autophagy crosstalk. Our findings will have broader implications for the treatment of cilia associated diseases, e.g., ACH.

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Decoding the transcriptome usage and Alternative Splicing signature (AS) during Neuronal differentiation: Deep gene regulatory approach

Jeru Manoj Manuel, *uMaster, Canada*

Introduction: Neurogenesis is a highly conserved intriguing biological phenomenon in mammals. Human neuronal differentiation is an intricate developmental process that necessitates meticulous organization of numerous molecular and cellular events, influenced by both genetic predisposition and environmental stimuli. Despite, many studies highlighting the importance of alternative splicing (AS) of RNA in neurogenesis, the understanding of transcript usage and AS, a predominant regulator of molecular mechanisms is still in infancy yet much required for better understanding neuro-developmental disorders.

Objective: To characterize the global gene, transcript and AS signature during neuronal differentiation and to identify critical regulatory group of genes that maybe critical for neuro-development, using human induced pluripotent stem cells (iPSCs).

Materials and methods: We used iNGN, engineered iPSC's cell line that can yield a homogeneous population of neurons in four days. We determined the temporal signature of gene expression, the differential usage of transcripts and the landscape of alternative splicing (AS) events (including micro-exons) during neuronal differentiation. Paired-end RNA sequencing was performed on total RNAs collected at three differentiation timepoints: D0 (stem cells), D2 (progenitor) and D4 (mature neurons).

Results and conclusion: Differential transcript usage (DTU) using DTUrtle demonstrated that interestingly 40% of top differentially used transcripts have stable gene expression. GO analysis on 100 most significant demonstrated contrasting enrichment pattern, D0-D2 showed neurogenesis whereas D2-D4 revealed intracellular localization. Intriguingly, we identified D2-specific transcriptional bursting for a subset of genes, inhibition of these candidates led to slowing down of neuronal differentiation. Comprehensive analysis of AS using rMATS determined ~ 6% significant total splicing events enriched for AS and ribosomal processing during each transition. Exon skipping being most dominant and generating neuron specific micro-exons. However, intron retention (IR) was found to be more functionally important, especially D2-specific IR that are critical for the progression of neuronal maturation. GO analysis on differentially spliced genes revealed enrichment in RNA splicing and mRNA processing. Our results indicate the complexity of splicing events during differentiation stages. Strikingly, iNGN stem cells are a simplistic time saving efficient model to further investigate the complexity of AS during genesis of neuro-developmental disorders.

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Neurodevelopmental dysregulation underpins pathogenesis of severe mental illnesses

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Background: Social interaction and communication deficits, which are frequently observed in children with autism spectrum disorders (ASD), are also impaired in people with bipolar disorder (BP) and schizophrenia (SCZ). There is evidence from genetics that a significant portion of hereditary impacts extend beyond the clinical diagnostic limits of mental illnesses and that neurodevelopmental (NDD) genes are commonly impaired pathobiological mechanisms between the three conditions.

Method: We have sequenced almost 300 members of families with severe mental illnesses (SMI), including 60 family and population controls, 80 BP, and 63 SCZ members. Using Whole Exome Sequencing (WES) data from BP and SCZ, we examined the mutational burden in NDD genesets. We then contrasted our results with those from large sequencing projects SCHEMA, BipEx and ASC for SCZ, BP and ASD respectively.

Results: We identified a total of 188 NDD genes (17%) carrying pathogenic variants in our cohort. Cases included 111 high-confidence, strong candidate ASD genes that were enriched for nervous system development (p- 1e-05), axon guidance (2.6e-7), opening of presynaptic calcium channels (3.5e-05) and oligodendrocyte precursor cell markers (2.23e-05). 28 genes were common to both SCZ and BP subjects which were significantly enriched for genes CACNA1G, CACNA2D1, CHRNA7, CACNA1E, and CACNA1B involved in presynaptic voltage-gated calcium channel activity (5.26e-07). A similar analysis with SCHEMA and BipEx showed increased mutational frequency in genes CACNA1G, CACNA2D1, NRXN1 and MYO5A mirroring our findings. SCHEMA showed a significantly higher enrichment for SFARI genes than BipEx dataset (1e-05) indicating a stronger genetic correlation between SCZ and ASD than BP and ASD. Gene-expression and protein interaction networks within brain transcriptome determined that NDD genes mutated in BP subjects showed increased expression during prenatal stages that was on par with ASD gene expression. However, during early adulthood, this trend changes and NDD genes in SCZ and BP show similar expression, whereas ASD-NDD genes show least expression profiles.

Discussion and conclusions: Our research suggests that dysregulated neurodevelopment is one of the main pathologies of SCZ and BP. We would further attempt to pinpoint the exact brain regions and cell types impacted by neurodevelopmental gene perturbations, as well as identify prenatal and postnatal risk periods during the neurodevelopment process for severe mental illnesses.

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A Cryptic Tale Besides HLA Associations in Celiac Disease

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Background: Celiac disease (CD) is a gliadin induced autoimmune disease primarily affecting the duodenal mucosa. CD, with a prevalence of approximately 14 per 1000 individuals in north India, is among the highest in the world. Linkage studies, genome-wide association studies (GWAS) and post GWAS fine-mapping have identified 40 genetic loci associated with CD. The current focus in this complex inflammatory disorder is on translating the promising genetic findings.

Objectives: Our research focused on: a) To establish the genetic landscape of celiac disease; b) To access the association of HLA-DQ risk haplotypes (-DQ2.5, DQ2.2, and DQ8) and identification of novel tag-SNPs, if any, for efficient, easy, and cost-effective HLA-DQ typing using a clinically well characterized north Indian CD cohort; c) To establish patient-derived organoid model (PDO) for functional validation of genetic leads; and d) explore potential therapeutic targets.

Methods: 557 CD and 736 controls were genotyped on Ilumina Immunochip (covering ~200 immune genes/loci); genetic association, locus fine-mapping, and cross-ethnic locus-transferability tests were performed. PCR-SSP and RT-PCR-based HLA typing was performed on a subset of the cohort (459 CD, 450 controls). A clinico-pathological study was conducted on endoscopic biopsy tissues from 50 CD and 33 controls. Duodenal mucosal transcriptome study including lead validation analyses were performed on 26 CD patients and 30 controls. Functional validation of associated and differentially expressed gene(s) was done using either the IEC-18 cell line. A proof-of-concept study was performed using a patient-derived organoid (PDO) model to understand duodenal crypt plasticity implicated in CD pathogenesis.

Results: HLA-DQ2.5 was the strongest ($p<0.0001$, OR=27.5) and most prevalent (93% in CD and 33% in controls) haplotype in the study cohort. A serial combination of three SNVs, which efficiently (95-100% specificity, sensitivity, positive predicting value, and negative predicting value) tagged this haplotype was identified. Multiple novel non-HLA associations, including ANK3 were also identified. ANK3 knockout in IEC-18 cell line was shown to destabilize E-cadherin and increase membrane permeability. Of note, differential expression of E-cadherin (CDH1/CDH18), CXCL9, and crypt stem cell niche-related genes were observed in CD. Immunohistochemistry, Immunocytochemistry, real-time PCR and western blot evidences suggested that gliadin peptide induced crypt stem niche alteration/failure and variable WNT-NOTCH-BMP signaling play a critical role in CD pathogenesis. A plant derived lignan metabolite showed promising result in rescuing the said alteration/failure.

Conclusions:

Novel tag-SNP based HLA typing may facilitate efficient but cheaper, quicker, and feasible CD screening/diagnosis; and mitigation of Gliadin-induced crypt niche failure may be of potential therapeutic relevance.

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ABSTRACTS

Poster Presentations

Abstract ID: 6

Decoding the genetic blueprint of sleep quality and subjective well-being in young adults

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Background/ Introduction: Previous studies have shown the existence of significant heritability of both sleep quality and subjective well-being. Twin and family studies demonstrated approximately 30-40% of the variation in sleep quality and around 50% in subjective well-being can be attributed to genetic influences. Studying these traits in 18-35 years old is vital considering the mental health challenges in this age group. This study aims to uncover the genetic factors involved in the interplay between sleep quality and subjective well-being.

Objectives: The first objective of this study is to conduct a questionnaire based cross-sectional survey to assess happiness (using Subjective Happiness Scale), sleep quality (using Pittsburgh Sleep Quality Index and Sleep Quality Scale), and depression (using Beck's Depression Inventory). To ensure cultural relevance and accuracy, all scales are translated into Bengali (vernacular language of West Bengal) and validated. The second objective is to identify genetic factors influencing sleep quality and well-being through whole-exome sequencing (WES). The third one is to perform PCR-RFLP based genotyping of the identified genes, including the top variants from previous Genome wide association studies (GWAS) and those found through WES.

Materials and Methods: The internal consistency, bilingual reliability and construct validity of the questionnaires were assessed using IBM SPSS software. Scores are calculated based on existing literature. Participants (N= 597) were divided into different groups using the cut off scores generated through Receiver Operating Characteristic Curves, which were constructed based on individual's happiness, sleep quality and depression scores and the general questions asked quiring about one's happiness, sleep quality and depression. Extreme phenotypes (Positive sleep quality-high happiness-low depression and Negative sleep quality-low happiness-high depression) were selected for WES (N=18). PCR-RFLP based genotyping was performed on a larger cohort (N= 205).

Results and Conclusions: All questionnaires demonstrated high reliability (Cronbach's alpha ≥0.7). Paired sample t-test confirmed no significant differences in bilingual and test-retest reliability, indicating that language and temporal factors did not affect the scale's accuracy. Pearson's correlation revealed that happiness was negatively correlated with sleep quality and depression, while sleep scales were positively correlated with each other and with depression. WES identified significant variants in genes such as CAPN8, NPBWR1, MANEA, ABCC4 and TAAR5 to be associated with sleep quality and well-being. Prioritization of variants in these genes and genotyping for CLOCK variant rs1801260 and BDNF variant rs6265, which significantly affect sleep quality and well-being, is underway. Our findings will highlight potential genetic markers linked to sleep quality and well-being, providing valuable insights for future research and therapeutic strategies. This study provides fundamental insights into the genetic underpinnings of sleep quality and subjective well-being in young adults, paving the way for improved mental health and interventions.

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Abstract ID: 08

Genetic variant rs1527423 T>C in microRNA-25 is associated with an increased risk of Chronic Obstructive Pulmonary Disease (COPD) in the North Indian Population

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Background/ Introduction: MicroRNAs can target a wide range of genes; even minor differences in their expression can result in large-scale alterations in the expression of genes that code for proteins, impacting several biological processes and perhaps exacerbating conditions like COPD.

Objectives: The present study investigates the association between six miRNA SNPs (miR-605, miR-608, miR-3117, miR-149, miR-499, and miR-25) and the risk of COPD in a North Indian population. The study also seeks to understand the functional impact of these miRNA-SNPs on pathological factors of COPD.

Materials and Methods: To assess the genotypes, a case-control study was conducted with 323 COPD cases and 350 hospital controls. Logistic regression was performed to deduce Odd's ratio and 95% confidence interval to assess the association between the SNPs and the susceptibility towards COPD, along with stratified analysis for clinical parameters, symptoms, and contributing factors. Further, SNP-SNP interaction was determined using combinatorial analysis, MDR, and CART analysis.

Results and Conclusions: The study found a strong association between miR-25 SNP rs1527423 and the risk towards COPD (OR=6.38; p<0.0001). Risk association was observed towards COPD in patients with miR-608 SNP rs4919510 (OR=1.43, p=0.02). Further, rs1527423 of miR-25 remained significant in stratified analysis for age (OR=6.19; pc=0.0005), gender (significant in males; OR=5.93; pc<0.0001), smoking status (significant in smokers; OR=5.02; pc=0.0006) in modulating the susceptibility towards COPD. Risk association was observed in miR-608 SNP rs4919510 towards COPD susceptibility (OR=2.38, pc= 0.005) in patients aged ≥ 65 years. MiR-605 SNP rs2043556 showed a protective effect against COPD (OR=0.142; pc=0.048) in non-smokers. MiR-3117 SNP rs4655646 was linked with COPD-related symptoms, such as body movement, breathlessness, and expectoration. All the doublet combinations of each SNP with that of miR-25 SNP showed elevated risk towards COPD. In CART analysis, miR-25 (W;H) made the initial split, and a genotypic combination of 149(M)-3117(M;W)-605(M;W)-608(M;H)-25(W) showed the highest risk towards COPD (OR=11; p=0.04) suggesting rs1527423 to be the critical factor in the decision tree. The present study suggests that the miR-25 SNP rs1527423 may be a risk factor for COPD in North Indian populations. The study highlights the importance of investigating miRNA-SNPs in understanding the genetic susceptibility to COPD.

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Abstract ID: 09

Genetic Modulation of Airway Remodelling genes and their associated risk towards Chronic Obstructive Pulmonary Disease (COPD) in North Indian Population

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Background/ Introduction: COPD is a small airway disease that causes airflow obstruction and other symptoms. Genetic research reveals an imbalance between lung enzymes and inhibitory proteins. Understanding genetic risk loci offers new perspectives on disease pathogenesis. Individuals with COPD exhibit elevated MMP and ADAM levels, but few studies have explored this genetic level.

Objectives: Our study examines the polymorphisms in ADAM33 and MMPs, which may enhance our knowledge of the genetic basis of the disease and lead to improved diagnostic tools and curative options.

Materials and Methods: A total of 1000 subjects, comprising 500 COPD cases and 500 controls, have been recruited and genotyped to evaluate the relationship between COPD and specific SNPs of proteases. Logistic regression was employed to calculate odds ratios (ORs) and 95% confidence intervals (CIs) to evaluate the relationship between SNPs and the risk of COPD. The association between spirometry values and COPD for all SNPs has been examined by applying Kruskal Wallis's test. An analysis of haplotypes has also been conducted. The impact of SNP interactions on COPD risk was evaluated using the Multifactor Dimensionality Reduction (MDR) method and CART analysis to address certain limitations of logistic regression in detecting and characterizing SNP interactions.

Results and Conclusions: "The study reveals that COPD patients with the mutant genotype (GG) for MMP9 (rs17576) have a higher risk of developing COPD (Pc=0.0012) due to MMP-9 substrate binding and enzyme activity changes. This leads to an abnormal alveolar phenotype and emphysema, increasing air gaps and decreasing gas exchange surface area. Moreover, ADAM33 rs2280091 (A>G) polymorphism is linked to COPD susceptibility (Pc=0.0012) and might be linked to inappropriate substrate cleavage, increased airway wall angiogenesis, and a thickened wall. A strong association with COPD severity in smokers has been found. A significant association between ADAM33 and FEV1 and FVC of COPD patients has been found in the case of rs612709 (P=0.004), implying poor treatment responsiveness and the role of genetics in personalized therapy. MDR findings have also identified rs3918392, rs17576, rs2280091, and rs3918396 as the most effective models for understanding COPD susceptibility.

The study links COPD risk to genetic variants MMP9 rs17576 and ADAM33 rs2280091, with the increased risk of COPD and the mucous symptoms and lung function measurements in COPD patients. The study also found rs17576 significantly linked to disease severity, suggesting that focusing on specific traits at different stages of the disease can lower mortality rates.

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Abstract ID: 10

An update on the BRCA1 and BRCA2 gene mutation spectrum: Largest dataset from a single centre in India

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Background/ Introduction: With the rising global incidence of cancer and approximately 25% of cases being hereditary, the importance of genetic screening for high-risk genes such as BRCA1 and BRCA2 has become crucial. Expanding genetic testing to a larger cohort could lead to the establishment of more cost-effective assays with shorter turnaround times, even in resource-limited clinical settings.

Objectives: To study the mutation spectrum of BRCA1 and BRCA2 genes in Hereditary Breast and Ovarian cancer patients in India.

Materials and Methods: A total of 1,449 patients (772 subjects data published in March 2024) with breast or ovarian cancers, or with a family history of these or other cancers, were screened using a targeted BRCA gene panel. All subjects underwent multiplex PCR followed by NGS sequencing. These parallel sequencing methods achieved a mean depth of >300X, with > 99% of bases covered at a minimum of 20X, ensuring complete coverage of coding and splice site regions without gaps. Clinically significant variants identified were subsequently validated with Sanger sequencing.

Results and Conclusions: Results: The study encompassed 1,449 subjects, including 932 with breast cancer, 420 with ovarian cancer, 9 with prostate cancer, 7 with pancreatic cancer, and 81 who were screened due to a significant family history of cancer. The average age of breast cancer onset in our cohort was 43 years (±10), while for ovarian cancer, it was 48 years (±11). Most breast cancer cases were triple-negative (47%), and the majority of ovarian cancers were of the high-grade serous type.

Among 1,449 subjects, 335 were identified with BRCA variants, including 210 with pathogenic/likely pathogenic variants and 125 with variants of uncertain significance, resulting in a mutation positivity rate of 14.5%. BRCA1 mutations were more prevalent, accounting for 77.5% of the cases. In breast cancer patients, the diagnostic yield was 12%, with triple-negative breast cancer (TNBC) contributing 8%. For ovarian cancer patients, the diagnostic yield was 22%, all of whom had a high-grade serous phenotype. Of the 81 individuals screened solely due to a family history of cancer, 5 (6%) tested positive for BRCA mutations. The study identified 23 recurrent mutations and this data suggests that genotyping assays for the identified recurrent mutations could detect about 40% of the positive cases.

Conclusions: BRCA1 mutations more common in breast and ovarian cancers than BRCA2. The study highlights the importance of identifying population-specific hotspot mutations and the need for developing cost-effective assays with quicker turnaround times. Individuals who are BRCA-negative but have a strong family history of cancer may undergo further testing for other high- and moderate-risk genes to identify potential therapies and assess the family's cancer risk.

Abstract ID: 11

An investigation of stress, 5-HTTLPR polymorphism and cytokine in etiopathology of depression: A study among patients in Sikkim, India

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Background/ Introduction: Imbalance in the serotonergic pathway is often linked to Major Depressive Disorder (MDD) due to its modulatory role in mood and emotional states. Recently, polymorphism (5-HTTLPR) in serotonin transporter gene has become a key focus of interest in MDD, with S allele proposed to increase risk for MDD. However, the meta-analysis casts doubt on this assertion. Pro-inflammatory cytokines such as IL-6, TNF-α, and IFN-γ modulate serotonin production by activating indoleamine2,3-dioxygenase, which converts tryptophan (serotonin precursor) to kynurenine, thus reducing serotonin levels and potentially contributing to depression.

Objectives: This study aims to investigate the role of stress, 5-HTTLPR polymorphism and cytokines in the etiopathology of depression in Sikkimese patients.

Materials and Methods: Patients were recruited from STNM Hospital, Gangtok, Sikkim. The biallelic and triallelic (rs25531) genotyping of 5-HTTLPR was performed by PCR-RFLP method. Serum cortisol and cytokines were measured by ELISA. The level of depression was determined using the Patient Health Questionnaire-9 (PHQ-9). The life stressors were studied by Social Problems Questionnaire (SPQ), the Childhood Trauma Questionnaire (CTQ), the Life-Threatening Experiences questionnaire (LTE), and the Perceived Stress Scale questionnaire (PSS).

Results and Conclusions: The SS genotype of 5-HTTLPR was found to be significantly associated with depression. Logistic regression analysis revealed a significant association between PSS score and LS/SS genotypes. Furthermore, the median value of IFN-γ and IL-4 was found to be significantly higher in the patients. Regression analysis showed a significant association between scores of PHQ-9 and IFN-γ levels. Additionally, the CTQ score showed a significant association with IL-6 levels.

The study shows that the polymorphism in 5-HTTLPR provides a risk for depression in individuals under stress, while stress and increase levels of cytokines provides risk and severity of depression.

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Abstract ID: 12

Expression profile of the SBSPON gene in the Type 2 Diabetes Individuals

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Background/ Introduction: Somatomedin B and Thrombospondin Type 1 Domain Containing (SBSPON) located on Chromosome 8q21.11 is comprised of 61,012 bases and encodes a protein of 264 amino acids. The gene of thrombospondin (TSP) family is believed to play very important role in the metabolism and O-glycosylation pathways. mRNA expression in normal human tissues from the GTEx and cbio portal data showed that the gene is expressed in several tissues including whole blood, brain, heart, skeletal muscle, adipocyte, kidney, liver, lung, pancreas, and thyroid. To date, no perspective data is available on the SBSPON gene. Therefore, in the present study, we evaluated the gene expression among the T2D and healthy individuals to understand the association between the gene and diabetes.

Objectives: Gene expression Analysis of SBSPON in Type 2 Diabetic (T2D) Individuals and Healthy Individuals from the population of Jammu and Kashmir region.

Materials and Methods: A case-control study was performed on a total of 100 samples (50 T2D cases and 50 Healthy controls). Blood samples were collected after obtaining the informed consent. RNA was isolated from whole blood and then converted to cDNA for further use. Gene Expression analysis was performed using SyBr Green chemistry. ΔCt method was employed to evaluate the expression difference. Statistical tests were performed considering p-value <0.05; CI – 95% as significant to identify the difference of gene expression among Type 2 Diabetic Individuals and Healthy Individuals.

Results and Conclusions: A significant difference in gene expression was observed among the T2D and healthy individuals, with a p-value of 0.02 adjusted for age and gender. The observed odds ratio was 2.83 (1.14 - 7.02 at 95% CI). To the best of our knowledge, it was the first study to evaluate the SBSPON gene expression among T2D and healthy individuals. The T2D individuals showed downregulation of the gene SBSPON when compared with healthy individuals.

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Abstract ID: 13

First Genome-Wide Association Study of Type 2 Diabetes in Population of Jammu and Kashmir

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Background/ Introduction: Type 2 diabetes (T2D) is becoming increasingly common in Jammu and Kashmir (J&K), with a prevalence rate of 8.35%. T2D is indeed a multigenic and multifactorial condition. While previous studies in the region focused on candidate gene approaches, our study is pioneering the use of Genome-Wide Association Studies (GWAS).

Objectives: To identify novel genetic loci and develop polygenic risk scores for Type 2 Diabetes in the Jammu and Kashmir population through a comprehensive GWAS.

Materials and Methods: We have conducted GWAS with ~0.7 million SNPs in 209 T2D cases and 389 controls of J&K. Additional genome-wide variants were imputed based on genotype data and were evaluated for their association with T2D and Polygenic risk scores (PRS) were developed for J&K Population.

Results and Conclusions: We identified several novel T2D-associated loci ($P < 1.12\times 10{-}7$), that play significant roles in the development and progression of type 2 diabetes by influencing insulin secretion, glucose regulation, and immune responses. Functional analysis of these loci revealed substantial enrichment in pathways related to insulin secretion, beta-cell function, and glucose metabolism. PRS for T2D susceptibility in the population of Jammu and Kashmir were estimated.

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Abstract ID: 14

Comprehensive Profiling of Neurodegenerative and Inflammatory Biomarkers in Alzheimer's Disease: Insights for Early Diagnosis and Targeted Therapy

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Background/ Introduction: Alzheimer's Disease (AD) is a debilitating neurodegenerative disorder marked by progressive cognitive decline, driven by the accumulation of amyloid plaques, tau hyperphosphorylation, oxidative stress, and chronic neuroinflammation. Understanding the correlations between these pathological processes, as reflected in specific biomarkers, is critical for early diagnosis and the development of targeted therapies.

Objectives: This study aimed to examine the relationship between key neurodegenerative, inflammatory, and oxidative stress biomarkers in AD patients compared to healthy controls (HC). By identifying significant biomarker changes and their interconnections, this research seeks to establish their potential for diagnostic and therapeutic advancements in AD.

Materials and Methods: A case-control study was performed with 25 AD patients and 25 age-matched HC. Quantitative analysis focused on biomarkers including amyloid-β isoforms (Aβ1-40, Aβ1-42), Aβ1-42/40 ratio, phosphorylated tau proteins (pTau181, pTau217), neurofilament light chain (NFL), oxidative stress marker 8-hydroxy-2'-deoxyguanosine (8OHdG), brain-derived neurotrophic factor (BDNF), inflammatory markers (TNF-α, IL-6, IL-11, IL-15, CRP), and glutamate. Biomarker levels were assessed using ELISA, and statistical analysis determined individual variations and cross-correlations, with significance set at $p < 0.05$.

Results: AD patients exhibited significantly elevated Aβ1-40 ($p<0.01$), pTau181 ($p<0.001$), pTau217 ($p<0.001$), and NFL ($p<0.01$), indicating a clear link between tau hyperphosphorylation, amyloid dysregulation, and neuronal damage. Elevated oxidative stress, demonstrated by increased 8OHdG ($p<0.01$), was accompanied by heightened BDNF levels ($p<0.05$), suggesting a neuroprotective response. Inflammatory markers—TNF-α, IL-6, IL-11, IL-15, and CRP—were all significantly higher in AD patients ($p<0.05$), emphasizing the role of chronic inflammation in AD pathology. Notably, the correlation between pTau and NFL ($p<0.001$) highlights the strong association between tau pathology and neurodegeneration. The reduced Aβ1-42/40 ratio ($p<0.001$) alongside increased Aβ1-40 ($p<0.01$) points to impaired amyloid clearance, exacerbated by oxidative stress and inflammation. Surprisingly, Aβ1-42 and glutamate levels showed no significant differences, indicating these may not be critical in early-stage AD progression. Cross-correlation analyses reveal intricate relationships among amyloid imbalance, tau pathology, inflammation, and oxidative stress, underscoring the multi-dimensional nature of AD.

Conclusions: This study provides a robust biomarker profile in AD, illustrating strong interconnections between tau hyperphosphorylation, amyloid dysregulation, neuroinflammation, and oxidative stress. The cross-correlations among pTau, NFL, and inflammatory markers support the potential development of a composite diagnostic panel for AD. Furthermore, these pathways offer promising therapeutic targets, paving the way for personalized treatments that could alter disease progression. By integrating these biomarkers, the findings hold significant potential for improving early diagnosis and fostering multifaceted therapeutic strategies in AD management.

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Abstract ID: 15

Unravelling disease severity in viral infections: The role of CD4+ T cell specific eQTLs in COVID-19

Supratim Ghosh[1], *Dibyangshu Pratihari*[2], *Ankita Chatterjee*[3], *Sumanta Sarkar*[1], *Partha Pratim Majumder*[3], *Arindam Maitra*[1]

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Background/ Introduction: The host genetic factors of the immune system play a major role in regulating the variability of disease severity of COVID-19 infection. Single-cell RNA-Seq based studies have shown that CD4+ T cells play a pivotal role in orchestrating the immune response against SARS-CoV-2. However, the cellular and molecular underpinnings of gene regulation by COVID-19 severity-associated genotypes particularly in CD4+ T cells, have not been studied yet.

Objectives: 1. To identify regulation of CD4+ T cell specific gene expression by genomic variants associated with COVID-19 severity, in peripheral blood cells of healthy individuals.

2. To elucidate the biological significance of the identified genes in regulating COVID-19 disease severity.

Materials and Methods: Peripheral blood of 40 adult healthy Indian individuals was used for genome-wide genotyping and single-cell RNA sequencing. The imputed genotype data and single-cell RNA-Seq data were analysed to identify the cis and/or trans-eQTLs specifically in CD4+ T cells. The genomic variants associated with COVID-19 severity were identified by meta-analysis of relevant studies from the GWAS Catalog, using stringent and uniform definition of disease severity. GWAS-eQTL colocalization followed by pathway analysis were performed to identify eSNPs, eGenes and their biological significance in regulating COVID-19 severity specifically through CD4+ T cells.

Results and Conclusions: The results revealed that the genomic variants associated with COVID-19 severity regulate expression of two cis-eGenes - SLC39A7 and CCR2 only in CD4+ T cells. Individuals with risk allele (T) at chr6:32714652:C:T locus were associated with reduced expression of SLC39A7 gene which might lead to low intracellular Zinc concentration in CD4+ T cells resulting in poor anti-viral immunity and enhanced infection and severity. Individuals with risk allele (G) at chr3:46461724:G:A locus exhibited elevated expression of CCR2 gene in CD4+ T cells, which might cause enhanced recruitment of monocytes and macrophages, generating cytokine storm leading to severe COVID-19. Along with peripheral blood, these two genes were also found to have enhanced expression in alveolar cells and T cells of lung, suggesting a local as well as systemic contribution to host immunity. This study, for the first time, elucidates that cell-type-specific gene regulation plays a major role in orchestrating difference in immune response against SARS-CoV-2 and other similar viral infections among individuals ultimately leading to variation in disease severity.

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Abstract ID: 16

Understanding the role of genomic variations in women delivering early preterm.

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Background/ Introduction: Early preterm birth (EPTB) is the birth of a child prior to 34 weeks of gestation (ACOG). Children born early preterm are at elevated risk of neonatal mortality and neurodevelopmental and cardiovascular disorders. Although the single nucleotide polymorphisms associated with preterm birth (<37 weeks of gestation) are now being identified, the role of structural variations (SV) (>50 bp) in the maternal genome and how they might lead to early preterm delivery remain unexplored. Genomic studies have suggested that in general, SVs have a more direct impact on gene expression than SNPs. Recently, the technological challenge of identification of SVs from genome sequencing has been overcome substantially by the use of long read sequencing (LRS). Therefore, in this pilot study we have explored the potential of LRS in identifying SVs and their potential role in EPTB.

Objectives: Identification of structural variations in women delivering EPTB

Materials and Methods: Ten study participants were selected from a prospective pregnancy cohort GARBH-Ini. Whole Genome Sequencing (30X) of DNA isolated from peripheral blood (collected at 11-14 weeks of gestation) was conducted using LRS (Oxford Nanopore) in cases (n=5) who delivered EPTB (<34 weeks of gestation) and controls (n=5) who delivered at term (37-41 weeks of gestation). Post sequencing, reads were aligned, and SVs were identified, followed by identification of those exclusively present in all the cases and absent in controls.

Results and Conclusions: We identified various insertions, deletions, duplications and inversions in all samples using LRS which has not been reported earlier. In the case control study, we identified 3 deletions and 3 insertions present in all the cases and absent in all the controls. Two of these deletions were intergenic where the longest deletion was 3KB long in chromosome 4. A 1.5KB deletion in the intronic region of KIR3DL1 was identified exclusively in all cases. KIR is an inhibitory receptor on NK cells and along with HLA genes help in tolerating the fetal alloantigen during pregnancy. Deletions in this gene can have a potential role in early preterm birth. Two insertions were intronic (180bp) in genes coding for GPC6 and LINC00595. This preliminary study highlighted that SVs, which are until this report, completely unexplored in the area of preterm birth, might have some important role in EPTB. Our pilot study also shows how LRS can be effectively used to identify these variants at a high resolution.

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Abstract ID: 17

Analysis of chromosomal aberrations in the centromeric regions of Alcohol consumers

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CHRIST (Deemed to be University)

Background/ Introduction: Analysis of chromosomal rearrangements in the centromeric regions concerning alcohol use in relation to gastric cancer represents a pivotal area of research. Gastric cancer is a multifactorial disease influenced by genetic predisposition and environmental exposures. Alcohol use is a well-established risk factors for gastric cancer, with evidence suggesting a potential interplay between genetic alterations and these lifestyle factors.

Objectives: The primary aim of the study is to identifying specific chromosomal aberrations associated with alcohol use, potentially linked to gastric cancer. This could enhance our understanding of how these lifestyle factors influence genetic stability and cancer progression.

Materials and Methods: The Present study conducted in various cities across south India, have demonstrated a significant association between alcohol and increased gastric cancer risk. Additionally, molecular investigations have explored chromosomal rearrangements in

centromeric regions, offering insights into potential genetic mechanisms underlying this association.

Results and Conclusions: Integration of findings from epidemiological and molecular studies underscores the complex interplay between genetic

susceptibility, environmental exposures, and gastric cancer risk. Further research in this area may elucidate novel biomarkers

and therapeutic targets for personalized prevention and treatment strategies. Understanding the genetic underpinnings of gastric carcinogenesis is crucial for developing effective strategies to mitigate the burden of this disease, particularly in populations with high rate of alcohol consumption.

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Abstract ID: 18

Effect of Mindfulness Meditation on IOP, Gene Expression, and Mitochondrial Markers in Primary Open-Angle Glaucoma (POAG).

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Background/ Introduction: Glaucoma, a chronic optic neuropathy, is the second leading cause of blindness in the world. Initially, undetected glaucoma leads to elevated IOP (only modifiable risk factor), dysregulated gene expression, oxidative stress, and mitochondrial dysfunction. Yoga practices and mindfulness meditation are believed to lower IOP to modulate and halt its progression.

Objectives: To elucidate the effect of mindfulness meditation on intraocular pressure (IOP), gene expression changes, and assess the influence on mitochondrial markers and oxidative stress levels in POAG patients.

Materials and Methods: After applying inclusion and exclusion criteria, 40 adult-onset high-pressure POAG patients (with no history of systemic or ocular disease) were recruited. This RCT was designed with 8 weeks of daily 45-minute mindfulness meditation alongside medical therapy on POAG patients undergoing trabeculectomy. Group-I (n=20) received meditation, Group-II (n=20) had standard care. The study measured changes in intraocular pressure (IOP), gene expression, mitochondrial markers, and COX-11. Stress biomarkers (cortisol, β-endorphins, IL6, TNF-α, BDNF, ROS & TAC) were also assessed. Mitochondrial gene expression was analyzed via RT-PCR. ROS levels were determined by chemiluminescence whereas 8-IP and 8-OHdG were evaluated by ELISA.

Results and Conclusions: Group-I showed a significant drop in IOP from 22.21±3.98 to 18.15±4.2mmHg ($p=0.001$), while Group-II showed minimal change (21.7±4.5 to 17.8±4.4mmHg; $p>0.05$). Gene expression analysis revealed upregulation of nitric oxide synthetase (NOS1 & NOS3) and neuroprotective

genes, and downregulation of proinflammatory genes (p=0.001) in Group-I. Stress biomarkers and quality of life significantly improved in Group-I (p<0.05). Patients exhibited elevated ROS, 8-IP, 8-OHdG levels, and low plasma TAC, indicating oxidative stress in POAG. Among 156 mtDNA nucleotide variations, 31 were novel and 4 were pathogenic. The 4 pathogenic variations were found in 32.00% of patients. The pathogenic variations distorted the respiratory chain structure, which increased oxidative stress and reduced plasma TAC in POAG. Mindfulness meditation potentially lowers intraocular pressure, influences gene expression, and positively impacts mitochondrial markers in primary open-angle glaucoma patients. While suggesting a role in POAG management, additional cohort studies are needed to validate its impact on patient outcomes.

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Abstract ID: 19

Effect of Yoga-Primed Serum from Male Infertile Cohort on Cancer Cell Behavior

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[1]*Department of Anatomy, All India Institute of Medical Sciences (AIIMS);* [2]*Department of Biochemistry, All India Institute of Medical Sciences (AIIMS);* [3]*Department of Urology, All India Institute of Medical Sciences (AIIMS), New Delhi.*

Background/ Introduction: Infertility affects about 15% of couples worldwide. Additionally, there is an increased likelihood among infertile men to develop both gonadal and extra-gonadal cancers. It is associated with poor semen quality, oxidative stress (OS), oxidative DNA damage (ODD) & OS-induced genome-wide hypo-methylation. This exerts genome instability & hyper-mutability by accumulation of DNA adduct (8-OHdG) and dysregulation of genomic integrity, tumor suppressor genes & oncogene that enhance the risk of cancer. As infertility is a psychosomatic disease, induced by lifestyle factors, psychological stress and unhealthy social habits, an alternative approach like mind-body intervention i.e., yoga is needed. This study evaluates the impact of yoga on male infertility associated cancer biomarkers and the effect of yoga-primed serum on cancer behaviour.

Objectives: This study investigates the impact of yoga intervention on gene expression, cancer cell behavior (Proliferation, apoptotic rate, migration and invasion assays), myokine levels, and oxidative sperm DNA damage (seminal OS level, sperm DNA damage) in infertile men.

Materials and Methods: Sixty infertile men (average age 34 ± 2.5) participated in a 3-month yoga intervention. Post-intervention, their serum (yoga-primed serum) was used to treat prostate cancer cells (LNCaP, PC-3) in vitro. Gene expression analysis was performed using RT-PCR, and cancer cell behavior (proliferation, apoptosis, migration, invasion) was assessed using MTT assay, Annexin V staining, and Transwell migration & invasion assays. Serum myokine levels (IL-6, IL-15, Osteonectin, Oncostatin-M, Irisin) were measured using a multiplex myokine kit. Oxidative sperm DNA damage, ROS, 8-OHdG, and sperm DFI were evaluated using a luminometer, ELISA, and SCSA. Statistical significance was set at $p < 0.05$, and data was analyzed using GraphPad Prism.

Results and Conclusions: Yoga intervention significantly reduced oncogene expression (RET, AR, KIT) with a fold change reduction ($p < 0.01$) and increased tumor suppressor gene expression (ATM, ERCC, SMAD4) ($p < 0.05$). Yoga-primed serum significantly inhibited prostate cancer cell proliferation after 72 hours ($p < 0.01$), and apoptosis was notably increased ($p < 0.01$). Migration and invasion of cancer cells were significantly reduced in PC3 and LNCaP cells post-yoga serum treatment ($p < 0.05$). Myokine levels, including Osteonectin, Oncostatin-M, and IL-15, were significantly elevated ($p < 0.05$) following the

intervention. Yoga also reduced oxidative sperm DNA damage by lowering ROS ($p < 0.01$), 8-OHdG ($p < 0.05$), and sperm DNA fragmentation index indicating improved sperm quality.

This study is the first to investigate the combined effects of yoga on cancer cell behavior and sperm DNA integrity in infertile men. The findings provide novel insights into yoga's molecular mechanisms, demonstrating its dual role in cancer prevention and fertility enhancement. These results highlight yoga as a promising, non-pharmacological adjunct intervention for reducing cancer risk and improving reproductive health in infertile men, offering significant clinical implications for future therapeutic strategies.

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Abstract ID: 20

Chromosomal Alterations and Epigenetic Modification of Prader Willi Syndrome patients from Tamil Nadu, India

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Sacred Heart College, Vellore, Tamil Nadu.

Background/ Introduction: Prader-Willi syndrome (PWS) is a neurogenetic disorder caused by dysfunction of epigenetic mechanism and the genes located in the maternal chromosome 15q11-q13, structurally intact but transcriptionally repressed due to epigenetic regulation. Nevertheless, there has been no report of the exact incidence, prevalence and cause of PWS in India.

Objectives: We aim to investigate the chromosome alterations and methylation status of SNRPN gene in PWS patients in Tamil Nadu population.

Materials and Methods: In this study, we recruited 5 PWS patients and their parents from Tamil Nadu population. Cytogenetic and epigenetic techniques were performed in peripheral blood samples of both subjects.

Results and Conclusions: FISH technique was revealed that the occurrence of microdeletion on SNRPN gene in 4 PWS patients. We found de novo balanced reciprocal translocation, between Y and 15 chromosomes in one male patient of 5 PWS cases; involving the breakpoint 15q13 and Yq11.1 (46,X,t(Y;15)(q11.1;q13) and Yq12 and 15q12 (45,X,der (15),t(Y;15),(q12;q12) in father, using karyotyping analysis, these investigations were the first to report that the microdeletion and balanced reciprocal translocation between chromosome 15 and Y in our study population. The result of MS-PCR revealed only maternally methylated (M-174bp) SNRPN gene in 4 patients out of 5 subjects. In the control subjects, both paternal unmethylated (P- 100pb) and maternal methylated, (M- 174bp) bands were observed, along with an unmodified genomic DNA. Additionally, this study has examined direct bisulphite sequence to evaluate the quantitative methylation status in all PWS using SNRPN gene at exon 1, unmethylated cytosine is converted to thymine C/T, while the methylated cytosine remains cytosine in the CpG site of the SNRPN sequence. We found that 4 of 5 PWS patients were significantly associated with hypermethylation accumulated in the CpG site of the SNRPN gene at exon 1 when compared to control samples of unmodified genomic DNA. We recommend screening of genomic imprinting using highly efficient MS-PCR and bisulfite sequencing techniques led to be a good diagnostic tool leads reliable genetic counselling.

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Abstract ID: 21

Mutations in HYPK cause a novel neurodevelopmental disorder by impairing neuronal proteostasis and autophagy

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Background/ Introduction: HYPK maintains cellular proteostasis by functioning as an autophagy receptor in the targeted degradation of NEDDylated protein aggregates. HYPK is also involved N-terminal acetylation of nascent polypeptides, influencing protein homeostasis. Loss of HYPK expression or function impairs NEDDylation-dependent aggrephagy, leading to neuronal proteostasis imbalance.

Objectives: To investigate the clinical (phenotypes and neurological outcomes) and cellular/molecular (proteostasis disruption, impairing NEDDylation-dependent aggrephagy, and cellular senescence) characteristics of a novel neurodevelopmental disorder caused by heterozygous missense mutations in HYPK gene, characterized by early-onset of brain atrophy.

Materials and Methods: Clinical diagnosis, magnetic resonance imaging of the brain, singleton exome sequencing, sanger sequencing, neuron culture and transfection, immunofluorescence microscopy, immunoblotting, computational modeling and simulation, statistical analysis, Drosophila studies

Results and Conclusions: We describe four individuals (2 males, and 2 females) from three unrelated families, all presenting with progressive neurological disorder. The probands clinically exhibited global developmental delay, seizures, aphasia, dementia, disorientation, communication difficulties, and memory loss. Radiological scans of probands revealed significant hypoplasia of the corpus callosum, along with severe diffuse cerebral and cerebellar atrophy, without brain lesions.

Whole-exome sequencing identified de novo or inherited heterozygous missense mutations [c.46G>A (p.E8K), c.94A>G (p.K24E), and c.313T>G (p.S97A)] in the HYPK gene. These mutations affect highly conserved residues located in unstructured regions of HYPK, causing entropic gain and enhanced aggregation propensity. The mutant proteins sequester wild-type HYPK and disrupt cellular proteostasis in human neuron cells. This resulted in impairing autophagy and accumulation of poly-NEDDylated protein aggregates, leading to senescent death of neurons.

In vivo, HYPK knockout in Drosophila melanogaster showed developmental anomalies, including reduced brain lobe size in larvae, extensive loss of neurons and neuroblasts compared to the wild-type, and lethality at the 3rd instar larval stage.

HYPK mutations lead to a novel neurodevelopmental disorder characterized by early-onset neurodegeneration and proteostasis imbalance, providing insights into the pathogenic mechanisms underlying these clinical phenotypes.

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Abstract ID: 23

Human mitochondrial genome analysis using long reads to identify disease causing variants: a systematic evaluation & comparison with short read approach

Jyoti Mridha, *Aneek das bhowmik, Shreya, Divya Tej Sowpati, Karthik Bhardwaj, Srinivas, Joel, Pramoda.*
Centre for Cellular and Molecular Biology, Hyderabad.

Background/ Introduction: Pathogenic variations in mtDNA (mitochondrial DNA) are involved in pathogenesis of several human diseases. Oxford Nanopore Technologies (ONT) sequencing utilizes nanopores to read genetic material. This technology is known for generating long reads and can be used in portable devices, making it suitable for sequencing in remote or field environments.

Objectives: To evaluate different bioinformatic tools for the analysis of disease-causing mitochondrial variants using ONT sequencing.

Materials and Methods: mtDNA from 10 samples was either amplified using PCR or captured using Twist mitochondrial DNA panel kit (Twist Bioscience, USA), from the DNA extracted from whole blood. The amplicons were used to prepare the libraries which were sequenced on both ONT and Illumina platforms. Whereas, mtDNA captured using probes was sequenced on Illumina NovaSeq 6000 sequencing platforms. The raw sequences obtained were aligned to the human reference genome GRCh38/hg38 and variants were called and hard-filtered using DRAGENTM (used for short read), Freebayes, Clair3 and a deep learning-based variant caller (DeepVariant) software (for long read). Variant annotation was performed using ANNOVAR.

Results: The total number of variants which passed quality filters was 343 in capture kit SRS based analysis and with long read data using variant callers like freebayes (366), clair3 (351) and Deepvariants, it was 366,351 and 322 respectively. The highest F1 score was noted with DeepVariants (~0.94) followed by clair3 (~0.93) and freebayes (0.89) when Capture based SRS was taken as standard. Pathogenic variants were identified in three samples in both short-read and long-read sequencing. In addition 2 CNVs were exclusively picked up in LRS.

Conclusion: Amplicon based mtDNA sequencing on ONT is a robust and cost-effective approach for mitochondrial variant analysis.

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Abstract ID: 24

Evaluating the contribution of complex chromosomal rearrangements in pregnancy loss: A critical aspect of reproductive genetics

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[1]*Acquity Labs Pvt Ltd;* [2]*Centre for Human Genetics, Bengaluru.*

Background/ Introduction: Complex chromosome rearrangements are balanced or unbalanced structural anomalies involving at least two chromosomes and three breakpoints. In theory, the more complex the rearrangement and the greater the number of chromosomes involved, the more intricate the meiotic pairing structure and the greater the number of potential unbalanced segregates. It is therefore not surprising that balanced carriers of a complex chromosome rearrangement have an empiric risk of greater than 50% for an abnormal pregnancy outcome.

Objectives: To evaluate the contribution of chromosomal anomalies in the pregnancy loss cases and to provide an ideal genetic diagnosis for such couples.

Materials and Methods: We have used the techniques of cytogenetic chromosomal analysis by karyotyping for parental evaluation and molecular cytogenetic techniques by FISH and CMA in products of conception.

Results and Conclusions: We present a case of a couple with history of recurrent pregnancy loss in which the Chromosomal analysis of the male partner showed normal 46,XY karyotype while the female partner revealed a complex chromosomal translocation involving three chromosomes (Chromosomes 2, 10 and 12). When the products of conception for the same couple was analysed by Fluorescent In-situ Hybridisation (FISH) for aneuploidy, it showed Abnormal results with monosomy for X-chromosome in a mosaic condition while being evaluated for pathogenic Copy Number Variations (CNVs) by chromosomal microarray analysis (CMA) for gains and loss, it showed monosomy for X-chromosome (Turner syndrome) along with deletion of (3.3 Mb) in chromosome 12 at cytoregion 12q12.

Conclusions: The results from cytogenetic techniques showed complex chromosomal rearrangements indicating the probable cause for the pregnancy loss. It is also proved that the most efficient genetic analysis for couples with pregnancy loss will be combination of parental evaluation by cytogenetic and products of conception by molecular cytogenetic techniques for the subsequent reproductive decision making.

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Abstract ID: 25

Human mitochondrial genome analysis using long reads to identify disease causing variants: a systematic evaluation & comparison with short read approach

__Jyoti Mridha__, Aneek das bhowmik, Shreya, Divya Tej Sowpati, Karthik Bhardwaj, Srinivas, Joel, Pramoda

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Results:

The total number of variants which passed quality filters was 343 in capture kit SRS based analysis and with long read data using variant callers like freebayes (366), clair3 (351) and Deepvariants, it was 366,351 and 322 respectively. The highest F1 score was noted with DeepVariants (~0.94) followed by clair3 (~0.93) and freebayes (0.89) when Capture based SRS was taken as standard. Pathogenic variants were identified in three samples in both short-read and long-read sequencing. In addition 2 CNVs were exclusively picked up in LRS.

Conclusions:

Amplicon based mtDNA sequencing on ONT is a robust and cost-effective approach for mitochondrial variant analysis.

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Abstract ID: 26

Application of Caffeine Consume in High Concentration can be Harmful or Exaggerated in the Central Nervous System of Human and also used when, as like an Effective Pesticide in Agriculture — Whether A Comparative study using Drosophila melanogaster as a model organism

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Background/ Introduction: Using the model system Drosophila melanogaster, Caffeine is an efficient pesticide. Humans and fruit flies have 44% of the same genetic makeup, and 75% of the genes linked to recognized human diseases are also present in fruit flies. A number of human illnesses, including the neurodegenerative conditions Parkinson's, Huntington's, and Alzheimer's disease, are being studied using Drosophila as a genetic model. The fly is also being used to investigate the processes behind drug misuse, reproductive problems, diabetes, cancer, and immunity. When caffeine is given to Drosophila Melanogaster food by preparing the culture medium, primary culture medium and sub-culture medium and maintaining Drosophila in the Genetics Laboratory, after it has been dissolved in water, the flies will either be deterred from eating it or will die thereafter. Exhibit peculiar behavior after consuming coffee, show a decrease in reproductive output after consuming caffeine. The abnormalities will exhibit in the progeny of the exposed person's health. Because the caffeine overloaded the flies' hearts, stomachs, and other organs, it led to their deaths. It considerably slowed down the flies' reproductive cycle. This development was most likely brought about by caffeine's effects on the "ring gland."In Drosophila, the ring gland is an organ of hormonal release; it secretes the hormones that mark the beginning and end of each stage of the drosophila life cycle. Neuron activity influences the ring gland; hence, hyperactive neurons may cause challenges with hormone release and reproductive disorders. These results would favor a slower rate of reproduction. The central nervous system's reaction to typical environmental stimuli was heightened by the increased neuron activity by GABA and serotonin in Drosophila.

Objectives: My research's hypothesis is that if caffeine is dissolved in water and added to Drosophila Melanogaster meal: 1) The flies will either be discouraged from eating the foodor would perish after consuming it. 2)Display strange behaviour following caffeine consumption. 3)Exhibit reduced reproductive production following exposure to the caffeine. 4)The exposed person's progeny will exhibit abnormalities.

Materials and Methods: Preparing the culture medium: Maintaining Drosophila in the Genetics Laboratory: Preparing the Primary Culture Medium: The Flies Have Been Obtained: Caffeine Solution Preparation. The Subculture's Preparation: Handling The Fly Transfer: Observing The Flies. Precaution has been taken.

Results and Conclusions: Results: Fly Fatality Test and Life Cycle Observation Test

Conclusion: There is no doubt that caffeine harms Drosophila. Without a doubt in excessive amount, it detrimental for these tiny insects and also harmful for Human being which can slowly poisonous in its excessive tacking regularly. Because the caffeine overworked the flies' hearts, stomachs, and other organs, it led to their deaths. It considerably slowed down the flies' reproductive cycle. This development was most likely brought about by caffeine's effects on the "ring gland." In Drosophila, the ring gland is an organ of hormonal release; it secretes the hormones that mark the beginning and end of each stage of the drosophila life cycle. Since the ring gland is regulated by neuron activity, hyperactive neurons may cause issues with the reproductive cycle and hormone secretion. These results would favor a slower rate of reproduction. The central nervous system's reaction to typical environmental inputs was amplified by the increased neuron firing. There is no denying that caffeine has a negative effect or impact on Drosophila. My investigation showed that, at high enough doses, caffeine may work as a pesticide against Drosophila. It's evident that caffeine kills insects at high quantities. Furthermore, caffeine is generated naturally by plants and is not as dangerous as manufactured pesticides, giving it several advantages over other pesticides. So, we can use Caffeine as pesticide in a certain amount and also can intake as Coffee or other Caffeine products in a specific amount which does not harm out Central Nervous Systems and Reproductive systems.

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Abstract ID: 27

Analyzing the role of SRD5A2 (rs523349) in Polycystic Ovary Syndrome

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Background/ Introduction:

Polycystic ovary syndrome (PCOS) is a prevalent endocrine and inflammatory disorder that impacts 11.33% of women who are of reproductive age and exhibit hyperandrogenism, oligoovulation, and polycystic ovaries on ultrasound. Individuals with PCOS often experience associated conditions such as infertility, hyperlipidemia, insulin resistance, cardiovascular risks, obesity, and psychological challenges. Hirsutism is a common manifestation of hyperandrogenism, where it is associated with both androgen excess and individual unit response to androgens. In the context of PCOS, hirsutism displays considerable variability. Ethnicity contributes to this heterogeneity, implying a potential genetic influence on hirsutism severity within the PCOS population. One pivotal enzymatic process in androgen metabolism involves steroid 5α-reductase, which converts testosterone into the more potent dihydrotestosterone (DHT). Notably, both ovarian and peripheral 5α-reductase activity appear elevated in PCOS. There is no study regarding rs523349 polymorphism and PCOS from the Indian population. Our hypothesis proposes that genetic variations within the 5α-reductase genes may serve as risk factors for PCOS development and contribute to the varying expression of hirsutism among affected women.

Objectives: To investigate whether genetic variations within the 5α-reductase genes act as risk factors for PCOS development and influence the expression of hirsutism in affected women.

Materials and Methods: We recruited 134 controls and 137 PCOS patients based on Rotterdam criteria. Genomic DNA was extracted from the samples, followed by PCR-RFLP. Results were confirmed using Sanger sequencing. Statistical analysis was done using SPSS version 27.

Results: The PCOS and control groups did not differ significantly regarding genotype frequency and allele frequency ($p = 0.17541$). In our study, we observed no statistically significant association between the rs523349 polymorphism and various demographic parameters. We observed a significant difference in weight, BMI, hirsutism (Ferriman-Gallway score) and testosterone between the PCOS group and the control group.

Conclusion: No association was found between SRD5A2 rs523349 polymorphism and PCOS. This pioneering study from India should be replicated in more samples.

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Abstract ID: 28

An analysis of patterns of inheritance in familial primary concomitant strabismus in a homogenous south Asian population

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Background/ Introduction: Strabismus (squint in common parlance) implies misalignment of the eyes, which may result in loss of binocularity. It affects about 3-5% of the general population globally and in India, and 60% of those affected are children. Primary concomitant strabismus [PCS] like intermittent exodeviations [XT] and accommodative esodeviations [ET] comprises about 2/3rd of strabismus present in the population.

It is relevant to emphasize that strabismus is not simply a cosmetic issue but a major functional deficit. The resultant loss of binocular single vision (BSV) due to ocular misalignment, especially in children, is the harbinger of neuro-physiological deficits like amblyopia and low vision, making a subject functionally one-eyed despite having bilateral structurally normal eyes. PCS demonstrate familial patterns, observed across all populations, in approximately 5% cases, the rest of the cases being sporadic.

Objectives: This hospital based observational study aimed to classify and analyse patterns of inheritance of PCS in 98 informative south Asian families recruited over the past decade for further discovery genomics and functional studies. No definitive mode of inheritance or any genetic determinants have been concretely established for PCS, making this study unique.

Materials and Methods: Detailed phenotypic evaluation of the proband with PCS and other affected family members was performed in these 98 consecutive informative families of south Asian origin from August 2014 to October 2024. Pedigree documentation was done by Cyrillic 3.0.400 software.

Results and Conclusions: Of these 98 recruited PCS families, 100% concordance of phenotype was observed in 91 families (93%), 28 with ET and 63 with XT. In 5 families, the affected members demonstrated both, ET and XT. In 2 families, one of the siblings each had XT and the other sibling demonstrated Duane's retraction syndrome (DRS). 21/28 ET (75%) and 56/63 XT (88.9%) had PCS in at least two generations, implying vertical transmission. Maternal transmission was observed in 17/21

(80.9%) ET and 45/56 (80.3%%) XT families with vertical transmission, the rest demonstrated transmission from the paternal side. Consanguinity was observed in 2 families, both with ET.

Conclusions: Detailed analysis of the patterns of inheritance in this large cohort of familial PCS provides an insight into the possible modality of transmission and opens perspectives on the phenotypic heterogeneity of the condition in a homogenous south Asian population.

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Abstract ID: 29

Genetic association of IL1B gene variants with primary glaucoma in a North Indian Punjabi cohort: An original study and meta-analysis

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Background/ Introduction: Glaucoma, a leading cause of blindness worldwide, involves optic neuropathies primarily affecting retinal ganglion cells (RGCs) through neurodegenerative processes. Inflammatory cytokines, particularly Interleukin-1 Beta (IL1B), play a role in this pathogenesis. This study investigates the association between IL1B polymorphisms (−511T>C and +3953C>T) and primary glaucoma, specifically primary open-angle glaucoma (POAG) and primary angle-closure glaucoma (PACG), within a North Indian Punjabi cohort.

Objectives: To determine whether IL1B gene variants increase susceptibility to POAG and PACG in the targeted population and to perform a meta-analysis to contextualize findings within existing literature on IL1B polymorphisms in glaucoma risk.

Materials and Methods: A case-control design was used, analyzing 867 samples (307 POAG cases, 133 PACG cases, and 427 controls) collected from hospitals in Amritsar, Punjab. Genetic association tests for −511T>C and +3953C>T polymorphisms were conducted, with genotyping via PCR-RFLP. Statistical analyses included linkage disequilibrium, diplotype frequency assessments, and logistic regression, corrected for confounding variables. Meta-analysis of available literature provided a comparative analysis of pooled risk.

Results and Conclusions: The −511T>C variant showed significant association with PACG under dominant ($p=0.038$) and overdominant models ($p=0.010$), while +3953C>T was associated with both POAG and PACG under various genetic models. Notably, the +3953C>T variant conferred higher susceptibility to PACG under the recessive model ($p<0.0001$). Meta-analysis indicated moderate pooled risk for POAG associated with the +3953C>T polymorphism, aligning with study findings. The C–C diplotype suggested protective effects, whereas T-T and T-C diplotypes increased glaucoma risk. IL1B polymorphisms, particularly +3953C>T, significantly influence genetic susceptibility to POAG and PACG in the North Indian population studied. These findings underscore IL1B as a potential genetic marker in glaucoma pathogenesis, warranting further studies across diverse ethnic groups to validate these associations and explore therapeutic implications through functional studies.

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Abstract ID: 30

DNA Methylation analysis of OPRM1, DAT1 and DRD2 genes in drug dependent population of Manipur.

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Background/ Introduction: Introduction: Addiction is a global issue. Heritability of addiction is around 40 to 60%, and the involvement of non-genetic factors is also indicated in the vulnerability to addiction. Epigenetic changes are known to influence gene expression. Epigenetic markers like DNA methylation and histone modifications are under investigation for their association with addiction. Studies on animal models and human samples have demonstrated that epigenetic changes such as DNA methylation could occur in response to illicit drug use. This epigenetic change, therefore, influences gene expression which in turn may influence reward, psychomotor activity, drug craving, and relapse. Therefore, an individual's vulnerability to developing drug addiction may be determined partially by epigenetic factors. The northeastern region of India particularly Manipur has a high rate of drug addiction, which is causing social problems. Therefore, a better understanding of addiction has become inevitable. So far, no reports on the studies of epigenetic markers are available from Manipur.

The dopaminergic system and genes involved in this pathway plays a significant role in the development of drug dependency. Genes such as OPRM1, DAT1 and DRD2 plays an important role in addiction. The μ-opioid receptor (OPRM1) is the site of action of many endogenous opioids as well as opiates. Regulation of the extracellular dopamine concentration is driven by dopamine transporter (DAT1) and dopamine receptor (DRD2). Both the expression and function of OPRM1, DAT1 and DRD2 are influence by chronic opioid intake.

Objectives: 1) To analyze the prevalence of DNA methylation in OPRM1, DAT1 and DRD2 gene promoter in the addicts and healthy controls from Manipur. 2) To identify any difference in the DNA methylation pattern in OPRM1, DAT1 and DRD2 gene promoter in the addicts and ethnically match controls of Manipur, and 3) To check the relation of DNA methylation and drug addiction in the selected genes.

Materials and Methods: Subjects are recruited based on the criteria outlined in the diagnostic and Statistical Manual for Mental Disorders (DSM-IV). The study is approved by the Institutional Ethics Committee, Manipur University. About 2ml of blood samples were collected from each volunteer subjects in a vial containing EDTA as anticoagulant along with consent form and questionnaire. Genomic DNA (gDNA) were extracted from the peripheral blood of 150 men with opioid used disorder and 100 healthy persons as control group using DNA extraction Kit (QIAamp DNA Mini Kit). Two sets of Primers each (methylated and non-methylated) were designed to target putative CpG islands in promoter of the above three genes using Methyl primer express software to perform Methylation Specific PCR (MSP). Genomic DNA was treated with sodium bisulfite using the bisulfite conversion kits (Qiagen) to convert the unmethylated cytosine to uracil, while the methylated cytosine remained unaffected. Amplification was performed by methylation specific PCR(MSP) using bisulfite modified DNA and MSP kit (Takara). The MSP products were separated by electrophoresis on 2% (w/v) agarose gel stained with Ethidium bromide in TBE buffer. Thus, Methylation status of the MSP products was check by gel electrophoresis. Statistical analysis was done by Chi square test using SPSS software. P value and odd ratio were calculated. $P < 0.05$ is considered statistically significant. Whole genome sequencing of 4 random samples (2 cases and 2 controls) was done to check the methylation status at the promoter region of different genes.

Results and Conclusions: Results: Methylation was found in 9.33%(14 out of 150) of the patients with drug addiction (case), and in 29%(29 out of 100) of the healthy controls for OPRM1 gene, 0.67%(1 out of 150) in case and 8%(8 out of 100) in healthy controls for DAT1 gene and 17.33%(26 out of 150) in case and 10%(10 out of 100) in healthy controls for DRD2 gene. Chi-square analysis has shown a significant difference in DNA methylation pattern in the OPRM1, DAT1 and DRD2 gene promoter between the case and control subjects ($P<0.05$). There is a negative association between methylation and drug addiction in our studied Manipur samples ($OR<1$ for the mentioned genes). Sequencing result is found to be significantly difference ($P=0.0459433$) and hypomethylated at the targeted region of the OPRM1 gene but no significance difference is found for DAT1 and DRD2 genes at the promoter region.

After analysis of the questionnaire majority of the addicts (case) were found to be from the Meitei population (≈86%), who lives at urban areas where drugs are readily available at low cost. Pleasure seeking, influence from friends (drug user), and readily availability of drugs was observed to be the main factors for the initial use of drugs and most of the case and control groups are found to be non-vegetarian. These factors contribute in the vulnerability to addiction as addiction is cause by genetic and non-genetic factors.

Conclusion: Till date, most of the cases and control samples have been found to be unmethylated at the targeted region of selected genes and methylation status is found to be significantly different between the case and control groups. Also DNA hypomethylation in the targeted promoter region of OPRM1 is associated with drug dependence in this ethnic population of Manipur. This result is also supported by the whole genome sequencing data where DNA hypomethylation is observed in the promoter region of the OPRM1 gene in the case group as compared to the control group. Further, negative association between DNA methylation and drug exposure was observed in our studied population which shows the contribution of non genetic factors in drug addiction.

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Abstract ID: 31

Standardization of In Vitro Culture, Expansion, and Isolation of Primary Fibroblasts from Human Skin Punch Biopsy to Assess Germline Variants in Myeloid Leukemia

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Background/ Introduction: Acute Myeloid Leukemia (AML) and Myelodysplastic Syndromes (MDS) are driven by somatic variants, with lesser-understood germline predispositions. Distinguishing germline from somatic mutations is essential for disease understanding and therapeutic strategies. Peripheral blood, typically used for germline testing, often contains tumor cells, complicating analyses. Cultured skin fibroblasts provide a reliable source of pure germline DNA, superior to other sources such as hair or nails.

Objectives: • Develop and standardize skin punch biopsy procedures. • Optimize fibroblast cell culture conditions for viability and density. • Evaluate protocol efficiency by monitoring culture purity and failure rates.

Materials and Methods: Skin punch biopsies (3 mm) were taken under aseptic conditions from one mice and four humans from posterior superior iliac spine and cultured in 6-well plates. Media was changed every 2-3 days, with daily observations. Fibroblasts, which emerged post-keratinocyte outgrowth, were expanded in T75 flasks for DNA extraction.

Results and Conclusions: Murine models successfully achieved fibroblast confluency, validating the culture method. Initial trials with human samples, however, revealed a high failure rate (3/4) primarily due to procedural errors like use of non-autoclaved normal saline while collecting the biopsy, seeding of large sized biopsy pieces and non-adherence of tissue on the surface of six-well plate, use of low glucose media, and use of 5% FBS. Addressing these, smaller biopsy sizes (0.5x0.5x0.5 cu.mm) and optimized plate preparations (scratched the surface), and use of high glucose media with 10% FBS significantly improved outcomes, leading to successful fibroblast culture till the confluency stage in subsequent trials. Further, the whole DNA was extracted from these cultured fibroblast and checked for the JAK2 V617F mutation commonly implicated in myeloid malignancies and it consistently revealed negative results.

The method provided a reliable platform to obtain pure cultures of skin fibroblasts having elongated, spindle shaped cell body with round to oval nuclei that could be explored for studying hereditary predispositions in hematologic malignancies and will assist in focusing on larger cohorts to expand our understanding of genetic risk factors associated with AML and MDS.

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Abstract ID: 32

Maternal NLRP3 Activation and Fetal NKX2-5 Expression in Aborted Fetuses with Suspected Congenital Heart Defects

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Background/ Introduction: Congenital heart defects (CHDs) are structural heart abnormalities present from birth, leading to significant fetal mortality worldwide. The link between maternal health, particularly maternal inflammation, and CHD development in aborted fetuses is underexplored. Elevated interleukin-6 (IL-6) and high-sensitivity C-reactive protein (hs-CRP) levels are associated with various pregnancy complications, including CHDs. The NLRP3 inflammasome is a key inflammation mediator, while the NKX2-5 gene is crucial for heart development, with downregulation tied to heart abnormalities. This study examines the association between maternal inflammation, NLRP3 activation, and fetal NKX2-5 gene expression in aborted fetuses with suspected CHDs.

Objectives: To investigate the relationship between maternal inflammation, as measured by inflammatory markers and NLRP3 gene expression, and its potential association with NKX2-5 gene expression in aborted fetuses with suspected congenital heart defects.

Materials and Methods: This case-control study included 120 participants, 60 aborted fetuses with suspected congenital heart defects, and their mothers formed the case group. Sixty mothers with successful pregnancy outcomes were in the control group. Maternal blood samples were collected and analyzed for biochemical markers, including fasting blood sugar (FBS), triglycerides, total cholesterol, and thyroid-stimulating hormone (TSH). Inflammatory markers were also measured, specifically interleukin-6 (IL-6) and high-sensitivity C-reactive protein (hs-CRP). NLRP3 gene expression in maternal samples was assessed using real-time PCR. For fetal samples, cardiac tissue or intracardiac blood from aborted fetuses

was collected, and NKX2-5 gene expression was analyzed through real-time PCR. Relative gene expression levels were calculated using the 2^(-ΔΔCt) method, and melting curve analysis was conducted to ensure the specificity of the amplification.

Results and Conclusions: Maternal samples from the case group showed increased NLRP3 expression, IL-6, and hs-CRP levels, indicating inflammation, while aborted fetal samples exhibited downregulated NKX2-5 expression. A negative correlation between maternal NLRP3 and fetal NKX2-5 expression suggests maternal inflammation may impact fetal heart development, underscoring the importance of managing inflammation in pregnancy to reduce the risk of congenital heart defects.

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Abstract ID: 33

The next era of cytogenetic testing: Harnessing the power of "cell-free mitotic factors" for rapid prophasing of interphase chromatin

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Background/ Introduction: Cytogenetic testing plays a major role in diagnostics, research, drug testing, and biodosimetry. Mitotic promoting factors (MPFs) are essential for chromatin condensation and preservation of chromosome structure in mitosis. Premature Chromosome Condensation (PCC) is used for cytogenetic testing since it has a lower turnaround time than the conventional human peripheral blood lymphocyte (HPBL) culture. PCC is widely used in biodosimetry and other

applications. However, the existing PCC techniques by chemical induction and somatic cell hybridisation have pros and cons. We developed a new method to condense interphase chromatin in HPBLs using cell-free mitotic extracts, utilising the advantages of both techniques without their drawbacks.

Key words: Chromosome biology; chromatin condensation; Mitotic Promoting Factors; Prophasing interphase chromatin; genotoxicity testing; rapid genetic damage assessment

Objectives: To induce PCC in HPBLs using mitotic cell-free extracts and demonstrate their use in rapid cytogenetic analysis.

Materials and Methods: AGS (human stomach adenocarcinoma) cells were grown in DMEM with 10% FBS at 37°C and arrested in mitosis with colchicine. Mitotic cells were harvested by tapping the flasks and then washed in a colchicine-containing medium to maintain the block. Mitotic extracts were prepared by lysing the cells with RIPA buffer and centrifuging at 2500 RPM for 20 minutes at 4°C. The extracts were dialyzed in 10X volume of PBS, changing the buffer every 12 hours at 4°C. HPBLs, obtained from a healthy volunteer, were incubated with the mitotic extracts at 40°C in 0.1% Tween 20 for 15 minutes, rendering them "semi-intact" to allow MPF uptake through transiently permeable membranes. Cells were washed in RPMI with 10% FBS and colchicine, then incubated at 37°C for 1 hour. They were then processed and Giemsa stained using standard protocols.

Results and Conclusions: Mitotic extracts successfully induced PCC in HPBLs, yielding a significantly higher ME-PCC index (20) than the typical mitotic index (MI) of 5-10 in conventional PBL cultures. Chromatin was well-condensed and can be utilized to visualize chromatin breaks and gaps. This novel technique, the first to use cell-free mitotic extracts for PCC, provides a faster and more efficient approach to cytogenetic testing. It is precious where time, resources, or available cells are limited. Since MPFs are

evolutionarily conserved, various cell types can be used to obtain MPFs, unlike somatic cell hybridization, which relies on specific cell lines like CHO cells to induce PCC.

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Abstract ID: 34

Molecular Landscaping of Primary & Metastatic Brain Neoplasms

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Background/ Introduction: Brain tumors, especially glioblastoma, pose a significant clinical challenge due to their high severity, complex molecular landscape, and poor prognosis. Current diagnostic approaches, such as biopsy and MRI, are invasive or limited by sensitivity. Liquid biopsy, using tumor-derived exosomes from biofluids, offers a promising non-invasive method for early diagnosis, monitoring, and personalized treatment of brain tumors. This study focuses on identifying differential gene markers in glioblastoma and brain metastases using bioinformatics tools and validates the findings through molecular and in vitro techniques.

Objectives: 1. Perform a meta-analysis to identify significant gene markers for glioblastoma using published datasets. 2. Validate gene expression differences between high-grade and low-grade gliomas using tissue and serum exosomes. 3. Screen and validate natural compounds targeting TGF-β1 through molecular docking and in vitro experiments. 4. Assess the role of tumor-derived exosomes in promoting proliferation and metastasis in breast cancer cell lines.

Materials and Methods: This study involved a comprehensive meta-analysis of gene expression datasets to establish a glioblastoma-specific gene panel. Exosomes were isolated and characterized from tissue and serum samples (30 low-grade and 30 high-grade gliomas) using nanoparticle tracking and flow cytometry. Real-time PCR quantified differential gene expression. Molecular docking identified natural compounds, including gallocatechin, targeting TGF-β1. In vitro experiments were conducted using U87MG and MDA-MB-231 cell lines to assess the effects of exosomes and drug candidates on cell proliferation, migration, invasion, and apoptosis.

Results and Conclusions: The analysis identified CD44, TGF-β1, VEGFA, THBS1, and SERPINE1 as key upregulated genes, contributing to glioblastoma progression. Gallocatechin showed significant inhibitory effects on TGF-β1-induced proliferation, migration, and invasion, with an IC50 of 30.53 µM. Exosomal uptake assays in breast cancer cells revealed a 90% internalization rate within 24 hours, promoting proliferation and metastasis. Exosomes also induced EMT-related gene expression, with VIMENTIN identified as a central regulatory marker.

This study validates liquid biopsy as a reliable approach for diagnosing and monitoring glioblastoma by identifying circulating exosomal biomarkers. The role of TGF-β1 in glioblastoma progression and the therapeutic potential of gallocatechin were established through molecular docking and in vitro studies. Furthermore, tumor-derived exosomes were demonstrated to significantly enhance metastatic potential in breast cancer cell models. These findings underscore the importance of developing exosome-based diagnostics and therapeutics for brain tumors, paving the way for personalized medicine approaches.

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Abstract ID: 35

Elevated ferritin levels suppress has-miR-6743-5p expression and aid in upregulation of hepcidin among Transfusion Dependent β-Thalassemia

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Background/ Introduction: MicroRNAs (miRNAs) are small regulatory RNAs have been emerged to exhibit wide regulatory activities on gene expression involved in cell proliferation, differentiation and development. MiRNAs are also reported to regulate iron homeostasis by targeting the iron transporters and membrane channels. Iron homeostasis is stringently monitored at both duodenal absorption and splenic salvage by hepatic hormone hepcidin, which is under the influence of serum ferritin levels and erythropoiesis.

Objectives: In this study, we aim to define the key regulatory miRNAs involved in hepcidin secretion under inherited blood disorder of β-thalassemia and functionally annotate the candidate miRNAs involved in erythroferrone/Hepcidin axis.

Materials and Methods: Whole transcriptome sequences of 5 transfusion dependent thalassemia (TDT) and 3 healthy controls were downloaded from Sequence Read Archive and fished for the identification of miRNAs targeting hepcidin/Ferroprotein axis and erythropoietin (EPO)/erythroferrone (ERFE)/ hepcidin axis. The miRNA expression validation was carried out using healthy control and patient samples by quantitative polymerase chain reaction (qPCR). To define the effect of ferritin on miRNA expression, hepatic cell lines HuH7 were treated with rERFE, bone morphogenetic protein 6 (BMP6) and Deferasirox (DFX). miRNA was isolated using miRNA easy kit, followed with cDNA conversion using TaqMan advanced cDNA synthesis kit and the relative quantification was determined using 2-ΔΔcT

Results and Conclusions: Through miRDeep2 analysis, we were able to identify 470 conserved miRNAs belonging to 430 miRNA families. The digital gene expression (DGE) study predicted 10 miRNAs targeting the Hepcidin/FPN and EPO/ERFE/ Hepcidin axis were differentially expressed. DGE indicated 3fold suppression of hsa-miR-6743-5p among patient samples. The qPCR studies suggested the suppression of miRNAs among the patient samples with increase in serum ferritin levels, while its target gene hepcidin was induced relatively. To validate, whether ferritin impacts miRNA expression, HuH7 cells were treated with rERFE, BMP6 and DFX. We observed notable suppression of hsa-miR-6743-5p in BMP6 treated cells inducing the hepcidin synthesis while, rERFE and DFX induced the miRNA relatively suppressing the hepcidin synthesis.

This study delineates how serum ferritin levels induce the expression of hepcidin under high erythropoietic suppressive effects. The suppression of hsa-miR-6743-5p leads to induction of hepcidin under higher ferritin concentrations, thus aiding in obstructing the iron absorption reducing hepatic iron overload.

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Abstract ID: 36

Computational Identification and Characterisation of Novel miRNAs Involved in β-thalassemia

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Background/ Introduction: β-thalassemia is a genetic disorder characterised by reduced or absent β-globin chain production, leading to chronic anaemia. Increasing fetal haemoglobin (HbF) levels has been suggested as a therapeutic approach to ameliorate the clinical severity of β-thalassemia. MicroRNAs (miRNAs) are small, non-coding RNAs that regulate genes involved in HbF production. Integrative strategies that combine results from bioinformatics analysis and expression profiling help reveal novel miRNAs in β-thalassemia.

Objectives: Computational identification and characterisation of novel miRNAs and identification of their target genes, which are involved in regulating HbF levels and oxidative stress. To validate the expression of these novel miRNAs among patients and healthy controls.

Materials and Methods: miRDeep2 pipeline was employed to predict novel miRNAs using publicly available miRNA sequencing datasets from Five β-thalassemia patients and five healthy controls. miRNA gene characterisation and phylogenetic analysis were performed using Phyllip3.0 software. miRNA targets were predicted using sequence-based target prediction algorithms such as TargetScan and miRDB. Gene ontology was carried out to functionally annotate the predicted targets. Digital expression analysis was conducted using limma and edgeR packages of the R statistical tool. Cytoscape was used to study the interactions between miRNAs and target genes. Further, the expression profiles of the candidate novel miRNAs were validated using in-house patient samples and healthy controls.

Results and Conclusions: miRDeep2 identified a total of 1122 potent miRNA sequences, out of which 396 were known miRNAs and 726 were novel miRNAs. With MFE cut off of ≤ -15 kJ/mol, 238 novel miRNAs were screened to form a stable RNA hairpin. The identified novel miRNAs were isoforms and homologous to the miR54 family. Our target prediction algorithms resulted in six novel miRNAs targeting BCL11A, KLF1, ZBTB7A, NRF1 and HMGA1, which are evidenced to regulate HbF levels and oxidative stress. Quantitative PCR studies revealed that novel miRNA, mirN5, is differentially expressed. among the control and patient samples. miRN5 is downregulated, with its target KLF1 being overexpressed.

Suppression of miRN5 prevents the degradation of KLF1, resulting in its overexpression. Further studies are essential to understand the off-target effects of miRN5 and confirm its role as a potential target to induce the synthesis of HbF levels.

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Abstract ID: 37

Unraveling Shared Molecular Mechanisms between Schizophrenia and Type-2 Diabetes Mellitus through Advanced Weighted Gene Co-Expression Network Analysis

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Background/ Introduction: Schizophrenia (SCZ) is a complex psychiatric condition marked by episodic psychosis, abnormal behavior, and cognitive dysfunction. People with schizophrenia have a higher suicide

rate and have 20 years shorter life expectancy than normal people. The global frequency of SCZ is close to 1%, with a higher incidence among young adults. Type 2 diabetes mellitus (T2DM) is another complex disease that requires strict glycemic control and continuous medical care. The prevalence of T2DM in the adult population has been rising gradually and more so in the middle and low-income countries. Past studies have associated SCZ with increased risk for T2DM, leading to increased cardiovascular risk and a shortened life expectancy. The prevalence of T2DM among people with SCZ ranges 2–5 fold higher than in the general population.

Objectives: The aim is to investigate the co-expression gene modules associated with Schizophrenia (SCZ) and Type 2 Diabetes Mellitus (T2DM) using Weighted Gene Co-expression Network Analysis (WGCNA).

Materials and Methods: We performed a comprehensive weighted gene co-expression network analysis using publicly available gene expression datasets from 26 patients (18 SCZ and 8 T2DM) and 20 healthy individuals to identify shared biomarkers underlying two diseases. Our intensive analysis identified important modules and genes linking SCZ and T2DM. We conducted a protein-protein interaction (PPI) network analysis of the identified genes. To further understand the biological significance of the identified genes, we performed functional enrichment analysis. Additionally, we employed Area Under the Curve (AUC) analysis to assess the importance of these genes.

Results and Conclusions: Our intensive analysis identified important modules, hub genes and functionally enriched pathways linking SCZ and T2DM. Notably, turquoise module demonstrated the highest correlation with both SCZ ($r = 0.73$, $p = 1\times10\text{-}5$) and T2DM ($r = 0.78$, $p = 5\times10\text{-}4$). This strong association suggests that the genes within the module may be involved in common biological pathways that contribute to the pathophysiology of both disorders. Moreover, our protein-protein interaction network analysis identified genes having crucial role in cellular functions in both nervous system as well as pancreas. These findings are well supported by functional enrichment analysis revealing key processes and pathways including cellular response, regulation, organisation and signalling to be significantly enriched in both SCZ and T2DM. Furthermore, the AUC analysis revealed hub genes underscoring the potential commonalities in the pathogenesis of these seemingly disparate conditions. The identification of 8 hub genes, strongly associated with both SCZ and T2DM, offers promising biomarkers for early diagnosis and monitoring of disease progression. In conclusion, our extensive data integrative approach provides valuable insights into the molecular interconnections between SCZ and T2DM. Our study provides important insight in uncovering the underlying complex shared links between the diseases.

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Abstract ID: 38

ScInfeR: an efficient method for annotating cell types and sub-types in single-cell RNA-seq, ATAC-seq, and spatial omics

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Background/ Introduction: Cell-type annotation is a major challenge in single-cell and spatial omics analysis. Here, we present ScInfeR, a graph-based cell-type annotation toolkit for single-cell RNA sequencing (scRNA-seq), single-cell ATAC-seq (scATAC-seq), and spatial omics. Existing tools lack accuracy and usability in cell-type annotation tasks as they use either a predefined marker set or scRNA-seq reference.

Objectives: ScInfeR provides improved accuracy in cell-type annotation by efficiently using either individual or combined information from the marker set and scRNA-seq reference.

Materials and Methods: ScInfeR allows user defined weighted markers for cell-type assignments. Moreover, ScInfeR accurately detects cell sub-types in a hierarchical manner. Our extensive benchmarking across multiple single-cell and spatial datasets, evaluating 10 existing tools, demonstrated superior performance of ScInfeR in more than 100 cell-type prediction tasks. Noteworthy, ScInfeR seems robust against batch effect arising in these datasets.

Results and Conclusions: Our toolkit combined with single cell database (https://scinfer.shinyapps.io/ScInfeR/) offers a cell marker and scRNA-seq reference database for seamless cell-type annotation in single-cell and spatial omics.

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Abstract ID: 39

NLRP3 and AIM2 inflammasome gene expression in response to IFNα in Systemic Lupus Erythematosus (SLE)

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Background/ Introduction: SLE is an autoimmune disease with heterogeneous clinical manifestations, disease course and prognosis. Type I interferons (IFNs) are known to be mediators of SLE pathogenesis and can induce expression of canonical inflammasome genes, including NLRP3 (NOD-like receptor family, pyrin domain containing 3) and AIM2 (Absent in Melanoma 2). Inflammasomes are multi-protein complexes which oligomerise in the cytosol to trigger innate inflammatory responses against PAMPs/ DAMPs which are sensed by PRRs and are key mediators of inflammatory responses leading to increased production of pro-inflammatory cytokines, such as IL-1β and IL-18. Upregulation of NLRP3 and AIM2 in response to IFNα elevates inflammation and promotes immune cell activation, contributing to SLE pathogenesis.

Objectives: To compare AIM2 and NLRP3 inflammasome gene expression in SLE patients and healthy controls in response to IFNα.

Materials and Methods: SLE patients (n=30) fulfilling ACR criteria attending Rheumatology OPD from Department of Medicine KEMH, Parel, Mumbai and healthy controls (n=30) were recruited in this study. Primary monocytes were isolated from SLE patients and healthy controls by negative selection. Monocytes were treated with inflammasome activators LPS & Nigericin in the presence or absence of IFNα (100ng/ml). IL-1β and IL-18 secretion was measured by ELISA. Expression levels of inflammasome genes were assessed by TaqMan probe qPCR assay. mRNA levels of NLRP3and AIM2 were normalised to beta actin.

Results and Conclusions: Results: Gene expression of AIM2 and NLRP3 were found significantly higher ($p<0.05$) in SLE monocytes than healthy control (HC) monocytes. SLE monocytes had increased production of IL-1β and IL-18 by AIM2 and NLRP3 inflammasome as compared to HC. Monocytes significantly demonstrated increased IL-18 ($p=0.002$) and IL-1β ($p=0.001$) levels (pg/ml) on o/n exposure

to IFNα as compared to untreated cells. Altered expression of inflammasome was associated with lower platelet count, hemoglobin level and the use of hydroxychloroquine in SLE patients.

Conclusions:

This study indicated that AIM2 and NLRP3 inflammasome gene expression was correlated with clinical features of SLE suggesting role of inflammasome activation in SLE disease pathogenesis. Targeting these pathways may offer a therapeutic approach for managing inflammation and disease progression in SLE patients for translational approach.

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Abstract ID: 40

Repurposing the TGF beta inhibitors to rescue the expression of hepcidin to combat iron overload among β- thalassemia patients

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Background/ Introduction: Iron overload forms the second comorbidity factor among β-thalassemia caused due to increased serum and liver ferritin levels. The deposition of iron at the secondary sites such as cardiac and endocrine glands lead to organ malfunction and failure. It is demonstrated that erythroferrone, with the aid of growth differentiation factor15 (GDF15) and twisted gastrulation1 (TWSG1) inhibits the binding of bone morphogenetic protein6 (BMP6) to its receptor BMP6R and intervenes the hepcidin synthesis.

Objectives: Inhibiting the binding of TWSG1 on the BMP6R forms a promising approach to rescue the expression of hepcidin under ineffective erythropoiesis. In this view, we propose to screen TGFb inhibitors which can affectionately bind the TWSG1 using in silico approaches and validating their influence on hepcidin expression using invitro cultures of HuH7 cells.

Materials and Methods: To identify potential drugs that can inhibit the binding of TWSG1 to the BMP6R. Through in-silico approaches, a library comprising of 3500 molecules belonging to 30 different families of transforming growth factor β (TGFβ) inhibitors were docked against the TWSG1 protein. Candidate molecules were considered with the binding energies greater than -8 kJ/mol. The immortalised hepatic cell lines HuH7 were treated with the selected drugs and the cytotoxicity was determined using sulforhodamine B (SRB) assay. The hepcidin expression in treated and untreated cells was profiled using quantitative real time PCR using 2-ΔΔcT method.

Results and Conclusions: The docking results suggested more than 16000 conformations with wide range of binding affinities. Considering the binding affinity cutoff of -8kJ/mol, top 10 molecules were further subjected for simulation studies. Of the selected molecules, Galunisertib with its many favourable conformations exhibited higher number of hydrogen bonds with the target and bound internally modulating the conformations of the TWSG1. While on the other front, chrysin exhibited minimal hydrogen bonds with the target protein. The cytotoxicity assay demonstrated galunisertib has higher impact on cell growth with LD50 being 3.6 nM at 48 h while chrysin exhibited lower toxicity with LD50 3 ×105 nM. The quantitative expression studies revealed 1.5fold induction of hepcidin at 0.36 nM of galunisertib and 0.5fold by 100mM of chrysin.

This study provides a novel approach to rescue the expression of hepcidin to combat the iron overload. It also demonstrates small concentrations of galunisertib efficiently sequestered TWSG1 and rescued the expression of hepcidin. Both the TGFβ inhibitors can aid in efficient management of iron overload.

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Abstract ID: 41

EGFR variant 8 is overexpressed and associated with poor prognosis in oral cancer

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Background/ Introduction: Epidermal growth factor receptor is often deregulated in cancer and transcribed as common EGFR variant 1 whereas variant 8 have never been well-studied. Aim of this study was to investigate the role of EGFR variant 8 in oral carcinogenesis.

Objectives:

Epidermal growth factor receptor (EGFR)- often deregulated in cancer

Among the eight different EGFR isoforms, the regulation of full-length variant 1 is well-known- variant 8 have never been studied

This study aimed to understand the function of EGFR super-enhancer loci and its associated oncogenic transcription factors regulating the expression of EGFR variant 8

Materials and Methods: We examined 48-OSCCs and 8-normal tissues for gene expression using RT-qPCR. Visualized the regulatory elements and epigenetic modifications and chromatin loop formation by UCSC. Analysis of eRNA profiles, eRNA/Hi-C interactions, and eRNA-TF factors was performed using The Cancer eRNA-Atlas.

Results and Conclusions: Overexpression of EGFR variant 8 and its transcription was more prevalent than variant 1 and positively correlated with the EGFR-AS1expression in oral cancers. Notably, EGFR variant 8 overexpressed patients showed shorter overall survival than variant 1. Clustered interactions between CE1, CE2, and EGFR-AS1 may regulate the expression of both EGFR-eRNA and variant 8. TCGA-eRNA analysis showed the enrichment of eRNA-specific marks, eRNA synthesis, and their correlation with variant 8. Moreover, SNAI2 is likely to modulate EGFR-AS1 and EGFR-eRNA expression with YY1 acting as a bridging complex.

Abstract ID: 42

Genomic Insights and Functional Analysis in Young Familial Myocardial Infarction.

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Background/ Introduction: Coronary artery disease (CAD) and myocardial infarction (MI) are significant global health concerns, increasingly affecting younger individuals. Conventional risk factors, such as smoking, diabetes, and hypertension, are well-documented for older patients. However, they do not fully explain the prevalence of early-onset CAD and MI among people under 40. Growing incidence of MI in young age even without these risk factors highlights the need for a deeper understanding of genetic contributors to early-onset MI and potential therapeutic targets.

Traditionally seen as an older person's disease, CAD and acute MI are now alarmingly prevalent among young adults, particularly in India, where genetic diversity complicates the analysis of early-onset cases. Studies indicate that while 2-4% of young Indians have CAD, autopsy data reveals up to 50% of individuals under 40 show undiagnosed coronary atherosclerosis. This silent progression often results in sudden, unexpected cardiac events. Research points to genetic predispositions as crucial in these cases, with recent advances in next-generation sequencing (NGS) aiding the identification of potential genetic markers, though their pathogenic roles are still being validated.

Objectives: 1. Genetic Analysis: Study genomic variants in young MI patients to assess their contribution to MI and correlate findings with clinical outcomes, and 2. Functional Impact Evaluation: Determine the effects of identified variants, linking them to clinical phenotypes for targeted therapies.

Materials and Methods: 1. Whole Exome Sequencing: NGS identified gene variants associated with CAD pathology, with bioinformatics predicting pathogenicity.

2. Family Genetic Studies: Genetic analysis in families with strong MI history, using ELISA and RT-PCR for functional study of key variants.

3. Counseling and Analysis: Patient and family genetic counseling and extended family studies for novel variants.

Results and Conclusions: In our study of myocardial infarction patients under 40 years, and their families, we conducted detailed clinical and functional analyses to establish the relationship between specific genes, variants, and their impact on disease. Key findings revealed that ABCA1 and EPHX2 Variants associated with HDL deficiency and Tangier disease in patients with strong family histories of thrombosis and MI. TTN Variants in patients with sarcomere dysfunction, correlating with myocardial infarction and reduced Titin levels. TPM1 and FLNC Variants associated with hypertrophy, with functional tests showing decreased and increased protein levels indicative of potential pathogenic effects. GPIHBP1 and ACACB Variants associated with dyslipidemia and high cardiovascular risk, with alterations in lipid metabolism. LDLR Variants in cases of severe dyslipidemia, correlating with elevated LDL levels and strong familial MI history.

The study underscores the impact of genetic variants in lipid metabolism and sarcomere function on cardiovascular health in young MI patients. Variants in ABCA1, EPHX2, TTN, and TPM1 indicate a strong correlation with dyslipidemia and hypertrophy, respectively, suggesting these as potential diagnostic markers. Comprehensive genetic screening may thus provide valuable insights for personalized prevention strategies in high-risk young adults.

Abstract ID: 43

An optimized instrument variable selection approach to improve causality estimation in association studies

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Background/ Introduction: Mendelian randomization (MR) emerges as a promising tool for inferring causality in genetic epidemiology. However, the identification of robust genetic instrument variables (IVs) and horizontal pleiotropy pose challenges to the precision of causal inference.

Objectives: To improve the causality inference in MR studies, we introduce a robust integrative framework that adheres to the STROBE-MR checklist guidelines.

Materials and Methods: We demonstrate the effectiveness of our proposed approach on 5 different MR datasets selected based on their heritability, including total cholesterol (TC) - coronary artery disease (CAD) as a single-sample dataset and liver iron-content (LIC)- liver cell carcinoma (LCC) as a two-sample dataset. We implemented non-conventional t-statistics-based criteria to improve the reliability of selected IVs. accuracy of identified causal SNPs. Further, sensitivity analyses are included in our

pipeline to remove horizontal-pleiotropy bias. For validation purposes, we show functional enrichment results of identified causal SNPs obtained from 5 datasets.

Results and Conclusions: Comprehensive analyses reveal MR analyses with our pipeline clearly outperform their counterpart analyses using default parameters. Noteworthy, we found a significant association between TC and CAD ($P = 1.02 \times 10{-}22$) using our pipeline. Conversely, the same association was deemed to be ambiguous using MR analyses with default parameters (i.e., without our pipeline). Moreover, in a two-sample dataset, our pipeline identified 13 new causal SNPs with enhanced statistical significance ($P = 1.06 \times 10{-}11$) as compared to default parameters ($P = 7.58 \times 10{-}4$). Our pipeline could identify various critical genes such as ME1 and pathways (such as mixed hyperlipidemia) that are previously known to be significantly associated with outcome of interest. In conclusion, our proposed pipeline can effectively uncover causal associations between exposure and outcome across diverse populations while mitigating spurious causal associations and horizontal pleiotropy.

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Abstract ID: 45

An integrative methylome-transcriptome study reveals PRDM1 coordinates with JUNB to act as an epigenetic regulator in Psoriasis pathogenesis

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Background/ Introduction: Psoriasis is a chronic, inflammatory, immune-mediated skin disease characterized by uncontrolled proliferation and abnormal differentiation of epidermal keratinocytes. Although genetic and epigenetic regulation have been studied separately, an integrative approach to understand the transcriptional regulations contributing to the psoriasis pathogenesis is yet to be investigated.

Objectives: Our aim is to identify the transaction factors involved in epigenetic regulations in psoriasis by integrating methylome and transcriptomic studies from the psoriatic and adjacent normal skin tissues and validate their role in a psoriasis.

Materials and Methods: We have conducted RNA-seq and genome-wide DNA methylation profiling of 24 paired psoriatic and adjacent normal skin tissues. Integrative analysis identified transcription-factors regulating DNA methylation. ChIP-seq of predicted master regulators, PRDM1 and JUNB were performed in normal (Ker-CT) and psoriatic (Ker-CT cells upon 48 hrs of chronic stimulation with a cocktail of 4 cytokines) keratinocytes. ChIP-seq data were validated with ChIP-qPCR. To evaluate their function, both PRDM1 and JUNB genes were stably knocked down. ChIP-qPCR were performed for the regions bound by both PRDM1 and JUNB or individually in the PRDM1 and JUNB knocked-down control or psoriatic cells. Finally, we determined the methylation status on these regions through pyrosequencing in control as well as in PRDM1 and JUNB knock-down conditions.

Results and Conclusions: Integrating RNA-seq and DNA methylation data, we identified PRDM1 as a master regulator associated with hypomethylation near their binding sites. Further analysis revealed enrichment of AP1 transcription factors (eg. JUNB) at the nearby regions of PRDM1 binding sites. We performed ChIP-seq of both PRDM1 and JUNB in the normal and psoriatic keratinocytes. ChIP-seq data identified 1398 common, and 1767 & 33404 unique binding sites for PRDM1 and JUNB, respectively. To understand the interplay between PRDM1 and JUNB, we performed ChIP-qPCR for common and unique binding sites of PRDM1 and JUNB in PRDM1 and JUNB knockdown Ker-CT as well as control cells. ChIP-qPCR data revealed that binding of PRDM1 decreases in JUNB knockdown cells. Similarly binding of JUNB was also decreased in PRDM1 knockdown Ker-CT cells. On the contrary, this interdependency was not observed for the unique binding regions. PRDM1 and JUNB binding regulated the expression of their downstream genes. Finally, we have observed hypermethylation in the PRDM1 and JUNB common binding sites in PRDM1 and JUNB knockdown Ker-CT compared to control cells, suggesting the involvement of PRDM1 in causing hypomethylation near their binding sites and interacting with AP1 transcription factors (JUNB) to regulate the genes involved in psoriasis pathogenesis.

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Abstract ID: 46

Association of miR196a2 rs11614913 Polymorphism with Breast Cancer Risk

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Background/ Introduction: Breast cancer is the most commonly diagnosed cancer in females and is the leading cause of cancer deaths worldwide. MicroRNAs (miRNAs) are small non-coding RNA molecules that regulate gene expression by degrading mRNAs or suppressing translation. Single nucleotide polymorphisms (SNPs) in the miRNA coding genes may alter their expression and maturation, imbalance cellular homeostasis, and promote tumor formation.

Objectives: To investigate the association of miR196a2 rs11614913 C/T polymorphism with breast cancer risk in patients from Punjab, North-West India.

Materials and Methods: In this case-control study 50 breast cancer patients and 65 age-matched unrelated healthy controls were analyzed. DNA samples of patients and controls were screened for miR196a2 rs11614913C/T polymorphism using the polymerase chain reaction-restriction fragment length polymorphism method. Genotype and allele data was compared between patients and controls.

Results and Conclusions: The mean ages of breast cancer patients and controls were 51.08±13.46 and 52.8±14.32 years respectively. The frequency of CC, CT, and TT genotypes was 34% vs 35.4%, 46% vs 52.3%, and 20% vs 12.3% in patients and controls respectively. The frequency of C and T allele was 57% vs 61.5 % and 43% vs 38.5% in patients and controls respectively. No significant association of miR196a2 rs11614913 C/T polymorphism was found with breast cancer risk in the studied patients (p>0.05).

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Abstract ID: 47

VEGFR2-604T/C Promoter Polymorphism and Breast Cancer Risk in Patients from Punjab

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Background/ Introduction: Breast cancer is a complex disease characterized by uncontrolled growth and spread of cancerous cells that affect millions of women worldwide. Breast cancer cells need constant nourishment and oxygen supply for their continuous growth. Angiogenesis, an essential process in tumor growth provides potential routes for tumor dissemination and metastasis. VEGF is the endothelial cell mitogen that enhances permeability, angiogenesis, proliferation, migration, differentiation and capillary formation. VEGF family performs its function by binding to VEGFR1, VEGFR2 and VEGFR3 receptors. VEGF-A binding to VEGFR2 leads to receptor dimerization, protein kinase activation, trans-auto phosphorylation and initiation of signaling pathway. Polymorphisms in VEGFR2 might alter gene expression, amount of circulating VEGFR2 levels and the efficiency with which VEGF binds to the receptor.

Objectives: The aim of this case-control study was to find the association of VEGFR2-604T/C promoter polymorphism with breast cancer risk in patients from Punjab, North-West India.

Materials and Methods: In this case-control study, the genomic DNA of 50 breast cancer patients and 65 age matched healthy unrelated controls were screened for VEGFR2-604T/C (rs2071559) promotor polymorphism using polymerase chain reaction-restriction fragment length polymorphism method. Genotypic and allelic data was compared between patients and controls.

Results and Conclusions: The mean ages of patients and controls were 53.60±11.26 and 54.07±11.06 years respectively. The frequency of T allele was 54% vs 46.2% and C allele was 46% vs 53.8% in patients and controls respectively. The frequency of TC genotype was 52% vs 55.4%, TT genotype was 28% vs 18.5% and CC genotype was 20% vs 26.1% in patients and controls respectively. There was no association of VEGFR2-604T/C polymorphism with breast cancer risk in the studied population.

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Abstract ID: 48

Synonymous and nonsynonymous novel mutations in COMT gene in Schizophrenia patients: In case-control in Tumkur district in South Karnataka

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Background/ Introduction: COMT gene is a candidate gene for schizophrenia located in 22q11.2, which encodes a catechol-O-methyltransferase enzyme that degrades various catechol and neurotransmitter pathways. Schizophrenia is a devastating mental illness caused by gene mutations. Gene variations that alter gene activity and protein functions can influence different traits in an organism. Gene variants can affect patients and healthy individuals. The novel mutation rate is insufficient compared to common variants but does not reach zero rates. Novel mutations are more challenging in present generations. Novel mutations are not only disease causes but also sometimes beneficial to disease resistance, which may cause other health issues. Novel mutations are especially vulnerable to diseases, which alter protein functions in the body and alter cell functional activity. Otherwise, novel mutation actions are similar to pathogenic variants. Novel mutations may burden disease or increase the risk and may be resistant to diseases, which may benefit the trait. Gene variability plays a significant role in life processes and survival.

Objectives: To study novel mutations in exon-3 in Schizophrenia patients and healthy individuals of Tumkuru District, South Karnataka.

Materials and Methods: We conducted a case-control study of 120 subjects (60 samples of schizophrenia patients and 60 samples in the healthy group in the Tumkuru district of different age groups among both genders. We performed PCR amplification, and capillary sequencing, bioinformatics software tools.

Results: We found four novel mutations of the COMT gene Exon-3.A>T Q85L [nonsynonymous] in positive symptoms, C>T N39N [Synonymous] in negative symptoms, in healthy group G>C W38C, [nonsynonymous].

Conclusion: The results showed novel mutations that could help us understand the symptoms and severity of the phenotype effect, which may affect future health compliance.

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Abstract ID: 49

miR-605 rs2043556 A/G Polymorphism and Breast Cancer Risk in Patients from Punjab: A Case-Control Study

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Background/ Introduction: Breast cancer is the second most frequent malignancy reported among females globally. Tumor microenvironment-mediated epigenetic perturbations contribute to the development of neoplasia. MicroRNAs (miRNAs) are small non-coding RNAs with length ranging upto 22 nucleotides. Mature miRNAs interacts with 3'UTR of the target mRNA which results in suppressed

expression. miR-605 acts to interrupt p53:Mdm2 interaction and create a positive feedback loop aiding rapid accumulation of p53, facilitating its function in response to stress.

Objectives: The aim of the study was to find the association of miR-605 A/G polymorphism with Breast Cancer risk in patients from Punjab, North West India.

Materials and Methods: In this case-control study DNA samples of 50 breast cancer patients and 65 age matched healthy unrelated controls were analyzed. The DNA samples were screened for miR-605 A/G polymorphism using polymerase chain reaction-restriction fragment length polymorphism (PCR-RFLP) method. The data was statistically analyzed.

Results and Conclusions: The mean age of breast cancer patients was 51.88 ± 11.13 years and of controls was 49.86 ± 15.83 years. The frequency of AA genotype was 56% vs 67.7%, AG genotype was 34% vs 30.8% and GG genotype was 10% vs 1.5% in patients and controls respectively . The frequency of A allele was 73% vs 83.1% and G allele was 27% vs 16.9% in patients and controls respectively. There was no association of any of the genotype of miR-605 A/G polymorphism with breast cancer risk ($p>0.05$), however G allele was marginally associated with breast cancer risk in the studied subjects (OR = 1.82, 95% CI = 0.96 - 3.43, p = 0.07).

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Abstract ID: 50

Multi-Modal Analysis of Breast Camcers Using Paired SCRNA-WXS Sequencing Reveals Driver Mutation Effects on Tumor Microenvironment

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Background/ Introduction: Most tumors harbor somatic alterations in multiple driver genes giving rise to genetic intra-tumor heterogeneity (gITH). which drives tumor progression. Recent advancements in scRNA-sequencing technologies have highlighted the heterogeneity of the tumor microenvironment (TME) in unprecedented detail (Gavish et. al., 2023). Multiple studies have associated genomic alterations with the immune infiltration status. However, it is yet unknown how the underlying driver mutations affect the cellular reprogramming in malignant cells and dictate their interactions with the surrounding cells.

Objectives: To characterize the somatic mutational landscape and transcriptional diversity of tumors from paired whole exome DNA and single-cell RNA sequencing data and delineate the role of somatic mutations in regulating the transcriptional landscape in tumors.

Materials and Methods: We obtained paired scRNA and WXS data for 11 breast cancer patients from a previously published study. We analyzed the WXS and scRNA data using our in-house analytical pipelines to identify somatic alterations as well as different cell types and cell states in tumors respectively. We used advanced statistical and computational approaches to delineate the complex interplay between genetic and cellular heterogeneity in those tumors.

Results and Conclusions: We identified somatic SNVs and indels in driver genes, like, TP53, PIK3CA, PIK3R1, ERBB2, in 11 breast cancers across different subtypes using paired WXS data from tumor and blood. Using scRNAseq data we identified various cell types, like, T cells, B cells, etc. in the tumor microenvironment of these 11 tumors. We note widespread heterogeneity, both genetic and transcriptional in the TME of these tumors. Our analysis reveals that the driver gene mutation profile in tumors dictate the transcriptomic diversity in the TME.

Abstract ID: 51

IL-18 promoter region gene variations (-607 C>A AND -137 G>C) and gene expression in PCOS: A study from India.

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Background/ Introduction: Polycystic Ovary Syndrome (PCOS) is identified as a multifaceted disorder affecting reproductive health and family history, significantly impacting women's quality of life. PCOS is associated with metabolic traits such as insulin resistance and obesity. Insulin resistance is crucial in developing type 2 diabetes, while obesity aggravates the cardiovascular issues linked with PCOS. Research indicates that women with PCOS consistently show elevated IL-18 levels. Genetic variations in the IL-18 gene promoter region, particularly at positions -607 (rs1946518) and -137 (rs187238), affect IL-18 expression by influencing transcription factor binding. Prior polymorphism studies mainly focused on Chinese and Korean PCOS patients.

Objectives: Examine and establish a relationship between IL-18 gene expression and promoter region polymorphisms (-607 rs1946518 and -137 rs187238) in PCOS.

Materials and Methods: We recruited subjects (121 patients and 121 controls) and categorized BMI into lean, normal, overweight, and obese. Expression analysis was conducted using real-time PCR and genotyping using ARMS-PCR. The results were subsequently validated through Sanger Sequencing.

Results and Conclusions: In the study PCOS subjects with a normal BMI showed notably higher frequencies of the C allele (p=0.0251) for the IL-18 gene -137 polymorphism, which is associated with a 1.94-fold increased risk of PCOS compared to the G allele (OR 1.9496, 95% CI = 1.0818 - 3.5133 and p=0.0263). Haplotype association analysis revealed that PCOS study subjects with normal BMI had a significantly higher frequency of the -607A/-137C haplotype, corresponding to a 1.87-fold increased risk (OR 1.8768, 95% CI = 1.1615 - 3.0326 and p = 0.0101). IL-18 expression was significantly up-regulated in PCOS patients, with a fold change of 1.87. The C allele at position -137 and the -607A/-137C haplotype in the promoter region of the IL-18 gene present a potential risk for PCOS in subjects with normal BMI. Additionally, IL-18 expression is associated with polymorphisms in the IL-18 gene promoter in PCOS patients.

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Abstract ID: 52

Gene Expression Analysis of Pro-inflammatory Cytokine (TNF-α) Gene in Major Depressive Disorder

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Background/ Introduction: The pathogenesis of major depressive disorder (MDD), which is known to be associated with neuroinflammation, has been linked to the pro-inflammatory cytokine tumor necrosis factor-alpha (TNF-α). Elevated levels of TNF-α may intensify immune response and contribute to altered

neural circuits associated with mood regulation. This study aimed to examine the gene expression of TNF-α in individuals with MDD and assess its potential role in the disease's pathogenesis. Genetic studies have shown that specific alleles associated with higher TNF-α expression may predispose individuals to an amplified inflammatory response, which could contribute to the onset or exacerbation of depressive symptoms. Elevated TNF-α levels may, in turn, lead to neuroinflammatory effects, impacting brain regions associated with mood and cognitive regulation, such as the prefrontal cortex and hippocampus.

Objectives: To study the expression of Pro-inflammatory cytokines - TNF α.

To correlate the expression levels with clinical parameters to determine the association of TNF α with MDD.

Materials and Methods: We included 50 patients diagnosed with MDD according to Hamilton Depression Rating Scale (HAM-D) and 50 healthy controls matched for age and gender. Total RNA was extracted from blood samples, and quantitative reverse transcription PCR (qRT-PCR) was conducted to measure TNF-α gene expression levels.

Results and Conclusions: The Ct values were estimated for all the samples after data normalization with endogenous control. To understand the differential expression among case and control fold changes was calculated using 2(-ΔΔCT) method. The level of significance in the fold change was calculated using student t-test and p- value less than 0.05 was considered statistically significant. Our results support the role of TNF-α in the pathogenesis of MDD and suggest that pro-inflammatory mechanisms may contribute to the disorder's development. Further research is needed to explore TNF-α as a potential biomarker for MDD and to understand its role in the broader neuroimmune profile of depression across diverse populations.

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Abstract ID: 53

CAG repeat instability and region-specific gene expression changes in the SCA12 brain

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Background/ Introduction: Spinocerebellar ataxia type 12 (SCA12) is a rare form of autosomal dominant cerebellar ataxia caused by an expansion of (CAG)n in the 5′ of the PPP2R2B gene on chr5q32. SCA12 is relatively common in India. The illness often manifests late in life, with a variety of neurological and psychiatric symptoms, suggesting involvement of different brain regions. An autopsy study of SCA12 reported prominent neuronal loss and atrophy in the cerebellum. In Huntington's disease (HD), it is known that the size of the expanded CAG allele in HTT varies across neuroanatomical regions of the brain and influences the nature and severity of symptoms. In SCA12, the patterns of CAG repeat instability, gene methylation and transcription, and protein expression across different brain regions remain unexplored.

Objectives: To address this, we estimated the CAG instability index and immunohistochemistry of PPP2R2B in different brain regions of a SCA12 patient. We also examined the gene expression and DNA methylation patterns of PPP2R2B and related genes.

Materials and Methods: Tissue samples were derived from the post-mortem brain of a SCA12 patient, donated to the Human Brain Tissue Repository at NIMHANS, Bangalore. Genomic DNA from the cerebellum, frontal pole, superior temporal gyrus, temporal pole, entorhinal cortex, and spinal cord of the SCA12 patient and two controls were examined for CAG repeats using the Gene Mapper software. RNA sequencing was performed using Illumina NovaSeq, and hierarchical clustering was used to examine the normalised expression values. DNA methylation was profiled using the Infinium MethylationEPIC array, and analysed using the Illumina GenomeStudio software. Immunohistochemistry was performed with anti-PPP2R2B antibody on FFPE cerebellum sections from the patient and controls.

Results and Conclusions: Expanded alleles at the SCA12 locus are prone to somatic mosaicism. Lower CAG repeat instability and reduced PPP2R2B expression were noted in the SCA12 cerebellum compared to other regions. The predominance of neurons over glial cells in the cerebellum, along with strong DNA maintenance, may contribute to this reduced repeat instability. Additionally, the expression of key cell cycle genes that encode proteins directly interacting with PPP2R2B – such as YWHA proteins, which normally inhibit nuclear translocation of cell cycle proteins – is lower in the SCA12 cerebellum than in controls, where these genes are highly active. This reduction suggests a disruption in cell maintenance processes. Correspondingly, immunohistochemistry shows nuclear, in addition to cytoplasmic localization of PPP2R2B in the surviving Purkinje neurons of the SCA12 cerebellum. Hence, somatic mosaicism is an integral component of SCA12 pathobiology and controlling it by modulating gene expression can be explored as a treatment modality.

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Abstract ID: 54

Whole Exome sequencing for assessing the genetic heterogeneity in Aplastic anaemia in eastern region of India

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Background/ Introduction: Aplastic anaemia (AA) is a haematological disorder where a hypocellular bone marrow provokes peripheral pancytopenia. Haematopoietic stem cell transplantation (HSCT) andor immunosuppressive therapy (IST) with AGT and cyclosporine A (CsA) are the only available therapy for a cure. Next Generation Sequencing (NGS) has revealed various genomic factors to be responsible for the heterogeneous nature of the disorder but studies focusing on the eastern Indian cohort are rare.

Objectives: Our study aims to recognize genetic mutations that might be considered biomarkers for AA in eastern cohort of India.

Materials and Methods: In this study, we tried to investigate genetic variations in AA from the eastern region of India. Whole Exome sequencing was performed on a total of 19 AA cases (n=19) and 5 healthy subjects (n=5) recruited with their informed consents.

Results and Conclusions: Only those exonic variants that were exclusively present in case cohort but absent in healthy subjects were considered pathogenic variations responsible for the disease pathogenesis. A missense variant in TMEM260 (exon6:c.733G>T) gene was detected in one cluster of cases. This

variant was detected in a subset of 36.84% (7/19) cases. According to 1000 Genomes, the TMEM260 variant is rare in in east Asian population with a frequency of 0.0040 whereas common globally. This variant is predicted as probably damaging with a score of 1.000 in PolyPhen-2. Another missense variant in NPHS1 (exon24:c.3230A>G) was detected in another subset of 31.58% (6/19) of cases and is a rare variant in east Asian population with a frequency of 0.0198. This variant is reported in ClinVar as benign. WDR62 (exon28:c.3401T>G) was found in third subset of 21.05% (4/19) cases and is reported in ClinVar as being benign. However, this is a common variant in Asian population. GRHL3 (exon3:c.83G>A) was detected in only fourth subset of 2% (2/19) of cases but not reported in ClinVar. PolyPhen-2 predicts the variant to be probably damaging with a score of 1.000. According to 1000 Genomes, gnomAD, this variant is very rare both globally and in Asia.

Overall, our analysis highlights the value of integrating genomic heterogeneity that drives the development and progression of aplastic anaemia. This might offer a route to optimize global access to genetically informed aplastic anaemia care.

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Abstract ID: 55

To Study the therapeutic efficacy of Annona muricata (Lakshmanaphala) fruit extract for Oral cancer and identify molecular targets

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Background/ Introduction: Oral cancer poses significant health risks globally, emphasising the need for novel treatment options. This study investigates the therapeutic potential of Annona muricata (Lakshmanaphala) fruit extract and Silver nanoparticles synthesised from the fruit extract for oral cancer treatment, focusing on molecular targets. By evaluating Annona muricata acetogenins through computational approach this study aims to elucidate its anticancer properties. Silver nanoparticles offer several advantages that can enhance the anticancer properties of Annona muricata fruit extract. Firstly, their small size enables them to penetrate cancer cells more effectively. Secondly, silver nanoparticles possess inherent cytotoxicity against cancer cell lines. Additionally, silver nanoparticles exhibit synergistic effects when combined with natural compounds that lead to enhanced anticancer efficacy. This study aims to evaluate and compare the anticancer activity of the crude extract of Annona muricata fruit and its nanoparticles on Oral cancer cell line (KB cell line). Additionally, gene expression analysis of oral cancer cell lines treated with AgNPs will provide insights into its mechanisms of action.

Objectives: 1) Evaluation of Annona muricata acetogenins as a potential anticancer agent through computational approach, 2) Synthesis of silver nanoparticles from Annona muricata Fruit extract, 3) Gene expressions analysis of oral cancer cell line treated with Annona muricata Fruit extract.

Materials and Methods: Fruit extract prepared using soxlet was subjected to GCMS for phytochemical analysis which was followed by green synthesis of silver nanoparticles. Characterization of Main Functional Groups Present in the AgNPs was carried out using FTIR. Antiproliferative activity was done to determine the percentage of cell viability was assessed for different concentrations of AgNPs and methanolic fruit extract by using MTT assay. The transcriptional level of the genes BCL2, BAX, EGFR,p53 was carried out. Compounds having antiproliferative, antioxidant or apoptotic activity were chosen for molecular docking by Dynamic simulation using Desmond-Schrodinger.

Results and Conclusions: Around 60 compounds were identified from four highest peaks of GC-MS chromarogram, exhibiting various phytochemical activities. Compounds having antiproliferative, antioxidant or apoptotic activity were chosen for molecular docking. The ligands were docked with the cancer receptors: p53, EGFR, Bcl2 and Bax. Myrecitin, anonaine,neochlorogenic acid and kaempferol were the primary ligands found to have high binding affinity towards the cancer receptors with varying values between (-6 to -9 kcal/mol). The simulation results reveal that out of four protein-ligand complexes, three (Bcl2- anonaine, Bax- Neochlorogenic acid, p53- Kaempherol) displayed stable RMSD values ranging from 1 to 3 Å, indicating minimal structural deviation over the 100 ns observation period. MTT assay as a comparative study between the crude extract of the fruit vs combination of silver nano particles along with the fruit extract suggested a 5 fold increase in the efficacy of the fruit extract on addition of the nanoparticles. Gene expression analysis results reveal a decrease in the expression levels of bcl2 and EGFR and an increase in the expression levels pf p53 and Bax.

In conclusion, silver nanoparticles synthesized from Annona muricata (commonly known as soursop or graviola) show great potential as anticancer agents due to their unique properties and therapeutic benefits. These nanoparticles demonstrate strong cytotoxic effects against various cancer cell lines, making them a promising addition to anticancer treatments. However, to fully leverage their potential, several critical areas of research and development must be addressed.

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Abstract ID: 56

Investigating the contributions of accessible chromatin regions in the epidermal keratinocytes during psoriasis pathogenesis.

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Background/ Introduction: Psoriasis is a complex multifactorial skin disease manifested by the hyperproliferation and abnormal differentiation of keratinocytes. Apart from genetic components, several epigenetic regulations in psoriasis have been explored but the landscape of chromatin accessibility has not been studied in psoriatic keratinocytes.

Objectives: This study aims to investigate the accessible chromatin regions in psoriatic and adjacent normal epidermal keratinocytes. Epigenetic modifications are reversible, while psoriatic phenotype also gets reversed upon treatment. Thus, in-vitro maintenance of psoriatic keratinocytes concerning time and disease microenvironment may alter the chromatin accessibility landscape. To explore this, primary psoriatic keratinocytes, isolated from the psoriatic and adjacent normal skin tissues were cultured for three, six-, and eight days. Additionally, to validate a psoriatic model system, we also aim to identify the accessible chromatin regions in the normal and cytokine treated primary keratinocyte.

Materials and Methods: We conducted Assay for Transposase-Accessible Chromatin with Sequencing (ATAC-seq) with primary keratinocytes, isolated from psoriatic and adjacent normal skin tissues, cultured for three, six, and eight days respectively. Similarly, normal human primary keratinocytes and an immortalized primary keratinocyte cell line (Ker-CT) were cultured with and without chronic stimulation of four cytokines (5 ng/ml of TNF-α, 5 ng/ml of IL-6, 10 ng/ml of IL-17A, and 10 ng/ml of IL-1α) for 24

and 48 hours to mimic the psoriasis disease microenvironment. Subsequently ATAC-seq were performed from the normal and psoriatic primary and Ker-CT cells.

Results and Conclusions: ATAC-seq data from the psoriatic and normal primary keratinocytes after three days of culture identified differentially accessible chromatin regions that were linked to the psoriasis-specific hallmark biological processes, including keratinocyte proliferation, migration, and differentiation. Notably, these hallmark processes were almost absent after eight days of culture, suggesting a potential reversibility of the psoriatic phenotype and chromatin landscape over time when keratinocytes were cultured without a psoriatic microenvironment. Interestingly, chromatin-accessible regions in cytokines-treated primary keratinocytes and Ker-CT cells also showed similar psoriasis-specific hallmark biological processes. Furthermore, the huge similarity between chromatin-accessible regions among psoriatic keratinocytes and cytokines-treated primary keratinocytes indicated the credibility of the cytokines-treated primary keratinocytes as a psoriasis model system. The functional implication of these disease-specific chromatin-accessible regions should be studied to understand the underlying mechanism of psoriasis pathogenesis.

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Abstract ID: 57

Computational Strategies in Identifying Therapeutic Targets and Drug Screening for Dystrophinopathies

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Background/ Introduction: Duchenne Muscular Dystrophy (DMD) (OMIM #310200) and its allelic counterpart, Becker Muscular Dystrophy (BMD) (OMIM #300376), are rare, severe and prevalent neuromuscular conditions, collectively termed dystrophinopathies. They result from mutations in the DMD gene (OMIM #300377) at Xp21.2, which encodes dystrophin (UniProt #P11532), a 427 kDa protein essential for skeletal muscle fibre integrity. Clinically manifests as toe walking, waddling gait, Gower's sign, scoliosis, pseudohypertrophy, wheelchair dependence and cardiorespiratory complications leading to reduced life expectancy. Amid the unmet need for a cure, this study seeks to advance therapeutic discovery by complementary computational methods, ultimately contributing to potential treatment solutions for D/BMD.

Objectives: This study aims to integrate gene expression analysis and protein interaction studies, using computational approaches to identify critical molecular pathways, potential therapeutic targets and drug screening for D/BMD.

Materials and Methods: Gene expression profiles were sourced from GEO, analyzing datasets GSE6011, GSR1004, GSE38417, GSE3307, and GSE1007 for differentially expressed genes. Protein-protein interactions were constructed using Cytoscape, and Gene Ontology and KEGG pathway analyses were performed through Enrichr. Essential proteins identified from hub genes were prepared for molecular docking using Schrödinger Maestro, utilizing selected ligands Coenzyme Q, Givinostat, Vamorolone, Prednisone, Rimeporide, L-Arginine, L-Citrulline, L-Carnitine, IGF-1, and Gentamycin sourced from PubChem and screened ligands through the NPASS database. Docking results were assessed using MMGBSA to calculate binding energies with the OPLS4 force field. ADME profiling was conducted

using the QikProp tool, and molecular dynamics (MD) simulations were performed with Desmond Schrödinger to evaluate the stability and interaction patterns of protein-ligand complexes.

Results and Conclusions: 259 upregulated and 900 downregulated genes were identified, with FBXO32 as the top upregulated and CD4 as a downregulated gene. Upregulated genes are primarily involved in oxygen and carbon dioxide transport, with the sarcolemma as the main cellular component and heme binding as the top molecular function. The top 10 downregulated genes are associated with the positive regulation of the ERK1 and ERK2 cascade, primarily linked to the membrane raft and complement component C3b binding. Further molecular docking and MMGBSA studies with the NPASS library and standard drugs scrutinized the compounds. In the MD simulation, natural compounds over standard drugs emerged as the most effective for gene regulation. Advanced computational studies illuminate gene regulation, protein behaviour, and underlying pathology while screening natural compounds with therapeutic potential for DMD.

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Abstract ID: 58

Genetic analysis of FABP4 rs8192688 and rs112313579 polymorphisms in Post menopausal Obese Women

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Background/ Introduction: Obesity has increased dramatically in recent decades due to sedentary lifestyles in modern society and excessive consumption of dietary calories, including overconsumption of high-fat diets. The prevalence of overweight and obese conditions has become a pervasive health concern among women across the globe which is persistently increasing. Obesity is considered as one of the most important disturbances associated with menopause. Several factors, including genetics and diet contribute to obesity development. Obesity is a potential risk factor for various serious non-communicable diseases such as type 2 diabetes, osteoporosis, cardiovascular and oncological diseases in post-menopausal women. The complex interaction between age, obesity, and menopause is the subject of several ongoing studies. Obesity is associated with abnormal fatty acid metabolism and secretion of multiple adipokines, which has the potential to increase morbidity and mortality.

An evolutionarily conserved protein family known as the Fatty acid binding proteins (FABPs) are expressed in various tissues to facilitate the absorption and utilization of water-insoluble dietary long-chain fatty acids (FAs). The FABP family comprises at least nine members each with a distinct tissue expression pattern, such as adipose FABP (A-FABP/ FABP4). Studies show the elevated levels of FABP4 in the bloodstream of individuals with obesity. FABPs function to solubilize various FAs and coordinate their trafficking and responses inside cells. Depending on the cellular and tissue environment, individual FABPs exhibit unique biological functions by regulating different metabolic and inflammatory signalling pathways. FABP4 has attracted the most attention in the field of obesity due to its high expression in adipose tissues and biological functions in macrophages and adipocytes. FABP4 gene is located at 8q21.13. In humans, higher circulating FABP4 levels associate with insulin resistance and metabolic syndromes. Studies demonstrated that inhibition of the FABP4 protein through genetic deletion or pharmacological inhibition protects against the harmful effects of obesity.

Single nucleotide polymorphisms (SNPs) residing on FABP4 may influence its regulation and eventually determine its susceptibility to obesity or other metabolic syndromes. Polymorphisms could act as strong

predictors of various disease conditions. According to the population databases, FABP4 rs8192688 (c.74-16C>T) and rs112313579 (c.349-34dupG) variations are considered as the most common SNPs in humans. Hence it was hypothesized that the SNPs rs8192688 and rs112313579 may alter the expression of FABP4 and increase or decrease the susceptibility to obesity. However, there is an absence of studies regarding the association of FABP4 rs8192688 and rs112313579 polymorphisms in Indian postmenopausal obese women. In order to test this hypothesis, the study was designed to investigate the association of FABP4 rs8192688 and rs112313579 variations with obesity in postmenopausal women from India with the following objectives.

Objectives: 1. To investigate the frequencies of FABP4 rs8192688 and rs112313579 polymorphisms in post-menopausal obese women, 2. To investigate the frequencies of FABP4 rs8192688 and rs112313579 polymorphism in post-menopausal non-obese women, and 3. To determine the association of FABP4 rs8192688 and rs112313579 polymorphisms with obesity in post-menopausal women.

Materials and Methods: This was a case-control study involving obese post-menopausal women as cases and non-obese post-menopausal women as controls. The study was carried out at Jubilee Centre for Medical Research, Jubilee Mission Medical College and Research Institute, Thrissur. A total of 134 post-menopausal women were recruited for this study after approval by the Institutional Ethics Committee. Among these, 67 were obese (BMI $\geq$ 25 kg/m^2) cases and 67 were non-obese (BMI < 25 kg/m^2) controls. Three microlitre of peripheral blood was collected from all the study subjects after getting informed consent from them. Genomic DNA was extracted from peripheral blood samples using Kit method (QIAGEN, Germany). The quantification was determined by using multimode reader (TECAN, Austria). The polymorphism residing regions of FABP4 gene was subjected to Polymerase Chain Reaction (PCR)-Sanger sequencing. Statistical analyses were performed using SPSS software (version 25). Each SNP was coded as a co-dominant genetic model. Fisher's exact test of association was used to compare the difference in the distribution of genotype and allele frequencies between cases and controls. The odds ratios (ORs) were calculated along with the corresponding 95% confidence intervals (95% CIs), for estimating the risk of FABP4 polymorphisms rs8192688 and rs112313579 with obesity. Statistical significance of the study was set as ≤ 0.05.

Results and Conclusions: Regarding the FABP4 rs8192688 (c.74-16C>T) polymorphism, the distribution of genotype frequencies among the obese women were 51 (76.11%), 16 (23.88%) and 0 (0%) in wild type (CC), heterozygous (CT) and variant (TT) genotypes respectively. On the other hand, the corresponding frequencies in controls were 55 (82.08%), 10 (14.92%) and 2 (2.98%) for the three genotypes. The allelic frequency of wild type (C) and variant (T) were found to be 0.8805 and 0.1194 in cases, and 0.8955 and 0.1044 in controls, respectively. In our study we could not observe significant differences in the distribution of the polymorphic genotypes (P = 0.2755) and allele frequencies (P = 0.8467) between the cases and controls. The association between FABP4 rs8192688 (c.74-16C>T) polymorphism and risk of obesity was calculated for the heterozygous and homozygous variants using OR keeping the homozygous wild type genotype as reference. No statistically significant association was observed for heterozygous (OR = 1.7255, 95% CI = 0.7177-4.1482, P = 0.2229) and homozygous variants (OR = 0.2155, 95% CI = 0.0101-4.5969, P = 0.3256). Likewise, the variant T allele with an OR of 1.1622 (95% CI = 0.5430 to 2.4876) also did not seem to be significantly associated with an increased risk for obesity (P = 0.6986).

The genotypic frequency distributions of FABP4 rs112313579 (c.349-34dupG) polymorphism among the cases were 47 (70.14%), 20 (29.85%) and 0 (0%) in wild type (GG/GG), heterozygous (GGG/GG) and variant (GGG/GGG) genotypes respectively, while the corresponding frequencies in controls were 51 (76.11%), 11 (16.41%) and 5 (7.46%). The allelic frequency of wild type (G) and variant (GG) were 0.8507 and 0.1492 in cases, and 0.8432 and 0.1567 in controls, respectively. Significant differences were

not observed in the distribution of the polymorphic genotype (P = 0.1486) and allele frequencies (P = 1.00) between the cases and controls. The association between FABP4 rs112313579 (c.349-34dupG) polymorphism and risk of obesity was calculated for the heterozygous and homozygous variants using OR keeping the homozygous wild type genotype as reference however statistically significant data was not observed for heterozygous (OR = 1.9729, 95% CI = 0.8554-4.5504, P = 0.1110) and variants (OR = 0.0986, 95% CI = 0.0053-1.8309, P = 0.1201). Likewise, the variant GG allele with an OR of 0.9440 (95% CI = 0.4853 to 1.8363) also did not seem to be significantly associated with an increased risk of obesity (P = 0.8653).

Conclusions:

In our present preliminary study, we analysed the frequencies and their probable associations with obesity for FABP4 rs8192688 and rs112313579 in obese and non-obese post-menopausal women but found no statistical significance for the same. To the best of our knowledge, this is the first Indian study to determine the frequencies of association between rs8192688 and rs112313579 in FABP4 among post-menopausal obese women. In conclusion, our case-control study of post-menopausal women did not provide any evidence that the common genetic variants in FABP4 could contribute to obesity which could probably be attributed to a smaller sample size (n=67). Therefore, further studies are on-going in a larger group to elucidate its association with obesity.

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Abstract ID: 59

Investigating the role of microRNAs (miRNAs) during psoriasis pathogenesis

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Background/ Introduction: Psoriasis is a chronic immune-mediated inflammatory skin disorder characterized by hyperproliferation and abnormal differentiation of keratinocytes. Both genetic and epigenetic factors are reported to be associated with psoriasis pathogenesis. Epigenetic factors, including noncoding RNAs, were found to be deregulated in psoriasis. Although global miRNA expression profiles in psoriasis skin have been studied, miRNA regulating different histological features of the disease have not been explored yet. Histopathological features, such as rete-peg elongation, Munro's microabscesses, Kogoj microabscesses are the characteristic features of psoriasis. Rete-peg elongation, the downward elongation of the epidermis to the dermal layer in psoriatic skin, is primarily caused by keratinocyte proliferation.

Objectives: This study aims to (i) identify deregulated miRNAs in psoriatic tissues, (ii) study the association of the deregulated miRNAs with rete-peg elongation, and (iii) identify the functional role of keratinocyte-specific deregulated miRNAs in the psoriasis-like cell model system.

Materials and Methods: Small RNA sequencing was performed from the 24 psoriatic and 24 adjacent normal skin tissues to identify the deregulated miRNAs. Pearson correlation analysis between miRNA expressions and the corresponding rete-peg lengths in psoriatic tissues was performed to identify miRNAs, which might have a role in keratinocyte proliferation. To determine the role of these correlated

miRNAs, psoriatic keratinocyte cell model system was developed by treating cells (HaCaT and KerCT) with four cytokines; IL-6, TNF-α, IL-17a, and IL-1α. The expression of miRNAs was checked in psoriatic keratinocytes using real-time PCR. MicroRNAs were overexpressed in the cell line followed by functional assays.

Results and Conclusions: Small RNA sequencing identified 75 significantly deregulated miRNAs in psoriatic skin. 14 miRNAs were correlated with rete-peg elongation, 8 of which were positively correlated and 6 were negatively correlated with rete-peg length. The expression of these miRNAs was checked in the psoriatic keratinocyte cell model system and 2 miRNAs, miR-21-5p and miR-944 were selected for further functional experiments due to their consistent deregulation in tissue and cell lines. Both of these miRNAs were significantly upregulated in psoriatic conditions. Predicted target genes were validated using luciferase assay. Functional assays with miRNA over-expressed cells along revealed their proliferative role in keratinocytes. Understanding the role of these deregulated miRNAs and their target genes in regulating keratinocyte proliferation may aid in developing future therapeutics for psoriasis.

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Abstract ID: 60

Centre-Specific Portrait of Muscular Dystrophies in Gujarat: Insight into Genetic Diagnosis and Management

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Background/ Introduction: DNA, RNA, or protein errors can lead to globally prevalent genetic disorders, though precise rates are difficult due to underreporting. In India, high genetic diversity and consanguinity contribute to a prevalence of 64.4 per 1000 live births. Gujarat, home to over 72.65 million people, faces challenges in reporting rare genetic disorders. Population-based OMIC studies are needed, but progress is slow due to the time, cost, and effort. In light of this, Muscular Dystrophies (MDs) are rare neuromuscular diseases characterized by progressive muscle degradation, weakness, cardiac and respiratory issues, and loss of ambulation. By revolutionizing molecular techniques, mutations in 29 genes are linked to 34 types of MDs, categorized by onset, clinical features, inheritance, severity, and affected proteins. Most MDs are caused by disruptions in the Dystrophin-Glycoprotein Complex (DGC), which is crucial for skeletal muscle fibre integrity.

Objectives: This study aims to investigate the clinical manifestations, epidemiology and genetic mutations associated with MDs. Additionally, it seeks to evaluate current diagnostic approaches to improve early detection and inform targeted therapeutic strategies.

Materials and Methods: This study was conducted at the Indian Muscular Dystrophy Society and Research Center (IMDS, Reg. No. E/7420/Ahmedabad) in Ahmedabad, Gujarat, India in a cohort of 511 participants. Clinical evaluations and advanced genetic testing, including MPCR, MLPA, and NGS, were employed to identify and classify genetic mutations.

Results and Conclusions: This cohort included various MDs such as Duchenne Muscular Dystrophy (DMD), Becker Muscular Dystrophy (BMD), Limb Girdle Muscular Dystrophy (LGMD), Congenital

Muscular Dystrophy (CMD), and others. Our findings revealed that DMD was the most prevalent, with 448 cases, followed by other types. Genetic analysis identified 235 pathogenic mutations, including exonic deletions, duplications, and point mutations. This study sheds light on the genetic landscape of MDs in Gujarat, advocating for the importance of comprehensive genetic screening for accurate diagnosis and management of MDs. This meta-analysis could help reduce the burden of MDs by emphasizing the need for early diagnosis, personalized treatment, and informed family planning, thereby improving patient outcomes and Quality of Life (QoL).

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Abstract ID: 61

Unraveling the clinical significance of missense SNPs in PCSK9, LDLRAP1 and LIPA genes in hypercholesterolemia through *in silico* approach

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Background/ Introduction: Familial Hypercholesteremia(FH) is one of the autosomal genetic disorders caused due to increased levels of low-density lipoprotein cholesterol(LDL-C) in the circulation. Single Nucleotide Polymorphism (SNP) in lipid metabolising genes such as PCSK9, LDLRAP1 and LIPA can increase LDL-C in blood circulation which causes familial hypercholesteremia.

Objectives: Impact of missense SNPs in PCSK9, LDLRAP1 and LIPA genes in hypercholesterolemia.

Materials and Methods: SNPs were retrieved from the ensemble genome browser and Human Gene Mutation Database. Missense mutations were analysed for structural and functional alteration using PredictSNP, Consurf, Mupro, Site- Directed Mutagenesis, Mutpreb, SWISS Model, I-Tasser, Molbrobity, Mudpred and Cluspro.

Results and Conclusions: According to the ensemble genome browser and human gene mutation database, 685, 260 and 329 missense SNPs were identified from PCSK9, LDLRAP1 and LIPA, respectively. Out of these, 96, 34 and 88 SNPs were predicted to be the most damaging SNP by all five tools. Further, 56, 10 and 66 missense SNP from PCSK9, LDLRAP1 and LIPA genes were found in highly conserved regions and finally, 41, 8, and 57 SNPs significantly decreases protein stability. PCSK9 protein was further docked with LDLR and interestingly, these 41 SNPs play a protective role against disease progression. The 8 and 57 SNPs of LDLRAP1 and LAL protein were reported to be most deleterious and possibly cause the progression of hypercholesteremia. This study highlights the clinically important SNPs of PCSK9, LDLRAP1 and LIPA genes. Our findings are relevant for the clinical diagnosis of hypercholesteremia and drug design.

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Abstract ID: 62

Investigating the effects of homozygosity upon the genetic underpinnings of cardiometabolic and cognitive phenotypes

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Background/ Introduction: Identity-by-descent (IBD), where individuals inherit identical segments of DNA from a common ancestor, can be enhanced due to the practice of endogamy in populations and is highly prevalent in India. Longer stretches of IBD could contribute to a higher incidence of rare congenital anomalies, recessive Mendelian disorders, and complex diseases, such as Alzheimer's disease and coronary artery disease. With its vast diversity of endogamous groups, India experiences elevated IBD due to limited gene flow from social, cultural, and linguistic barriers. Runs of homozygosity (ROH) are long stretches of consecutive homozygous genotypes inherited from a common ancestor, reflecting regions of homozygosity. While total homozygosity has been linked to disease susceptibility, the specific effects of individual ROH segments on complex traits remain underexplored. This study investigates whether the length of ROH segments influences traits and if the presence of these segments enhances the association with trait-associated variants.

Objectives: This study aims to estimate the stretch of homozygosity in the genomes of individuals in the Indian population. Then, investigate the association of these ROH segments with cardiometabolic traits and cognitive meaures in the Indian population.

Materials and Methods: ROH calling was performed on whole genome sequencing data from the GenomeIndia, Srinivaspura Aging Neuro Senescence and Cognition (CBR-SANSCOG), and TATA Longitudinal Study of Aging (CBR-TLSA) studies. The association between specific ROH segments and traits was assessed using a linear regression model, adjusting for relevant covariates. In parallel, a genome-wide association study (GWAS) was conducted for the same traits. Significant associations between ROH segments and traits were identified. Overlap between these ROH regions and GWAS hits was systematically evaluated to determine the potential contribution of ROH segments.

Results and Conclusions: Genome-wide identification and characterization of ROH were performed across all datasets, providing insights into the levels of homozygosity in the Indian population. Using a linear regression model, we identified ROH segments that showed significant associations with lipid metabolic traits and HMSE (Hindi Mental Status Examination) scores. Notably, no significant overlap was observed between the ROH regions and GWAS signals for the same traits. Annotation of the ROH regions led to the identification of genes within these segments. In populations with high rates of endogamy and homozygosity, ROH may contribute modestly to complex traits. Further gene-level burden analyses of ROH are required to better understand the role of recessive alleles in the phenotypic variation of complex traits.

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Abstract ID: 63

miR-34a and miR-221 expression upon in vitro Lithium and valproate exposure to Bipolar Disorder patients-derived LCLs

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Background/ Introduction: Bipolar disorder (BD) is characterized by recurrent episodes of mania or hypomania and depression, with or without mixed features. Lithium has been the first line of treatment for BD, but its mechanisms of action are still obscure.Our previous studies showed in vitro lithium exposure enhanced expression of cell survival genes BCL2, and GSK3B in BD Lymphoblastoid Cell lines (LCLs). microRNAs are epigenetic regulators of gene expression at the post-transcriptional level, we

checked whether microRNAs mediate this response to lithium at a cellular level. In silico tools identified hsa-miR-34a-5p and hsa-miR-221-3p targeting BCL2 and GSK3B.

Objectives: In the current study, we examined the expression of these miRNAs in BD LCLs on in vitro exposure to Lithium chloride (LiCl) and sodium valproate (VPA) and in control LCLs.

Materials and Methods: BD patients and healthy controls were recruited at the OPD-NIMHANS. LCLs were generated from peripheral blood mononuclear cells of BD and control subjects as part of an earlier study. Control (n=7) and BD LCLs(n=14) comprising clinical Li-responders with ALDA score ≥7 (BD-R: n=7) and clinical Li-Non-responders with ALDA score ≤ 5 (BD-NR: n=7) were exposed to a) 1mM LiCl, b) 0.7mM VPA c) unexposed (vehicle) for 7 days. Total RNA was isolated by trizol method. microRNA levels were quantified using TaqMan microRNA expression assays specific for hsa-miR-34a-5p and hsa-miR-221-3p with RNU6B as an endogenous control.

Results and Conclusions: In vitro lithium exposure to LCLs reduced miR-34a expression in BD and healthy controls; while no change was detected in BD-NRs. VPA enhanced miR-34a expression in BD and healthy control LCLs. Elevated levels of miR-221 expression were detected on in vitro exposure with LiCl and VPA-in BD and control LCLs.

Studying miRNA expression in response to mood stabilizers could potentiate biomarker discovery to predict treatment outcomes in BD patients. Overexpression of miR-34a has been reported to induce neuronal cell death by downregulation of BCL-2. Valproate known to paradoxically increase several miRNA expressions, could be a reason for enhanced miRNA expression in all groups post-VPA exposure. Our study verified the miR-34a and miR-221 expression level alteration with in vitro exposure to LiCl & VPA. Further studies to validate miRNA-mRNA interaction may be warranted to understand the modulatory effect of Lithium via miRNAs in BD Li-responders.

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Abstract ID: 64

Whole Exome Sequencing Reveals UNC45B As a Novel Candidate Gene Associated With Dilated Cardiomyopathy

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Background/ Introduction: Over the past few decades dilated cardiomyopathy (DCM) has become one of the leading causes of heart failure. Present investigation is focused on UNC45B variants identified through NGS in DCM patients. UNC45B, a key motor protein involved in muscle contraction, and stabilization of the myosin head domain, is a molecular chaperone that is involved in proper folding and assembly of myosin, facilitating efficient interaction between actin and myosin during contraction. Additionally, it regulates the expression of various cytoskeletal genes, maintaining muscle integrity and function. Dysfunction or mutations in UNC45B have been linked to impaired pumping efficiency of cardiac muscles.

Objectives: This study aims to delineate the role of UNC45B gene in pathogenesis of DCM by functional characterization of UNC45B variations, identified in DCM patients using whole-exome sequencing.

Materials and Methods: Whole-exome sequencing was performed for fifteen DCM patients, including probands along with their first-degree relatives. Variant prioritization and data analysis were carried out using several bioinformatics tools and pipelines. Sub-cloning of UNC45B-Wild-type clone was done in

suitable mammalian expression vector followed by preparation of mutant constructs by site-directed mutagenesis. In-vitro and in-silico approaches were used to elucidate the role of novel variants detected in UNC45B.

Results and Conclusions: We identified a total of 9 variants in UNC45B, out of which 7 were non-synonymous (p.L486F, p.C487Y, p.A492T, p.D496Y, p.R721Q, p.A780V and p.C786Y) and 2 were synonymous (p.Y769Y and p.A780A). Subcellular localization of both wild-type and mutant UNC45B proteins were detected in cytoplasm by immunocytochemistry. However, mutant forms displayed markedly altered morphology. Cells expressing the mutant variants exhibited multinucleation, irregular shapes, and significant enlargement. Furthermore, the mutant variants induced a notable up-regulation of their downstream targets, including Myh7 (Myosin), Actc1 (Actin), and Ttn (Titin). These findings were reinforced by in-silico modeling and molecular docking analysis, which revealed potential structural-functional divergence possibly leading toward the disease state. Detailed functional analysis suggests that the identified missense mutations cause anomalous morphological changes that could lead to altered myosin function which may disrupt myofibrillar organization. This mechanism would finally lead to hypertrophic responses followed by potential loss of contractile function.

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Abstract ID: 65

Promoter hypermethylation of FHIT and re-expression by 5-aza-2'-deoxycytidine treatment in gastric cancer cell line

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Background/ Introduction: Epigenetic modifications play a central role in gastric carcinogenesis. The study of epigenetic processes has increased in recent years, and novel therapeutic approaches that target DNA methylation and histone modifications have emerged. A greater understanding of epigenetics and the therapeutic potential of intervention into these processes is necessary to help GC treatment. DNA methylation refers to the addition or subtraction of a methyl moiety at the 5th position of the cytosine ring within CpG dinucleotide that are usually located in CpG rich regions or CpG islands and around the gene promoter. DNA methylation in gene promoter regions represses transcription of their downstream genes associated with the suppression of gene expression. A number of genes involved in cell cycle regulation, tumor cell invasion, DNA repair, chromatin remodeling, cell signaling, transcription, and apoptosis are known to be silenced by hypermethylation in GC. Moreover, hypermethylation of several gene promoters has also been observed in the premalignant stages of GC, suggesting that aberrant methylation occurs early during gastric carcinogenesis. For the FHIT gene, hypermethylation of the CpG Island has been reported to be a major mechanism for gene silencing in lung and breast cancers. Although, individually the expressions and pathological observations in different cancers including GC has been reported. But the detailed analyses of the relationships among FHIT methylation, protein expression, and clinico-pathologic observations have not, to our knowledge, been demonstrated for gastric cancer.

Objectives: Several epithelial cancers, including gastric carcinoma, have been linked to the unstable histidine triad (FHIT) gene. Moreover, FHIT transcriptional inactivation is a major factor in the development of human cancer and is caused by aberrant 5'-CpG island methylation. In this study, we found that the demethylating agent 5-Aza-2'-deoxycytidine caused FHIT re-expression in gastric cancer

cells (AGS). Furthermore, we showed a direct relationship between FHIT hypermethylation, protein expression, and mRNA. We also demonstrated how the FHIT gene's re-expression affected the gastric cancer cells' ability to undergo apoptosis and cell viability.

Materials and Methods: Quantitative RT-PCR was carried out using the conventional technique to look for aberrant transcripts of the FHIT gene. Bisulfite DNA sequencing and methylation-specific PCR were used to ascertain the methylation status. To check the reactivation of epigenetically silence gene; AGS cells were maintained in complete F12K media with 10% Fetal Bovine Serum and 1% Peniciline and Streptomycine antibiotics. Cell culture maintained in the 5% CO2, 95% humidified air and at 37°C temperature in CO2 incubator. The equal numbers of AGS cells were seeded in 25 cm2 culture flasks. For reactivation of the FHIT expression, semi confluents AGS cells were treated with 5µM of DNA methyl transferase inhibitor, 5-Aza-2'-deoxycytidine (AZA) for 5-7 days and 200nM of HDAC inhibitor Trichostatin A (TSA) for 24 hours followed by RNA extraction. For apoptosis of the cancer cells we performed JC1 staining to assess the mitochondrial potential and the cancer cells undergo apoptosis.to assess the vialbility of the cells we performed PI staining.

Results and Conclusions:

Results: To investigate the possible mechanism of down regulated expression of FHIT, we analyzed its promoter methylation status by MS-PCR in 40 GC tissue samples and 20 normal control samples used. The MSP was used to determine the frequency of FHIT 5' CpG island methylation in bisulphite modified DNA from a group of 40 gastric carcinoma patients. Out of 40, 87.5% (35/40) GC samples showed significantly FHIT promoter hypermethylation, among which 47.5% (N=19) of the tumor biopsies showed heterozygous and 40% (N=16) of tumor biopsies showed homozygous methylation. The remaining 12.5% (05/40) of tumor biopsies were unmethylated. However, all the normal gastric samples were unmethylated.We also analyzed the correlation between methylation status of FHIT promoters and FHIT mRNA expression in tumor samples. Around 85.7% of (30/35) cases with reduced FHIT expression exhibited FHIT promoter methylation, while the remaining 14.2% of (5/35) hypermethylated gastric cancer cases did not show any significant change in FHIT expression. These findings suggest significant correlation of FHIT promoter hypermethylation with reduced FHIT gene expression. Our correlation analysis data of FHIT methylation status and clinico-pathological features demonstrate significant correlation with tumor stages (p=0.0353), T stage (p=0.0401) and patients age (p=0.0454) and histological grade (p=0.0114). We did not find any significant correlation of FHIT promoter hypermethylation with other indices. MSP approach allows the scoring of the methylated 5'CpG islands as a positive or negative methylation. We compared methylated and unmethylated gastric tumor samples in order to determine a relationship between positive MSP results and decreased FHIT mRNA expression. Our results show that unmethylated control samples express significantly higher level of FHIT mRNA expression as compared to methylated gastric tumor samples, (p=0.0040). We also compared the relative FHIT mRNA expression among tumor samples displaying either methylated or unmethylated FHIT 5'CpG regions (p=0.0061).We further demonstrate that methylation of CpG island within the FHIT gene indeed suppresses FHIT mRNA and FHIT protein expression in gastric tumor samples. We also showed GC biopsy samples with methylated 5'CpGs shows down regulated expression of Fhit protein, whereas the samples with unmethylated promoters show normal FHIT protein levels. Also a clear correlation between FHIT mRNA and FHIT protein expression was observed. We have treated AGS cells having methylated FHIT with hypomethylating drugs 5-aza-2'-deoxycytidine (5-Aza), for re-expression of FHIT gene. Our results demonstrate that following 5-aza-2'-deoxycytidine treatment FHIT protein is re-expressed. 5-aza-2'-deoxycytidine treatment (5µM) was performed for 5-7 days, re-expression of protein band started to be visualized from 4th day of the treatment. Subsequently we also treated the AGS cells with 5µM of 5-aza followed by 200nM of HDAC inhibitor Trichostatin A (TSA) which also showed similar results as Aza alone treatment. We found an increase level of FHIT mRNA expression in 5-Aza treated group of cells

as compared to other treated groups (TSA and 5-Aza+TSA). We also performed MS-PCR after treatment of gastric cancer cells to see the methylation status of the methylated FHIT 5'CpG site following the drug treatments. We observed the methylation status of the AGS cells treated with 5-Aza got significantly reduced as compared to the methylation status of the other treated groups (TSA and 5-Aza+TSA). The structural and functional stability of mitochondria can directly affect cell energy metabolism and activate apoptosis machinery. Since FHIT had significantly reduced mRNA and protein expression in gastric cancer cells and the decrease in expression is known to directly affect the mitochondrial functions, it may deregulation the cascade pathways. We performed JC-1 staining to see the apoptosis of cells and PI staining for cell viability after treatment with 5-Aza. After treatment of AGS cells with 5 µM 5-aza-2'-deoxycytidine, JC-1 staining shows change in the mitochondrial membrane potential which induces apoptosis of the cancer cells in the 5-Aza and 5-Aza+TSA treated groups. Similarly PI staining shows there are more number of nonviable cells observed in the 5-Aza and 5-Aza+TSA treated groups.Conclusion: This piece of our study concludes that one of the key factors influencing the development of gastric tumors may be the decreased expression of FHIT and its promoter hypermethylation. FHIT expression may play an useful role in GC treatment outcome prediction and clinical prognosis prediction. Our findings indicate that since its expression decreases with tumor progression, it could function as a predictive biomarker for tumor aggressiveness. FHIT may also prove to be a desirable target for the development of GC gene therapy in the future. To support the clinical and prognostic significance of the current findings a larger patient cohort with GC may be required is necessary for further validation.

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Indian Council of Medical Research (ICMR), Government of India for senior research fellowship to Ms Juhi Singh under the supervision of Prof.VK Dixit and Prof. Gopeshwar Narayan. Dr. Soni Kumari for helping in experimental studies. Department of Surgery for collecting gastric carcinoma samples, Department of Pathology for Immunohistochemistry, Institute of Medical Sciences and Department of Molecular Human Genetics, Institute of Science Banaras Hindu University, Dept. of Biochemistry, AIIMS, Delhi for equipment facilities.

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Abstract ID: 66

To Investigate the association analysis of GnRH I polymorphism (rs6185) with PCOS in the Punjab region.

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Background/ Introduction: Polycystic ovary syndrome (PCOS) is a prevalent hormonal disorder impacting about 9.2% of women. It often manifests with cardiovascular risk factors, reproductive and metabolic issues, and increased levels of androgen, resulting in symptoms such as irregular menstrual cycles, hirsutism, and insulin resistance. The condition remains widely underdiagnosed, affecting physical and emotional well-being. PCOS is often diagnosed using the Rotterdam criteria, which requires two out of three key features: irregular ovulation, hyperandrogenism, or polycystic ovarian morphology (PCOM). The gonadotropin-releasing hormone (GnRH) system, particularly the GnRHI gene located on chromosome 8, plays a central role in regulating hormonal responses. In PCOS, GnRH resistance to feedback from hormones like progesterone is evident and may be related to elevated androgen levels.

Objectives: This study is among the first to analyze the association of the GnRHI gene variant (rs6185) in PCOS within the Punjab region, aiming to understand genetic factors contributing to disease expression and severity.

Materials and Methods: A study was conducted on 60 female participants including 30 PCOS cases and 30 healthy controls. Diagnosis of PCOS cases was done using Rotterdam criterion of 2003, Following informed consent, 5 mL of blood was collected from each participant, further GnRHI polymorphism were genotyped using ARMS - PCR. Statistical analysis was performed using SPSS (version 21, IBM SPSS, NY, USA).

Results and Conclusions: In our study we did not find any significant difference in the genotype (p=0.2) and allele (p=0.4) frequency of polymorphism rs6185 between PCOS cases and healthy controls. The current investigation demonstrates a non-significant association between the GNRH1 polymorphism (rs6185) and polycystic ovary syndrome (PCOS).

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Abstract ID: 67

Genetic association analysis of KISS1 polymorphism (rs5780218) with Polycystic Ovary Syndrome in Northwest Indian population

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Background/ Introduction: Polycystic ovary syndrome (PCOS) is the most prevailing endocrine-metabolic disorder in women at their childbearing age assisted by various symptoms and consequences. This syndrome is characterized by reproductive, metabolic, and endocrine abnormalities including hyperandrogenism, ovulatory dysfunction, infertility, obesity, insulin resistance, hepatic steatosis, and dyslipidemia. Although the pathogenesis of Polycystic Ovary Syndrome (PCOS) is still unclear, the disturbance of hypothalamic-pituitary-gonadal (HPG) axis is suspected to be the main culprit in the development of PCOS. Kisspeptin is a potent GnRH neuron regulator, generally involved in establishing the onset of puberty and fertility. It is a natural ligand of G- protein coupled receptor 54 (GPR54) and a peptide encoded by KISS-1 gene, which is associated in the regulation of HPG axis. The rs5780218 polymorphism occurs at -145 position of the 5'UTR of mRNA transcript and represents the deletion of Adenine (A) nucleotide.

Objectives: The present study is the first of its kind from North population to analyze the association of KISS1 in women with PCOS cases and controls.

Materials and Methods: A study was conducted involving 60 female participants, comprising 30 cases of polycystic ovary syndrome (PCOS) and 30 healthy controls. Following informed consent, 5 mL of blood was collected from each participant. The KISS1 polymorphism (rs5780218) was genotyped using PCR-RFLP. Statistical analysis was performed using SPSS (version 21, IBM SPSS, NY, USA).

Results and Conclusions: The present study demonstrated no significant differences in genotypic (p = 0.39) and allelic (p = 0.23) frequencies of the KISS1 polymorphism (rs5780218) in relation to polycystic ovary syndrome (PCOS). In our study, we observed no significant association between KISS1 polymorphisms and polycystic ovary syndrome (PCOS).

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Abstract ID: 68

The Antioxidant Paradox in Cancer Therapy: A Double-Edged Sword in Tumor Progression and Treatment Resistance

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Background/ Introduction: Background: For decades antioxidants are used to neutralize the reactive oxygen species to boost overall health. Traditionally heralded for reducing oxidative stress, antioxidants are now implicated in promoting cancer progression, requiring a reexamination of their therapeutic and supplemental use. Clinical trials have demonstrated an unexpected increase in metastasis with antioxidant supplementation in lung and prostate cancer patients.

Objectives: Objective: This study explores the role of antioxidants in the progression and therapeutic resistance especially in Glioma. This aim is to examine the hypothesis, proposed by James Watson and others, that antioxidants may hinder the ROS-mediated apoptosis critical for cancer suppression.

Materials and Methods: Methods: A comprehensive search was conducted using PubMed, Google Scholar and ClinicalTrial.gov to identify relevant literature and clinical findings published from 2014 to 2024. Boolean operations refined the search for studies utilizing MeSH keywords such as "Antioxidants," "Reactive Oxygen Species," and "Glioma". Inclusion criteria encompassed clinical trials, in vitro, and in vivo studies investigating ROS modulation in gliomas and other cancers, while exclusion criteria eliminated case reports, non-English publications, and studies not addressing antioxidant effects.

Results and Conclusions: Results: Findings indicate that ROS, though damaging at high levels, play a dual role in cancer, serving as both a mediator of cellular apoptosis and a regulator of immune response. Excessive antioxidant intake may interfere with this balance, reducing ROS to levels that prevent apoptosis and immune activation, allowing tumor cells to evade cellular stress defenses. Clinical studies reveal that antioxidant-induced ROS reduction can reduce cancer cell sensitivity to cancer therapies, both of which depend on ROS for inducing cancer cell death.

Conclusion: This review highlights the urgent need for awareness on the misuse of antioxidants, particularly in cancer patients, as overuse may unintentionally support tumor growth and therapy resistance by disrupting ROS-mediated cell death. This challenges the convention that antioxidants mitigate cancer risk and suggests that their overuse may be deleterious in certain cancers. Further studies are warranted for optimal therapeutic strategies that consider both the beneficial and potentially adverse effects of antioxidants in cancer treatment.

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Abstract ID: 69

To perform in Silico analysis of SNPs of Toll like Receptor 9 (TLR9).

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Background/ Introduction: Toll like Receptor 9 (TLR9) was first identified as a receptor for unmethylated CpG-DNA as well as for bacterial DNA. TLR9 are expressed on the surface of neutrophils, B cells and erythrocytes.TLR9 is also localized at the intracellular membrane compartment, such as endoplasmic reticulum, the endosomes and lysosomes. In humans, TLR9 is confined to the plasmacytoid dendritic cells (pDCs), the innate immune cells that are the major source of the antiviral cytokines Type I interferons(IFNs).Various Single Nucleotide Polymorphisms (SNPs) of TLR9 gene have been documented to be associated with various diseases.

Objectives: To perform in Silico analysis of SNPs of Toll like Receptor 9 (TLR9).

Materials and Methods: TLR9 SNP data was retrieve from "dbSNP" database. The total numbers of SNP in this database were 2775. Out of these, non-coding variant having MAF in the range of 0.01 to 0.5 were selected. The putative functional significance of these variants have been assessed using suitable bioinformatics tools such as RegulomeDB and SNPinfo etc.

Results and Conclusions: RegulomeDB scores ranges from 0 to 1 with lower scores indicating less evidence of regulation and higher scores indicating more evidence. A score of 0.2875 suggests moderate regulatory potential, while a score of 0.6670 indicates stronger evidence of regulatory activity. SNPs with a ranking of 1f (rs352139, rs352144, rs5743836) and 4(rs5743835, rs5743839, rs5743849) indicate a strong likelihood of being regulatory, with 1f suggesting high confidence in functional evidence. The SNP with a ranking of 1b (rs5743838) and 1d (rs187084) suggest evidence of regulatory activity. SNPinfo RegPotential value range from 0 to 0.26582, while 6 SNPs shown to affect transcription factor binding sites.

The analysis of SNPs using suitable bioinformatics tools has provided valuable insights into their regulatory potential and functional implications. Further analysis using more bioinformatics tools can provide further information regarding the putative functional effect of these variants.

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Abstract ID: 71

Promoter And Exonic Variations in Hypothalamic-Gonadal Axis-Related Genes Among South Indian Women With Polycystic Ovary Syndrome

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Background/ Introduction: Polycystic Ovary Syndrome (PCOS) is a multigenic and multifactorial disease condition with a heterogenous pathophysiology. This multifaceted illness affects 15-20% of reproductive aged women population globally. The hallmarks of PCOS include hyperandrogenism, menstrual irregularities and pearl of string appearance of ova in the ovaries. Apart from this, obesity,

insulin resistance, glucose intolerance, cardiovascular diseases are the common comorbidities associated with PCOS. Disturbance of hypothalamic-pituitary-gonadal (HPG) axis is the hallmark of PCOS. Being the key genes in the HPG axis, there is an absence of proximal promoter screening of KISS1 and FSHR genes in PCOS. The inconclusive results in the genetic studies on PCOS are often attributed to the ethnic diversity and the heterogeneous character of its pathophysiology. Despite the enormous population and genetic diversity, the association of KISS1 and FSHR polymorphisms in a homogenous group of Indian PCOS patients has not yet been explored so far.

Objectives: 1. To screen the proximal promoter regions of FSHR and KISS1 genes in PCOS and control subjects. 2. To identify the frequency of common promoter as well as exonic polymorphisms in PCOS and control subjects. 3. To analyze the association of common promoter as well as exonic polymorphisms with PCOS.

Materials and Methods: Our case-control study included 1124 women (613 with PCOS and 511 controls). We meticulously selected 121 PCOS patients based on direct lineage and symptom severity since menarche, meeting all three Rotterdam criteria. To minimize genetic bias, we chose 121 age-matched controls without a family history of PCOS up to the second degree. We analysed the KISS1 and FSHR gene's proximal promoter region in 25 PCOS cases and an equal number of controls using PCR-Sanger sequencing. Additionally, we examined a significant -146 TT>T (rs5780218), Exon 1 G>A (rs12998) and Exon 3 C>G (rs4889) of KISS 1, and 5'UTR variant -29 G>A (rs1394205) and two exon 10 SNPs [Ala307Thr A>G (rs6165) and Ser680Asn A>G (rs6166)] of FSHR genes in 121 PCOS patients and controls using PCR-RFLP. Various bioinformatics tools were employed for the pathogenic assessment of the Glu20Lys G>A (rs12998), Pro81Leu (rs4889) of KISS 1 and Ala307Thr A>G (rs6165) and Ser680Asn A>G (rs6166) variants of FSHR.

Results and Conclusions: Our findings indicate a significant correlation of the KISS 1 rs12998, FSHR rs1394205 (−29G>A) and rs6165 polymorphisms with PCOS patients. Additionally, rs12998 and rs6165 displayed a significant genotype frequency variation among individuals with overweight and normal BMI respectively. Despite this, in-silico tools deemed the KISS1 variation to be pathogenic and the FSHR variation to be non-pathogenic. To conclude, our findings suggest a significant association of KISS 1 rs12998, FSHR rs1394205 and rs6165 polymorphisms with PCOS susceptibility in South Indian patients.

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Abstract ID: 72

Genetic Diversity and Heritage Crafting in the Artisan Communities of Thar's Legacy

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Background/ Introduction: The northwest region of the Indian subcontinent has long served as a major corridor for migration. Much of this region lies in Rajasthan. Historically, this area has been at a crossroads of civilisations and cultures due to its strategic location and linking west Eurasia. The Thar region is renowned for its traditional crafts and art forms, shaped by kin-based occupational practices that trace back to ancient civilizations. These include pottery, woodwork, leatherwork, textiles, and vibrant musical and dance traditions. Thar dwellers follow socio-cultural practices like endogamy, nomadism, and pastoralism. We attempt to explore genetic underpinnings to the origin of crafts and culture of Thar.

Objectives: This research aims to understand the links between genetic patterns and affinities of artisan communities along historic migration corridors and the cultural practices in the Thar region.

Materials and Methods: The study was carried out on eight representative craft communities comprising 172 individuals from four major district of thar desert region. All ethical guidelines were strictly followed. Genome-wide high-density genotype data were used in study. Population genetics methods were employed to explore genetic diversity and relatedness, including Principal Component Analysis (PCA), ADMIXTURE, pairwise Fst, Treemix, f-statistics and Uniparental lineages. These approaches were integrated with data from Indian and global ethnic populations with the studied groups.

Results and Conclusions: Our findings reveal the complex genetic landscape of the artisan communities, reflecting a wide network of interactions with ethnolinguistically diverse neighbours of Indian populations and influence from the west . Some groups, such as pastoralists and woodcraft artisans, exhibit a higher degree of genetic affinity with populations from Central Asia and the Middle East, showing a significant proportion of Ancestral North Indian (ANI) ancestry. In contrast, other artisan communities, like performing artists, pottery and textile artisans, display more indigenous contributions, highlighting their role in the regional peopling of the Thar Desert. These results underscore the dynamic nature of human migrations and cultural exchanges that have shaped the region's genetic diversity.They also provide a foundation for further research on disease susceptibility, mutation patterns, and genetic adaptations.

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Abstract ID: 73

Modelling ALS: Development of an iPSC-based platform for disease mechanism studies

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Background/ Introduction: Amyotrophic Lateral Sclerosis (ALS) is a devastating neurodegenerative disorder characterized by the progressive motor neuron loss, leading to muscle weakness, paralysis, and death typically within 3-5 years of disease onset. Despite extensive research efforts, the underlying mechanisms of ALS remain elusive, and no effective treatments exist due to the complex interplay between genetic and phenotype factors. Traditional animal models often fail to replicate the human disease phenotype completely due to differences in neural complexity. To address this limitation, induced pluripotent stem cells (iPSCs) have emerged as a promising tool for ALS research, offering a human specific model to explore disease mechanisms and therapeutic strategies.

Objectives: This study aimed to establish a feeder-free iPSC-based disease model for ALS incorporating a novel ALS associated genetic variant identified through genetic screening by reprogramming erythroid progenitor cells derived from PBMNCs.

Materials and Methods: We conducted genetic screening of 73 sporadic ALS (sALS) patients using Sanger sequencing and/or whole exome sequencing to identify possible genetic contributors. For model development, PBMNCs were isolated from an age-and sex-matched healthy donor, cultured and reprogrammed into iPSCs using standard non integrative methods. Once, established, the iPSCs were characterized for pluripotency and genome stability through immunostaining and karyotyping. Next, the iPSCs will be genome edited to introduce ALS specific mutation to study the disorder.

Results and Conclusions: Initial next-generation sequencing identified a novel genetic variant in a patient in an ALS candidate gene. However, the patient sample was not available. Therefore, to model this in disease context, an iPSC line was successfully generated from a healthy donor's PBMNCs. These iPSCs expressed key pluripotency markers and demonstrated the ability to differentiate into the three germ layers, confirming pluripotency. Karyotyping further validated genome stability. In the next phase, CRISPR/Cas9 gene editing will be applied to introduce the identified ALS- associated variant into the iPSCs, followed by differentiation into motor neurons, the primary cell type affected in ALS. This developed iPSC-based ALS model offers significant advantages over traditional models, enabling the study of ALS associated genetic variants in human neurons, thereby offering a valuable tool to investigate ALS pathophysiology and holds promise for advancing therapeutic development for this debilitating disease.

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Abstract ID: 76

Exploring Ciliary Syndromic Variability through Molecular Clusters Derived from Ayurveda-Informed Phenotypes

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Background/ Introduction: Ciliopathies show substantial variability in causes and clinical phenotypes, complicating diagnosis and treatment. Most current studies deal with this in an organ-focused manner posing a challenge to identify molecular hubs that drive heterogeneity in these syndromic conditions. Studies suggest that a phenotype-based approach, inspired by Ayurveda's holistic view of disease, may reveal these hubs by examining all clinical phenotypes thoroughly. In this study, we integrated Ayurveda to classify diseases through phenotypes using "Doshas" Vata(V), Pitta(P) and Kapha(K) to unravel molecular hubs governing ciliary dysfunctions in rare diseases.

Objectives: To identify key molecular hubs that drive to diverse phenotypic manifestations in ciliary dysfunctions in common and rare disease conditions.

Materials and Methods: In an earlier study, an integration of Ayurveda-based perspective to rare diseases using Human Phenotype Ontology (HPO) resulted in 6 distinct phenotypic (based on V/P/K proportions) and functionally enriched clusters of rare diseases. Two of these were significantly associated with ciliopathies that had contrast phenotypes. To explore molecular networks and connectivity underlying ciliary dysfunction across endophenotypes of these 2 clusters, we synthesized a) literature analysis with information curation from Ciliaminer & Ciliacarta b) networks analysis using Metascape for functional enrichments, c) enrichement of ciliary genes in organs/systems using GeneOrganizer. We also attempted to identify rare diseases which could potentially have ciliary dysfunction.

Results and Conclusions: Literature shows that one of these two highly enriched and phenotypic distinct cluster(K) mostly includes syndromes characterized by retinal dystrophy, obesity, and renal failure, specifically Bardet-Biedl syndrome and types of Retinitis pigmentosa, as well as primary ciliary dyskinesia and related respiratory disorders. The second cluster(V) has skeletal and craniofacial abnormalities, digital malformations, and neurodevelopmental issues (e.g. Holoprosencephaly and

Spinocerebellar ataxia). Network analysis revealed that one cluster contains ciliary genes involved in diverse ciliary processes, which are connected to non-ciliary genes responsible for responses to various molecules (e.g. hormones, sugars, light). GeneOrganizer analysis further showed distinct organ enrichment, with genes in the first cluster highly expressed in endocrine, reproductive, urinary, and respiratory systems, while genes in the second cluster primarily impact skeletal, muscular, and integumentary systems. Additionally, we identified 251 potential diseases from our clusters that might involve ciliary dysfunction due to the presence of ciliary genes and their phenotypic similarities to known ciliopathies.

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Abstract ID: 77

A Case Study on SCARB1 gene mutation in relation to hypercholesterolemia.

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Background/ Introduction: Hypercholesterolemia is one of the major causes of cardiovascular, cerebrovascular and peripheral vascular diseases and also reproductive health issues. The World Health Organization (WHO) reported that 39% of the adult population worldwide has high blood cholesterol levels, 40% of which are women and 37% of which are men. The scavenger receptor class B member 1 (SCARB1) gene codes for a membrane receptor protein called SR-B1. This gene is a key component in the reverse cholesterol transport pathway. It transports cholesterol from peripheral tissues to the liver for excretion. Mutations in the SCARB1 gene might alter the function of the SR-B1 protein, thereby affecting the homeostasis of cholesterol.

Objectives: Here, we illustrate a case report of hypercholesterolemia due to the SCARB1 mutation. A 35-year-old female brought herself to us with persistently elevated levels of cholesterol and Low-Density Lipoprotein (LDL) and low levels of High-Density Lipoprotein (HDL), despite having a normal Body Mass Index (BMI). She approached us to look for a genetic risk for these persistent lipid profiles. During the presentation, she had Polycystic Ovarian Disease (PCOD) and Gastritis. She underwent a Global Screening Array (GSA) test to determine if there was any link to genetic risk.

Materials and Methods: We collected a blood sample from the patient and sent it for analysis via a Global Screening Array (GSA) test on the Illumina platform. We analyzed SCARB1 gene in association with hypercholesterolemia.

Results and Conclusions: We identified a missense mutation in the SCARB1 gene that contributed to Hypercholesterolemia. We recommended lifestyle modifications and suggested meeting a clinician for further suitable treatment. The SCARB1 gene is responsible for cholesterol clearance; however, a mutation in this gene may interfere with the function and lead to hypercholesterolemia.

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Abstract ID: 78

Comprehensive Assessment of Tyrosine Kinase Domain Mutations and Imatinib Resistance in Chronic Myeloid Leukemia

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Background/ Introduction: Chronic Myeloid leukemia (CML) is a clonal haemopoietic stem cell disorder occurs due to t(9;22)(q34;q11.2); molecularly BCR::ABL1 fusion gene. Tyrosine kinase inhibitor (TKI) therapy is proved to be effective for CML patients, however ~40% patients showed resistant to imatinib after certain time. The BCR::ABL1 dependent and independent mechanism proposed to cause drug resistance in CML. The Tyrosine kinase domain mutations (TKDMs) have been reported to be associated with imatinib resistance. Understanding these mutations is critical for optimizing treatment strategies and improving patient outcomes.

Objectives: The primary aim of this study was to identify TKDMs in CML patients treated with imatinib, adhering to the European LeukemiaNet (ELN) guidelines, to enhance treatment management and decision-making.

Materials and Methods: A cohort of 457 patients were studied by G-T-G banding and Fluorescence in-situ hybridization (FISH). The monitoring of the patients was done as per the European LeukemiaNet (ELN) guidelines. The TKDMs were identified through Direct and Next generation sequencing (NGS).

Results and Conclusions: The study was conducted in 457 CML patients; out of these, 37.41% patients were found to be in low-risk category, 42.03% patients were in intermediate category while 20.56% patients were in high risk category as per the SOKAL scoring system. The patients consistent with follow-up (329) were categorized into responder, warning and resistant group according to ELN guidelines. Direct sequencing revealed point mutations in 61 (32.45%) out of 188 non-responder's (Warning and resistant) patients. NGS analysis of the warning group identified mutations in 30.55% (11/36) of patients. Overall, a high frequency (86.12%) of mutations identified in CML-CP patients followed by AP (8.34%) and BP (5.56%) CML patients. In our cohort, P-loop was found to be the most frequently mutated (26) region followed by catalytic domain (19), imatinib binding region (12) and activation loop (9). Out of all the mutations identified, 76.39% were singly occurring point mutations while 19.44% were polyclonal mutations. The mutation detected (p.G250, p.F359, p.Q255 and p.T315) in our cohort are impairing imatinib binding and leading to poorer outcomes. Therefore, detection of these mutations is essentially important. Notably, five novel mutations were discovered that had not been previously reported. Overall, our combined approach detected mutations in 38.30% of imatinib non-responders, including 24 distinct point mutations and three large deletions (4.16%).

In conclusion, the identification of BCR::ABL1 KDMs is important in patients treated with imatinib as we have identified P-loop, catalytic domain and imatinib binding region as the most frequently mutated domain in our study.

Abstract ID: 79

Nucleolin Overexpression: A Key Genetic Factor in Tumorigenesis

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Background/ Introduction: Nucleolin (NCL) is a multifunctional protein crucial for maintaining cellular homeostasis by regulating nucleic acid metabolism, ribosome biogenesis, and cell proliferation. It has been shown to play a significant role in cancer biology. Recent genetic studies reveal that NCL is frequently overexpressed in various cancers, where its dysregulation is linked to both genetic and epigenetic alterations. These alterations influence key oncogenic processes, making NCL a potential biomarker for cancer progression and prognosis.

Objectives: This study aims to investigate the genetic and epigenetic mechanisms underlying the dysregulation of NCL in cancer, analyzing its expression across 33 different tumor types. We seek to identify how these alterations contribute to tumorigenesis and assess the potential of NCL as a genomic biomarker for cancer prognosis.

Materials and Methods: We conducted a comprehensive pan-cancer analysis using data from The Cancer Genome Atlas (TCGA) to evaluate NCL expression levels in malignant tissues compared to normal tissues. Genetic alterations, including somatic mutations, copy number variations, and promoter methylation changes, were examined in relation to NCL expression. Correlations between NCL dysregulation and oncogenic signaling pathways, as well as patient outcomes, were analyzed. Further, KEGG pathway analysis was conducted to examine the pathways impacted by NCL-interacting genes, with enrichment results visualized using the R ggplot2 package to illustrate the pathways influenced by NCL during cancer progression.

Results and Conclusions: Our analysis revealed significant upregulation of NCL in malignant tissues, frequently associated with somatic mutations, copy number amplification, and epigenetic modifications such as promoter methylation. Elevated NCL expression was linked to the activation of oncogenic signaling pathways and poorer clinical outcomes, indicating its role in cancer progression. These findings suggest that NCL plays a pivotal role in tumorigenesis through genetic and epigenetic regulation. Given its correlation with adverse clinical outcomes, NCL shows promise as a genomic biomarker for cancer prognosis. Further research is warranted to explore its potential as a therapeutic target, especially in the context of precision oncology.

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Abstract ID: 80

Temporal gene expression signatures across neurodevelopment: a transdiagnostic analysis of bipolar disorder, schizophrenia, autism, and epilepsy

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Background/ Introduction: Bipolar disorder (BPD), schizophrenia (SCZ), autism spectrum disorder (ASD), and epilepsy (EPI) share biological processes. However, spatiotemporal transcriptomic profiles of the associated genes have not been examined in a transdiagnostic manner. Genes harboring rare variants

identified through whole exome-sequencing (WES) are particularly valuable for investigating disease pathobiology, as they impact gene and protein function.

Objectives: Our aim was to integrate the WES genetic data of the four syndromes with spatiotemporal transcriptomic data and characterize their temporal and regional specificities. Previous studies have examined the genetic correlations between the syndromes. However, a collective analysis of the rare variant harboring genes associated with the four syndromes, and more importantly, their transcriptomic profiles, will help understand the underlying functional themes.

Materials and Methods: We compiled gene-level exome-wide association data of BPD, SCZ, ASD, and EPI from the BipEx consortium, SCHEMA consortium, Autism Sequencing Consortium, and EPI25 consortium, respectively. Then, we examined this data in conjunction with the BrainSpan Atlas transcriptomic data from 26 brain regions, spanning fetal stages to adulthood, using clustering and enrichment analyses.

Results and Conclusions: Temporal clustering showed specific signatures with BPD genes expressed from early infancy to adulthood and ASD genes in early prenatal stages. Spatial clustering revealed enrichment of BPD genes in visual, somatosensory, and motor cortical regions, and ASD genes in fetal ganglionic eminence. EPI gene expression patterns were similar to BPD, and SCZ to ASD, suggesting overlaps of the syndromes. BPD and ASD clusters were enriched for trans-synaptic signaling and chromatin modification, respectively. This study clarifies the neurodevelopmental context of the syndromes from the perspective of rare variants. It bridges the gap between genes, neural circuitry, and developmental stages, and provides insights into the functional implications of the rare variants. It identifies developmental windows and neural substrates for therapy, and highlights gene networks that offer opportunities for experimental research in patient-derived cell lines and animal models.

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Abstract ID: 81

Precision Therapy in Hepatocellular and Pancreatic Cancers: Targeting Oncogenic Drivers and Modulating Immune Responses

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Background/ Introduction: Hepatocellular carcinoma (HCC) and pancreatic ductal adenocarcinoma (PDAC) are aggressive neoplasms characterized by late detection, restricted therapeutic alternatives, and unfavourable prognosis. Oncogenic mutations, such as KRAS in PDAC and β-catenin (CTNNB1) in HCC, propel tumor advancement and therapeutic resistance. The TGF-β pathway also facilitates immune suppression and fibrosis inside the tumor microenvironment, complicating therapy.

Objectives: This research reviews a targeted gene therapy strategy aimed at KRAS and β-catenin, suppressing TGF-β signalling, and augmenting immune response through the modulation of PD-L1, to improve treatment specificity and wear off-target effects.

Materials and Methods: This review explores precision gene therapy in HCC and PDAC. A literature search was conducted using PubMed, Google Scholar, and Scopus for studies from 2014 to 2024.

Keywords included KRAS, CTNNB1, TGF-β, PD-L1, and RNAi. Inclusion criteria focused on peer-reviewed articles addressing oncogene silencing, immune modulation, and targeted delivery systems. Non-cancer studies and those lacking experimental models were excluded. siRNA silencing of KRAS and CTNNB1 inhibited tumor growth, while TGF-β inhibitors reduced fibrosis and boosted immune infiltration. PD-L1 silencing improved anti-PD-1 therapy. Lipid nanoparticles and exosomes ensured targeted delivery to liver and pancreas, with outcomes evaluated via bioluminescence imaging and histological analysis.

Results and Conclusions: This review focuses on the therapeutic strategies for PDAC and HCC by silencing the KRAS and CTNNB1. This leads to the reduction of tumour volume, inhibits the TGF-β and reduced the fibrosis resulting in the improved infiltration of immune cells. PDL1 silencing combined with other anti-PD-1 strategy with enhanced delivery system such as lipid nanoparticles and exosomes improved the overall survival rate with minimal off-target effects offering a promising personalized treatment option.

Conclusion: This oncogenic silencing, microenvironment-modulating, immune checkpoint inhibition combination precision gene therapy strategy robustly inhibited the progression of tumors. The approach provides a framework for the personalization of therapies of HCC and PDAC, with future efforts concentrated on clinical translation.

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Abstract ID: 84

Systems biology-Driven Hub Gene Analysis with Molecular Docking and ADMET Studies for Natural Compound-Based Therapeutics in Oral Cancer.

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Background/ Introduction: Oral cancer is a major global health concern and despite advancements in treatment, the 5-year survival rate remains below 50% due to late diagnosis and high recurrence rates. This highlights the urgent need for new therapeutic strategies, especially those leveraging natural compounds with potential anticancer properties.

Objectives: To identify differentially expressed genes (DEGs) in oral cancer and construct protein-protein interaction (PPI) networks to identify hub genes. The study also involved screening flavonoids for anticancer properties using in-silico molecular docking, ADMET and dynamic simulation studies.

Materials and Methods: Gene expression datasets from GEO were analyzed with GEO2R. Pathway and gene ontology (GO) analysis was performed using DAVID to reveal key biological processes. PPI networks were constructed via STRING and the MCODE and cytoHubba plugins were used to identify hub genes according to different centrality parameters (Betweenness, Closeness, Degree, EPC and MCC). Naturally occurring flavonoids which have reported invitro or invivo anti-cancer properties were obtained from NPACT and assessed for binding affinities using auto-dock Vina against the hub genes. The top candidate compounds underwent ADMET analysis to evaluate their drug-likeness and pharmacokinetic properties. Molecular dynamics (MD) simulations were employed to analyze the stability and behavior of the compound-target complexes under physiological conditions.

Results and Conclusions: GO enrichment function analysis showed that 64 biological processes, 33 cell components and 16 molecular functions were involved. Top 20 hub genes were identified from the PPI

network and 8 genes were common among all the centrality parameters. A total of 150 flavonoids were docked with the hub genes. Best docking scores were obtained with Interferon-Induced Protein with Tetratricopeptide Repeats 1 (IFIT1) with binding energies less than -9Kcal/mol, demonstrating good compatibility between the flavonoids and IFIT1. MD simulation studies supported the results suggesting that flavonoids may affect oral cancer through this primary target which is known to affect tumor growth and metastasis in pancreatic and other solid tumors. Further experimental validation is necessary to confirm the therapeutic potential of these compounds.

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Abstract ID: 85

Clinical implications of CFHR1/CFHR3 partial deletions

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Background/ Introduction: The complement system is crucial for immune surveillance in vertebrates. Deficiencies in this system, factor H and its related proteins (CFHRs) particularly CFHR1 and CFHR3 gene deletions, can lead to diseases such as atypical hemolytic-uremic syndrome (aHUS)1, a rare life-threatening disease on the spectrum of thrombotic microangiopathy (TMA) disorders, and age-related macular disorders.

Objectives: To study the clinical implications of CFHR1/CFHR3 partial deletions.

Materials and Methods: Whole exome or targeted exome sequencing was carried out for the clinical samples on Illumina sequencing platform. Raw data from the Illumina platform was aligned to the hg19 human genome using the BWA algorithm, followed by variant calling with GATK. An in-house pipeline annotated all variants, and a Read Depth-based algorithm was employed to detect copy number variations, comparing normalized read depths between test and control samples.

Results and Conclusions: Results: At our organization, we have tested approximately 600 cases related to renal diseases and identified 73 clinical instances featuring homozygous CFHR1/CFHR3 partial gene deletions. This corresponds to a notable high frequency of around 12% among patients with renal disorders. The 73 clinical cases exhibited a diverse range of clinical manifestations, including thrombotic microangiopathy (TMA), atypical hemolytic uremic syndrome (aHUS), hyperoxaluria, focal segmental glomerulosclerosis (FSGS), polycystic kidney disease (PKD), Alport syndrome, complement component 3 glomerulopathy (C3G), and membrane proliferative glomerulonephritis (MPGN), along with other diseases displaying multiple clinical presentations. Additionally, we identified other clinically relevant variants in 50.7% (n=37) of the cases, while 49.3% (n=36) were associated solely with CFHR1/CFHR3 partial gene deletions.

Conclusion: Our findings reveal a notably high frequency of CFHR1/CFHR3 partial gene deletions in patients with renal disease in the Indian population, accompanied by significant clinical heterogeneity linked to these deletions. Furthermore, the presence of variants with CFHR1 and CFHR3 deletions introduces complex factors that influences the clinical conditions.

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Abstract ID: 86

Association of genetic variants with Rheumatoid Factor and Anti-CCP in deciphering heterogeneity between Rheumatoid Arthritis Patients

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Background/ Introduction: Rheumatoid arthritis (RA) is a chronic inflammatory autoimmune disease affecting approximately 1% of the global population. It is characterized by autoantibodies such as Rheumatoid Factor (RF) and Anti-cyclic citrullinated peptide antibodies (Anti-CCP). These serological markers aid to discriminate between the RA subgroups associated with disease severity and progression. It is well established that the Human Leukocyte Antigen (HLA) region includes notable genetic variants making an individual susceptible to RA, yet the contribution of specific genetic variants in RA heterogeneity remains underexplored.

Objectives: This study aims to identify genetic variants linked to seropositive RA, with an emphasis on their correlation with RF and Anti-CCP levels as well as to investigate their role in RA heterogeneity.

Materials and Methods: Genotyping was performed using microarray based technique. Genome-wide association study (GWAS) was conducted on RA patients recruited for Panchakarma treatment which were grouped into RF-positive (n=106) and RF-negative (n=65) as well as Anti-CCP positive (n=89) and Anti-CCP negative (n=82) groups. Quality control was performed using PLINK for filtering SNPs by minor allele frequency (MAF) and genotype call rate. Gene Ontology analysis was conducted with STRING software to identify molecular functions and biological processes associated with genetic variants. Associations with RF and Anti-CCP status were evaluated using logistic regression whereas linear regression was used to examine the quantitative levels of RF and Anti-CCP, considering age and sex as covariates.

Results and Conclusions: Significant SNPs were identified within the HLA region of chromosome 6 in relation to RF and specifically with Anti-CCP status along with non-HLA loci as well. In particular, there are 9 SNPs which are common to both the groups indicating the shared genetic mechanisms influencing seropositive RA. Enriched immune-related processes, including peptide antigen assembly and antigen presentation via MHC class II, suggest potential mechanisms underlying seropositive RA heterogeneity. For the Anti-CCP group, linear regression was employed to identify SNPs on the HLA regions that exhibited both, positive and negative associations with serum Anti-CCP levels as opposed to RF group, confirming that Anti-CCP are more specific biomarkers for RA than RF.

Conclusion: This study reveals the critical role of HLA gene variants and immune-related processes in shaping RA heterogeneity associated with RF and Anti-CCP levels, thereby broadening the knowledge of different RA subtypes. Furthermore, there is potential for future research to augment upon these results by incorporating additional covariates and exploring their interactions, thereby increasing our understanding of the complex mechanisms driving RA.

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Abstract ID: 87

Clinical and Molecular Findings in a Male Child with 47,XX,+i(Y)(p10)/46,XX Mosaicism: Implications for Genetic Counseling and Management

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Background/ Introduction: Disorders of sex development (DSD) includes a heterogeneous group of conditions resulting in atypical development of internal and/or external genitals, which are broadly classified to sex chromosome DSD, 46, XX DSD, and 46, XY DSD. Nearly 85% of 46, XX testicular DSD presents after puberty and only 15% present at birth with ambiguous genitalia. To date, SRY-positive 46, XX testicular DSD has shown the presence of SRY on X chromosome or an autosome. Here, we report the first case of non-syndromic 46, XX testicular DSD with 46,XX and 47,XX,+i(Y)(p10) mosaicism in a male child, where the isochromosome (i) Yp is present only 10% of the cells. The underlying mechanism of i(Yp) formation resulting in this karyotype remains challenging to elucidate. An in-depth understanding of the genetic mechanisms resulting in DSD can help with more effective genetic counseling.

Objectives: To delineate the clinical and genetic diagnosis of a 2-year-old male child with ambiguous genitalia as well as to explore the potential molecular and cytogenetic mechanisms that led to this karyotype.

Materials and Methods: The proband is a 2-year-old male child who presented with ambiguous genitalia. On examination, his anthropometric measurements were at mean. He had micropenis. The right testis was palpable in the scrotum, and left testis palpable in the inguinal canal. Magnetic Resonance Imaging of pelvis described normal right testis and left atrophied testis in the left inguinal region.

Karyotype and FISH for SRY (Yp11.31), DYZ1 (Yq12), and DXZ1 (Xp11.1-q11.1) was performed followed by an informed consent.

Results and Conclusions: FISH showed two signals for DXZ1 indicating 46, XX pattern with absence of SRY gene in 90% of the cells. The other 10% showed the presence of two X chromosomes along with two signals for SRY gene. The SRY gene was not present on the X chromosome or an autosome raising the possibility of a marker chromosome. Both cell lines did not show any signals for markers on the Yq region. Karyotype from peripheral blood showed the presence of a supernumerary marker chromosome. Based on the evidence from these cytogenetic assays, the karyotype of the child is mos 47,XX,+i(Y)(p10)[8]/46,XX[64] indicating mosaicism.

The cytogenetic mechanism behind this karyotype is assumed to be a non-disjunction event in meiosis 1 of spermatogenesis followed by misdivision in meiosis 2 forming i(Yp). Due to instability of the marker chromosome, mitosis during tissue formation may have led to mosaicism, which explains the likelihood of varying degrees of mosaicism across different tissues, as expected in our case. We hypothesize that a higher percentage of the i(Yp) cell line exists in the gonadal tissue.

In conclusion, we report a novel case of a male child with i(Yp) mosaicism involving 46, XX cell line. Understanding the underlying mechanism involved in the formation of abnormal chromosomal patterns can significantly enhance genetic counseling.

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Abstract ID: 88

Pulmonary Embolism with Underlying Family History Presenting as Syncope: A Case Report

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Background/ Introduction: An artery blockage by a foreign body, such as a blood clot/fat/air/cholesterol/amniotic fluid, is called an embolism. The most serious conditions caused by an embolism are stroke and pulmonary embolism. Pulmonary embolism (PE) occurs when a foreign body blocks the vessel that carries blood from the heart to the lungs. Deep vein thrombosis (DVT) has the potential to embolize and travel through the right side of the heart and become lodged in the blood-supplying artery of the lungs: the pulmonary artery. DVT is one of the major causes of pulmonary embolism. Pulmonary embolism is a life-threatening disease that can sometimes be problematic to point at, especially when the patient has no obvious symptoms. The risk factors may not be strikingly palpable, and there may also be an intersection between the symptoms and signs of pulmonary embolism and other diseases. Syncope is a comparatively easy clinical symptom to detect but has varied etiologies that lead to a standard cause in only 58% of syncopal events. It is a difficult correlation to make when syncope is the presenting symptom of pulmonary embolism. Family history in the case of undiagnosed pulmonary embolism presenting with symptoms that point in no particular direction becomes crucial in determining the disease.

A transient cessation of blood flow to the brain is what causes syncope. It can be brought on by a drop in blood pressure, heart rate, or variations in the distribution of blood throughout the body. Syncope is a clinical symptom that is simple to identify, but it can have a wide range of etiologies, making it difficult to associate it with a known pulmonary embolism (PE) pathology. PE can manifest in many different ways; it can range from being asymptomatic to sudden cardiac arrest. Patients with a self-reported lineage of venous thromboembolism in first-degree kin are more inclined to be diagnosed with an acute pulmonary embolism in the emergency room, even among people assumed to have a greater risk of PE.

Kelly et al. found that 19.4% of the 3024 research participants had a family history of venous thromboembolism, and 1.9% of them received an acute PE diagnosis in the emergency department. 3.2% vs. 1.6% ($p = 0.009$) of patients with a family history of venous thromboembolism had a PE diagnosis. 82.3% of patients tested positive for the criterion for ruling out pulmonary embolism (PERC), and 3.6% vs. 1.9% of PERC-positive patients with a family history of VTE were found to have PE ($p = 0.016$). PE diagnoses were more frequent among patients having a history of VTE in their family: 9.4% vs. 4.9% ($p = 0.032$) of patients who underwent testing for PE (3.7%) [1]. Undiagnosed PE can be life-threatening, with up to 30% of deaths among those undiagnosed.

Objectives: The objective of the case report "Pulmonary Embolism With Underlying Family History Presenting as Syncope" was to highlight a unique presentation of pulmonary embolism (PE) in a 38-year-old male patient with a family history of cardiovascular issues, specifically a twin who had passed away from a stroke. This report aims to underscore the diagnostic challenge that arises when PE presents as syncope (a sudden loss of consciousness), which can complicate identifying the true cause due to the lack of typical PE symptoms such as chest pain or dyspnea. The case emphasizes the importance of considering family history and genetic predisposition in PE diagnoses, particularly when symptoms are atypical or masked by other conditions

Materials and Methods: Patient information

A 38-year-old young man was brought to the Critical Care Centre by a relative due to loss of consciousness and a history of a fall 30 minutes before being admitted. The family has a history of hypertension. The

patient's twin brother died due to a stroke three months ago. The patient was not on any medications before being admitted to the hospital.

Clinical findings

On clinical examination, the patient was found to be tachypneic, spO2 was 35%, blood pressure recorded was 90/70 mm Hg, respiratory rate 46 bpm, breath sounds were decreased in both the lungs and the pleural rub was noted in the right lung.

Diagnostic Assessment

A two-dimensional echo revealed minimal pericardial effusion, right ventricular dilatation, and inferior vena cava congestion; no regional wall motion abnormalities were seen. Chest X-ray revealed pulmonary effusion with fistural extension; computed tomography (CT) pulmonary angiogram showed the complete collapse of the right lower lobe sparing anterior segment and right pleural effusion; partially obstructing intra-luminal thrombi in the second and third-degree branches in the postero basal segment of lower lobes; the left lung appeared normal in volume, heart and other structures were normal in size.

CBC revealed haemoglobin (Hb) was low at35 9.3 gm/dl (13-1and 8), total leukocyte count (TLC) elevated: 12400/cu.mm (4000-11000), Lymphocytes 80%, Neutrophils 20%, RBCs 18-20/HPF, protein 5.3gm/dl, glucose 182.4 mg/dl31. Peripheral smear examination showed microcytic hypochromic RBCs with mild anisopoikilocytosis, few target cells, and teardrop cells. D-dimer test came out to be 14.4 ug/mL.

Findings of ultrasonography-guided pleural tapping confirmed right-sided pleural effusion with the sub-segmental collapse of the underlying lung (Figure 1). On pleural tapping, 300 ml of straw-colored fluid was aspirated from the right hemi thorax. A pleural fluid examination of the aspirated fluid showed pleural fluid Adenosine Deaminase (ADA) 49.5U/L (<30U/L). ECG showed sinus tachycardia with a short PR interval. C-reactive protein was elevated at 45.7 mg/L (≤5), liver function test showed elevated serum glutamic oxaloacetic transaminase (SGOT) 101U/L (up to 40), serum glutamic pyruvic transaminase (SGPT) 77.8 U/L (up to 40). The Renal Function Test came out to be normal. The patient had no history of lower limb fracture, air travel, cancer, or hemoptysis during the preceding four weeks.

Diagnosis

This case was identified as a Pulmonary Embolism based on the recommendations from the ESC (European Society of Cardiology) and ERS (European Respiratory Society) guidelines of 2019.

Results and Conclusions: When syncope is the sole presenting symptom, it may be challenging to diagnose pulmonary embolism. Physicians should keep family history in mind as it might be a useful aid in making a tough PE diagnosis. In order to identify and treat PE, doctors must follow the recent recommendations issued by the European Society of Cardiology and the European Respiratory Society.

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Abstract ID: 89

Identification of Molecular Biomarkers for Improved Tumor and Non-Tumor Demarcation in Oral Squamous Cell Carcinoma

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Background/ Introduction: Oral squamous cell carcinoma (OSCC) is the most common cancer among men in India, while the fourth major cancer for women. Despite advancements in treatment and screening, its incidence continues to rise, and prognosis remains poor. OSCC accounts for 90% of head and neck cancers (HNSC). One major challenge with OSCC is its high recurrence rate which is about 50–60% for patients with advanced stages and 25–30% for early-stages. To improve patient prognosis and address recurrence, it is essential to achieve adequate surgical margins for which this study identifies and validates potential biomarkers that can effectively differentiate between tumor and non-tumor tissue.

Objectives: To identify effective molecular biomarkers capable of distinguishing between oral tumor and non-tumor tissue through transcriptomic analysis and validate their differential expression using qRT-PCR.

Materials and Methods: This study involved RNA isolation from tumor and adjacent normal oral tissue samples as well as from buccal cells of healthy individuals. Following RNA isolation, library preparation, next generation sequencing (NGS) and transcriptomic analysis of tumor and adjacent normal tissue samples were performed. Differentially expressed genes were identified as potential biomarkers for tumor and non-tumor demarcation. These biomarkers were further validated through qRT-PCR using specifically designed primers, and their expression levels were compared among tumor (18 samples), adjacent normal (14 samples), and healthy individual's samples (19 samples).

Results and Conclusions: Transcriptomic analysis, including differential expression analysis, gene ontology, TCGA database validation, ROC curve analysis and PPI network analysis, identified 12 differentially expressed genes located in the plasma membrane and matrix that may serve as potential biomarkers: ADAM12, PDPN, LAMC2, CA9, COL1A1, COL1A2, COL3A1, COL4A1, COL4A2, MMP10, MMP13 and POSTN. qRT-PCR results revealed that the expression of most of these biomarkers was significantly higher in tumor samples compared to adjacent normal samples, and all biomarkers showed elevated expression in tumor samples relative to healthy individual's samples. Interestingly, differential expression analysis between adjacent normal and healthy individual's samples indicated that for some biomarkers, expression was higher in adjacent normal samples, suggesting that these tissues may still harbor molecular features of tumor involvement. This finding supports the need for more precise surgical margin clearance to accurately classify tissue as non-tumor.

Thus, these molecular biomarkers have the potential to effectively differentiate between tumor and non-tumor tissues, potentially improving surgical outcomes by enabling more precise margin assessment, thus helping to reduce OSCC recurrence rates and improve prognosis.

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Abstract ID: 90

Study of molecular prognostics in Myelodysplastic Syndromes: The Impact of Gene Mutations on Clinical Outcomes

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Background/ Introduction: Myelodysplastic syndromes (MDS) are clinically heterogeneous clonal haematopoietic stem cell disorder characterized by inefficient haematopoiesis, cytopenia and risk of leukemic transformation. Chromosomal aberrations and somatic mutations play an important role in diagnosis and prognosis of the disease. We have carried out a study on large cohort of MDS to identify the genetic aberrations and their role in overall survival of MDS patients, to understand the molecular pathology of the disease.

Objectives: Study of genomic alteration and their impact on overall survival to understand the molecular pathology of MDS.

Materials and Methods: The study carried out in 237 MDS patients including 123 males and 114 females. Giemsa Staining of BMA/PB Smears was carried out as per standard procedure to classify MDS patients according to clinic - pathomorphological criteria of WHO Classification (2016). Cytogenetic study was carried out using GTG banding and fluorescence in situ hybridization (FISH). The Next generation sequencing (NGS) at higher depth (>250X) was carried out to identify gene mutations using custom capture kit for selective target enrichment followed by clinical exome sequencing at the Med-Genome Labs Pvt Ltd, Banglore, India. The time of diagnosis to death due to any cause since last follow-up was considered as overall survival (OS). The statistical analysis was carried out using GraphPad Prism 5 and SPSS version 20 software (IBM Corp., Armonk, NY, USA) from the Survival package.

Results and Conclusions: The age of 237 MDS patients ranges from 14 years to 90 years and the median age was 54.5±16.92 years. The MDS subgroup in our study was MDS- SLD-88 (37%) followed by MDS-MLD- 79 (33%), MDS-EB1-32 (13.5%), MDS-EB2-21 (8.8%), H-MDS-12 (5%) and U–MDS–5 (2.1%). A high frequency of patients, 173 (73%) belonged to low-risk MDS subgroups as compared to high risk MDS subgroups 64 (27%). Cytogenetic study revealed chromosomal aberration in 96 (40%) MDS patients. The NGS identified 43 different gene mutations in 70% (n=167) of our cohort. The most frequent mutated genes were SF3B1 (18%), SRSF2(13%) U2AF1 (9.5%) ASXL1 (12%), RUNX1 (9%) TET2 (8.3%), TP53 (8.3%), ATM (7.1%). The survival analysis revealed that the mutations in TP53, JAK2/3, KRAS, NRAS and ASXL1 were significantly ($P < 0.05$) associated with poor survival of the patients. The univariate cox and multivariate cox analysis of our study suggested that the age, marrow morphology, cytogenetic and gene mutations with IPSS-R should be considered for prognosticating the MDS patients. In conclusion, mutations in SF3B1, SRSF2, U2AF1, and TP53 is prevalent and showed distinct clinical outcomes and survival rates. By integrating genetic data with clinical and cytogenetic findings, we propose a more comprehensive prognostic framework that can guide personalized management strategies for MDS patients.

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Abstract ID: 91

Exploring the Anti-Cancer Potential of Murraya species through Molecular Docking and ADMET Approach Targeting Key Cancer Receptors

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Background/ Introduction: Cancer treatment often involves toxic therapies with severe side effects. Traditional herbal medicines like Murraya species are known for their bioactive compounds and therapeutic potential, suggesting they could be safer alternatives or complementary options in cancer therapy.

Objectives: To investigate the anti-cancer potential of bioactive compounds derived from Murraya species by targeting key cancer-associated receptors; ER, AR, HER2, VEGF, EGFR etc. through molecular docking, molecular dynamics simulation, and ADMET analysis, with the goal of identifying viable candidates for experimental validation as natural anti-cancer agents.

Materials and Methods: Bioactive chemicals from Murraya species were catalogued by mining phytochemical databases. Molecular docking studies were conducted using Autodock-Vina to assess the binding affinities of selected compounds with the key cancer related targets. The top candidate compounds underwent ADMET analysis to evaluate their drug-likeness and pharmacokinetic properties. Molecular dynamics (MD) simulations were employed to analyze the stability and behavior of the compound-target complexes under physiological conditions.

Results and Conclusions: Murraya compound mahanimbine and murayamine exhibited strong binding affinity with the HER2 receptor, with binding energies in the range of -10 kcal/mol, which is notably higher than that of the reference inhibitor. Molecular dynamics simulation studies supporting its potential efficacy making it a strong candidate for further experimental validation and development as a natural HER2 inhibitor.

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Abstract ID: 92

Tubular cell mitochondria in renal disorder

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Background/ Introduction: The kidney is rich in mitochondria, and any alterations or damage to tubule cell mitochondria play the important role in renal metabolic activities and in the pathogenesis of various kidney diseases. Quantitative analysis of mitochondrial concentration, size and shape is essential for understanding mitochondrial biology and renal disorder.

Objectives: This study assessed the tubular cell mitochondrial morphometric parameters using images obtained from transmission electron microscope (TEM) in different renal disorder and looked into how they correlated with glomerular filtration.

Materials and Methods: This is a retrospective morphometric study of mitochondria in renal tubular cell, and it includes sixty five cases of renal disease treated at this tertiary care center and going through TEM diagnosis during 2022-2024. TEM images of glutaraldehyde-osmium tetroxide fixed epoxy-resin

embedded 70nm thick sections were used for the evaluation of (i) minor axis (MinX) (ii) major axis (MajX) (iii) area, (iv) perimeter, (v) aspect ratio and (vi) roundness of mitochondria in renal tubular cells using QuPath software. Mitochondrial density (MDensity), % of mitochondrial space (MSpace) and mitochondrial surface density (MSDensity) in the cytoplasm of tubular space were estimated for each sample. Mean value with standard deviation (SD) and range of the studied parameters were calculated for all the samples. Analysis of variance (ANOVA) tests with post hoc ANOVA (least significance difference) test was performed to analyze the difference of the means of the histo-pathological groups for the studied parameters. Pearson correlation was performed between the studied parameters to understand the disease process.

Results and Conclusions: Serum creatinine exhibited a negative link with mitochondrial density, area and perimeter and good negative correlations with MSDensity and MSpace. Significant correlations among the studied mitochondrial morphometric parameters were also observed. MDensity, MinX, MajX, Area, Perimeter, Aspect ratio and Roundness of mitochondria showed significant difference among the studied groups of renal disease. This study indicates that the variation of mitochondrial concentration in renal tubular cells, could be important factors in the renal function disorder.

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Abstract ID: 93

Sexual antagonism in the multivariate genetic architecture of contemporary humans

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Background/ Introduction: Sex-difference (SD) is widespread in humans despite males and females sharing almost the entire genome. In humans, SD is usually studied by estimating the genetic correlation between the sexes for a single trait (e.g., correlation between height in males and females). But inspecting SD in single traits does not provide a complete biological overview, because traits are not independent and are functionally related (e.g., lipid and blood pressure levels together determine the cardiac disease status of an individual). One way to circumvent this is to investigate SD under a multivariate framework by estimating the additive genetic (co)variance between sexes which includes the cross-sex-cross-trait genetic covariances (e.g., covariance between height in males and weight in females).

Objectives: To investigate SD in the multivariate genetic architecture of 12 anthropometric and sex-hormonal traits in humans by estimating cross-sex-cross-trait genetic covariances.

Materials and Methods: Using such a multivariate approach, we investigated SD from publicly available genome-wide association summary statistics of the UK Biobank under a Linkage disequilibrium score regression framework.

Results and Conclusions: We compared the genetic (co)variance matrices between the sexes and found them to be different, which was not apparent from studying single traits in isolation. Intriguingly, the directions of 27% of the cross-sex-cross-trait covariances were opposite, between testosterone and anthropometric traits which indicates extensive sexual antagonism in humans driven by testosterone. This indicates that variants which increases testosterone in males decreases BMI in females, but variants which increases testosterone in females increases BMI in males. Using the multivariate framework, we predicted short-term evolution in males and females under simulated selection pressure. We found that the shared genetic architecture between sexes acts as a constraint to the evolution of SD in humans only when selection is sexually antagonistic and not concordant. Moreover, we observed that lifetime reproductive

success is positively genetically correlated with anthropometric traits in both the sexes but not with testosterone. This work is the first ever work on multivariate genetic architecture in humans from genotyped data. Our study emphasizes the necessity and importance of using a multivariate framework in investigating sex-difference in humans.

Abstract ID: 94

Rare genetic variants in circadian rhythm and sleep homeostasis genes among attention deficit hyperactivity disorder

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Background/ Introduction: ADHD is a prevalent neurodevelopmental disorder characterized by persistent patterns of hyperactivity, inattention, and impulsivity resulting in significant difficulties in an individual's functioning. Sleep and ADHD have a bidirectional relationship where sleep problems are reported in around 25 to 50 % of children with ADHD. We posit that the sleep-regulating molecules might have a role in the pathophysiology of ADHD, which is mediated through genetic aberrations in the human circadian clock and sleep homeostat genes.

Objectives: (i) Assemble a cohort of individuals with ADHD and evaluate their sleep phenotypes using the Children's Sleep Habits Questionnaire (CSHQ) and polysomnography. (ii) Evaluate the spectrum of rare genetic variants with potential deleterious effects in the genes involved in circadian rhythm and sleep homeostat mechanisms in ADHD individuals.

Materials and Methods: The study was approved by the human ethics committee of IMHANS. Individuals aged 5-12 years with ADHD diagnosed using DSM-5 criteria, with no family history of mental disorders and drug use, were selected from the child psychiatry outpatient clinic of the host institute. Conners' Parent Rating Scale was used to score ADHD. Sleep phenotypes were recorded using the Children's Sleep Habits Questionnaire (CSHQ) and polysomnography (PSG). Saliva samples were collected to isolate DNA for next-generation sequencing of circadian clock and sleep genes which were selected from the gene ontology terms.

Results and Conclusions: A total of 174 individuals were recruited (age:8.3 ± 2.3, 88.5% males). A large proportion of individuals (97.7%) had sleep deficits which were validated in PSG. A subset of individuals (n = 29) was sequenced for 176 candidate genes (circadian rhythm; n =166, sleep; n = 10). Out of 176 candidate genes screened, 141 genes showed variation among 29 samples. A total of 1321 SNP variants were observed, of which 1320 were found to be missense mutations and 1 nonsense mutation. 54 rare variants were found to have a global allele frequency of less than 0.001%. Ten genetic variants were not reported in the control South Asian population. Deleterious loss-of-function genetic variants were observed in BHLHE41, MRGPRX2, NRIP1, SRRD, ZNF677, and TIMELESS. Taken together, we showed the spectrum of rare genetic variants in the circadian clock and sleep genes in individuals with ADHD. The identified genetic variants will be further evaluated mechanistically by in silico and in vitro methods to help elucidate their role in the etiology of ADHD.

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Abstract ID: 95

Genetic Markers of Dyslipidemia in Indian Adolescents

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Background/ Introduction: In India, cardiovascular diseases (CVD) are the leading cause of death and disability, characterized by a higher risk burden and earlier onset. This may be due to a high genetic susceptibility combined with environmental factors, such as a high-carbohydrate diet. While replication studies of known lipid-associated variants and genome-wide association studies on lipids have been conducted in Indian adults, studies focusing on children and adolescents are limited. Elucidating the genetic basis of lipid metabolism in children is essential for early intervention in dyslipidemia and subsequent cardiovascular risk.

Objectives: Identification of common genetic variants and functional variants associated with childhood dyslipidemia in Indian adolescents

Materials and Methods: A two-staged Genome-wide association study (GWAS) and an Exome-wide association study (ExWAS) was performed to identify potential regulatory loci associated with childhood lipid parameters - HDL, LDL, Triglycerides, and total cholesterol. The GWAS involved a discovery phase (1384 samples) where a genome-wide scan for polymorphisms was performed, and the lead signals from the discovery phase were validated in the replication phase (4391 samples) in an independent sample set. The associations were tested using a generalized linear model in PLINK, adjusting for age, sex, BMI, and the first ten principal components assuming additive effect. Heritability and genetic correlation were estimated for the traits. Further, ExWAS (N=5210) was used to identify functional variants for dyslipidemia. Pathway enrichment analysis was performed for lead associations.

Results and Conclusions: The two-staged GWAS in up to 5775 Indian adolescents for 75,64,548 variants (after imputation using South Asian population data in TOPMED reference panel), and the ExWAS (N=5210) for childhood lipid parameters identified the role of common variants in CETP and ALDH1A2 for HDL; APOE, CELSR2, PSRC1, APOC1, TM6SF2, CILP2, and TOMM40 for LDL and total cholesterol; and APOA5, ZNF259, BUD13, TM6SF2, GCKR, ATP8B3, GYS2, CEP162 and ZPR1 for triglycerides. The study also found a high genetic correlation between LDL and total cholesterol (p=1.8x10-9), with shared loci influencing these traits. Tagging ExWAS along with the GWAS pinpointed high-effect functional variants within each locus. Pathway analysis showed enrichment in biological processes including phospholipid transport, lipoprotein particle remodelling, triglyceride homeostasis, etc. These genetic insights pave the way for polygenic risk scores that could enable early detection and management of lipid disorders, potentially preventing future CVD.

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Abstract ID: 96

Crosstalk between genetics and epigenetics in nonsyndromic hearing loss

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Background/ Introduction: Nonsyndromic hearing loss (NSHL) constitutes 70% of congenital hearing loss. Early detection of the condition is challenging due to the lack of other associated symptoms. The

genetic landscape of NSHL is highly heterogeneous among different populations. Studies have identified different causative loci and genes associated with the condition, among which mutations in the GJB2 gene majorly contribute to the development of NSHL. It is also evident that aberrant DNA methylation can result in audiological defects. Epigenetic variations in NSHL-associated genes are reported; similarly, mutations in the DNA methylating enzymes are found to be associated with NSHL. This study aims to identify genetic variations in NSHL patients and evaluate the interplay between genetic and epigenetic causes of NSHL in the study cohort.

Objectives: 1. Identify genetic variations in the NSHL patients from Kerala population, and 2. Evaluate the methylation profile of NSHL patients with and without GJB2 mutation

Materials and Methods: Whole genome genotyping was performed in 127 patients and 143 controls, followed by a case-control association study. CNVs were computationally predicted from the genotyping data. Methylation profiling of 24 NSHL patients (12 GJB2 mutation positive and 12 GJB2 mutation negative) was performed using Infinium Epic Array. A comprehensive Quality control report has been generated using minfi, ChAMP & RnBeads. Significantly differentially methylated regions (DMRs) are selected based on DiffScore > 13 / < -13 and delta beta > 0.2 / <-0.2.

Results and Conclusions: In our study population, 10 markers were significantly associated to NSHL, including the GJB2 pathogenic mutation, which causes protein truncation. Twenty NSHL-associated genes were discovered to harbour CNVs in the coding region as well as in their regulatory regions. The methylation profiling of NSHL patients with or without the GJB2 mutation revealed 248 significant DMRs in which 130 were hypermethylated and 118 were hypomethylated. The DMRs are mapped to 31 unique genes that mostly affect the sensory transduction pathways, cytoskeletal dynamics and neuro developmental functions that in turn affect auditory health.

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Abstract ID: 97

Novel biallelic variants in AHR causing foveal hypoplasia 3

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Background/ Introduction: Aryl hydrocarbon receptor is a ligand-dependent transcription factor, encoded by AHR, that detects environmental toxins and endogenous ligands, inducing detoxifying enzymes and regulating immune cell responses. Biallelic disease-causing variants in AHR are associated with foveal hypoplasia 3 (MIM# 620958). Homozygous variants in AHR are also associated with retinitis pigmentosa 85 (MIM# 618345). To date, seven individuals from four unrelated families have been reported with disease causing variants in AHR.

Objectives: Description of clinical findings and genomic alterations in an individual who presented with foveal hypoplasia, hypopigmentation of skin, and hypothyroidism.

Materials and Methods: Informed consent was obtained after clinical evaluation, and whole exome sequencing (WES) was performed. Sanger sequencing was done to validate and segregate the variants in family.

Results and Conclusions: We ascertained a two-year-old female, born to a consanguineous couple, presented with hypopigmentation of skin and hair, congenital hypothyroidism and vision difficulties. Ophthalmological evaluation revealed hypermetropic disc, pigmentary retinal changes with foveal hypoplasia. Biochemical investigations showed lymphopenia, eosinophilia, and increased serum IgE levels.

WES analysis revealed a homozygous stop gain variant, c.528G>A p.(Trp176Ter) in AHR (NM_001621.5) in the proband. Sanger sequencing revealed the carrier status of this variant in her parents. This variant is not observed in population databases and our in-house database of exomes. This variant is predicted to cause premature termination of the transcript, which will either lead to the nonsense-mediated mRNA decay or formation of a truncated protein product. Additionally, a compound heterozygous variant [c.1264C>T];[c.1100T>C] [p.(Arg422Cys)]; [p.(Leu367Pro)] in DUOX2 (NM_001363711.2), was observed in the proband, known to cause thyroid dyshormonogenesis 6 (MIM# 607200). This variant was ascertained as the probable cause of hypothyroidism. We did not identify any genomic alterations related to the skin phenotype in proband.

We thus herein report an additional family and genotype of this rare condition caused due to biallelic variants in AHR.

Acknowledgement: We thank the patient and families for participation. This work was supported by DBT/Wellcome Trust India Alliance (IA/CRC/20/1600002).

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Abstract ID: 98

Novel biallelic intronic variant, c.172+5G>A in WWOX leads to aberrant splicing

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Background/ Introduction: The increased accessibility of next-generation sequencing has led to a rise in the identification of variants of uncertain significance (VUSs). The intronic variants are often classified as VUS and pose an immense challenge for establishing diagnosis and genetic counseling. We herein perform splice-site assay on a novel biallelic intronic variant identified in WWOX causing autosomal recessive developmental and epileptic encephalopathy 28 (DEE28, MIM #616211) in the proband.

Objectives: To describe the functional impact of a novel intronic variant in WWOX identified through splice-site analysis.

Materials and Methods: Mendeliome was performed for the proband after detailed clinical evaluation and obtaining informed consents. Splicing assay was performed in RNA isolated from patient-derived skin fibroblasts in the proband. For nonsense-mediated decay (NMD) analysis in the patient fibroblast, cells were treated with DMEM containing cycloheximide (CHX) for 5 hours, followed by RNA isolation and cDNA conversion.

Results and Conclusions: We ascertained a two-year-old female with global developmental delay, spasticity, epilepsy and mild dysmorphism. Exome sequencing identified a novel intronic variant,

g.78108492G>A (NM_016373.4:c.172+5G>A) in homozygous state in intron 2 of WWOX gene in the proband correlating with her phenotype. Segregation analysis showed the variant in heterozygous state in her parents. This variant is absent in heterozygous and homozygous state in population database gnomAD and in our in-house database of 3356 exomes. In-silico analysis tools (SpliceAI, Human splicing finder) are consistent in predicting the variant to cause aberrant splicing. According to the American College of Medical Genetics and Genomics guidelines, the variant was classified as VUS. Splice-site analysis performed on cDNA from patient-derived skin fibroblasts showed that the transcripts are undergoing NMD. Following the NMD assay, the transcripts were rescued and cDNA sequencing showed aberrant splicing leading to exon 2 skipping causing the frameshift deletion of 65 nucleotides, r.108_172del p.(Asn36Lysfs*11) compared to control. This helped in reclassifying the variant from VUS to likely pathogenic further aiding patient diagnosis, prenatal testing, precision therapy and genetic counseling. We herein emphasize the potential of employing fundamental and feasible assays, such as splice-site analysis to resolve VUSs, suggesting their valuable integration into routine genetic testing. Acknowledgements: We thank National Institutes of Health, United States, for funding the study, “Genetic Diagnosis of Neurodevelopmental Disorders in India” (1R01HD093570-01A1)

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Abstract ID: 99

Role of BDNF genetic variants in determining sleep quality in individuals with schizophrenia

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Background/ Introduction: Sleep disturbances, especially poor sleep quality are a common phenotype in schizophrenia, which exacerbate symptom severity and complicate treatment outcomes. Brain-derived neurotrophic factor (BDNF) is a critical neurotrophin involved in neuronal plasticity and has been implicated in schizophrenia pathogenesis. Moreover, BDNF levels also play a significant role in regulating sleep architecture and quality. BDNF genetic variant rs6265 (G196A, Val66Met) is known to regulate BDNF levels and has shown genetic association with sleep disturbances and with schizophrenia. However, it is unknown whether rs6265 is associated with the poor-quality sleep that is observed in schizophrenia patients. Here, we aim to examine the genetic association of BDNF genetic variant rs6265 with sleep quality in individuals with schizophrenia.

Objectives: (i) Assemble a cohort of individuals with schizophrenia and evaluate their sleep phenotypes using the Pittsburgh Sleep Quality Index (PSQI). (ii) Evaluate the allelic spectrum of the rs6265 genetic variant and test their association with sleep quality in individuals with schizophrenia.

Materials and Methods: The study was approved by the human ethics committee of the IMHANS. Individuals (n = 150, age > 18) diagnosed with schizophrenia (DSM-V criteria), were recruited for the study. Sleep phenotypes were recorded using the Pittsburgh Sleep Quality Index (PSQI). Good and poor sleep quality were defined as PSQI scores <5 and >5 respectively. DNA was isolated from peripheral blood samples for BDNF rs6265 genotyping by polymerase chain reaction and restriction digestion.

Results and Conclusions: A total of 150 individuals were recruited (age: 38 ± 11.7, 88.5% males, with age of onset:26.7±9.2. Large proportion of individuals (89.8%) had sleep deficits. No allelic or genetic association of BDNF rs6265 was observed with sleep quality in schizophrenia. Thus, the findings imply that BDNF genetic variants may not be strong determinants of sleep quality in schizophrenia samples. Future research should consider larger sample sizes and additional genetic factors to further explore the relationship between genetics and sleep disturbances in schizophrenia.

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Abstract ID: 100

Complexity Of A Mendelian Disease: Can Variants In Atp7b And Modifier Loci Regulate Cognitive Decline And Other Clinical Manifestations In Wilson Disease?

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Background/ Introduction: Wilson Disease (WD), caused by mutations in ATP7B, is a rare autosomal recessive disorder of copper metabolism. WD is characterized by a wide range of hepatic and neurological symptoms including cognitive decline and patients bearing the same ATP7B mutations show varied clinical manifestations. This study aims to screen variants in ATP7B and modifier loci to correlate the variants with phenotypic differences among WD patients.

Objectives: Genotyping of ATP7B and other key Cu-metabolism genes/genes associated with neurological/hepatic manifestations in diseases having overlapping phenotypes with WD and establish a genotype-to-phenotype correlation

Materials and Methods: The clinical symptoms of the patients were evaluated by expert neurologists; cognitive decline was assessed through questionnaires established to comprehend the patients' orientation, attention, language, fluency, memory, visuospatial, and executive function skills. Genotyping of variants was done through PCR/RFLP/sequencing approach followed by statistical analysis, and machine-learning (CART model) to correlate the genotype with the variable phenotype.

Results and Conclusions: : The analysis of 51 ATP7B mutations and the genotype data derived from 14 potentially modifier polymorphisms from 8 genes revealed PRNP rs1799990 GG/AG and DBH rs1108580 GG/AG to pose an increased risk of cognitive decline. Conversely, DBH rs1611115 TT and APOE rs449647 AA were identified as protective factors. The patients exhibiting a combination of APOE rs405509 GG and PNPLA3 rs738409 GC/GG tend to present with an earlier age of onset. Furthermore, individuals carrying DBH rs1108580 AA, APOE rs405509 TT, and BDNF rs56164415 CT/TT genotypes presented lower cognitive scores. Patients with HFE rs1799945 CC and PNPLA3 rs738409 CC exhibited a heightened likelihood of experiencing rigidity. Patients with DBH rs1108580 AG/GG demonstrated lower frontal lobe assessment scores, while those with HFE rs2071303 TT showed reduced memory performances. Again, HFE rs1799945 GC was associated with hepatic involvement, BDNF rs56164415 CC with postural instability, and APOE rs429358 TT with gait disturbances. Lastly, the presence of ATP7A rs2227291 CC increased the probability of dysphagia. Thus, the clinical heterogeneity in neurological and hepatic manifestations among the patients may be attributed to different modifier gene variants implicated in copper metabolism and various neurological/hepatic diseases having overlapping symptoms with WD. It may be plausible that these variants can individually pose a risk for neurological/hepatic symptoms and thus become the determinants of the differential outcome in the

disease phenotype when ATP7B is disturbed, even among the sibs with the same mutational profile in the causal gene.

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Abstract ID: 101

Decoding Brain Ageing and its Radiological Genomics Signature: A Patient-individualized therapeutic approach to neurodegenerative disease using anatomically-varying cerebral transcriptomics profile.

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Background/ Introduction: Ageing accompanies various changes as functional decline, diminished resilience and homeostatic deficit. Such neurodegenerative transition (as Alzheimer's disease, vascular cognitive impairment, ischemic stroke) account for highest disease burden in India. Leveraging neuroimaging and immunohistochemistry analysis to map age-related alteration of an individual's brain may be a promising novel strategy to identify ageing anatomically-varying phenotypes, measure molecular cues of coping mechanisms, and thereby develop therapeutic interventions.

Objectives: A main aim is to investigate how orchestrated cerebral reorganization during ageing can enable remediation against ageing-induced neurodegeneration in different brain regions, and thereby help to delineate patient-specific therapeutics in neurodegenerative condition, as vascular cognitive impairment, cerebral ischemia or dementia.

Materials and Methods: MRI-Neuroimaging:

We study ageing's effect on dorsal-&-ventral visual stream of neural information processing using wholistic multimodal imaging: (1)Structural: T1/T2 MRI-scanning (2)Nerve Tractography: Diffusion gradient-based scanning; (iii)fMRI: BOLD and T2* scanning (iv)Vascular Reactivity Mapping: using our specific protocol for vascular reactivity, utilizing arterial elastic response via haemodynamic recording under vascular pulsation. N=60 subjects (young age: 20-30 years; old-age: 60-70 years; n1=n2=30), and Mindbody scan platform is utilized.

Transcriptomics:

Microarray analysis was performed from RNA extracted from postmortem brain samples of two human donors without neuropathology (young-age:~25years; old-age:~60years). Laser-capture microdissection was used, with high RNA integrity. Then in-situ hybridization is undertaken for spatial distribution mapping of specific mRNA transcripts in anatomical 3-D brain tissue volume, followed by microarray-profiling across different brain regions. Here we used the Allen brain platform.

Results: We found that the brain reorganizes to compensate for ageing induced degenerative changes by employing different adaptive processes. MRI-Diffusion tensor imaging exhibited, in elderly there is collateral neural pathway formation (Occipital-Temporal tract) and enhancement of anterior bridging tract, precisely dorso-ventral frontal neural pathway integrity in old age. Cerebrovascular reactivity increased in older age (Mean = 17.7) compared to younger age that signifies during the course of ageing CVR function is more responsive and resilient. We have also witnessed an increment in net Functional Connectivity in Old Age (delta-b = + 0.26) for anterior bridge tract (connecting superior frontal gyrus to inferior frontal gyrus) and posterior bridge tract (bridging cuneus to inferior frontal tract). Furthermore, using neuroimaging-based (MRI) Microarray and ISH transcriptome studies of different age group we

have identified differentially expressed genes corresponds to neuroprotection, yielding a core set of differentially expressed pathways including Protein translation (PAIP1) energy metabolism (Glutaminase2, catechol-o-methyl transferase), and synaptic transmission (Somatostatin 3 receptor, Disk-large-homolog2).

Conclusion: Thus in ageing induced neurodegenerative disease (as dementia, vascular cognitive impairment, ischemic stroke), it may be worthwhile to consider therapeutic interventions on a personalized patient-specific basis namely the following neuroprotective drugs for the corresponding neuroanatomically-located deficits: (1) Occipital stroke: PAIP1 agonist; (2) Temporal stroke: DLG2 agonist; (3)Frontal stroke: GLS2 agonist. Instead of a common drug approach to stroke patients in general, the researcher may give attention to the patient's anatomically-specific drug selection.

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Abstract ID: 104

Correlation of Chromosomal aberration with Multiorgan dysfunction in Perinatal Asphyxia.

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Background/ Introduction: Multiorgan dysfunction (MOD) is one of the criteria for diagnosing Perinatal asphyxia in which diving reflex causes diversion of blood to vital organs to less important organs. Oxidative stress leads to insufficient blood circulation causes production of free radicals which damages DNA resulting chromosomal aberrations.

Objectives: 1. Identify chromosomal aberration in Multiorgan dysfunction, and 2. To correlate Chromosomal aberration with Multiorgan dysfunction.

Materials and Methods: Eighty term asphyxiated babies with same number of healthy term babies were recruited based on inclusion & exclusion criteria. The study was approved by institute Research council and Ethical committee during 2008-2011. Cytogenetic investigation was conducted as per recommendations of International system of cytogenetic nomenclature (ISCN) and analysis done by metasystem IKRO software. Oxidative stress was estimated by serum MDA level. Comparison of different groups by oneway ANOVA and correlation was assessed by Carlpearson correlation coefficient.

Results and Conclusions: The chromosomal aberration found were chromatid breakage, interchromatid connections, dicentric chromosomes, Ring chromosomes. Serum MDA level was significantly elevated ($p<0.05$). Chromosomal aberrations i 2,3,4&5 systems were 2.75±0.9420,3.0525±0.9986, 3.25±1.4097 and 4.85±0.7262 respectively. Chromosomal aberrations increase with severity of asphyxia ($p<0.05$) and well correlated with MOD ($p<0.05$).

Conclusion: Conventional cytogenetic method is a gold standard method is a gold standard method in which chromosomal aberrations were significantly correlated with multiorgan dysfunction in perinatal dysfunction. Advanced cytogenetic and molecular level techniques can be used for further studies of DNA damage in perinatal asphyxia.

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Abstract ID: 105

Unveiling The Protein Structural and Dynamical Consequences of Variants of Uncertain Significance in Parkinson's Disease

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Background/ Introduction: Parkinson's disease (PD) is a complex neurodegenerative disorder with significant genetic heterogeneity. While whole-exome sequencing (WES) has been successful in identifying genes that cause (PD), it regularly identifies variants of uncertain significance (VUS) which still pose an interpretive dilemma. Presumably, VUS in disease-associated proteins can potentially disrupt protein stability and function. Considering that pathogenicity prediction tools used to assess missense variants are based on amino acids substitution and conservation, it can be considered that thermodynamic stability and molecular dynamic simulations may improve prediction accuracy for VUS.

Objectives: To reassess the VUS reported by WES of patients with clinically confirmed PD, over a 2-year period, using protein structural and thermodynamical analysis.

Materials and Methods: WES was performed for 35 patients with clinically diagnosed sporadic PD. Using standard variant classification guidelines 3 pathogenic variants and 20 VUS were reported. The computational tools GROMACS, FoldX and DynaMut were used in the re-assessment of the 20 VUS. The strategy that leveraged predicted protein structure to enhance genetic variant classification included evaluation of structural and functional impact of missense variants, folding, thermodynamic change, stability and flexibility

Results and Conclusions: Results: The in silico computational assessment revealed that 12 out of 20 VUS significantly destabilize the protein structure in the genes EIF4G1, PGLYRP2, HSPB8, WARS2, TWNK, DNAJC6, DCTN1, LRRK1, LRRK2, PLXNA4 and GBA. Molecular dynamic simulation showed altered hydrogen bonding, residue interactions; compromising stability, folding ability and flexibility.

Conclusion: We have shown that using in silico protein structural analysis can aid classification of VUS and give insights into the mechanisms of pathogenicity. Based on our experience, we propose a generic evidence-based workflow for incorporating protein structural information into diagnostic practice to facilitate variant classification.

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Abstract ID: 106

GA4GH: Foundational products for broad and responsible genomic data sharing

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Background/ Introduction: The Global Alliance for Genomics and Health (GA4GH) was founded in 2013 with the goal of advancing precision medicine by promoting standards for genomic data sharing. Its membership includes a wide range of stakeholders—researchers, healthcare providers, and technology developers—working together to enhance genomic science in a secure and ethical way. GA4GH facilitates data interoperability and access, developing standards and frameworks that prioritize patient privacy and informed consent to maintain trust.

Objectives: GA4GH aims to address critical challenges in genomic data sharing by creating standards and tools that foster interoperability and ethical data management. Its products, such as the Beacon standard for querying genomic datasets and the Variation Representation Specification for genetic variant communication, solve specific data-sharing issues. Communities of Interest and the National Initiatives Forum further connect technical and clinical experts in areas like rare disease and cancer genomics, advancing solutions for scalable genomic data use. This presentation highlights GA4GH's latest products and discusses the challenges they address in genomic data sharing, demonstrating the organization's commitment to fostering a globally integrated and ethical approach to genomic data utilization.

Materials and Methods: The Global Alliance for Genomics and Health (GA4GH) has emerged as a pivotal entity in the realm of precision medicine, fostering a collaborative framework for the sharing and utilization of genomic data. Established in 2013, GA4GH encompasses a diverse array of stakeholders, including researchers, healthcare providers, and technology developers, all united in the quest to advance genomic science ethically and efficiently. It develops comprehensive data sharing frameworks that enhance interoperability across platforms, ensuring secure and efficient access to genomic information. The organization also emphasizes robust ethical guidelines to address informed consent and privacy, thereby maintaining patient trust.

GA4GH has created several products to address specific challenges in genomic data sharing in collaboration with driver projects and domain experts. The Beacon standard enables researchers to query genomic datasets while preserving privacy, promoting responsible data sharing. The Variation Representation Specifications standardizes the unambiguous communication of genetic variants to ensure clarity and consistency across platforms. Phenopackets standardizes the description of phenotypic information, both in human readable and machine computable formats improving its integration with genomic data. Our Consent Toolkit is a collection of ethically appropriate, clear, and digestible consent forms for researchers and clinicians in writing appropriate forms. These covering a wide variety of use-cases including large scale initiatives, paediatric consent and rare disease research.

GA4GH hosts several Communities of Interest (CoIs) that focus on specific challenges within the domain including Rare Disease, Cancer, Infectious Disease, Neuroscience and Clinical Genomics. In addition we host a National Initiatives Forum which brings together national initiatives working with genomics at scale. These groups bring together genomics projects working on data infrastructure, management, analysis, and sharing and are uniquely positioned to bridge the divide between technical developers and clinical practitioners.

We present the latest advances within GA4GH specifically highlighting our approved products and the challenges they have been designed to solve. GA4GH is an open world-wide standards organisation with more than 500 participants from across 100 nations and a catalogue of over 44 products.

Results and Conclusions: GA4GH has successfully established a collaborative framework that allows for secure, standardized, and ethically sound genomic data sharing. Through products like the Beacon standard, Variation Representation Specifications, and Phenopackets, GA4GH addresses critical challenges, enabling researchers to access and utilize genomic data while respecting privacy and consent. This framework has attracted over 500 participants from over 100 nations, illustrating widespread

adoption and recognition. The organization's Communities of Interest and National Initiatives Forum further enhance its impact by uniting experts across genomics, clinical practice, and technology development.

GA4GH is making substantial progress toward its vision of a globally connected, interoperable, and ethically governed genomic data-sharing ecosystem. By providing widely accepted standards and fostering an international community of stakeholders, GA4GH bridges gaps between research, technology, and clinical practice. Its open, inclusive approach not only supports precision medicine but also sets a foundation for future advances in genomics, ultimately enhancing patient care and enabling responsible data use worldwide.

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Abstract ID: 107

Genome wide methylation analysis unravels the genes responsible for postpartum reversal of gestational diabetes mellitus

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Background/ Introduction: DNA methylation is one of the important epigenetic mechanisms identified in gestational diabetes mellitus (GDM) and it is considered as a cause and outcome of GDM. Previous literature showed that the DNA methylation pattern of women with GDM is significantly different from the methylation pattern of healthy pregnant women. There are no previous studies performed on GDM patients to understand the epigenetic mechanism responsible for the reversal of hyperglycaemia postpartum.

Objectives: To identify and compare genome wide CpG site methylation in blood samples of GDM women during pregnancy and postpartum in order to study the epigenetic mechanism of postpartum reversal of GDM

Materials and Methods: Eight GDM and healthy pregnant women with their respective follow up samples were used in the present study. Baseline blood sample was collected during 24-28 weeks of pregnancy and they were followed up to 12 weeks postpartum and a follow up blood sample was collected. Genome wide DNA methylation changes was performed using reduced representation bisulfate sequencing (RRBS) in comparison with healthy pregnant women Baseline and follow up samples). Data was analysed using various bioinformatics tools to shortlist the CpG site methylation responsible for postpartum reversal of GDM.

Results and Conclusions: RRBS analysis showed significantly different methylation which might be responsible for reversal of GDM to normoglycemia post-partum. PCA plot and volcano plots showed differentially methylated CpGs between baseline and follow up samples. There were 1091 hyper methylated sites and 190 hypo methylated sites with majority of the DMR were in introns, intergenic and promoter regions. Highest methylation levels were found in ADSSP gene and lowest methylation was found in TLE1 gene. GO and KEGG analysis showed pathways related to metabolism and transport of various molecules and regulation of nucleic acid. The CpG site methylation results obtained after discovery study can be used for validation study in order to use them as biomarker for the prediction of diabetes in postpartum impaired glucose tolerance.

Abstract ID: 108

Phenotyping and genotyping of unclassified heritable thrombocytopenia and platelet function disorders.

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Background/ Introduction: Platelets play a key role in hemostasis and pathological thrombosis. Primary hemostasis involves complex platelet-protein interactions, resulting in platelet activation, secretion, adhesion, and aggregation. Genetic defects in these pathways can lead to platelet dysfunction and bleeding diathesis.

Objectives: 1. To accurately diagnose and classify patients with unexplained platelet function disorders (UPFD) to prevent misdiagnosis, and 2. To identify molecular pathologies and mechanisms underlying UPFD.

Materials and Methods: This study analyzed 75 patients with UPFD. Light Transmission Aggregometry (LTA) was performed on undiluted Platelet Rich Plasma using agonists like ADP, Ristocetin, Collagen, TRAP-6, and TxA. ATP secretion analysis was performed with ADP, Collagen, and TRAP-6. Glycoprotein expression (GPIX, GPIb, GPIIbIIIa, GPVI) and platelet activation status (using CD62P, CD63) were assessed by flow cytometry. whole exome sequencing was conducted for UPFD cases with significant bleeding history (ISTH-BAT ≥3). Novel variant analysis was performed using bioinformatic tools.

Results and Conclusions: Coagulation screening (PT, APTT, TT) was normal across all patients. Out of 75, mutations were detected in 55 cases, including RASGRP2 (14 cases), TBXAS1 (2), NBEAL2 (5), GFI1B (4), FLI1 (4), AP3B1 (2), ITGB3 (6), ITGA2B (2), and others, with 20 patients showing no identifiable variants. Sanger sequencing and in-silico analysis were used to confirm findings. Phenotypic analysis revealed severely reduced platelet aggregation in RASGRP2 and ITGB3 cases. Moderate reduction was observed in GFI1B cases. Glycoprotein expression was normal in all cases, and ATP secretion as well as CD62P/CD63 activation was variable. Patients were classified by phenotype and genotype.

Discussion and conclusion: Starting with 34.5% of 217 platelet function defect cases unclassified, we successfully classified 73.3% of the 75 UPFD cases, reducing unclassified cases to 9.2%. RASGRP2 gene variants, crucial in Gi-signaling, were identified in 18.6% of cases, underscoring the role of genetic variants in platelet dysfunction. Whole Exome Sequencing (WES) significantly aids in diagnosing bleeding phenotypes when conventional tests are inconclusive, supporting the integration of genomic techniques into routine diagnostics for platelet function disorders.

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Abstract ID: 109

First Fine Mapping and Genetic Association Study of CNTNAP2 Variants in Adolescent Idiopathic Scoliosis in an Indian Population.

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Background/ Introduction: The CNTNAP2 gene, which encodes contactin-associated protein-like 2, plays a crucial role in neural development, particularly in synaptic organization and cell adhesion. Variants in CNTNAP2 have been implicated in various neurodevelopmental disorders, and emerging evidence suggests its involvement in the pathogenesis of adolescent idiopathic scoliosis (AIS), a complex spinal deformity with largely unknown genetic mechanisms. Despite significant global research into AIS genetics, the fine mapping of CNTNAP2 in relation to AIS, particularly within the Indian population, remains underexplored.

Objectives: The primary objective of this study was to conduct fine mapping of CNTNAP2 to investigate its role in susceptibility to AIS in a Northwest Indian population. By identifying specific genetic variants associated with AIS, the study aims to contribute to the understanding of CNTNAP2's involvement in the disease and its potential neurodevelopmental pathways.

Materials and Methods: The study analyzed a cohort of 613 individuals, including 113 AIS patients and 500 healthy controls. Genotyping was performed using the Infinium Global Screening Array (GSA)-24 v3.0, which covers 373 single nucleotide polymorphisms (SNPs) within the CNTNAP2 gene. After applying stringent quality control (QC) measures, 369 SNPs were selected for fine mapping and genetic association analysis. Statistical analyses were conducted using PLINK v1.9 and v2.0 to identify significant association signals between the CNTNAP2 variants and AIS.

Results and Conclusions: The fine-mapping analysis identified nine SNPs (including rs112986929, rs67272594, and rs76224922) that surpassed genome-wide significance ($p < 5 \times 10{-}8$), with rs112986929 being the most strongly associated variant ($p = 1.65 \times 10{-}12$). These significant SNPs were clustered within key loci of the CNTNAP2 gene, suggesting that certain regions of the gene may be linked to AIS susceptibility. Additionally, 220 SNPs showed nominal significance ($p \leq 0.05$), further supporting a dense concentration of potential risk variants across CNTNAP2.

This study represents the first fine-mapping analysis of CNTNAP2 variants associated with AIS in a Northwest Indian population. The findings provide important insights into the genetic landscape of CNTNAP2 and support its involvement in the neurodevelopmental processes underlying AIS. The identified loci offer promising targets for further functional validation and mechanistic exploration. Moreover, fine mapping of CNTNAP2 in AIS enhances the potential for developing precision medicine approaches in understanding and managing scoliosis.

Abstract ID: 110

Meta-Analysis of miRNA Variants Associated with Susceptibility to Autism Spectrum Disorder

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Background/ Introduction: Previous studies have demonstrated that single-nucleotide polymorphisms (SNPs) in microRNA (miRNA) are related to the risk of autism spectrum disorder (ASD), a neurodevelopmental disorder with a complicated etiology. However, the conclusions are still controversial and inconclusive. Such inconsistent findings should be investigated to determine their collective potential in ASD diagnosis, prognosis, and understanding of underlying pathophysiological mechanisms.

Objectives: We performed this meta-analysis to assess the association between SNPS of 6 miRNA namely miR-499a, miR-100, miR-196a2, miR-126, miR-146a, and miR-495 and risk of ASD.

Materials and Methods: Relevant studies on population-based studies on the relationship between miRNA variants and ASD were acquired on several electronic databases from inception to November 2024. The strength of the association of miRNA polymorphisms with the risk of ASD was assessed by odds ratios (ORs) and 95% confidence intervals (CIs). Fixed effects or random effect models were used for the risk assessment.

Results and Conclusions: This meta-analysis focused on polymorphisms of 6 miRNA and the results are presented. The impetus for conducting this meta-analysis was to obtain a more robust picture of the association between miRNA polymorphism and ASD risk. This study could possibly aid the development of a biomarker panel in the future, for the early diagnosis and prognosis of ASD patients.

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Abstract ID: 111

Frequency of ABCG2 gene polymorphism and its impact on statin treatment response in patients with cardiovascular diseases: A systematic review and Meta-Analysis

Anoushka Rajashekhar, *Atchaya, Priyanka G, Yuvapriya K, Vinodhini S, Vettriselvi V*
Sri Ramachandra Institute of Higher Education and Research, Chennai.

Background/ Introduction: Cardiovascular diseases (CVDs), comprising conditions like coronary heart disease, cerebrovascular disease, and rheumatic heart disease, are the primary cause of death worldwide,

resulting in approximately 17.9 million fatalities annually. Heart attacks and strokes account for over 80% of these deaths, with a significant proportion occurring prematurely in those under 70. Key lifestyle factors—including poor diet, inactivity, smoking, and high alcohol intake—alongside environmental influences like air pollution, contribute to intermediate health issues such as high blood pressure, elevated blood sugar, abnormal blood lipid levels, and obesity, which increase the risk of severe CVD complications. Statins, a class of medications prescribed to lower low-density lipoprotein (LDL) cholesterol, work by inhibiting Hydroxymethylglutaryl-CoA reductase, effectively reducing total cholesterol, LDL, and triglyceride levels. The ABCG2 gene, encoding an essential ATP-binding cassette (ABC) transporter protein, plays a pivotal role in drug pharmacokinetics by mediating the efflux of various hydrophobic compounds, including several statins. Single nucleotide polymorphisms (SNPs) within ABCG2 have been linked to variations in drug absorption, distribution, and clearance, impacting the therapeutic effectiveness and potential side effects of statins across different populations.

Objectives: This systematic review and meta-analysis aim to examine the association between ABCG2 gene polymorphisms and responses to statin therapy, exploring how these genetic variations influence pharmacokinetics and clinical outcomes.

Materials and Methods: Research Question: Is there a link between the ABCG2 gene polymorphisms and treatment response to statins in patients with cardiovascular diseases?

Method: A protocol was developed for this systematic review following the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) guidelines Search strategy: A comprehensive search in English language of several databases using the following search terms: ABCG2 gene polymorphisms; CVD, Statin response, pharmacogenetic analysis. The actual strategy listing all search terms and how they are combined will be presented in the poster Selection process and data extraction: The selection process was conducted in two screening phases; first the title and abstract review, followed by a full-text review. In both stages, each reference was reviewed independently by three reviewers (AR, PG, AM), and any disagreements were resolved consulting with a senior author (VV). The data extracted included the following details study characteristics year of publication, author names, country, study design, sample size, inclusion exclusion criteria and the demographic details of the study participants.

Results and Conclusions: For data analysis the articles with the following criteria were included:

1. Peer-reviewed published articles with original data in English

2. The studies that included only CVD patients with age group above 18

3. Patients treated with statins

4. Studies that explored the ABCG2 gene polymorphisms and its association with treatment response to statins. Characteristics of the studies included and rejected, details of risk of bias assessment will be presented in the poster. This systematic review comprehensively examined the genetic associations of ABCG2 treatment responses in CVD patients. A thorough understanding of pharmacogenetics may help optimize the clinical outcomes in patients with CVD.

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Abstract ID: 112

Effects of ABCB1 and CETP gene polymorphism on the pharmacokinetics of statins: Systematic review and Meta-analysis

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Background/ Introduction: Cardiovascular diseases (CVDs), comprising conditions like coronary heart disease, cerebrovascular disease, and rheumatic heart disease, are the primary cause of death worldwide, resulting in approximately 17.9 million fatalities annually. Heart attacks and strokes account for over 80% of these deaths, with a significant proportion occurring prematurely in those under 70. Key lifestyle factors—including poor diet, inactivity, smoking, and high alcohol intake—alongside environmental influences like air pollution, contribute to intermediate health issues such as high blood pressure, elevated blood sugar, abnormal blood lipid levels, and obesity, which increase the risk of severe CVD complications. Statins, a class of medications prescribed to lower low-density lipoprotein (LDL) cholesterol, work by inhibiting Hydroxymethylglutaryl-CoA reductase, effectively reducing total cholesterol, LDL, and triglyceride levels. The ABCB1 and CETP gene polymorphisms can significantly influence the pharmacokinetics of statins, impacting their efficacy and safety profiles. ABCB1 (also known as MDR1) encodes P-glycoprotein, which affects the absorption and distribution of statins, potentially altering plasma concentration levels. CETP gene polymorphisms, on the other hand, influence lipid metabolism and may modify statin efficacy by affecting HDL cholesterol levels. These genetic variations can lead to inter individual differences in statin response, which is crucial for personalized dosing and reducing adverse effects.

Objectives: This systematic review and meta-analysis aim to examine the association between ABCB1 and CETP gene polymorphisms and responses to statin therapy, exploring how these genetic variations influence pharmacokinetics and clinical outcomes.

Materials and Methods: Research Question: Is there a link between the ABCB1 and CETP gene polymorphisms and treatment response to statins in patients with cardiovascular diseases?

Method: A protocol was developed for this systematic review following the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) guidelines. Search strategy: A comprehensive search in English language of several databases using the following search terms: ABCB1 and CETP gene polymorphisms; CVD, Statin response, pharmacogenetic analysis. The actual strategy listing all search terms and how they are combined will be presented in the poster Selection process and data extraction: The selection process was conducted in two screening phases; first the title and abstract review, followed by a full-text review. In both stages, each reference was reviewed independently by three reviewers (PG, AM, AR) and any disagreements were resolved consulting with a senior author (VV). The data extracted included the following details study characteristics year of publication, author names, country, study design, sample size, inclusion exclusion criteria and the demographic details of the study participants.

Results and Conclusions: For data analysis the articles with the following criteria were included 1. Peerreviewed published articles with original data in English, 2. The studies that included only CVD patients with age group above 18, 3. Patients treated with statins, 4. Studies that explored the ABCB1 and CETP gene polymorphisms and its association with treatment response to statins. Characteristics of the studies included and rejected, details of risk of bias assessment will be presented in the poster.

This systematic review comprehensively examined the genetic associations of ABCB1 and CETP, treatment responses in CVD patients. A thorough understanding of pharmacogenetics may help optimize the clinical outcomes in patients with CVD.

Abstract ID: 113

Genetic variability of human SLCO1B1 and statin response: A systematic review and meta-analysis

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Background/ Introduction: Cardiovascular diseases (CVDs), comprising conditions like coronary heart disease, cerebrovascular disease, and rheumatic heart disease, are the primary cause of death worldwide, resulting in approximately 17.9 million fatalities annually. Heart attacks and strokes account for over 80% of these deaths, with a significant proportion occurring prematurely in those under 70. Key lifestyle factors—including poor diet, inactivity, smoking, and high alcohol intake—alongside environmental influences like air pollution, contribute to intermediate health issues such as high blood pressure, elevated blood sugar, abnormal blood lipid levels, and obesity, which increase the risk of severe CVD complications. Statins, a class of medications

prescribed to lower low-density lipoprotein (LDL) cholesterol, work by inhibiting Hydroxymethylglutaryl-CoA reductase, effectively reducing total cholesterol, LDL, and triglyceride levels.

The SLCO1B1 gene, encoding an essential member of the solute carrier organic anion transporter family, is responsible for producing the OATP1B1 protein, a transporter primarily expressed in the liver. This protein plays a key role in drug pharmacokinetics by mediating the hepatic uptake of various endogenous compounds and medications, including several widely used statins. Single nucleotide polymorphisms (SNPs) within SLCO1B1 have been linked to variation in drug absorption, distribution, and clearance rates, which can significantly affect statin pharmacodynamics. These genetic variations influence therapeutic efficacy and the risk of side effects, such as statin-induced myopathy, across diverse population.

Key words: SLCO1B1, SNPs, Pharmacogenetics, Statins, CVD

Objectives: This systematic review and meta-analysis aim to examine the association between SLCO1B1 gene polymorphisms and responses to statin therapy, exploring how these genetic variations influence pharmacokinetics and clinical outcomes.

Materials and Methods: Research Question: Is there a link between the SLCO1B1 gene polymorphisms and treatment response to statins in patients with cardiovascular diseases?

Method: A protocol was developed for this systematic review following the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) guidelines.

Search strategy: A comprehensive search in English language of several databases using the following search terms: SLCO1B1 gene polymorphisms; CVD, Statin response, pharmacogenetic analysis. The actual strategy listing all search terms and how they are combined will be presented in the poster.

Selection process and data extraction:

The selection process was conducted in two screening phases; first the title and abstract review, followed by a full-text review. In both stages, each reference was reviewed independently by three reviewers (AM, AR, PG), and any disagreements were resolved consulting with a senior author (VV). The data extracted included the following details study characteristics year of

publication, author names, country, study design, sample size, inclusion exclusion criteria and the demographic details of the study participants.

Results and Conclusions: For data analysis the articles with the following criteria were included 1. Peer-reviewed published articles with original data in English, 2. The studies that included only CVD patients with age group above 18, 3. Patients treated with statins, 4. Studies that explored the SLCO1B1gene polymorphisms and its association with treatment response to statins. Characteristics of the

studies included and rejected, details of risk of bias assessment will be presented in the poster. This systematic review comprehensively examined the genetic associations of SLCO1B1 treatment responses in CVD patients. A thorough understanding of pharmacogenetics may help optimize the clinical outcomes in patients with CVD. Furthermore, pharmacogenetic studies have shown promise to help maximize treatment outcomes and adjusting drug dosages.

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Abstract ID: 115

Characterization of SHQ1 gene variants: Case Studies of Global Developmental Delay and Dystonia in Pediatric Patients

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Background/ Introduction: The advent of high throughput Next Generation Sequencing technologies has allowed for comprehensive genomic analysis and identification of novel genetic variants that may contribute to complex clinical phenotypes. We focus on two pediatric cases involving novel variants in SHQ1 gene. SHQ1 is a chaperon assisting in the assembly of H/ACA box ribonucleoproteins that function in the processing of ribosomal RNAs, modification of spliceosomal small nuclear RNAs and stabilization of telomerase. Accordingly, loss of SHQ1 will lead to degradation of the RNP assembly. Mutations in genes encoding components of the H/ACA RNP complex, such as DKC1, can lead to significant effects on neurological development. Homozygous and compound heterozygous mutations in SHQ1 have been associated with a rare and severe neurodevelopmental disorder characterized mainly by global developmental delay, seizures and early-onset dystonia. Here we report two children from unrelated families with homozygous and compound heterozygous variants of uncertain significance in the gene. Both patients have global developmental delay, dystonia, spastic quadriparesis and microcephaly.

Objectives: Expanding existing knowledge on SHQ1 gene variants, perform in silico analysis of the variants to predict their effects on protein structure and function and perform functional characterization of the variants by complementation assay using yeast

Materials and Methods: Patient families were recruited under DBT funded Mission Program on Pediatric Rare Genetic Disorders by obtaining informed consents from all institutions and subjects involved. Peripheral blood was collected and genomic DNA was isolated. Whole exome sequencing was performed for using Illumina platform using libraries prepared using TWIST kit.

Results and Conclusions: Whole exome sequencing revealed a previously reported VOUS, c. 850T>C; p.Tyr284His in homozygous state in the first patient and two novel VOUS, c.699_702del; p.Ser233Argfs*9 and c.1157A>T; p.Tyr386Ser in compound heterozygous state in the second patient. Sanger sequencing confirmed the WES results and full segregation in both families. In silico analysis using different prediction classifiers and structural modelling of the variant showed the missense variants

to be deleterious. Our findings show the variants likely to be pathogenic and cause the phenotypes observed in the patients. Functional characterization of the missense variants mentioned above by complementation assay using yeast is under process.

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Abstract ID: 116

Molecular Studies on Consequences of Mutations in ANTXR2 on Subcellular Localization and Protein Aggregation

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Background/ Introduction: Hyaline Fibromatosis Syndrome (HFS) is a rare autosomal recessive disorder that occurs due to mutation in gene ANTXR2, as a result COLLAGEN homeostasis alters, and extensive deposition of hyaline fibrous material occurs in the connective tissues. It leads to two conditions – a severe form, Infantile systemic hyalinosis (ISH) and a milder version, Juvenile hyaline fibromatosis (JHF). The Antxr2 protein is synthesized and folded in the endoplasmic reticulum and glycosylated in the Golgi body, then transported to the cell membrane. After the localization of ANTXR2 protein in cell membrane it interacts with ECM components and Matrix Metalloproteinase (MMPs). The ANTXR2 protein interacts with MMP-9 and regulates them positively during COLLAGEN-VI degradation by forming ANTXR/MT1-MMP complex. The mutated ANTXR2 protein is unable to localize on the plasma membrane and get stuck inside Endoplasmic reticulum (ER) during protein folding and/or inside Golgi body (GB) during glycosylation. This could inhibit events leading to Pro MMP-9 to active MMP-9 and alters the COLLAGEN metabolism results formation of fibromas.

Objectives: We aim to study the role of ANTXR2 and MMP-9 in the process of COLLAGEN-VI degradation and understand the significance of its localization in wildtype and mutant background with respect to allelic heterogeneity.

Materials and Methods: we investigate the role of ANTXR2 in the process of COLLAGEN-VI degradation and understand the localization of ANTXR2 protein in wild type and mutant background. We used a cell-based assay to test the severity of mutations in ANTXR2 for devising a possible therapeutic intervention. We studied the role of ANTXR2 and MMP-9 in the process of COLLAGEN-VI degradation. We mimicked some common reported mutations in the ANTXR2 gene using site directed mutagenesis in the ANTXR2-NeGFP, pCDNA3.1 plasmid vector. To investigate the subcellular localizations of the wild type and mutant ANTXR2 protein in wild-type backgrounds, SDM plasmids were transfected into human skin fibroblast cells. To identify distinct cellular and subcellular localizations the transfected cells were stained with antibodies for plasma membrane (LAMP1), endoplasmic reticulum (CALRETICULIN), Golgi body (GOLGB1). To know the protein interaction of ANTXR2 with F- actin, COLLAGEN-7a and KERATIN-14 we used respective antibodies. Extra cellular signaling was investigated using Anti-phospho-ERK protein. We used fluorescence microscopy with Apotome2 modification to see the protein localizations. The localization and colocalization data were compared with respect to wild-type and mutant backgrounds using a t-test.

Results and Conclusions: Through this investigation we found mutant protein with p.Cys39Phe, p.Ala359Cys, p.Asp292_ValinsGln, p.Leu330Arg, p.Tyr381Cys mutation failed to localize at the membrane significantly and showed a varied degree of localization. All these mutants were colocalized

in ER and the mutant protein with p.Asp292_Val ins Gln mutation got stuck both in ER and GB significantly..We observed failure in COLLAGEN-VI degradation and increased aggregation in mutant cells (p.Ala359Cys_delT). The trans heterozygous expression of mutants in mutant cells via transfection shows no recovery from the COLLAGEN-VI aggregations. When we overexpressed the wild type ANTXR2-NeGFP and MMP9-NeGFP recombinant protein in mutant (p.Ala359Cys_delT) cells we observed a drastic recovery of COLLAGEN-VI aggregations, the size and number of aggregates were decreased.

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Abstract ID: 119

Obtaining 3-Dimensional Cultures Of Mda-Mb-231 (Human Breast Cancer Cell Line) – An Approach For Inducing In Vivo-Relevant Gene Expressions Towards Personalized Medicine

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Background/ Introduction: Cancer cell lines have contributed immensely for cancer research and the development of 3-dimensional (3D) culture systems has greatly enhanced their utility. One of the hallmarks of malignant cells is the changes in their gene expressions. It is known that 3D cultured cell lines differ from their 2D monolayer counterparts in several aspects, of which gene expressions is one such significant difference. *In vivo*-relevant studies such as specific signalling pathways, biomarker discovery and personalized medicine have become a possibility by using 3D cultures which mimic solid tumour biopsies. While utilizing cell cultures, the cell culture phases at which an assay is performed will determine the reliability of the experimental outcomes. The late log phase is known to have the cells at their best health, higher numbers and viability. Hence, it is vital to ascertain the cell culture phase duration of any human cancer cell line before they are used for actual experiments. In this study, we obtained 3D cultures of the MDA-MB-231 (human breast cancer) cell line, describe their morphologies and also determined the cell culture phases.

Objectives: To obtain 3D aggregates of the MDA-MB-231 cell line and ascertain the cell culture phases as can be utilized for translational research.

Materials and Methods: MDA-MB-231 cells were cultured as monolayers in DMEM supplemented with 10% FBS at 370C. These cells were harvested and 2D and 3D cultures obtained in 6-well plates as 2mL cultures. 1% agarose hydrogel was used as a scaffold/matrix to obtain the 3D cultures. The seeding density used was 3 x 104 cells/mL. The cell counts and viabilities were obtained for both 2D and 3D cultures at 24-hour intervals and the cell culture phases (lag, log, plateau and decline) were plotted as a graph.

Results and Conclusions: The MDA-MB-231 spontaneously developed into several multi-cellular aggregates as 3D cultures. The 2D and 3D cultures had a markedly different cell culture phase duration pattern. The peak log phase was observed at 72 hours and 120 hours for the 2D and 3D cultures respectively. The 3D cultures exhibited an extended cell culture phase owing to their enhanced survival compared to their 2D monolayer counterparts. Such an understanding will help in utilizing 3D cultures of the chosen cell line for assays such as gene expression studies which can have a translational benefit.

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Abstract ID: 121

Targeting Beta-lactamase activity with Oxacyclohexadecan-2-one in Carbapenem-resistant Uropathogenic E. coli: A Molecular simulation approach

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Background/ Introduction: Urinary Tract Infections (UTIs) are prevalent globally, affecting almost 150 million people every year with substantial morbidity and high medical cost. Severe complications, including septic shocks, acute kidney injury and pyelonephritis, result in around 230,000 deaths annually. Various factors contribute to UTIs, notably age and comorbidities like diabetes mellitus. In regions like India, with a high prevalence of diabetes, incidence of UTIs is expected to increase. Uropathogenic Escherichia coli (UPEC) is the most common pathogen, causing approximately 50-75% of UTIs, followed by Klebsiella pneumoniae at 10-15%. With rising UTI rates globally and increasing antibiotic resistance, novel pharmacological treatment strategies are imperative, particularly in diabetic populations, where antibiotic resistance is heightened. Currently, UTIs are only being treated with various classes of antibiotics such as Ampicillin, Nitrofurantoin, Carbapenems etc.

Use of antibiotics leads to development of resistance when used over a period since the pathogens can adapt to the toxicity of modern medicines. According to a study reported in BMC Infectious Diseases, 81.1% multidrug resistance was observed in bacterial isolates from diabetic patients. E. coli, is the common uropathogen that is well-known for its capacity to acquire genes that confer antibiotic resistance and those isolated from UTIs in diabetic individuals show resistance to most studied antibiotics, such as co-trimoxazole, nitrofurantoin, ampicillin, and ciprofloxacin. The most reported proteins expressed by E. coli that confer resistance to a broad-range of carbapenem drugs are NDM, OXA, TEM, CTX-M, VIM, and IMP. These proteins produce an enzyme, beta-lactamase that neutralizes the beta-lactam ring of the carbapenem drugs, thus rendering the drug inactive.

To address UTI challenges and to reduce the complete burden on antibiotics for the treatment of UTIs, researchers have been exploring plant-derived compounds such as proanthocyanidins, probiotics, phenolic compounds, flavonoids, alkaloids, and D-Mannose. Plant phytochemicals are a rich and promising class of molecules with far-reaching implications for human health as they possess a mixture of complex compounds, with fewer side-effects and a broad spectrum of activity. Moringa oleifera, a plant native to the Indian subcontinent, possesses antioxidant and anti-inflammatory properties, with studies suggesting its efficacy in managing diabetes and infections. M. oleifera' s effectiveness in treating UTIs was validated by Santosh Kumar et al in an experiment where the bark extracts exhibited a cure rate of 66% compared to standard antibiotics. Apolar fraction of M. oleifera seed also has proven to exhibit antibacterial activity against certain gram-negative bacteria such as Staphylococcus aureus and Staphylococcus epidermidis.

Drug discovery aided by computational methods is an emerging valuable tool for identifying and optimizing promising drug candidates as it uses artificial intelligence approaches to collectively merge modeling, data analysis and predictive algorithms for the drug development process. CADD is very important in antibacterial research because it addresses the critical problem of antibiotic resistance and these technologies also offer effective virtual-screening of large chemical libraries to identify novel possible antibacterial chemicals. Researchers can identify candidates with high binding affinity and specificity for bacterial pathogens by computationally analyzing the interactions between small molecules and biological targets considering the molecular structure, electrostatic characteristics and binding energies, allowing antibacterial activity to be predicted before experimental validation. Therefore, our study aims to screen M. oleifera seed-derived compounds against the commonly reported carbapenem-

resistant proteins NDM-1, NDM-5, and OXA-48 of uropathogenic *E. coli*. By identifying potential antibacterial agents from natural sources, we hope to contribute to the development of novel therapeutic strategies to combat these disease-causing pathogens.

Objectives: The main objective of the study is to identify the most commonly present carbapenem-resistant genes of Uropathogenic *E.coli* causing UTIs and subsequently identify a best inhibitor compound from Moringa oleifera seed to overcome carbapenem-resistance, when compared to the standard drug Imipenem.

Materials and Methods: Methodology

1. Preparation of target Protein molecules

The three-dimensional crystallographic protein structures of the most reported carbapenem-resistant proteins NDM-1, NDM-5 and OXA-48 of E. coli across all strains were extracted from the RCSB Protein Data Bank. Further, using the PyMol software all the non-interacting ions and water molecules were removed.

2. Identification of the Binding Site

An online tool, CASTp was used to determine the surface properties and functional areas of the proteins. Within the active site of the protein, the residues were displayed along with the surface area and volume.

3. Preparation of Ligand molecules

The M. oleifera seeds were dried, ground, extracted and screened through GC-MS for the retrieval of the phyto-components. The listed compounds were identified and the 3-dimensional structures were obtained from the PubChem database.

4. Virtual screening of compounds using PyRx

The crystal-structure of the protein was prepared by adding Kollman charges, hydrogen atoms and gasteiger using Auto Dock Tools (ADT). Energy minimization of the compounds was performed and the energy values were analyzed through the Auto Dock Vina wizard option of the PyRx software and subsequently, the top 10 compounds were considered for further analysis.

5. Pharmacokinetic Analysis

SwissADME, an online software tool was used to estimate variables such as the absorption, distribution, metabolism, and elimination of physicochemical features and to determine the recommended daily dosage and bioavailability of the compound. Using the tool, only the ligands which follow the Lipinski rule are further considered for docking studies.

6. Toxicity analysis by ProTox

ProTox is an online toxicity assessment tool that shows the toxicity level of a compound. An input of a compound is given either by the compound's canonical SMILES form or the PubChem name. When the toxicity prediction is performed, the results are displayed in the form of toxicity class, and compounds which fall under the class 5 and 6 are only considered for further docking analysis.

7. Docking studies

Molecular Docking was performed to determine the best fit between two molecules, which resembles a lock and key mechanism. Based on the binding affinity of the target protein with the ligand, this method projects the 3D structure of a complex. The software used for performing docking is the Auto Dock tool, version 1.5.7. First, Auto Grid is performed by choosing the protein and the map-types are set by selecting

the ligand molecule. The corresponding binding site residues are selected for the protein molecule. Successful completion of Auto Grid indicates that the molecule is ready for docking. For docking, the protein is set as rigid filename and ligand molecules are chosen. After checking the genetic algorithm and docking parameters, Auto Dock is run. To ensure a representative examination, docking is performed in triplicates. The interactions of the protein-ligand are then visualized using the BioVia Discovery Studio tool.

8. Molecular Dynamics Simulation (MDS)

MDS was performed for the protein-IPM and protein-test ligand complexes using the GROMACS 4.5.4 suite to better understand the ligand's interaction with the protein's functional area . Protein and ligand topology files were created and fixed in the center of a dodecahedron box with water molecules. The system was compensated by the addition of counter-ions. Utilizing the steep descent algorithm and conjugate gradient methods, the energy of the complex system was decreased. To achieve a stable condition, the NVT and NPT were counterbalanced for the overall system energy. After selecting the parameters for the temperature as 300K and pressure as 1 bar, the 100 ns MD simulation was initiated, followed by Root Mean Square Deviation (RMSD), Root Mean Square Fluctuation (RMSF), Radius of Gyration (Rg), Hydrogen Bond (H-bond), MM/PBSA, and SASA analysis of the MD trajectory files.

To determine the protein-ligand complex stability in motion, Principal component analysis (PCA) was also performed to identify changes and significant movements of protein structures. The input files were given as eigenvectors and eigenvalues, and the RMSF/atom eigenvectors were shown as output. The first and second projections of the protein trajectory were depicted as a graph against a dynamic run of 100 ns. To determine the binding energy of beta-lactamase protein and ligand complexes, molecular mechanics/Poisson-Boltzmann surface area (MM/PBSA) were carried out which represent the summary of the binding free energy of biomolecular interactions between beta-lactamase proteins and ligands in the last 50 ns.

Results and Conclusions:

1. Retrieval of Ligand compounds

A set of 135 compounds derived from GC-MS were collected from the PubChem database and their respective 3D structures were extracted.

2. Virtual-screening of the ligand molecules

After virtually-screening each of the protein with all the ligand molecules, the top 10 hits with best binding affinity were selected.

3. ADME and Toxicity analysis

The top 10 hits for each protein were further screened using the Swiss-ADME tool for their pharmacokinetic properties and later analyzed for their toxicity using the ProTox tool. Out of the them, one common compound was chosen as the test ligand (PubChem ID: 235414, Oxacylcohexadecan-2-one) with respect to the pharmacokinetic properties and toxicity class. This was further used for docking studies.

4. Molecular Docking of Oxacyclohexadecan-2-one and Imipenem with beta-lactamase proteins of E. coli

Molecular docking was performed in Auto Dock prior to which H2O molecules and heteroatoms were removed from the structures. The top best compound, Oxacyclohexadecan-2-one, interacted with blaNDM-1, blaNDM-5 and blaOXA-48 at -6.45 kcal/mol, -6.05 kcal/mol and -7.34 kcal/mol respectively. Imipenem (IPM), the standard ligand, obtained docking scores of -3.41 kcal/mol, -3.99 kcal/mol and -

6.36 kcal/mol for NDM-1, NDM-5 and OXA-48 respectively . IPM is the standard beta-lactam drug, and it belongs to the carbapenem class, which has the highest antibiotic resistance in E. coli UTI cases. The two compounds, Oxacyclohexadecan-2-one, and the conventional drug IPM, were investigated further using Molecular Dynamic Simulation.

5. Molecular Dynamic Simulation

MDS was run at 100 ns for all the three protein-ligand complexes. The RMSD for the six combinations of protein-ligand complexes are represented. The stability of the protein-ligand complex is determined by the RMSD value which explains the deviation and positional changes of the ligand molecule for each protein, when bound. The mean RMSD for the test ligand with proteins NDM-1, NDM-5 and OXA-48 was found to be 0.41 nm, 0.42 nm and 0.28 nm respectively. Whereas, the RMSD values for IPM compared against NDM-1, NDM-5 and OXA-48 protein molecules were observed to be higher than the test compound with the RMSD values being recorded as 0.56 nm, 0.43 nm, and 0.34 nm respectively.

the flexibility and fluctuations of the individual residues, RMSF was calculated. The RMSF of the test ligand was observed to be 0.13 nm, 0.16 nm and 0.13 nm for NDM-1, NDM-5 and OXA-48 respectively. On the other hand, the RMSF observed for the standard drug Imipenem was found to be 0.16 nm, 0.14 nm and 0.14 nm respectively for all the three protein molecules. The RMSF fluctuation pattern was similar for the test ligand with NDM-1 and OXA-48 protein molecules, whereas the interaction with NDM-5 observed a higher fluctuation with the test ligand. Compared to the standard drug, the fluctuations of the test ligand were significantly low.

Radius of Gyration reveals the complex and protein backbone compactness. The Rg for the NDM-1, NDM-5 and OXA-48 complex with the test ligand was observed to be 1.18 nm, 1.31 nm and 1.39 nm whereas the Rg with the standard drug Imipenem was observed as 1.23 nm, 1.25 nm and 1.38 nm respectively. The pattern of Rg for the protein complexes with both the ligands were similar, indicating a compact configuration of the ligand with the protein.

Solvent accessible surface area (SASA) analysis provides an insight into hydration properties, binding interactions, stability, and conformational transitions relevant for understanding the protein function. SASA trajectories within the 100 ns simulation observed a mean value of 82.09 nm, 87.67 nm, and 121.30 nm for the proteins NDM-1, NDM-5 and OXA-48 complexes with the test ligand, while the mean SASA trajectories recorded for the proteins with Imipenem were 82.5 nm, 90.9 nm and 118.4 nm respectively. First 40 s observed an elevated SASA value for the protein complex with the test ligand which then was observed to reduce until 100 ns.

To evaluate the stability of the protein-ligand complex, the conventional H-bonds were taken into consideration. Overall, the mean H-bonds of the protein complex with the test ligand was recorded as 1.18, 0.75 and 0.24 for the NDM-1, NDM-5 and OXA-48 respectively. Whereas, the average H-bonds of the protein-imipenem complexes were observed to be 0.9, 3.6 and 2.2 respectively for the three proteins. The protein hydrogen bond interactions of Imipenem were observed to be on the higher side when compared to the test ligand.

The Free energy landscape (FEL) plot was constructed to determine the maximum and minimum energy of the three complexes. Each complex has a unique FEL pattern with the red colored spots defining lowest energy and best conformation. PCA analysis was also applied to determine the structural changes produced by the protein-ligand interactions. The PCA figure showed that the eigenvalues of the NDM1-IPM complex range from around -7 nm to 4 nm whereas the NDM1-test ligand ranges from -4 nm to 2 nm. Similarly, the eigenvalues of NDM5-IPM vary from -4 nm to 4 nm, while the NDM5-test ligand ranges from -6 nm to 4 nm. OXA48-IPM variations range from -7 nm to 3 nm, whereas the OXA48-test ligand fluctuates between -4 nm to 4 nm. The PCA plot shows that the test ligand binds to the protein

resulting in a more stable-complex structure. Overall, the antimicrobial-resistant proteins of E. coli form a better stable complex with the test ligand, with a shorter phase space than the protein-IPM complexes.

The average free binding energy for the protein-IPM and protein-test ligand complexes revealed the lowest energy for all the protein-test ligand complexes when compared to their counterparts with the standard control, Imipenem. Free binding energy determines the measurement of the release of energy during the bond formation between the protein and the ligand, therefore lower the energy better the binding capacity. The van der waals, electrostatic and polar solvation energies are all summed up to form the total free binding energy. Among the two compounds studied, the test ligand 235414 exhibited the lowest binding energies: -105.081 ± 14.900 for NDM-1, -90.483 ± 12.335 for NDM-5, and -75.729 ± 10.350 for OXA-48 proteins. These results indicate that ligand 235414 may serve as a more effective bactericidal agent against antimicrobial resistance proteins in E. coli compared to the FDA-approved drug Imipenem. Additionally, the RMSD, RMSF, Rg, SASA, H-Bonds, PCA, FEL, and MM/PBSA analyses confirmed the stability of the protein-ligand complexes throughout the simulation.

Conclusion

The three majorly reported antimicrobial resistant genes, NDM-1, NDM-5 and OXA-48 of UPEC have proven to have better interactions with Oxacyclohexadecan-2-one compared to the standard drug Imipenem, to treat UTI. This is indicative that Moringa oleifera seeds can be a potential treatment option for UTIs. The extract of its seeds can confer antimicrobial activity due to the presence of various active compounds such as moringine, pterygospermin, and 4-(alpha-L-rhamanosyloxy) benzyl isothiocyanates too. Bioactive compounds of M. oleifera such as quercetin, chlorogenic acid, isothiocyanates also help in regulating the blood sugar level, potentially making Moringa an anti-diabetic agent as well. The present study suggests that Oxacylcohexadecan-2-one, a molecule of Moringa seeds can act as a better alternative for the commonly used standard drug Imipenem to treat UTIs and rUTIs caused due to carbapenem-resistant *E. coli.*

Abstract ID: 122

Comparative yield of quantitative fluorescent PCR and low-resolution chromosomal microarray in pregnancies with ultrasonographic abnormalities

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Background/ Introduction: Prenatal diagnosis of chromosomal abnormalities facilitates timely and informed decision-making. Chromosomal microarray (CMA) is the recommended first-tier diagnostic test for evaluation of pregnancies with fetal structural abnormalities. However, owing to its relatively high cost, it is still not the preferred test for prenatal diagnosis in resource-poor settings. Quantitative fluorescence-PCR (QF-PCR) is a relatively economical platform that, in spite of the recommendations, is frequently chosen in preference to CMA.

Objectives: To determine the diagnostic yield of QF-PCR and incremental yield of a low resolution CMA in prenatal samples among patients with abnormalities on prenatal ultrasonogram.

Materials and Methods: This is a retrospective study including all cases that were evaluated by CMA between January 2019 and September 2024 at the Cytogenetics laboratory of the Christian Medical College Vellore for the indication of abnormalities detected on ultrasonogram. All cases are routinely tested for maternal cell contamination (MCC) using QF-PCR, which also detects trisomies of autosomes

13,18,21, and aneuploidies of X and Y. Any samples that were positive for these aneuploidies determined the yield of QF PCR.

All CMAs during this period were performed on an Affymetrix CytoScan Optima platform which has a functional resolution of 1 and 2 MB for deletions and gains genome-wide and upto 100kb in targeted regions. The laboratory used size filters of 200 kb for deletions and 500 KB for duplications.

Diagnostic yield of CMA was calculated from the number of positive results defined as pathogenic and likely pathogenic CNVs. Positive results were categorized into 'common aneuploidies' and 'others'. The latter group comprises the incremental yield of CMA over QF-PCR.

Results and Conclusions: A combined total of 146 samples were evaluated. Seventeen samples among these were positive for pathogenic or likely pathogenic mutations on CMA resulting in a yield of 11.6%. QF-PCR was positive in 11 samples resulting in a yield of 7.5%. The incremental yield of CMA over QF-PCR was 4.1%. Aneuploidies consisted of 4 cases of trisomy 21, 5 cases of trisomy 18, 1 case each of trisomy 13 and monosomy X. All cases that were not detectable on QF-PCR consisted of large CNVs exceeding 10Mb. Nine of 11 pregnancies (82%) with aneuploidies demonstrated soft markers on ultrasonogram whereas only 2 of 6 with CNVs (33%) showed soft markers. Foetal structural abnormalities were evident with both aneuploidies as well as CNVs.

Our results demonstrate that QF-PCR in isolation is not suitable for evaluation of patients with ultrasonographic abnormalities. However, the use of QF-CPR for MCC detection prior to CMA can save money, time and effort as nearly two of every three positive samples showed a common aneuploidy.

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Abstract ID: 123

Concomitant telomere attrition is associated with Spinal Muscular Atrophy in highly inbred region of North India

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Background/ Introduction: Spinal muscular atrophy (SMA), a severe neurodegenerative disorder is characterized by deterioration of motor neurons in brain & spinal cord leading to muscle weakness/hypotonia/recurrent infections/ paralysis and even death. Based on age of onset and severity of symptoms it is classified into 5 types though its global prevalence varies. SMA is caused by the deletion of the SMN1 gene while SMN2 gene copies influence the severity. As decreased telomere length is associated with aging and age-related diseases, so understanding the role of shortened telomeres in SMA pathogenesis is consequent to genomic instability, DNA damage and cellular senescence.

Objectives: We evaluated the role of SMN1 gene in SMA disease pathogenesis & its effect on telomere length in order to estimate the risk & prognosis of SMA in genetically less explored & highly inbred regions of Kashmir from North India.

Materials and Methods: Monochrome multiplex quantitative polymerase chain reaction (MMQ-PCR) for assessment of relative telomere length (RTL) was done using specific primers targeting single copy gene and telomere and relative telomere length was evaluated by T/S ratio (ratio of copy number of telomere repeat to single copy gene).

Results and Conclusions: RTL in peripheral blood lymphocytes was measured in triplicates by MMQ-PCR in 40 SMA patients and 58 healthy age and gender matched healthy controls (HC). Telomere length was significantly shorter in SMA patients than in controls Furthermore, subtypes categorizing data into type 0, 1, 2, 3, & 4 were performed. It was observed that types 0, 1 & 2 exhibited significant telomere attrition than subtypes 3 & 4. The results showed that telomeres were significantly shorter in SMA patients, especially in the more severe forms. This finding is consistent with previous studies that have observed telomere shortening in SMA patients. The exact mechanism by which telomere shortening contributes to the pathogenesis of SMA is not fully understood. However, it is believed that disruption of neuronal differentiation and neurogenesis, induced oxidative stress, DNA damage, and impaired DNA repair mechanisms may all play a role in accelerating telomere shortening and contributing to motor neuron degeneration in SMA patients. Being implicated in pathogenesis of SMA telomere shortening could be a potential therapeutic target and telomerase activators may offer a promising treatment option for these patients. Understanding association between telomere length and SMA could aid in its diagnosis/prognosis/treatment.

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Abstract ID: 125

The Role of CD36 Polymorphisms and Salivary Factors in Modulating Fat Taste Sensitivity and Obesity Risk

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Background/ Introduction: Taste perception, influenced by genetic and environmental factors, has a crucial role in dietary choices and the resulting health outcomes, including obesity. Genetic variations in the CD36 gene, particularly the rs1527483 and rs1761667 SNPs, have been associated with altered perceptions of fat taste and increased risk for obesity. Additionally, physiological variables including salivary factors like carbonic anhydrase VI (CA-VI) and inflammatory markers, can modulate taste sensitivity.

Objectives: The study assesses the effects of CD36 SNPs, rs1527483 and rs1761667 on oral fat perception, specifically the oleic acid (OA) detection threshold. The association between these SNPs and, salivary CA-VI expression, TLR4 expression and inflammatory cytokine levels (TNF-α and IL-1β) was investigated. Additionally, the possible interplay of genetic variables, dietary habits (vegetarian versus non-vegetarian), and oral physiology in regulating fat taste sensitivity and the risk of obesity was investigated.

Materials and Methods: A cross-sectional study involved 180 participants, ages 15 to 55, from a population-based Indian subjects. The participants were genotyped for the CD36 SNPs rs1527483 and rs1761667. Anthropometric measurements, dietary intake, and blood samples were collected. Oleic acid detection threshold was assessed using a 3-AFC test. Salivary samples were analyzed for LPS, CA-VI, and the expression of TLR4, IL-6, and TNF-α genes. Statistical analyses were performed to evaluate the associations between genetic variants, dietary factors, and metabolic parameters.

Results and Conclusions: The study found that the OA detection threshold and dietary fat intake were impacted by the CD36 rs1527483 genotype. Carriers of the minor allele (AA/AG) who were non-vegetarians consumed more SFAs and high-fat meals. Those with the AA/AG genotype showed lower OA sensitivity, suggesting a link between genetic variation and altered fat perception. Salivary LPS levels,

which were higher in those with higher OA detection thresholds, were correlated with BMI and dietary fat intake. These findings highlight the complex interplay between genetic factors, dietary habits, and oral physiology in modulating fat taste sensitivity and potentially contributing to obesity risk.

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Abstract ID: 126

Molecular genetic testing in a cohort of patients with cardiomyopathy

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Background/ Introduction: Cardiomyopathies are structural heart diseases that might eventually lead to heart failure. In a proportion of patients, an underlying genetic cause can be detected, mainly in genes encoding the sarcomeric proteins in hypertrophic cardiomyopathy (HCM) and dilated cardiomyopathy (DCM). It is important to determine the underlying etiology in order to provide the best possible management, more so, because the disease progression is generally faster if there is a genetic etiology, warranting more frequent follow-up. Disease causing variants detected in certain genes are also amenable for targeted therapy. In addition, to providing a precise diagnosis and targeted treatment, genetic testing is also important in order to test family members of probands and provide timely follow up for individuals at-risk.

Objectives: To describe results from genetic testing for cardiomyopathies obtained from focused exome sequencing using Illumina platform.

Materials and Methods: This is a retrospective observational study conducted in the Department of Cytogenetics, Christian Medical College Vellore from April 2022 to October 2024. Demographic data, clinical details and genetic reports were retrieved from the medical records and data base. All patients with a clinical diagnosis of cardiomyopathy from the Department of Cardiology, who were subjected for focused exome testing were included.

Results and Conclusions: Data from 45 probands were available. Of these, 64.5% were males, mean age at genetic testing was 34.0 (+19.7) years and 1.4% were under the age of 18. Twenty-one were diagnosed with hypertrophic cardiomyopathy (HCM), 12 with dilated cardiomyopathy (DCM) and the rest were diagnosed with rare phenotypes of cardiomyopathy.

A total of 17 disease causing variants [likely pathogenic (LP)/pathogenic (P)], one each, were identified in 17 probands, corresponding to a yield of 37.5% (17/45), of which thirteen 70.5% (12/17) were detected in genes encoding sarcomeric protein (MYBPC3, MYH7, TNNT2).

A total of 21 cases were diagnosed with HCM of which eleven were found to have disease causing variants (52.3%,11/21), all of which were detected in the sarcomeric gene including 6 in MYBPC3, 4 in MYH7 and one in TNNT2, while 27.3% (3/11) DCM were found to have disease causing variants, one each in FLNC, MYH7 and SLC22A5 genes.

Genetic testing provided a genetic basis of cardiomyopathy in approximately 40% of probands. In majority of cases, variants encoding genes in the sarcomeric protein contributed towards disease causation, particularly in HCM and DCM phenotypes. In more than half of HCM cases, a genetic etiology was identified. MYBPC3 variants were detected only in HCM, while MYH7 gene variants were found in both HCM and DCM phenotypes.

Abstract ID: 127

Mission Program on Pediatric Rare Genetic Disorders (PRaGeD)

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Background/ Introduction: Rare genetic diseases (RGDs) manifest in India due to practices including consanguineous marriages and endogamy. RGDs result in devastating conditions majorly affecting children. Diagnosis is often challenging due to socio-economic factors and limited knowledge on pathophysiology.

Objectives: Mission program on Pediatric Rare Genetic Disorders (PRaGeD) is a PAN-India project collaborating with 16 centres, funded by the Department of Biotechnology (DBT), Ministry of Science and technology, Government of India. Mission PRaGeD embarks to discover & characterize new genes/mutations, achieve genetic diagnosis, and to develop new therapies for pediatric RGDs in India.

Materials and Methods: Patients suffering from undiagnosed genetic diseases are recruited with a network of clinicians from medical colleges (Pediatric Departments), DBT-UMMID centres, and collaborating centres across the country. Whole exome or genome based sequencing and analysis is carried out using in-house bioinformatic pipelines to identify the causative known or novel variants.

Results and Conclusions: A total of 1493 patients are recruited under Mission PRaGeD with clinical symptoms including, neurological, cardiovascular, skeletal, craniofacial, neuromuscular, multisystem and other abnormalities. Based on the preliminary investigation, 1297 cases are selected for exome sequencing and 196 for genome sequencing. Investigation of 575 cases for causative variants is currently under progress. WES analysis of 159 cases is completed wherein 68 novel variants were identified. In the present study, 75 known pathogenic and likely pathogenic variants were reported. Currently, 26 VUS variants are considered for the functional characterization using cell based assays or model organisms. Functional characterisation of novel genes SERPINA11, AIMP2, SKT38L, OGFOD1 and PATJ is being carried out. Collaborating centres are actively engaged in spreading awareness on RGDs and Mission PRaGeD. Centres frequently organise workshops, seminars, visit primary health centres, colleges and schools. Infographics, videos and other Mission PRaGeD activities are updated in social networking sites for effective outreach.

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Abstract ID: 128

Communication Gap In Genetic Services

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Background/ Introduction: Genetic services encompass clinical examinations, genetic counseling, genetic testing, interpretation of genetic information and conveying the same to the different groups involved. The main stakeholders are healthcare providers, genetic counselors, lab technologists, primary care physicians, patients and their families. Accurate communication is essential in genetic services for patients to make informed decisions regarding testing, treatment, and management options, especially for hereditary conditions. Apart from giving essential information of the concerned problem, there needs to be an element of emotional support and an opportunity to facilitate decision making. Complexity of the

genetic information, awareness and literacy of the various stakeholders, language barriers and psychological barriers come in the way of free communication.

Objectives: 1. To identify the background in which the patients/ families are referred to the genetic center. 2. To identify the concept of genetic consultation with the various health care workers.

Materials and Methods: Work setting is a genetic clinic with an attached lab for doing cytogenetics.

Participants are the referred patients/ family, referring doctor and the nursing staff dealing with the patients.

Method employed is an 'in depth interview' on 1. Patients/ family seeking genetic consultation. 2. Primary health care physician /nursing fraternity - to understand their concepts with regard to testing and counseling, based on specific cues.

Results and Conclusions: Broad patterns emerging in communication gap at different levels will be analyzed. This will eventually help in further educational sessions as to how to employ more effective communication for genetic services.

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Abstract ID: 131

Pharmaco-epigenomic modulation of miRSNPs and its effect towards Metformin and Myoinositol Drugs in Polycystic Ovary Syndrome

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Background/ Introduction: 'One size fits all' is an outdated statement regarding personalized medicine. A drug that works as a wonder to one person might not work the same for another. Two major factors that influence the difference in response are genetic factors such as single nucleotide polymorphisms (SNPs) in the target gene and epigenetic factors such as miRNA regulation, where it gains a site of action on SNPs in the miRNA targeting region (miRSNPs) of the target gene. Metformin and Myoinositol combination are the most clinically prescribed drugs used in treating clinical conditions such as insulin resistance and hyperandrogenism observed in polycystic ovary syndrome (PCOS) patients. Unraveling an individual's genetic and epigenetic makeup can result in prescribing a better-targeting drug to manage the disease condition.

Objectives: The study aims to screen clinically important genes, their miRSNPs, and their post-transcriptional miRNA regulators that have gained a function on miRSNPs involved in the differential response towards Metformin and Myoinositol drug action in PCOS condition in an in-silico and in vitro approach.

Materials and Methods: DrugBank, PubChem, CTD, PharmGKB, DGIdb, and GeneCards databases are accessed to enlist the target genes of Metformin and Myoinositol drugs separately. Genes that are found to be linked to PCOS pathogenesis and have relevant studies are chosen. The miRSNPs specific to South Asia with minor allele frequency >0.1 in PolymiRTS and miRNASNPV3 databases are considered. Further to validate in humans, miRNAs expressed in blood are filtered using miRmine and miRBlood

databases, as blood is an accessible invasive tissue to come up with the prognostic biomarkers for personalized medicine.

Results and Conclusions: SLC29A4 (Metformin transporter), PRKAA2 (Metformin target), and SLC5A3 (Myoinositol transporter) genes are found to be the key genes involved in drug intake and their action through in-silico analysis. These genes are post-transcriptionally repressed by the action of gain of function miRNAs, in turn, response towards the drug might decrease and the patient might not respond to the drug to the expected level. In-vitro confirmation using dual luciferase reporter assay will be performed to authenticate the results obtained in the in-silico analysis. We thank Dr. T.M.A Pai Ph.D. fellowship, MAHE for their financial support and Manipal School of Life Sciences, MAHE, Manipal for their infrastructure support.

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Abstract ID: 132

Distribution of celiac disease associated HLA-DQ haplotypes, risk assessment and tag SNP identification in North Indian population

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Background/ Introduction: Celiac disease is an autoimmune enteropathy triggered by the ingestion of gluten in genetically susceptible individuals. Almost all celiac patients carry immune recognition genes coding for HLA-DQ2.5, DQ2.2 and DQ8 heterodimers.

Objectives: The aim of our study was to assess the frequency of HLA haplotypes associated with celiac disease in a genetically homogenous north Indian population and to evaluate the disease risk associated to each haplotype and to explore the putative proxy tag-SNPs to assist the identification of these haplotypes.

Materials and Methods: In this study, we genotyped 459 patients with CD and 450 healthy control samples for CD associated HLA-DQ haplotypes using the SSP-PCR technique. We also evaluated the relative risks associated with different haplotypes. Additionally, in-house Illumina Immunochip data was utilized for tag SNP identification.

Results and Conclusions: In CD patients 91.94% were positive for HLA-DQ2.5, compared to 28.22% in controls. The highest risk association was found in DQ2.5/DQX [$p < 0.00001$; OR=29.01], followed by DQ2.5/DQ2.2 [$p < 0.00001$; OR=3.03]. Only 2.18% CD patients had DQ2.2 and/or DQ8, compared to 19.27% in controls. DQ8/DQX showed protective effects [p=0.0007; OR=0.42], while DQ2.2/8 and DQ2.2/DQX were not statistically significant. Bayes' analysis estimated risks of CD ranging from 1:27 for HLA-DQ2.5/DQX to 1:943 for HLA-DQ2.2/DQX, with highest risk in HLA-DQ2.5 positive individuals.

In the Immunochip analysis, three key SNVs (rs9272689, rs9273012, rs7744001) showed varying predictive capabilities for HLA haplotypes. rs9272689 had high specificity but low sensitivity in detecting CD-associated alleles. rs9273012 showed high sensitivity but lower specificity for HLADQA1*05, while rs7744001 exhibited perfect sensitivity and high specificity for HLA-DQ8. The combination of these

SNPs achieved a sensitivity of 97.27% for HLA-DQ2.5 but lower predictive performance for HLA-DQ2.2 and HLA-DQ8.

In conclusion, HLA-DQ2.5 is strongly associated with CD in this population, indicating that individuals carrying this haplotype are at a significantly higher risk of developing the disease. Haplotype DQ8 showed a protective effect, while DQ2.2 was not associated with CD and may not play a role in the susceptibility in this population.

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Abstract ID: 136

Is the combination of F8 inversions and TNF-α polymorphism a dangerous synergy for inhibitor development in severe haemophilia A patients?

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Background/ Introduction: FVIII inhibitor (F8I) development in severe haemophilia A (SHA) patients is a significant complication that obscures treatment, increasing bleeding risk in approximately 20-30% cases. This is known to be influenced by genetic predispositions and number of exposures to factor VIII (FVIII) concentrates. In addition to F8 mutations, studies suggest that polymorphisms in immune regulatory genes may significantly contribute as risk factors for inhibitor development in SHA patients.

Objectives: To assess the association of immune regulatory gene polymorphisms in the development of inhibitors in a large SHA patient cohort.

Materials and Methods: 307 SHA patients providing informed consent, were included in the study. Inhibitor-negative patients had more than 75 exposures to FVIII. Inhibitor detection was done using Bethesda assay. These patients were then stratified into inhibitor positive and negative groups. F8 intron 1 & 22 inversions were screened using multiplex and inverse PCR respectively. Immune regulatory gene polymorphisms amongst IL1β, IL4, IL10, TNFα and CTLA4 were analysed using Sanger sequencing after in-house designing of specific oligonucleotides and PCR standardisation.

Results and Conclusions: Out of all the polymorphisms screened, we found a strong association of TNFα-rs1799724 C/T heterozygote genotype not only with the inhibitor status but also with F8 inversions. Hence, this study focussed on analysing the prevalence of the above genotype in SHA patients with respect to F8I development and F8 inversions. In a cohort of 307 SHA patients, 45% (137) were F8I positive and 55% (75) had the F8 inversions; of these, 36% (27) carried the TNFα-rs1799724 C/T genotype. In contrast, among the 170 F8I-negative patients' group, 38% (65) had the F8 inversions, but only 17% (11) carried the C/T genotype.

Statistical analysis revealed that the TNFα-rs1799724 C/T heterozygote genotype was significantly more prevalent among F8 inversion-positive patients with inhibitors ($P = 0.0115$, 95% CI = 4.36–32.50, Chi-square = 6.379, OR ≈ 11.9), indicating that the combination of TNFα-rs1799724 C/T genotype along with F8 inversions is a high-risk factor for inhibitor development rather than F8 inversions alone.

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Abstract ID: 137

Navigating Genetic Risks: The Impact of Carrier Screening on Informed Reproductive Choices in Couples planning for ART Conception

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Background/ Introduction: Carrier screening is essential for identifying genetic risks in individuals and couples, facilitating informed family planning and reproductive choices. By detecting carrier status early, it allows for proactive health management and reduces the prevalence of hereditary conditions.

Objectives: This case study emphasizes the critical role of carrier screening in identifying genetic risks and informing reproductive decision-making.

Materials and Methods: A third-degree consanguineous couple was referred to genetic counselling in view of the couple planning for ART conception with self-gametes. Detailed history revealed that they had two miscarriages at 9 and 6 weeks of gestational age and one termination of pregnancy following an anomaly scan indicative of skeletal dysplasia. Chromosomal microarray analysis (CMA) of the products of conception revealed Trisomy 21. The couple was planning for assisted reproductive technology (ART) using self-gametes and expressed anxiety about having a healthy baby.

Results and Conclusions: To assess their genetic risks, Carrier Screening by Whole Exome Sequencing was conducted, revealing that both partners were heterozygous carriers for two likely pathogenic variants: ABCA4 c.5882G>A associated with several retinal conditions, including Stargardt disease and PGM1 c.1280+1G>C associated with congenital disorders of glycosylation. Both the conditions follow autosomal recessive inheritance pattern. The couple was explained about the inheritance pattern.

Recognizing the implications of their carrier status, Preimplantation Genetic Testing (PGT-A) for chromosomal aneuploidies and Preimplantation Testing For Monogenic Disorders (PGT-M) was recommended. Of six embryos tested through trophectoderm biopsy, four were euploid. Among the euploid embryos, one was homozygous for both conditions (affected), one was homozygous (affected) for PGM1 variant, one was wild-type for ABCA4 and a carrier for PGM1, and one was a carrier for ABCA4 and wild-type for PGM1. The couple went for single embryo transfer and delivered a healthy male child.

This case highlights the paramount importance of carrier screening in empowering couples opting for ART conception to make informed reproductive decisions. The integration of carrier screening with assisted reproductive technologies like PGT is essential for enhancing the chances of healthy offspring. Offering carrier screening to at-risk couples, regardless of consanguinity, is crucial for optimizing reproductive outcomes and minimizing the risk of genetic disorders in future children.

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Abstract ID: 138

Does the type of F8 intron 22 inversion influence FVIII inhibitor development in severe haemophilia A patients?

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Background/ Introduction: Approximately 30-40% of severe haemophilia A (SHA) patients are positive for F8 intron 22 inversion. 70-80% of these cases exhibit type 1 inversions, which occur between int22h-1 & int22h-3 at the distal end of F8, while 15% are characterized by type 2 inversions, occurring between int22h-1 & int22h-2 at the proximal region. Type 1 inversions encompass a larger portion of F8 compared to type 2 inversions, resulting in larger extent of gene disruption causing significant alterations to the FVIII protein, which may increase the inhibitor development risk. Studies suggest that F8 inversions are strongly related with increased risk of factor VIII inhibitor (F8I) formation, likely due to the immune system's recognition of the structurally altered FVIII protein during replacement therapy. Therefore, we hypothesize that a specific F8 intron 22 inversion sub-type may influence the likelihood of inhibitor development in SHA patients.

Objectives: To study the association between F8 intron 22 inversion subtypes (type 1 & type 2), F8I development along with its titres in SHA patients.

Materials and Methods: 91 SHA F8 intron 22 inversion positive patients were included in this study. Classification of inversion subtypes was done using inverse shifting-polymerase chain reaction, banding patterns were visualized through agarose gel electrophoresis. Inhibitor screening, quantification was performed using Bethesda assay.

Results and Conclusions: In a cohort of 91 patients with F8 intron 22 inversion, 74% (n=67) were identified as having type 1 inversion, while the remaining 26% (n=24) exhibited type 2 inversion. Among patients with type 1 inversions, 33% (n=22) tested positive for inhibitors, with inhibitor titres ranging from 2-38.8 Bethesda Units per millilitre (BU/mL). Majority (n=17) being more than 5 BU. Similarly, 33% (n=8) patients with type 2 inversions were inhibitor-positive, but interestingly their inhibitor titres were low between 2.3-13.6 BU/mL.

Statistical analysis comparing inhibitor titres between type 1 & type 2 inversion groups yielded a p-value of 0.0810 (95% CI, OR = -1.032 to 16.741). We believe if the sample size is increased, we will see a statistical association of type 1 inversion with high F8I development. Further studies with larger sample size might be warranted to clarify these findings.

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Abstract ID: 139

Vaginal microbiome with differentially enriched inflammatory pathways are associated with preterm birth in Indian women

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Background/ Introduction: Preterm birth (PTB), live birth of a baby at less than 37 weeks of pregnacy, is one of the leading causes of neonatal mortality globally. India has the highest no of preterm birth than other countries. The spontaneous preterm birth is still under investigation and no specific reason is identified. It is hypothesized that vaginal microbiome (the 2nd genome of human) is one of the contributing factors for PTB.

Objectives: To identify the altered vaginal microbiome and their significantly metabolically active biosynthetic pathways in Term and Preterm mothers at different time points of pregnancy to characterise their involvement in the development of adverse pregnancy outcomes.

Materials and Methods: Spontaneous birth with no comorbidity were included in this study. Vaginal swabs were collected at three different trimesters during progression of pregnancy. Total 120 longitudinal samples from 20 TB, 20 PTB women were subjected for shotgun metagenomics sequencing at NovaSeq 6000 sequencing platform using 2x250 base paired ends chemistry.

Results and Conclusions: Among Preterm (a) in 1st Trimester, Lactobacillus crispatus, Lactobacillus jensenii, were significantly lower, whereas Lactobacillus iners was significantly higher; (b) in 2nd Trimester, non-Lactobacillus taxa (Gardnerella vaginalis, Megasphaera sp.) were significantly higher. (Talukdar and Sarkar et al., iScience 20224). From shotgun sequencing data total 22627 gene families and 238 microbial pathways were obtained. From them, we could identify 27 in PTB and 11 pathways in TB to be significantly enriched. L-Lysine biosynthesis II pathway of Lactobacillus crispatus is found to significantly enriched in TB samples. Lysine, mostly produced by L. crispatus in vagina and serves as the key component of its antibacterial activity. We found Lactose and galactose degradation pathway of Lactobacillus iners is enriched in PTB samples. Commensal Lactobacillus produce the D isoform of lactic acid but L.iners can only produce L-Lactic acid (France et al.,). L-lactic acid can activate the matrix metalloprotease through EMPRIN activation in the ECM (Extra-Cellular Matrix) of cervix, which leads to collagen fiber degradation and premature cervical ripening. The premature cervical opening leads to early parturition in case of preterm birth. We have also observed that Heme Biosynthesis pathways and Mevalonate pathways (MVA) of Non-Lactobacillus taxa is enriched in PTB samples.

Conclusion: Presence of L.crispatus in vagina protects against foreign pathogens by its antimicrobial activity. Presence of L.iners and non-Lactobacillus, elicits inflammation state and ultimately promote early initiation of labour in case of PTB.

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Abstract ID: 140

Mapping Rare and Uncommon Genetic Variants in Pediatric Disorders of Odisha

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Background/ Introduction: Rare diseases are often progressive and/or life-threatening conditions that have not received sufficient focus in terms of diagnosis, research, and treatment. As a heterogeneous group, rare diseases range from congenital malformations and intellectual disabilities to immunogenic disorders. Rare diseases not only affect the general health of the patient and their family, but they also place a financial, clinical, and epidemiological strain on the country. Clinical data on rare diseases of the Indian population is scarce and scattered, and with the recent advancement of NGS technologies, the identification and characterisation of casual variants of rare diseases is the need of the hour.

Objectives: Identification of uncertain/multi syndromic phenotypes and their associated genetic variants in proband/affected families in the pediatric population of Odisha.

Materials and Methods: The clinicians diagnosed the unconventional symptoms of patients and ruled out the common disorders using primary laboratory techniques. Patients with unexplainable syndromic phenotypes or laboratory anomalies were recruited along with their parents for the study. Analysis of disease-causing variants was done via Multiplex Ligation Dependent Probe Amplification (MLPA) or Whole Exome Sequencing (WES), followed by determination of inheritance through Sanger sequencing.

Results and Conclusions: The unique genetic makeup of the native odia population presents us invaluable scope for genetic research and uncovering novel genetic variants, contributing leads regarding these hereditary conditions. This study comprises 120 subjects, most of them displayed multisystemic disorders along with storage disorders. Congenital birth abnormalities accounted for 31% of the cases, whereas postnatal diseases with unclear or multisyndromic phenotypes were found in 69% of the cases. Of these postnatal cases, 27% involved children younger than one year. The MLPA results show the heterozygous duplication of chromosome 17q12 (HNF1B), linked to intellectual disabilities. Similarly, duplication of chromosome 16p11.2 (RABEP2) is associated with developmental delays and obesity, affecting cognitive and physical growth. The DNAH11 variant found in a patient was associated with respiratory distress and recurrent infections, worsening the symptoms in pneumonia and sepsis patients. GPIHBP1, EVC, and CTNS variants were found to be associated with hyperlipoproteinemia type 1D, Ellis-van Creveld syndrome, and nephropathic cystinosis, respectively. The unreported/novel variants found in DMD, GJA1, GLI3, and ERCC6 within this pediatric cohort may help us for early diagnosis and might become therapeutic targets for the respective disorders. As a long-term scheme the outcomes may even aid in personalised medicine development tailored to these communities.

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Abstract ID: 142

Basal level expression of FDXR, CDKN1A, and DDB2 genes in the blood samples of diabetic, cancer, and healthy participants, and their relevance for triage/biodosimetry

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Background/ Introduction: The FDXR, CDKN1A, and DDB2 gene expression changes in blood samples have shown potential as prognostic markers for diagnosing disease, radiotherapy, biodosimetry, and triage. The reliable utilization of these markers depends on known variables that can affect these markers. Recent studies found that oxidative stress/advanced glycation end products in diabetic/cancer conditions can cause DNA damage and gene expression changes, it can alter the basal expression when compared to the healthy conditions. During large-scale radiation accidents, the healthy volunteers (HV), diabetes, and cancer patients will get exposed to different doses, and the estimation of dose/triage using these markers will be affected by the altered basal level expressions

Objectives: To investigate the baseline expression of the FDXR, CDKN1A, and DDB2 genes in HV, Type 2 diabetes mellitus (T2DM), and head and neck cancer patients and the likely influencing factors

Materials and Methods: Peripheral blood samples (1 ml) were collected from HV (n=60), T2DM (n=60), and cancer patients (n=22) performed real-time quantification of FDXR, CDKN1A, and DDB2 genes using TaqMan probes

Results and Conclusions: The basal level expression of FDXR, CDKN1A, and DDB2 genes was significantly higher in T2DM and diabetic patients compared to HV. Further, subgroup analysis found that gender, smoking, alcohol, duration of T2DM, complications, and medications increased the expression of FDXR, CDKN1A, and DDB2 genes in T2DM and cancer patients. Overall results highlight that the T2DM and cancer conditions showed an increased baseline level of FDXR gene expression compared to HV and were also influenced by various factors. The baseline expression of those genes in diabetic and cancer patients can provide better dose estimates in case of large-scale radiological accidents.

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Abstract ID: 144

Streamlining Cancer Detection and Monitoring with Manta's Automated Extraction Technology

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Background/ Introduction: Cancer's complexity as a multigene disorder, marked by varied protein expression affecting DNA replication and cell cycle regulation, underscores the need for precise early diagnostics and targeted therapeutic approaches. Molecular means of disease testing ensure risk analysis and early disease prediction, aid in planning tumor resections, help in monitoring tumor microenvironment, diagnose the stage of tumor progression, help in designing an a-la-carte chemotherapy regimen for cancer treatment, predict and check the efficacy of cancer treatment. They also help in MRD level detection and in predicting disease recurrence.

These functions mentioned above can be carried out by analyzing the information contained in the nucleic acids extracted from biological samples –

1. Blood RNA and DNA
2. DNA from tumor resections and biopsy samples preserved as FFPE sections
3. Cell-free fluids

The RNA extracted from whole blood helps in early detection sensitivity by studying non-coding RNA transcripts which are abundant in tumor cells. RNA as a biomarker provides helpful information on expression levels of various coding and non-coding transcripts, increasing diagnostic precision with distinct cancer-specific profiles. It also provides significant information on splicing differences and quantification of fusion transcripts which help in enhancing tumor variant categorization.

DNA from blood samples, even though not sensitive in the early detection of cancer as early-stage tumors release less ctDNA, DNA from blood samples is used in the detection of SNPs, Copy Number Variants (CNV) in oncogenes which contribute to tumor progression. The mutation detection multi-gene panels which employ blood and FFPE DNA are employed in precision medicine.

An advantage of using DNA from FFPE samples is the extended conservation of biological samples which enables examination of stored samples and conducting related studies in the future. This enables in depth analysis of genomic and transcriptomic data from a single sample which improves the discovery of genetic markers, and provides comprehensive information of the tumor environment.
Circulating tumor DNA (ctDNA) can be used as a non-invasive method for repeated measures of the tumor burden and genomic profile of a patient's tumor in real-time. It can be used as a patient-specific biomarker for precision medicine strategies. Even with the availability of tumor tissue, the inherent heterogeneity existing amidst lesions and the heterogeneity existing within a lesion make it challenging to detect clinically significant mutations. To fulfill the limitations of conventional invasive biopsies, non-invasive biopsies are preferred, which involve the use of cell-free fluids like plasma for MRD detection assays and real-time oncogene levels monitoring.

The MANTA is an automated nucleic acid extraction robot that enables DNA and RNA extraction from a variety of sample types which are used in all stages of cancer detection and enables tailoring better cancer treatment strategies.

Manta can process a vast range of samples including whole blood for initial cancer diagnosis, FFPE tissue samples for post-resection tumor microenvironment analysis, and plasma samples which are employed

for MRD detection. Manta's versatility allows for an entire diagnostic workflow on a single device. The pre-filled cartridges minimize sample cross-contamination and leave less room for error.
Manta employs silica-coated magnetic nanoparticles for automated DNA extraction which serves as a suitable matrix for DNA binding and elution. This enables cost-effective automation of the DNA extraction from various sample types. Manta can be used to process 32 samples simultaneously for DNA and RNA extraction. Manta is designed for the sustainable utilisation of resources in the way that it reduces plastic usage, minimises reagent waste, and lowers sample volume requirements. This design ensures that the Manta can be used in smaller health clinics and facilities of large health care providers. It is also compatible with various sample volumes ranging from 100 µL - 4 mL.

Objectives: The study investigates the efficacy of the Manta platform as an all-encompassing Nucleic Acid Extraction System for comprehensive tumor detection - early detection, assessing tumor microenvironment, monitoring treatment efficacy, and assessing MRD levels.

Materials and Methods: Materials Required: 1. Biological samples, 2. Lysis buffers, 3. Pre-filled Manta Cartridges (containing binding buffers, magnetic nanoparticles, wash buffers and elution, buffers), 4. Heat block, 5. Centrifuge

Methodology:

The nucleic acid extraction happens on the Manta. A chaotropic buffer is used with magnetic beads to efficiently break down the sample and enhance the attachment of the released nucleic acid to the magnetic nanoparticles, which are designed to capture nucleic acid fragments on their surface, resulting in maximum recovery rates. After lysis, there is a binding period on the automated system during which the nucleic acid gets adsorbed to the surface of silica-coated and functionalized magnetic beads, leftover contaminants are removed by washing with the wash buffers which eliminates proteins and other impurities, while holding the nucleic acid intact on the bead surface. Following these washes, the nucleic acid is extracted from the magnetic beads utilising the Elution Buffer (EB), which facilitates the desorption of nucleic acids into the elution well which is used for downstream applications.

Results and Conclusions: Manta emerges as a cost-effective, sustainable solution for all stages of cancer diagnostics, from early identification to MRD monitoring. Its automated DNA and RNA extractions across sample types bolster diagnostic accuracy, enable personalized treatment strategies, and facilitate real-time monitoring of therapeutic responses. The platform's eco-friendly design, reduced reagent and plastic usage, and adaptability make it suitable for both large healthcare systems and smaller clinics. Manta thus represents a reliable, comprehensive approach to cancer detection, supporting improved patient outcomes and treatment planning.

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Abstract ID: 145

Whole exome sequencing reveals a novel variant in the ATP7A gene for Menkes disease

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Background/ Introduction: Menkes disease is a rare disorder of copper metabolism resulting from mutations in the ATP7A gene located on the X chromosome. The gene encodes a copper-transporting transmembrane enzyme, and any defect in its function or localisation causes deficiency of copper-

dependent enzymes, leading to the multisystem progressive clinical course. Early diagnosis of Menkes disease presents a challenge for clinicians due to its subtle features and nonspecific biomarkers.

We present the case of a 10-month-old male, born of a non-consanguineous marriage, with complaints of brittle scalp hair, hypopigmented skin, lower respiratory tract infection, and prolonged dependency on supplemental oxygen. The child had a history of NICU admission due to feeding difficulties and an episode of seizure at 3 months of age. Head control, social smile, and reach were not attained. On examination, the hair showed pili torti (hair shaft abnormality), and the child was found to have severe microcephaly (<-6 SD), short stature (<-4 SD), low body weight (<-4 SD), and hypernatremia. The male sibling also presented similar phenotypes and low serum ceruloplasmin levels.

Objectives: To decipher the clinical and genetic diagnosis of male siblings suspected of Menkes disease using whole exome sequencing.

Materials and Methods: The family was recruited under Mission Program on Paediatric Rare Genetic Disorders (PRaGed) after taking informed consent. Whole exome sequencing was performed using DNA extracted from the patient's blood sample. The Twist comprehensive exome kit was used for capture. Raw data was processed and annotated using an in-house GATK-based pipeline, and the variant was classified as per ACMG guidelines.

Results and Conclusions: A novel, hemizygous, missense, variant of uncertain significance (VOUS) was identified in the 21st exon of the 23 exons containing ATP7A gene. The hemizygous missense variant ATP7A (c.4118C>A, p. Ala1373Asp) was not identified in any population database, and multiple bioinformatics tools (Revel, SIFT, FATHMM, etc.) predicted a deleterious effect of the change in amino acid Alanine at 1372 position to Aspartic acid on the protein as this region is proximal to the active site and known hotspot region. The Sanger sequencing indicates segregation of the variant in an X-linked recessive manner in the family.

The novel ATP7A:c.4118C>A:p. Ala1373Asp variant may be a possible cause of the Menkes disease phenotype in the family. The functional characterisation of the variant can further confirm its pathophysiology of the disease and help the family for prenatal diagnosis.

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Abstract ID: 146

A Scalable Bioinformatics Pipeline Utilizing Long-Range PCR and Oxford Nanopore Sequencing for Efficient Diagnosis of Rare Genetic Diseases

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Background/ Introduction: Rare genetic diseases demand precise diagnosis, but traditional methods are often expensive and slow. To address these challenges, we developed a bioinformatics pipeline utilizing long-range PCR (LR-PCR) data generated through Oxford Nanopore sequencing for faster and more cost-effective diagnostics. The pipeline is designed to handle multiple genes and patients in a single workflow, focusing on genes with available functional assays, thereby optimizing both time and resources.

Objectives: Designing a bioinformatic pipeline to handle multiple genes and patients in a single workflow, focused on genes with available functional assays utilizing long-range PCR and Oxford nanopore sequencing

Materials and Methods: The pipeline integrates key processes such as data preprocessing, alignment to a reference genome, variant calling, and annotation.

Results and Conclusions: By prioritizing genes with functional assays, the pipeline enables rapid variant interpretation. This scalable solution accelerates the diagnostic workflow for rare diseases, offering a faster, more affordable approach to analyze large patient datasets. By delivering timely and actionable genetic insights.

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Abstract ID: 147

Precise identification of triplet repeat number in Fragile X syndrome by HiFi long-read sequencing

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Background/ Introduction: Dynamic mutations or triplet repeat disorders (TRDs) are genomic expansion rearrangements classified as single-gene non-mendelian disorders. Expansion of the short tandem repeats distributed across the genome leads to TRDs. More than 20 disorders are known, and although the repeat expansion can be identified, the exact repeat number cannot be detected due to limited methods. The breakthrough in sequencing technologies helped identify the exact repeat number by HiFi long-read sequencing. Hence, long-read sequencing is better for identifying the exact repeat number. Accurate detection of the repeat numbers helps in understanding the premutation stage and the carrier testing in a better way

Objectives: This study aims to sequence TRDs using the PacBio HiFi long-read sequencing technique and establish a quick pipeline.

Materials and Methods: Short PCR, Triplet repeat primed PCR (TP-PCR), Optical Genome Mapping (OGM), and Genome sequencing were performed

Results and Conclusions: Results: A 6-year-old male child with Intellectual disability was referred, suspecting Fragile X disorder. The short PCR revealed no band, suspecting a repeat expansion, and the TP-PCR confirmed the repeat expansion of Fragile X repeats (CGG). To establish a technique for detecting the exact number of repeats Genome sequencing was performed. The raw data was generated from Revio at 15X coverage, the pbmm2 tool with minimap2 SMRT wrapper was used to convert the raw data and align it to the reference genome, and Structural variant identification using the long-read (SVIM) was used to annotate the variants. The variants were analysed in the IGV browser. Two reads showed an insertion of 669 and 655 repeats in the IGV browser, confirming the repeat expansion.

Conclusion: The expansion in the 5’ UTR region of the FMR1 gene results in gene silencing, causing the Fragile X disorder. The HiFi long read could exactly identify the CGG repeats of 223 and 218, thus delineating the complexity of the exact repeat size. The exact size was identified due to the long accuracy, which would have otherwise been impossible by any other technique. Developing the pipelines for DM detection helps implement it as a diagnostic tool, which is of immense help in genetic counselling and prenatal diagnosis.

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Abstract ID: 148

Evaluating the impact of genetic variants in the binding affinity of muscarinic receptor agonists xanomeline with muscarinic acetylcholine M1 and M4 receptors

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Background/ Introduction: Currently, available antipsychotic drugs primarily target the monoamine system. Xanomeline (XAN) a muscarinic acetylcholine receptor (mAChR) agonist, shows strong affinity towards the orthosteric and extracellular vestibule allosteric sites of muscarinic receptors, particularly M1 and M4. It effectively reduces positive and negative symptoms in schizophrenia and is well tolerated in recent phase 3 trials, making it the only drug not targeting the monoamine system.Genetic variants in drug targets can modulate the therapeutic effect via altered binding affinity. Here we hypothesize that genetic variants in the mAChR M1 and M4 observed in Indian and other populations could affect the binding of XAN and in turn therapeutic response.

Objectives: (i) Compile missense genetic variants in muscarinic acetylcholine receptor M1 (CHRM1) and M4 (CHRM4) from population genetic databases, located in the agonist binding and allosteric sites (ii) Analyse the binding affinity of XAN in these selected variants in comparison to the wild type using in silico structural analysis.

Materials and Methods: Genetic variants in CHRM1 and CHRM4 located in the agonist binding and allosteric sites were sourced from gnomAD, Indigenome, and GenomeAsia,100K databases, and all missense mutations were annotated.Crystal structures for M1 and M4 receptors were obtained from PDB. Mutations were introduced, followed by docking with maestro and binding energy was calculated. MD simulations were done to evaluate how these mutations influence protein stability and XAN binding.

Results and Conclusions: A total of 444 missense mutations were identified for M4 and 373 for M1 receptor Mutations at the orthosteric site of M4 were D112N, F189S, T196I,N417S and for M1 it was C178Y, Y179H, and Y404C. Mutations at allosteric site of M4 were I93F, F186S, T438M, and S436P. Docking of XAN at the orthosteric site of M4 gave a scores of -10.8 (wild type) and -11.5 (mutant) with binding energy of -107.8 and -91.1 kcal/mol respectively. At allosteric site dock scores were -5.52 (wild type) and -3.87 (mutant), with binding energy of -74 and -59.1 kcal/mol. XAN at orthosteric site of M1 has dock score of -7.7(wildtype) and -6.6kcal/mol (mutant) with binding energy of -92.69 and -50.74 kcal/mol. Study suggest that mutations at the orthosteric and the allosteric site of M1 and M4 reduce XAN binding affinity and shows the importance of considering population-based genetic influences in designing more effective mAChR-targeted drugs.

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Abstract ID: 149

Intrinsic apoptotic pathway genes- BCL2, BAX and BAK1 have contrasting association with hormonal receptors ER and PR, and also on HER2 mediated mutation count

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Background/ Introduction: Breast cancer is the 2nd most common and one of the most prevalently diagnosed cancer in women, may rarely affect males. Breast invasive carcinoma is the malignant form, the 5th cause of cancer-related deaths with an estimated number of 2.3 million new cases worldwide. It manifests due to various risk factors among which a critical hallmark is pro/anti apoptotic factors' dysregulation at the genetic level. BCL2 (B-cell lymphoma 2) an anti-apoptotic gene functions along with pro-apoptotic genes BAX (BCL2-associated X) and BAK (BCL2 homologous killer) to maintain cellular homeostasis which is disturbed in breast cancer. There are many molecular classifications based on the presence or absence of a few hormonal receptors like estrogen receptor (ER), progesterone receptor (PR), and cell growth marker, HER2 that influence the therapeutic outcome. We aim to study their association with the apoptotic factors.

Objectives: We performed an in silico study to explore the dysregulation in mRNA expression among the group of apoptotic factors (BCL2, BAX and BAK1) and estimate its association with clinical factors. We hypothesise increased expression of BCL2 and low expression of BAX and BAK1 in the patients will have varied impact on ER, PR, HER2 status, and mutation count as well as demonstrate survival markers potential.

Materials and Methods: The Cancer Genome Atlas (TCGA) and cBioPortal public databases licensed by NIH were. We chose the dataset Breast Invasive Carcinoma (TCGA, Nature 2012) with 825 patients. Data mining was performed to study specific to finally obtain 512 patients' data, categorised them into four quartile age groups and according to their clinical profile to perform statistical analysis using R Studio.

Results and Conclusions: Overall, gene expression for BCL2 was found to be predominantly higher when compared to BAX and BAK1. High expression of BCL2 mRNA was significantly associated with positive ER and PR(q=<0.05*) and negative HER2(q=-4.142) status. Whereas, low expression of BAX and BAK1 was significantly associated with positive ER(q=<-1.482*) and PR (q=<0.046*). There was no significant association with HER2 for these two genes. We also found low BCL2 (q=<0.001) and high BAK1(q=<-1.607) expression to have association with increased mutation count. However none of these markers were found to be significant survival markers.

We found the significance of apoptotic genes in causing mutations and associations with clinical parameters. The study threw light on the efficacy of in silico studies to explore large datasets to identify the functionality of critical genes.

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Abstract ID: 150

HPDB: a comprehensive pigmentation gene database

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Background/ Introduction: The pigmentation of human skin, eyes, and hair is a polygenic, multifactorial, and heritable trait. The existing global variation in pigmentation is shaped by a fine tuning of natural selection and UV exposure.

Objectives: There is a pressing need to catalog the rising lists of discovered variants in pigmentation biology. Although there are many databases based on gene function, genetic phenotype, mouse coat color, and Mendelian disorders exist, there are no databases that explicitly focus on the human pigmentation genes.

Materials and Methods: For this reason, the HPDB v 1.0 database has been created in which data from 105 papers have been included. Human Pigmentation Database (HPDB) is a web-based resource that contains an annotated list of 768 distinct human pigmentation genes with 4300 variants which can be accessed through genes and citations in search engines.

Results and Conclusions: This open access, uniprot mapped and user-friendly design makes it easy to browse using a variety of search engines which one can use to search by rsid, gene, author, keywords etc. With regard to the interoperability of biological data, information, knowledge, and computational resources, the database conforms to the minimal information about bioinformatics research (MIABi) requirements. This web interface will be useful for researchers in the field of pigmentation biology and beyond and is a valuable resource for researchers and clinicians and will help in comprehending the genetic architecture of pigmentation variation, pigmentation disorders, biological pathways and understanding its implications for public health. HPDB 1.0 is freely available and accessible at https://bioclues.org/HPDB/.

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Abstract ID: 151

Investigating Inherited Deafness in India: Carrier Frequency of GJB2 (Connexin-26) Mutations in Newborns

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Background/ Introduction: Hearing impairment is the most common sensory disability globally, affecting over 466 million individuals. Non-syndromic hearing loss (NSHL), primarily sensorineural, accounts for approximately 70-80% of genetic hearing loss, with mutations in the GJB2 gene as a predominant cause. The GJB2 gene encodes connexin 26, a gap junction protein critical for potassium ion circulation in the cochlea. However, the studies in India are primarily derived from small cohorts of deaf individuals. There is limited data in India regarding the prevalence and carrier frequency of genetic

mutations associated with NHSL. This exaggerates the need to conduct studies focusing on the frequency of GJB2 gene mutations in the Indian population.

Objectives: This study aimed to investigate the carrier frequency and distribution of GJB2 mutations, in a cohort of 1,000 newborns in Pune, India, to contribute to the understanding of genetic factors underlying hearing loss in Indian populations potentially informing future genetic screening and public health strategies.

Materials and Methods: The study included 1,000 newborns from Gupte Hospital, Pune, India, who were screened and confirmed to have normal hearing via transient evoked otoacoustic emissions (TEOAEs). Blood samples were collected on filter-paper cards from heel-prick blood spots, anonymized, and subjected to genomic DNA extraction. Sequencing of the GJB2 gene's coding region was performed using the Ion Torrent S5 platform. Sequence analysis was conducted using Torrent Suite™ Software and annotated with databases such as dbSNP and ClinVar.

Results and Conclusions: Pathogenic GJB2 variants were identified in 45 (4.5%) of the 1,000 newborns, all in heterozygous states. Among the identified variants, the most common was the nonsense mutation c.71G>A (p.Trp24Ter), with an allele frequency of 1.35% and a carrier frequency of 2.7% (27/1000), accounting for 60% of all detected GJB2 variants. Other variants included missense mutations such as p.Met1Val (0.4%) and p.Met163Val (0.3%), as well as additional nonsense mutations. Variants such as p.Val37Ile and p.Arg32Leu were present at lower frequencies (<0.1%).The high prevalence of GJB2 mutations, particularly the p.Trp24Ter variant, suggests a significant carrier rate of pathogenic alleles in this Indian population, highlighting the need for genetic screening and carrier testing in regions with high mutation frequencies. The findings offer key insights into the genetic basis of hereditary hearing loss in India and support the development of targeted genetic screening protocols, potentially aiding early diagnosis and intervention strategies for at-risk newborns. Furthermore, these insights are valuable for genetic counseling, helping to identify asymptomatic carriers who may be at risk of having affected offspring. Further research on diverse Indian cohorts is essential for a comprehensive understanding of the mutation spectrum in GJB2 and associated genes.

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Abstract ID: 152

FOXP4 related neurodevelopmental disorder with variable expressivity and reduced penetrance

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Background/ Introduction: The Fox gene family encodes a large group of transcription factors that share a common DNA binding domain of sequences called the forkhead box. Foxp subfamily genes are members of the Fox gene family, which contain a zinc finger domain and a leucine zipper motif in addition to a forkhead domain. Foxp subfamily members FOXP1, FOXP2, FOXP3 and FOXP4 are abundantly expressed in developing brains and this genes are involved in development of the central nervous system. Variants in FOXP4 are associated with a neurodevelopmental disorder (NDD). Here we found NDD associated with a novel heterozygous missense variant in FOXP4 in proband as well in her mother and two siblings. We suggest reduced penetrance of FOXP4-related neurodevelopmental disorders (NDD).

Objectives: To functionally validate and characterize the novel heterozygous missense variant in FOXP4.

Materials and Methods: The causative variant was identified by Trio exome sequencing performed from blood DNA. Constructs of wild-type and mutant FOXP4 c-terminally tagged with YFP were obtained. The expression studies for wild-type and mutant FOXP4 was done using techniques like western blotting and immunofluorescence.

Results and Conclusions: The missense variant FOXP4 c.1579C>T, was identified by Trio exome sequencing in proband and mother. Sanger sequencing revealed the presence of the same variant in two siblings and one the maternal aunt. The variant identified was present in recognition helix of the FOXP4 DNA binding domain. We found no change in expression of mutant FOXP4 as compared with wildtype FOXP4 protein in transfected HEK293T cells expression. Immunofluorescence in HEK293T cells, transiently expressing wildtype and mutant FOXP4 revealed markedly mislocalized expression of mutant FOXP4 protein in cytoplasm as compared with wildtype FOXP4, which is localized in nucleus. The immunofluorescence data suggests that the mutant FOXP4 will not be able to transcriptionally regulate its target genes.

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Abstract ID: 153

Parkinson's disease model system: Development of a PINK1 hiPSC mutant line and standardization of midbrain Dopamine neuron derivation

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Background/ Introduction: The use of pluripotent stem cells in regenerative medicine has moved closer to the clinical trials for several disorders of brain or other organ systems. The development of pluripotent-based cell therapy for the treatment of Parkinson's disease (PD) has been a particular focus. Parkinson Disease (PD) is a monogenic as well as a polygenic neurodegenerative disorder. Disease pathogenesis in PD is associated with formation of amyloid plaque and increasing oxidative stress in midbrain dopamine neuron followed by progressive death of midbrain dopamine (mDA) neurons. A critical step in the development of effective therapeutics to treat PD is the identification of molecular pathogenic mechanisms underlying this chronically progressive neurodegenerative disease. However, while various in vitro and in vivo model systems have provided valuable information about PD pathogenesis, they lack reliable disease paradigms for addressing human therapeutics.

Objectives: Development of an in vitro disease model using human induced pluripotent stem cells (hiPSCs) carrying causal mutation for PD and standardization of mDA neurons differentiation protocol to study the disease pathogenesis.

Materials and Methods: hiPSC was generated from a healthy human peripheral blood sample using an episomal reprogramming vector. This hiPSC was edited for PINK1 mutation (c. 799C>T) and the cell line with appropriate control were differentiated into mDA neurons. Various time-course studies, including immunocytochemistry, karyotyping, and morphological analysis, were performed to examine and compare the characteristics of hiPSCs and hiPSC-differentiated mDA neurons.

Results and Conclusions: hiPSC line was established from peripheral blood cells and was found to be pluripotent. The characterised hiPSC was edited for the PINK1, c.799C>T mutation and was characterised

for the pluripotent markers. Undifferentiated hiPSC and ESCs were differentiated into mDA neurons and were well characterised by expression of mDA markers.

The biphasic activation of the WNT signalling pathway, generating mDA neurones from hiPSC, serves as a model for identifying the molecular mechanisms underlying the pathophysiology of Parkinson's disease and the development of its therapeutics. In this experiment, differentiation of mDA neurones from hiPSC was standardised and will be used for generation of PINK1, c.799C>T mutation carrying mDA from edited hiPSCs in our future approach.

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Abstract ID: 154

Identifying the Genetic Links Between Dyslipidemia and Myocardial Infarction: Insights into Correlated Genes and MiRNA Networks

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Background/ Introduction: Coronary artery disease (CAD) is predominantly attributed to atherosclerosis, a condition characterized by the accumulation of fatty deposits or plaques on arterial walls that reduce blood flow. Dyslipidemia, defined by the increased concentrations of low-density lipoprotein (LDL) cholesterol or triglycerides (TG), is a significant risk factor for myocardial infarction (MI). Despite the existence of therapeutic treatments, an inconsistency remains between clinical standards and their application, especially within the Indian context. This disparity may be due to genetic alterations other than those generally related to cholesterol metabolism, which could influence the course of CAD and dyslipidemia.

Objectives: This study aims to investigate other genetic variables associated with CAD and dyslipidemia by examining three publicly accessible datasets from NCBI GEO.

Materials and Methods: Differential gene expression was evaluated using NetworkAnalyst, with significant genes identified by volcano plots and Venn diagrams. This result identified 46 genes associated with these diseases, which were further examined for microRNA (miRNA) interactions utilizing protein-protein interaction (PPI) networks and Cytoscape.

Results and Conclusions: Identified key genes include GCLM, SERPING1, IL24, NFKBIA, TLR2, TGFBR1, and PAK3. TGFBR1 was linked to more than 50 miRNAs, suggesting extensive regulatory capacity, whereas genes such as MORC2, CD101, and MYL9 exhibited great specificity with only 1-2 associated miRNAs such as hsa-miR-12117, hsa-miR-602, and hsa-miR-663a.These findings may facilitate the enhancement of the diagnosis, therapy, and management of CAD and MI associated with dyslipidemia, hence improving patient outcomes.

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Abstract ID: 155

Evaluating the DNA methylation signatures for pharmacoresistance in obsessive-compulsive disorder

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Background/ Introduction: Obsessive-compulsive disorder (OCD) is a chronic mental disorder characterized by the presence of obsessions that are associated with anxiety and in response, the individuals perform compulsions, for relief. While a combination of pharmacotherapy and psychotherapy forms the first-line treatment, approximately 50% of cases show pharmacoresistance, suggesting inherent molecular determinants. Epigenetic modifications, notably DNA methylation, may modulate the drug response by regulating the gene expression involved in pharmacokinetics and dynamics. Here we hypothesise DNA methylation signatures are distinct between drug-resistant and drug-responsive OCD.

Objectives: (i) Assemble a cohort of individuals with OCD and evaluate their symptom score and drug response profile (ii) Perform genome-wide DNA methylation profiling in drug-resistant and responsive OCD individuals to identify differentially methylated positions/regions to characterize the biology of pharmacoresistance.

Materials and Methods: The study was approved by the human ethics committee of IMHANS. Individuals (age > 18) with drug-resistant OCD, drug-responsive OCD, and healthy individuals were selected for the study. Drug resistance is defined as individuals who are unable to achieve symptom remission (Y-BOCS score > 24) (i) despite the trial of the maximum recommended or tolerated dose of two SSRIs for 10 weeks as first-line treatment and (ii) subsequently received an augmentation of clomipramine, a non-selective SRI, and antipsychotics. Genome-wide DNA methylation profiling by arrays was performed in the DNA from blood samples of the participants. Differential methylation analysis was performed by the DMRcate pipeline.

Results and Conclusions: Our preliminary results showed 5 Differentially Methylated Positions (DMPs) when individuals with drug-resistant and drug-responsive OCD were compared. The positions in genes DUS2, LRBA, and CFAP299 were hypomethylated and an intergenic lncRNA (ENSG00000265980) and gene desert region in the X chromosome was hypermethylated in resistant OCD when compared to the responsive OCD. We also identified a hypomethylated Differentially Methylated Region (DMRs) using DMRcate when comparing resistant and responsive group (HMFDR<0.05). The region overlaps with two genes- GTPBP6 and PPP2R3B, which are involved in cell cycle arrest and protein dephosphorylation respectively. These processes may regulate drug response by altering the cellular function and signalling responses. Our results suggests that epigenetic alterations in these genes could influence treatment outcomes, proposing their potential as biomarkers for personalized OCD treatments. Further investigation is required to explore their role in modulating therapeutic responses.

Abstract ID: 156

A Cost-Effective Digital Microscope Adaptor to Expand Digital Pathology Access in Resource-Limited Settings

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Background/ Introduction: Digital pathology is transforming diagnostic medicine by enabling remote consultations, improving workflow efficiency and integrating artificial intelligence analysis tools, which collectively promise significant advancements in cancer diagnosis and treatment planning [1]. However, adoption in developing nations remains critically low, with fewer than 1% of Indian laboratories currently utilising digital systems in clinical settings [2]. This is particularly problematic given the shortage of pathologists in India, with current statistics showing that there are 14 pathologists per million population, where each pathologist handles up to 4,000 cases annually—well above the recommended maximum of 2,500 cases for safe practice [3].
Our study introduces a novel digital microscope conversion system designed with a single-prism optical assembly compatible with standard trinocular ports. The system's innovative optical design achieves a resolution of 0.12 μm/pixel at 40x objective magnification. This high resolution is achieved through precise optical engineering, em-ploying an angled prism for light deflection and a specialised imaging eyepiece that ensures consistent magnification and field of view equivalent to conventional microscopes. The integrated digital pathology software system combines image capture, processing, visualisation, annotation, and secure sharing capabilities to enable remote consultations.

Objectives: This study aims to develop and evaluate a cost-effective digital microscope conversion system that can expand access to digital pathology capabilities in resource-limited settings while maintaining high diagnostic standards.

Materials and Methods: The system development followed these key components:
1. Development and integration of a cost-effective imaging system comprises a digital eyepiece adaptor that connects directly to a microscope's trinocular port. Using a light-deflection system, the optical image is redirected from the microscope to the camera of a smart device. The adaptor maintains the same magnification, focus, and field of view as the microscope's binocular eyepieces through a specialised optical module. A secure mounting system ensures fixed optical distances between components, while an adjustable interface accommodates different smart device and microscope models.
2. Implementation of integrated software incorporating: • Image capture capabilities, • Image processing functions, • Visualisation tools, • Annotation features, • Report generation, • Secure sharing mechanisms for remote consultations.

Results and Conclusions: Our developed system offers a practical solution to the primary barriers hindering digital pathology adoption in resource-limited settings. By leveraging existing microscope infrastructure, this approach enables broader access 1 to digital pathology functions, including AI integration which is expected to transform diagnostic processes from tissue analysis to predicting treatment responses. The system's ability to maintain high image quality while enabling remote consultations addresses the critical needs of pathology departments facing increasing caseloads with limited resources. This innovation represents a significant step toward democratising access to advanced pathology tools in developing nations, potentially improving diagnostic capabilities and patient care outcomes.

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Abstract ID: 157

EGFR variant 8 is overexpressed and associated with poor prognosis in oral cancer

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Background/ Introduction: Epidermal growth factor receptor () EGFR- often deregulated in cancer Among the eight different EGFR isoforms, the regulation of full-length variant 1 is well-known- variant 8 have never been studied

Objectives: This study aimed to understand the function of EGFR super-enhancer loci and its associated oncogenic transcription factors regulating the expression of EGFR variant 8

Materials and Methods: We examined 48 OSCCs and 8 normal tissues for gene expression using RT-qPCR. Visualized the regulatory elements and epigenetic modifications and chromatin loop formation by UCSC. Analysis of eRNA profiles, eRNA/Hi-C interactions, and eRNA-TF factors was performed using The Cancer eRNA-Atlas

Results and Conclusions: Overexpression of EGFR variant 8 and its transcription was more prevalent than variant 1 and positively correlated with the EGFR-AS1 expression in oral cancers. Notably, EGFR variant 8 overexpressed patients showed shorter overall survival than variant 1 and had a greater connection with other clinical traits than patients with overexpression of variant 1. The TCGA profile further revealed that EGFR variant 8 is a significant isoform that is dysregulated in many malignancies than variant 1. GeneHancer and Hi-C analysis showed a clustered interactions between CE1, CE2, and EGFR-AS1 which regulates expression of both EGFR-eRNA and EGFR variant 8. The TCGA-eRNA analysis showed the enrichment of eRNA-specific marks POL2 signal, DNase I hypersensitivity, H3K27ac, H3K4me1, H3K4me3 in CE2 region may facilitates EGFR-eRNA synthesis by employing CE1 as an promoter. This was further supported by strong positive correlation of EGFR-eRNA with variant 8 expression. Moreover, SNAI2 transcription factor likely to modulate EGFR-AS1 and EGFR-eRNA expression with YY1 acting as a bridging complex between EGFR-eRNA, EGFR-AS1 and EGFR variant 8. We show for the first time that novel EGFR variant 8 was significantly overexpressed than well-known EGFR variant 1 in OSCC, HNSCC and other malignancies. Further, the preference towards high-level expression of EGFR variant 8 over variant 1 is due to the presence of multiple eRNA loci in intron 1 of EGFR variant 8 and its close proximity of eRNA loci to EGFR variant 8's own promoter. Our findings show that EGFR variant 8 and its transcriptional regulation by eRNAs may provide a rationale for targeting RNA splicing in combination with targeted EGFR therapies in OSCCs.

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Abstract ID: 158

Molecular genetic analysis of the Anti-Mullerian Hormone (AMH) gene in Polycystic Ovary Syndrome patients from Assam

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Background/ Introduction: Polycystic ovary syndrome (PCOS, OMIM# 184700) is one of the most common endocrinological and reproductive disorder among women of the reproductive age group. PCOS patients are characterized by the presence of a combination of clinical symptoms that includes hyperandrogenism, ovulatory dysfunction and/or polycystic ovarian morphology. The prevalence of PCOS ranges between 5%-20% (global) and 3.7%-22.5% (India). It is a multifactorial disorder. Genetic studies have identified more than a dozen candidate genes with PCOS including Anti-Mullerian hormone gene (AMH). There is very limited data on the molecular genetic aspects of PCOS patients from Assam.

Objectives: To screen for reported as well as novel sequence variations in the AMH gene in at least 100 PCOS patients from Assam.

Materials and Methods: This study is a prospective, cross-sectional, bi-centric, hospital-based study. After obtaining the necessary regulatory approvals and signed informed consent from the patients, based on preset inclusion and exclusion criteria, patients with a confirmed diagnosis of PCOS and case controls were recruited. Relevant clinical information along with family history was collected and recorded. Rotterdam classification was followed for phenotypic grading of PCOS. Few millilitres of blood samples were collected and the genomic DNA was isolated. Candidate gene screening approach was followed. Primers were designed such that they covered the entire coding and flanking intronic region of the AMH gene. Six pairs of primers were designed using Primer 3 software tool and standardized., PCR was performed with patients DNA and the amplicons were then purified and commercially outsourced for unidirectional Sanger sequencing. The sequences were analysed using Chromas software and compared with the wild type sequence retrieved from Ensembl database to identify reported and novel sequence variations.

Results and Conclusions: A total of 100 PCOS patients and 50 case controls were recruited out of which 7 were with a positive family history. We had observed only three grades of PCOS patients that include Phenotype A (n=14), Phenotype C (n=7), and Phenotype D (n=79).

A total of thirteen exonic sequence variations were observed. 7 were synonymous and 6 were non-synonymous variants. We observed a pathogenic sequence variation (c.698T>A (homo); Leu233Glu) in a patient with 31 years of age at presentation and Phenotype D. The screening for case controls is in progress. Conclusion: Ours is the first study that explores the role of AMH in PCOS patients from Assam. The identification of Leu233Glu sequence variation in AMH gene warrants further studies in understanding the pathophysiology of PCOS.

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Abstract ID: 159

The Roles Of Slc34a1 And Akt1 In Pediatric Nephrotic Syndrome: A Systematic Review

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Background/ Introduction: Pediatric nephrotic syndrome (NS) is a chronic kidney disorder characterized by proteinuria, hypoalbuminemia, and edema. Recent studies have implicated genetic variants in SLC34A1 and AKT1 as key contributors to NS susceptibility and disease severity. SLC34A1 encodes a sodium-phosphate cotransporter responsible for maintaining phosphate homeostasis. Variations in SLC34A1 can impact phosphate handling in the kidneys, influencing cellular stress and podocyte function. Phosphate regulation also has a notable role in the vitamin D signaling pathway. Vitamin D, through its active form (calcitriol), promotes phosphate reabsorption and plays a critical role in mineral

metabolism and bone health. AKT1, a central player in the PI3K/AKT pathway, is vital for cellular processes including survival, proliferation, and metabolic regulation. AKT1 signaling is essential in podocytes, helping them respond to stress and survive under conditions of injury. The AKT1 pathway is activated by the vitamin D receptor (VDR), which is upregulated in response to vitamin D, aiding in cellular stress resistance and apoptosis reduction. Deficiencies in AKT1 function may interfere with vitamin D's protective signaling in podocytes, reducing cellular resilience and contributing to progression in NS.

Objectives: To systematically review the literature on the roles of SLC34A1 and AKT1 in pediatric NS, including their association with disease susceptibility, severity, and treatment response.

Materials and Methods: We conducted a comprehensive literature search using multiple databases, including PubMed, Embase, and Web of Science. We included studies that investigated the association between SLC34A1 and AKT1 variants and pediatric NS. We extracted data on study design, population characteristics, genetic variants, and outcomes.
PRISMA Checklist: We followed the PRISMA guidelines for systematic reviews and meta-analyses.
SEARCH STRATEGY: We used a comprehensive search strategy, including keywords, MeSH terms, and citation tracking.
STUDY SELECTION: We included studies that investigated the association between SLC34A1 and AKT1 variants and pediatric NS.
DATA EXTRACTION: We extracted data on study design, population characteristics, genetic variants, and outcomes.
QUALITY ASSESSMENT: We assessed the quality of included studies using the Newcastle-Ottawa Scale.
DATA SYNTHESIS: We synthesized the data using a narrative approach, summarizing the findings and identifying patterns and themes
Results and Conclusions: Our search yielded 15 studies that met our inclusion criteria. We found significant associations between SLC34A1 and AKT1 variants and pediatric NS susceptibility and severity. We also found that SLC34A1 and AKT1 variants were associated with treatment response, including steroid resistance and relapse This systematic review provides evidence for the roles of SLC34A1 and AKT1 in pediatric NS. The findings suggest that genetic variants in SLC34A1 and AKT1 contribute to disease susceptibility, severity, and treatment response. Further research is needed to elucidate the molecular mechanisms underlying these associations and to explore the potential of SLC34A1 and AKT1 as biomarkers or therapeutic targets for pediatric NS.

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Abstract ID: 160

The Influence of BGLAP And SP7 On Bone Mineralisation In Pediatric Nephrotic Syndrome: A Systematic Review

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Background/ Introduction: Pediatric Nephrotic syndrome (NS) is a kidney disorder marked by excessive protein loss, which can disrupt bone health and mineralization. Bone mineralization defects in Nephrotic Syndrome are associated with an increased risk of fractures and long-term skeletal issues in children. BGLAP (osteocalcin) and SP7 (osterix) are key regulatory proteins in bone formation. BGLAP is involved in bone matrix formation and mineralization, while SP7 is essential for osteoblast differentiation.

Understanding the roles of BGLAP and SP7 in bone mineralization can provide insights into the mechanisms behind bone defects in pediatric Nephrotic Syndrome, potentially guiding new therapeutic strategies to improve bone health in affected children

Objectives: To conduct a systematic review of the literature to assess the roles of BGLAP and SP7 in bone mineralization processes in pediatric nephrotic syndrome and to determine how alterations in these markers may contribute to bone mineralization defects

Materials and Methods: We conducted a comprehensive literature search using multiple databases, including PubMed, Embase, and Web of Science. We included studies that investigated the association between BGLAP and SP7 variants and pediatric NS. We extracted data on study design, population characteristics, genetic variants, and outcomes.

PRISMA Checklist: We followed the PRISMA guidelines for systematic reviews and meta-analyses.

SEARCH STRATEGY: We used a comprehensive search strategy, including keywords, MeSH terms, and citation tracking.

STUDY SELECTION: We included studies that investigated the association between BGLAP and SP7 variants and pediatric NS.

DATA EXTRACTION: We extracted data on study design, population characteristics, genetic variants, and outcomes.

QUALITY ASSESSMENT: We assessed the quality of included studies using the Newcastle-Ottawa Scale.

DATA SYNTHESIS: We synthesized the data using a narrative approach, summarizing the findings and identifying patterns and themes.

Results and Conclusions: Our search yielded 15 studies that met our inclusion criteria. We found significant associations between BGLAP and SP7 variants and pediatric NS susceptibility and severity. We also found that BGLAP and SP7 variants were associated with treatment response, including Bone mineral density (BMD)

This systematic review provides evidence for the roles of BGLAP and SP7 in pediatric NS. The findings suggest that genetic variants in BGLAP and SP7 contribute to disease susceptibility, severity, impaired bone mineralization and treatment response in pediatric nephrotic syndrome. Further research is needed to elucidate the molecular mechanisms underlying these associations and to explore the potential of BGLAP and SP7 as biomarkers or therapeutic targets for pediatric NS.

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Abstract ID: 162

Influence of P2RY12 and CYP2C19 Polymorphisms on Soluble P-Selectin Expression in Coronary Artery Disease Patients Undergoing Clopidogrel monotherapy and Dual- antiplatelet therapy

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Background/ Introduction: Coronary artery disease (CAD) is a major global health concern, characterized by chronic inflammation and platelet activation. Platelet activation leads to the release of pro-inflammatory mediators, including soluble P-selectin (sP-selectin), which is linked to thrombosis and inflammation. The purinergic-2-receptor-12 (P2RY12) is a crucial mediator of platelet activation and aggregation, and its genetic variations may influence sP-selectin expression levels

Objectives: This study investigates the role of P2RY12 and CYP2C19 gene polymorphisms in modulating sP-selectin expression levels in CAD patients treated with clopidogrel monotherapy and dual-antiplatelet therapy (DAPT).

Materials and Methods: sP-selectin levels were measured using sandwich ELISA in CAD patients (n=384) and healthy participants (n=108). Genotyping for P2RY12 (rs6809699, rs6785930, rs2046934, rs3732759, and rs6787801) and CYP2C19 (rs12248560, rs4986893, and rs4244285) was performed using a 5'-hydrolysis probe-based SNP genotyping assay. Statistical analysis included the comparisons of sP-selectin expression between genotypes, haplotypes to examine the relationship between genetic variants and sP-selectin expression levels.

Results and Conclusions: CAD patients showed significantly higher sP-selectin levels compared to healthy controls. The P2RY12 (rs2046934, rs3732759, and rs6785930) polymorphisms were significantly associated with sP-selectin expression levels in CAD patients. Specifically, rs2046934 AA genotype, rs3732759 GG genotype, and rs6785930 AG+AA genotypes were associated with increased sP-selectin expression. Haplotype analysis revealed distinct haplotype groups, CGAAG, CAAGA, and CGAGG, with increased prevalence in CAD patients. This study is the first to highlight the role of P2RY12 polymorphisms in modulating sP-selectin levels among CAD patients on clopidogrel, despite varying CYP2C19 metabolizer phenotypes suggesting a potential role for P2RY12 polymorphisms in modulating thrombo-inflammation. Hence, genetic testing for P2RY12 polymorphisms could enhance therapeutic strategies in the management of CAD, potentially improving patient outcomes.

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Abstract ID: 163

Survival of the sickest: Gaucher disease as a potential clinical modifier of COVID-19

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Background/ Introduction: Gaucher Disease (GD) is a lysosomal storage disorder caused by GBA1 mutations, resulting in beta-glucosidase deficiency and the accumulation of glucosylceramide. Research suggests that GD-related humoral immunity and disruption of the lysosomal immune axis offers protection against COVID-19. (Keywords: Gaucher Disease, COVID-19)

Objectives: To examine the severity of COVID-19 in individuals with homozygous and heterozygous GBA1 gene mutations.

Materials and Methods: A retrospective study (2021–2023) collected data from 41 mutation-proven GD patients at CHG. COVID-19 details were obtained from their families, and the data for the general

population was sourced from WHO. A chi-squared test assessed COVID-19 severity in GBA1 mutation carriers compared to the general population.

Results:

Of 41 GD patients under 18 (24 males, 17 females), 32 had type 3 GD (L444P, RecNcil, D409H), 8 had type 1 (R535C, W432C, V414M, G377S, IVS9-3G>C, R496C, H404D), and 1 had PSAP mutation. Nine had undergone splenectomy.

7 homozygous GBA1 patients tested positive for COVID-19 by RT-PCR. 2 were asymptomatic, 4 had mild symptoms treated conservatively, and 1 required supplemental oxygen for 3 days. One GD1 and one GD3 patient had undergone splenectomy.

Among 9 heterozygous carriers (ages 30-50, 6 females) who tested positive for COVID-19, 8 had mild symptoms treated conservatively, and 1 was hospitalized for dyspnea for 1 week.

Conclusions: During the pandemic, studies worldwide examined GD's chronic inflammatory nature and predisposition to severe COVID-19. A 2020 U.S. study of 181 GD patients did not find an increased risk of severe COVID-19, with 1.65% of the cohort having GD3. Studies using animal and cell models have explored the potential clinical advantage of GD patients against viral and bacterial infections, including COVID-19. Cholesterol accumulation in late endosomal and lysosomal compartments inhibits viral escape. Similarly, glucosylceramide accumulation in GD patients offers better defense against SARS-CoV-2 and Mycobacterium tuberculosis.

Our cohort consisted of 78% GD3 cases. We examined the impact of COVID-19 on GBA1 mutation carriers (homozygous and heterozygous) and found that among the 16 COVID positive cases, 14 had mild and 2 had moderate infections. Comparing this to WHO population data, our findings suggest an overall milder infection in GBA1 mutation carriers (chi-squared = 12.77, p-value = 0.0052).

Although the sample size is small, this study suggests a positive trend in clinical outcomes, and these findings align with other studies.

Abstract ID: 164

Meta-Analysis of PTH and VDR Gene Associations in Pediatric Nephrotic Syndrome

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Background/ Introduction: Pediatric nephrotic syndrome (NS) is a chronic kidney disorder characterized by significant morbidity. The roles of PTH and VDR in the vitamin D signaling pathway are critical for kidney health and Recent studies shown that vitamin D receptor VDR signaling pathway not only in bone health and mineral metabolism but also in regulation of immune responses and kidney functions.While individual studies have explored these associations, a comprehensive synthesis is lacking.

Objectives: To systematically review and perform a meta-analysis on the association of PTH and VDR gene expression with pediatric NS.

Materials and Methods: A protocol was developed for this systematic review following the preferred Reporting item for Systematic Reviews and Meta analysis (PRISMA) guidelines Search strategy : A systematic search was conducted in PubMed, Embase, and Web of Science using keywords and MeSH terms related to PTH, VDR, and pediatric NS. Data extraction: Studies were included based on defined

criteria, and data were extracted on study design, population characteristics, and outcomes. Study selection: we included Gene expression analysis of PTH and VDR in pediatric nephrotic syndrome.

Statistical analysis: The pooled allele and genotype frequencies for the gene were used to calculate the odds ratio with 95% confidence interval to assess the strength of the association. Heterogeneity test and evaluation of publication bias for all the selected studies were performed using software Stats Direct. Quality assessment: Quality was assessed using the Newcastle-Ottawa Scale. Data synthesis: Data synthesis involves a random-effects model to account for variability among studies.

Results: A total of 15 studies were included in the meta-analysis. Significant associations were observed between altered expression of PTH and VDR and increased RESULT:
susceptibility to NS (pooled OR: 2.5; 95% CI: 1.8-3.3). Heterogeneity was moderate (I^2 = 45%), suggesting consistency across most studies

Conclusion: This meta-analysis supports a significant association between PTH and VDR expression and pediatric NS. These findings reinforce the potential of targeting the vitamin D pathway in NS treatment and highlight the need for further mechanistic studies.

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Abstract ID: 165

Comparative transcriptome of normal and cancer-associated fibroblasts

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Background/ Introduction: The characteristics of a tumor are largely determined by its interaction with the surrounding micro-environment (TME). TME consists of both cellular and non-cellular components. Cancer-associated fibroblasts (CAFs) are a major component of the TME. They are a source of many secreted factors that influence the survival and progression of tumors as well as their response to drugs. Identification of markers either overexpressed in CAFs or unique to CAFs would pave the way for novel therapeutic strategies that in combination with conventional chemotherapy are likely to have better patient outcome.

Objectives: To identify the differentially expressed genes between normal and cancer-associated fibroblasts

Materials and Methods: Fibroblasts have been derived from Benign Prostatic Hyperplasia (BPH) and prostate cancer. RNA from these has been used to perform a transcriptome analysis in order to get a comparative profile of normal and cancer-associated fibroblasts.

Results and Conclusions: This study identified differentially expressed markers between normal and cancer-associated fibroblasts that would help in targeted therapy against CAFs/derived factors, in combination with conventional therapy. However, this would in future need more experimental validation.

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Abstract ID: 166

Automated DNA Extraction from Complex Metagenomic Samples Using the MANTA System

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Background/ Introduction: Efficient DNA extraction is essential for uncovering microbial community insights, developing microbiome-based applications, and guiding clinical trials in metagenomics. Yet, traditional workflows often introduce biases at various stages, such as sample collection, storage, DNA extraction, and bioinformatics, potentially compromising data quality and accuracy. This study explores automation's potential in optimizing metagenomic workflows, particularly in minimizing human error, enhancing reproducibility, and increasing sample processing efficiency.

Automated extraction systems using paramagnetic particles with DNA-binding surfaces have become pivotal for streamlining processes, as they eliminate the need for manual handling and simplify extraction steps. In this study, we evaluated several DNA extraction kits compatible with Cambrian Bioworks' MANTA system, designed to optimize workflows in metagenomic laboratories. With robotic precision, the MANTA system reliably extracts DNA from complex environmental samples, including those with diverse microbial compositions like soil, stool, and water. Such samples are prone to inhibitory substances and may contain low microbial biomass, which complicates traditional extraction methods.

Objectives: The primary objective was to establish an automated, magnetic bead-based workflow for high-yield, high-integrity DNA extraction from metagenomic samples using the MANTA system.

Materials and Methods: The metagenomic sample underwent mechanical lysis with glass/steel beads, followed by enzymatic treatment, and the resulting lysates were processed using magnetic beads within an automated system, facilitating DNA binding, washing, and elution while minimizing contamination risks and improving reproducibility. DNA quality was assessed via fluorometric measurements to confirm integrity.

Results and Conclusions: The MANTA system's automation enables high-throughput, contamination-free DNA extraction from a variety of complex sample types, promoting reliable and reproducible metagenomic analysis. The system's magnetic bead-robotic approach delivers high-quality DNA, capturing both common and rare taxa within microbial communities, making it highly suitable for environmental safety and large-scale studies. Our assessment highlights that automated magnetic bead technology is cost-effective for processing large sample volumes compared to manual methods. Additionally, the use of environmentally safe magnetic beads enhances safety for operators and reduces environmental impact, reinforcing its utility in metagenomic research.

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Abstract ID: 167

Variants in SNRPN/SmB cause a variable neurodevelopmental disorder with developmental and behavioral abnormalities in Drosophila melanogaster

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Background/ Introduction: Small nuclear ribonucleoprotein polypeptide N (SNRPN, MIM *182279) is a paternally expressed imprinted gene located within the Prader-Willi critical region (PWCR), 15q11.2-q13. Imprinting defects in PWCR cause Prader-Willi syndrome (PWS). Single nucleotide variants (SNVs) in SNRPN have recently been reported in two families with Prader-Willi-like syndrome (PWLS).

Objectives: To determine the developmental and behavioral phenotypes of the identified variants in SNRPN using Drosophila melanogaster as a disease model

Materials and Methods: A detailed clinical evaluation followed by exome sequencing was performed in two individuals from two unrelated families. Allele-specific RT-PCR was performed to determine the parental origin of the mutated alleles. In vivo studies using Drosophila melanogaster were carried out to determine the developmental and behavioral phenotypes of the identified variants.

Results and Conclusions: Proband 1 (P1) presented with speech delay, intellectual disability (ID), seizures, and truncal obesity. Proband 2 (P2) presented with clinical features overlapping with PWS such as neonatal hypotonia, speech delay, motor delay, ID, truncal obesity, sleep disturbances, voracity, and autistic behavior. The heterozygous missense variants, c.260C>A p.(Pro87His) in SNRPN (NM_003097.6) was present in de novo state in P1 and c.280C>T p.(Arg94Trp) was inherited from the asymptomatic father in P2. Allele-specific RT-PCR from peripheral blood sample in P1 and P2 confirmed the paternal origin of the mutated allele, and maternal origin in the father of P2. The impact of these variants on neurodevelopment and behavior were further investigated using Drosophila melanogaster as the disease model. Knockdown of SmB, fly homologue of SNRPN, leads to impaired locomotion, early developmental defects, and larval lethality. Moreover, we show that the variants identified in the study, c.260C>A and c.280C>T are loss-of-function alleles using rescue experiments in Drosophila. Additionally, knockdown of SmB in mature neurons led to increased inter-male aggression, increased food intake in males, and increased daytime sleep in isolated females. We provide further evidence of SNVs in SNRPN as a cause of variable neurodevelopmental phenotype overlapping with PWS.

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Abstract ID: 168

miRNAs: The gene regulators in paediatric nephrotic syndrome

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Background/ Introduction: MicroRNAs (miRNAs) are small non-coding RNAs that play crucial roles in gene regulation and have been implicated in various diseases, including neoplasia. This study aims to delineate the correlation between the expression patterns of selected miRNAs and their targeted genes in urine samples (NS) versus healthy controls. Specifically, miR-155 was investigated in relation to INF2, miR-424 with ANLN, miR-186 and PODXL, miR-221 and COL4A5, and miR-100 with E2F3. Quantitative real-time PCR (qRT-PCR) was employed to measure the expression levels of the miRNAs

and their corresponding target genes. The data were analysed using a correlation coefficient to ascertain the degree of association between miRNA expression and target gene expression in both cohorts. Preliminary results indicate significant downregulation of miR-100 and miR-424 in NS samples, which corresponded with upregulation of their respective target genes, E2F3 and ANLN, suggesting a potential role of these miRNAs in tumorigenesis. Conversely, miR-155, miR-186, and miR-221 showed contrasting expression patterns, highlighting the complexity of miRNA-target interactions. These findings underscore the importance of miRNA signalling in NS processes and provide insights into potential biomarkers for diagnosis and therapeutic targets.

Objectives: To correlate the expression pattern of selected miRNA and the targeted genes in NS and healthy controls.

Materials and Methods: Bioinformatic Analysis for Target Prediction and Correlation Analysis

To elucidate the interactions between selected miRNAs and their target genes in neoplastic samples, a robust bioinformatic analysis was performed, encompassing miRNA target prediction and correlation analysis. Initially, target genes for the miRNAs of interest (miR-155, miR-424, miR-186, miR-221, and miR-100) were identified using established online databases such as TargetScan, miRanda, and DIANA-microT, which utilize sequence homology and evolutionary conservation to predict potential miRNA targets based on the binding sites within the 3' untranslated regions (UTRs) of mRNAs. Selected target genes with significant binding affinity were cross-referenced across these databases to confirm consistency and enhance reliability. Following target identification, expression data obtained from quantitative real-time PCR (qRT-PCR) were subjected to correlation analysis using statistical software R or GraphPad Prism. Pearson's correlation coefficient was employed to quantify the relationship between miRNA and target gene expression levels in both cases and healthy control groups. A correlation value (r) was calculated to determine the strength and direction of the relationship; positive correlations indicated that as miRNA levels decreased, target gene levels increased, and vice versa. Statistical significance was established with a p-value threshold of <0.05. This methodological approach not only highlights the regulatory networks underlying miRNA-target interactions but also enables the identification of potential biomarkers and therapeutic targets relevant to disease progression.

RESULT:

In the correlation analysis between the microRNAs and their targeted genes, we found that in the SSNS group, miR-155, miR-424, miR-186, miR-221, and miR-100 exhibited decreased expression relative to target genes such as INF2, ANLN, PODXL, COL4A5, and E2F3, while the expression of these target genes increased. In contrast, in the SRNS group, miR-155, miR-424, miR-186, and miR-100 showed elevated expression, while their target genes decreased. Notably, miR-221 was found to be decreased, but its target gene COL4A5 exhibited increased expression.

• SSNS Group: Decreased miRNAs: miR-155, miR-424, miR-186, miR-221, miR-100. Increased target gene expression: INF2, ANLN, PODXL, COL4A5, E2F3

• SRNS Group: Increased miRNAs: miR-155, miR-424, miR-186, miR-100. Decreased target gene expression. miR-221: Decreased; COL4A5: Increased expression.

Overall observation: Divergent expression patterns of specific miRNAs and their target genes in SSNS and SRNS groups suggest distinct regulatory mechanisms in nephrotic syndrome.

Results and Conclusions:

Discussion: Our correlation analysis examining the relationship between specific microRNAs (miRNAs) and their target genes provides valuable insights into the molecular mechanisms underlying nephrotic

syndrome (NS). The differential expression patterns observed in the SSNS and SRNS groups suggest that these two subtypes of nephrotic syndrome may be driven by distinct regulatory pathways.

In the SSNS group, the decreased levels of miR-155, miR-424, miR-186, miR-221, and miR-100 correlate with an increase in their respective target genes, including INF2, ANLN, PODXL, COL4A5, and E2F3. This inverse relationship may indicate a compensatory response to the dysregulated state of renal cells in SSNS, where the upregulation of these target genes could contribute to cellular proliferation, repair processes, or potentially maladaptive responses leading to podocyte injury and subsequent proteinuria. The increase in target gene expression despite lower miRNA levels may suggest potential miRNA sponging or the presence of additional regulatory factors influencing gene expression in an SSNS context.

Conversely, in the SRNS group, the elevated expression of miR-155, miR-424, miR-186, and miR-100 implies a different regulatory landscape. The rise in these miRNAs is coupled with a decrease in their corresponding target genes, indicating that these miRNAs may suppress gene expression as part of a pathogenic process contributing to more severe forms of nephrotic syndrome. This suggests that miRNAs could play a crucial role in modulating pathways associated with podocyte function and response to injury.

Interestingly, while miR-221 is decreased in SRNS, its target gene COL4A5 is upregulated. This atypical expression pattern may point to a more complex regulatory mechanism affecting COL4A5, possibly involving other factors or compensatory pathways that promote its expression despite the reduced presence of the miRNA. This observation warrants further investigation to elucidate the specific interactions and pathways involved.

CONCLUSION: The correlation analysis of microRNA and target gene expression in nephrotic syndrome revealed distinct and divergent patterns between the steroid-sensitive nephrotic syndrome (SSNS) and steroid-resistant nephrotic syndrome (SRNS) groups. In SSNS, the observed downregulation of specific miRNAs—miR-155, miR-424, miR-186, miR-221, and miR-100—was associated with an increase in their target genes, including INF2, ANLN, PODXL, COL4A5, and E2F3. This suggests a compensatory mechanism wherein decreased miRNA levels may lead to enhanced expression of genes that could contribute to podocyte injury and proteinuria.

In contrast, the SRNS group exhibited elevated levels of these miRNAs alongside decreased expression of their associated target genes, indicating a potential suppression of critical regulatory pathways required for maintaining podocyte health and functionality. Notably, the unique finding of decreased miR-221 with increased COL4A5 expression in the SRNS group points to a complex interplay of regulatory mechanisms that warrants further investigation.

Overall, these findings underscore the importance of understanding the distinct regulatory networks involved in SSNS and SRNS. Identifying such differences not only enhances our knowledge of nephrotic syndrome pathophysiology but also paves the way for the development of targeted therapeutic strategies that could modulate miRNA expressions and restore their proper function, ultimately improving clinical outcomes for patients affected by this condition.

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Abstract ID: 169

IL-2 Gene Polymorphism: A Key Contributor to Steroid Resistance in Nephrotic Syndrome

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Background/ Introduction: Nephrotic syndrome (NS) is a common kidney disorder in children, characterized by excessive protein loss in the urine, leading to complications such as hypoalbuminemia (albumin levels below 30 g/L), edema, and hypocholesterolemia. The condition is associated with proteinuria levels exceeding 40 mg per square meter per hour. NS presents in two main forms: Steroid-Sensitive Nephrotic Syndrome (SSNS) and Steroid-Resistant Nephrotic Syndrome (SRNS). Histologically, NS can manifest as focal segmental glomerulosclerosis (FSGS), diffuse mesangial proliferation, or minimal change disease (MCD). The annual prevalence of NS is estimated at approximately 2-7 per 100,000 children, with a higher prevalence of 16 per 100,000 children under 16 years old. The pathogenesis of NS remains unclear, but it is believed to result from a combination of immunological dysregulation, systemic circulating factors, and genetic predispositions. Hereditary abnormalities in podocyte structure may also contribute to the development of NS. The response of NS to steroid treatment plays a key role in distinguishing between SSNS and SRNS. Steroid resistance, particularly in SRNS, remains a major challenge in the management of the disease. Cytokines, such as interleukin-2 (IL-2), play a critical role in immune regulation and have been implicated in various kidney diseases, including NS. These cytokines are produced in response to genetic variations, or gene polymorphisms, which can influence an individual's susceptibility to disease and its progression. IL-2, a strong pro-inflammatory cytokine, is particularly important in the immune response and has been associated with several autoimmune disorders. The IL-2 gene is located on chromosome 4q26-q27, a region known to be linked with autoimmune diseases. The IL-2 gene encodes a cytokine that regulates immune cell proliferation, and its production can be influenced by genetic variations, particularly single nucleotide polymorphisms (SNPs). One such SNP, IL-2 (rs2069763), is located 114 base pairs from the initiation codon in the first exon of the gene. This variation has been suggested to contribute to the development of autoimmune diseases and may play a role in the pathogenesis of NS. This study hypothesizes that variations in cytokine genes, particularly the IL-2 gene, may influence the susceptibility and progression of NS. Specifically, the study investigates the association between IL-2 gene polymorphisms and NS, comparing 50 children with NS (25 with SSNS and 25 with SRNS) to 50 healthy controls. The analysis was conducted using polymerase chain reaction (PCR) and restriction fragment length polymorphism (RFLP) techniques to assess IL-2 gene variations and their potential link to steroid resistance in NS.

Objectives: 1. To examine the association of IL-2 (rs2069763) gene polymorphisms with susceptibility to pediatric NS, and 2. To evaluate the statistical significance of genotype-specific variations (TT vs GG and TT vs TG) in relation to disease progression.

Materials and Methods: The study included 50 pediatric NS patients and 50 healthy controls, with ethical approval granted by the Institutional Ethics Committee of Sri Ramachandra Institute of Higher Education and Research, Chennai (IEC - NI/22/JUL/83/71). Approximately 3 ml of blood was collected from each participant after obtaining informed consent from their parents. Genomic DNA was isolated from whole blood using the Qiagen DNA isolation kit. IL-2 (rs2069763) gene polymorphisms were identified through a Polymerase Chain Reaction-Restriction Fragment Length Polymorphism (PCR-RFLP) assay, using the MwoI enzyme specific to rs2069763, and analyzed on a 4% agarose gel electrophoresis. Allele and

genotype frequencies were calculated using MedCalc statistical software. The association between genotypes and clinical characteristics was assessed by calculating odds ratios (OR) with 95% confidence intervals (95% CI).

Results and Conclusions: In the study of IL-2 rs2069763, participants were classified into genotypic categories based on band patterns observed in a gel electrophoresis assay. Those with a heterozygous genotype (TG) displayed three bands (111 bp, 151 bp, and 262 bp), individuals with a homozygous mutant genotype (GG) exhibited two bands (111 bp and 151 bp), and those with the wild-type homozygous genotype (TT) showed a single band (262 bp). The distribution of genotypes among the samples is as follows: In the control group, there were 33 individuals with the TT genotype, 7 with the GG genotype, and 10 with the TG genotype, totaling 50 healthy individuals. In the SSNS group, 4 individuals had the TT genotype, 12 had the GG genotype, and 9 had the TG genotype, with a total of 25 individuals. Similarly, in the SRNS group, there was 1 individual with the TT genotype, 16 with the GG genotype, and 8 with the TG genotype, also totalling 25 individuals. For the analysis comparing homozygous wild-type (TT) individuals with those homozygous for the mutant allele (GG), the odds ratio was calculated as 26.40. The 95% confidence interval (CI) ranged from 7.5389 to 92.448, and the Z statistic was 5.119. The p-value for this comparison was $P < 0.0001$, which is statistically significant. In contrast, when comparing homozygous wild-type (TT) individuals to those with the heterozygous genotype (TG), the odds ratio was significantly higher at 10.56. The 95% CI for this comparison ranged from 3.0915 to 36.0708, with a Z statistic of 3.761 and a p-value of 0.0002. Since the p-value is less than 0.05, this result is considered statistically significant, suggesting a meaningful association between the TT and TG genotypes in this context. In conclusion, our research emphasizes the critical role of immune factors in the development of NS. Specifically, we observed a significant association between the IL-2 (rs2069763) polymorphism and the (TT vs. GG) and (TT vs. TG) genotypes, with statistically significant p-values. This suggests a potential link between this genetic variant and increased susceptibility to NS. Although this study identifies an association, further research with larger sample sizes is necessary to validate these findings and gain a more comprehensive understanding of the genetic factors contributing to NS. In summary, our study highlights a strong association between the IL-2 gene polymorphism and steroid resistance in patients with NS, suggesting its potential role in the development of steroid-resistant forms of the disease.

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Abstract ID: 170

Podocyte gene expression pattern in Childhood Nephrotic syndrome

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Background/ Introduction: Nephrotic syndrome (NS) is a pediatric glomerular disease marked by various clinical manifestations, including significant proteinuria (>40 mg/m²/h or a urine protein-to-creatinine ratio >2 g/g), hypoalbuminemia (<2.5 g/dL), and edema. This study seeks to evaluate specific gene biomarkers in urine samples from children diagnosed with steroid-sensitive nephrotic syndrome (SSNS) and steroid-resistant nephrotic syndrome (SRNS) (N=50) along with samples from healthy controls (N=50) using RT-PCR. The study's objectives are to (i) predict clinical steroid resistance, (ii) clarify the molecular mechanisms contributing to steroid resistance, and (iii) distinguish between SSNS and SRNS. The identification of gene biomarkers such as NPHS2 and LIN28 in the urine of NS patients could potentially improve prognostic evaluations, differentiate between SSNS and SRNS, and assist in developing non-invasive diagnostic panels for monitoring the disease condition.

Objectives: To analyze the expression patterns of candidate genes NPHS2, CD2AP, and LIN28 in patients with nephrotic syndrome (NS) versus healthy control groups.

Materials and Methods: RNA Extraction and cDNA Synthesis

RNA was extracted using TRIZOL to ensure high integrity for future analyses. The RNA concentration and purity were assessed via a NanoDrop spectrophotometer and agarose gel electrophoresis. One microgram of RNA was reversely transcribed into cDNA using a specific reverse transcription kit.

Gene Expression Quantification

Real-time PCR was employed to quantify the expression levels of three target genes with GAPDH serving as a control gene. The thermal cycling protocol consisted of initial denaturation followed by 40 amplification cycles, including a melt curve analysis for specificity confirmation.

Statistical Analysis

Data were analyzed using Student's t-test with SPSS 16.0, with a significance level set at $p < 0.05$. Receiver Operating Characteristic (ROC) curves were created to evaluate the sensitivity and specificity of the potential biomarkers. Subsequently, functional analyses, including STRING and Gene Ontology (GO) analysis, were conducted to explore the functional aspects of the identified genes.

Results and Conclusions: The study revealed that NPHS2 and LIN28 genes exhibited increased expression levels in patients with SRNS when compared to SSNS. To identify reliable biomarkers, integrating existing biological data from both healthy and affected individuals was employed. ROC curve analysis is established as a favored method for evaluating diagnostic biomarkers, focusing on:

• Diagnostic Sensitivity: The percentage of true positives among those with the condition.

• Diagnostic Specificity: The percentage of true negatives among healthy individuals.

Biomarkers are evaluated based on area under the curve (AUC) values: 0.7–0.8 indicates fair, 0.8–0.9 indicates good, and values over 0.9 demonstrate excellent differentiation. STRING analysis was conducted to examine the interactions between NPHS2 and LIN28, unveiling a protein-protein interaction (PPI) network comprising 22 nodes, approximately 15 of which are associated with SRNS. Gene Ontology (GO) enrichment analysis identified 629 significant terms ($p < 0.05$), including 494 related to biological processes, 88 to molecular functions, and 47 to cellular components. Notable findings included differentially expressed genes (DEGs) related to transcription regulation, pre-miRNA processing, and Fc-gamma receptor signaling, alongside RNA binding and sequence-specific DNA binding. KEGG pathway analysis highlighted pathways associated with cancer, bacterial and viral infections, and signaling pathways linked to B-cell receptor, PDGF, and nephrin interactions, illustrating gene regulation by POU5F1 (OCT4), SOX2, and NANOG, key factors in differentiation and proliferation.

Conclusion: In summary, this study demonstrates elevated expression levels of NPHS2 and Lin28 in patients with SRNS, suggesting their potential role as biomarkers. The findings from STRING analysis, coupled with GO enrichment and KEGG pathway evaluations, emphasize their involvement in crucial biological processes and signaling pathways related to SRNS. The gene biomarkers identified in urine samples of children with nephrotic syndrome may significantly enhance prognostic evaluations, facilitate the distinction between steroid-sensitive and steroid-resistant forms of the disease, and support the development of non-invasive diagnostic and monitoring tools for pediatric SRNS patients.

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Abstract ID: 171

miRNA Cluster Dysregulation in Colorectal Cancer: Mapping Uncharted Paths to Diagnostic Biomarkers

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Background/ Introduction: Colorectal cancer (CRC) is a most common gastrointestinal malignancy. The clusters of miRNAs-transcribed as polycistronic units have gained recognition as they regulate gene expression and play a critical role in cancer. Dysregulation of these miRNA clusters in CRC exhibits differential expression patterns and shows potential as diagnostic biomarkers.

Objectives: This study aims to investigate the dysregulation of miRNA clusters in CRC, focusing on the genomic locations and regulatory roles in the pathophysiology of CRC. Furthermore, this study examined the mechanistic basis of dysregulation of these miRNA clusters.

Materials and Methods: A systematic analysis was done using the PubMed, Scopus, and Web of Science databases. The studies were retrieved using terms like "miRNA clusters," "colorectal cancer," "dysregulated miRNAs," and "biomarkers." The peer-reviewed articles included from 2020-2024, focused on clinical studies with in vitro and in vivo models. The identified studies were then synthesized to assess the involvement of dysregulated miRNA clusters in CRC.

Results and Conclusions: Several miRNA clusters have been found to be differentially dysregulated in CRC through analysis. Specifically, the downregulated miRNA clusters included miR-137, miR-143/145, and miR-375; whereas those upregulated were miR-21, miR-31, miR-17-92 (miR-17, miR-18a, miR-19a, miR-20a, miR-19b, miR-92a), miR-23 (miR-23a, miR-23b, miR-27a), miR-181 (miR-181a, miR-181b, miR-181c, miR-181d) and miR-200 (miR-200a, miR-200b, miR-200c, miR-141, and miR-429). These dysregulated miRNA clusters are important regulators of critical pathways related to CRC, such as cell proliferation, invasion, and apoptosis. All these clusters have a good promise for their use as diagnostic markers in the early detection of CRC and as a prognostic marker to predict disease progression. The current review has given an overview that dysregulated miRNA clusters hold the possibility to serve as diagnostic and prognostic biomarkers in CRC. Further elucidation of the underlying regulatory mechanisms may drive such novel biomarkers toward clinical use in diagnosis and prognosis. Efforts should be made for future research to translate findings into clinical application by unraveling the complex regulatory networks of miRNA clusters toward personalized management of CRC.

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Abstract ID: 172

Comprehensive Assessment of APOL1 Genetic Variants in Chronic Kidney Disease, Focal Segmental Glomerulosclerosis of South India

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Background/ Introduction: Over the last decade, investigators have established the basic population genetics and epidemiology of APOL1. Understanding the biology of APOL1 risk variants has been advancing at the molecular level. APOL1 mediated kidney disease (AMKD) is a type of kidney disease caused by variants (changes) in the APOL1 (apolipoprotein L1) gene. APOL1-related kidney disease typically presents in adolescents to young adults, and individuals harbouring risk variants (RVs) are more likely to progress to kidney failure than are those with kidney disease who lack APOL-1 RVs. Certain APOL1 variants have been linked with different types of kidney disease, including a higher risk of high blood pressure-related chronic kidney disease (CKD) or kidney failure, focal segmental glomerulosclerosis (FSGS).

Objectives: To study the prevalence of APOL1 gene polymorphisms in patients of chronic kidney disease (CKD) or kidney failure, focal segmental glomerulosclerosis (FSGS) of South India.

Materials and Methods: A total of 115 chronic kidney disease (CKD) patients were recruited The APOL1 genotype of G0 (rs73885319; A/G), G1 (rs71785313; -del/TTATAA), and G2 (rs60910145; T/G) polymorphisms loci were determined in patients by restriction fragment length polymorphism (RFLP) assay then genotypes were confirmed by Sanger sequencing using 2% of samples. Three patients with FSGS were selected to perform the sequencing of the risk variant of G1/G2 in exon 7 (full length) and the remaining exons (1 - 6) were also sequenced.

Results and Conclusions: Our results showed no significant changes in G0 wild type pattern in all 115 CKD cases and none of them exhibited the risk variants of G1/G2 genotypes. We found the south Indian CKD patients not associated with APOL1 (G1/G2) polymorphism variant. These results were confirmed by Sangers sequencing for Exon 7 in FSGS patients (n=3), exhibited absence of G1/G2 pathogenic variants. Then we explored other APOL1 variants from exons 1 to 6, 3'UTR, and intronic variants. None of our patients showed any pathogenic variants in APOL1 gene. Hence this study confirmed that APOL1 genetic variants may not be associated with CKD ad FSGS patients of South India. The study warrants confirmation in larger sample size in South Indian CKD patients.

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Abstract ID: 174

A rare case report: Idiopathic arterial calcification and hypercholesterolemia in a South Indian young man

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Background/ Introduction: A young man (22 years old) has been facing difficulty from intermittent claudication for the last 7 years and is unable to walk more than 100m without leg pain (Leriche-Fontaine IIB) at admission. The aetiology of claudication was unknown.

Objectives: We aimed to identify the genetic variants in the coding region of genes that may lead to claudication and to find the root cause of these defective genetic variants from his familial background.

Materials and Methods: The genomic DNA was isolated from the patient, sibling, and parents' blood. DNA purity and integrity were verified before doing whole exome sequencing using Next Generation Sequencing and their results were verified by the Sanger Sequencing method.

Results and Conclusions: Exome sequencing identified a homozygous deletion mutation in exon 3 of NT5E gene (located on chromosome 6) with autosomal recessive inheritance and LDLR heterozygous genetic variant (located on chromosome 19) in exon 10 with autosomal dominant/recessive inheritance. These mutations were confirmed by Sangers sequencing. We also screened the patient's parents and sibling for the genetic variants and observed heterozygous deletion mutation in exon 3 of NT5E in parents, and LDLR heterozygous genetic variant in exon 10 was present in the father. Sibling had only homozygous deletion mutation in exon 3 of NT5E and showed developmental disorders like Klippel-Feil syndrome (Short neck) and Pectus carinatum (Chest wall deformities). In conclusion, LOF variants in exon 9 of NT5E have been previously reported to cause arterial calcification. Whereas, the LDLR LOF genetic variant was reported close to the observed variant in the LDLR gene causing familial hypercholesterolemia. The deletion mutation in exon 3 may belong to the LOF of NT5E (which codes CD73 enzyme) leading to CD73 deficiency (essential for the conversion of AMP to adenosine). This may affect the mineralization mechanism in surface vascular cells and cause arterial calcification in patients. In addition, familial hypercholesterolemia may further enhance the risk of arterial calcification and may lead to claudication in the patient.

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Abstract ID: 176

Systematic review of transcriptomics studies in bladder cancer reveals the prevalent expression of growth factor receptor genes.

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Background/ Introduction: Background: Bladder cancer has high prevalence worldwide, characterized by its complexity and heterogeneity. Understanding the molecular mechanisms underlying bladder cancer is critical for predicting patient recurrence and informing treatment strategies. Transcriptomics has emerged as a key area of research, but its role in bladder cancer, particularly in relation to recurrence monitoring, needs more research.

Objectives: Objectives: This study aims to conduct a systematic review of the correlation between transcriptomic alterations in bladder cancer, with a particular focus on gene signatures that could help monitor potential recurrence.

Materials and Methods: Methods: A PICO framework was used to guide the study objectives. A systematic search was conducted for articles that included transcriptomic data obtained through next-generation sequencing (NGS) in bladder cancer, specifically focusing on studies that performed differential gene expression analysis. Studies involving other experimental methods were excluded. The search was performed across multiple electronic databases (PubMed, ScienceDirect, etc.). PRISMA guidelines were followed to select high-quality studies for inclusion in the review. The analysis aimed to identify key genes and pathways associated with bladder cancer based only on transcriptome data.

Results and Conclusions: Results: Preliminary findings indicate significant correlations between specific transcriptomic profiles and changes, particularly in Non muscle invasive cancer type of bladder.Although

the evidence is still emerging, these findings show that FGFR3 and EGFR genes have interactions between one another.It was also found out that both the genes involved in MAPK pathway actively played a role in resistance mechanism to targeted therapy and paved way to agressive tumor development.

Conclusion:The investigation highlights the potential of FGFR3 co-expressed genes and EGFR ligand genes for better understanding of our bladder cancer biology and identify novel pathway targets. However, there is a necessity for further research for the roles of these genes in bladder cancer and their implications for clinical recurrence management.

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Abstract ID: 177

Contribution of rare sequence variants in cilia and hedgehog pathway genes in Down syndrome associated atrioventricular septal defects

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Background/ Introduction: Down syndrome (DS) is the most common genetic disorder (worldwide incidence estimated to be between 1 in 650 to 1 in 1000 live births), caused as a result of trisomy of chromosome 21. While cognitive impairment is present in all DS individuals, albeit with variable severity, other phenotypic features may show incomplete penetrance, most notable being congenital heart defects (CHD). Approximately 40% of DS individuals present with some form of CHD, the most frequent being atrioventricular septal defects (AVSD), comprising 43% of the CHD cases. Individuals with DS have complete AVSD, corresponding to a 2,000-fold increased risk compared to euploid poplulation. Therefore, the increased dosage of genes on chromosome 21 explains only part of the increased risk for CHD in DS and suggests that additional variants throughout the genome may play a role.

We hypothesize that the 2,000-fold increase in risk for developing an AVSD due to trisomy 21 unmasks additional genomic events, and that rare genetic variants (CNVs and sequence variants), incompletely penetrant on a euploid background, act synergistically with trisomy 21 to increase risk for AVSD. Since recent studies have suggested a strong role for cilia genes in the causation of AVSD, we investigated the role of ciliome/Hedgehog singling genes in the development of atrioventricular septal defect in children with Down syndrome.

Objectives: To analyse the contribution of sequence variants in the ciliome/hedgehog signalling genes in causation of AVSD in children with Down syndrome

Materials and Methods: In the present study we recruited a well phenotyped cohort of 100 children, 50 with Down syndrome and AVSD (DS +AVSD) and 50 Down syndrome children with structurally normal heart (DS +NH). All children enrolled were between the age groups 0-16 years. Individuals diagnosed with a full, free or translocation trisomy 21 were recruited. Only those children ascertained to have AVSD were assigned to DS +AVSD group. Subjects with mosaic and partial trisomy of chromosome 21 were excluded. To minimize phenotypic heterogeneity of cases, we focused on AVSD as the most severe heart phenotype seen associated with DS. Cases (DS + AVSD) included Down syndrome children with a complete, balanced AVSD with or without an additional CHD. Unbalanced AVSDs (those requiring a single ventricle repair) and partial AVSDs (inlet VSD only or primum ASD only) were also excluded. Control cohort included Down syndrome children with a structurally normal heart, patent foramen ovale, or patent ductus arteriosus (DS+NH).

Genomic DNA from all samples was enriched for the complete coding regions and splice site junctions using a custom bait- capture system. Paired End Sequencing was performed with 2x100/2x150 chemistry, on an Illumina platform. Reads were assembled and were aligned to reference sequences based on NCBI RefSeq transcripts and human genome build GRCh37/UCSC hg19. Appropriate softwares were used to align the raw data to hg19, to sort and mark the duplicated reads. Then, local realignment, base quality score recalibration, single nucleotide polymorphism calling, and short insertion/ deletion calling was performed. Variants were prioritised using appropriate softwares and tools. Variants in AVSD-associated ciliaome/hedgehog signalling genes were selected for further analysis. Genes were identified that are known to be associated with CHD forward genetic mouse screen, the PCGC, MGI, CHDGene, and the literature. Of these, cilia-genes were identified from known ciliopathy genes, the CiliaCarta database, the PCGC, the CPLANE network, and the literature. In total, 1307 genes were selected of which 150 genes were considered to be cilia-CHD related. Variants with gnomAD Allele frequency <0.001 were considered for further analysis.

Results and Conclusions: From the list of curated 150 cila-CHD related genes, 57 genes were found to be exclusively present only in the DS+AVSD group, FUZ, MYH10, PKD1L1, DNAH11 and DNAH5 being the top five. Gene set enrichment analysis to compare the effect sizes of the cilia-CHD gene set, suggests that individually, cilia-related genes have relatively small contributions to CHD. However, considering possible interactions between variants, where a variant in more than one gene is required to cause disease, gene set enrichment analysis specifically of the associated cilia genes shows that these are enriched for the GO biological process cilium assembly. There are 34 DS+AVSD children with known pathogenic or likely pathogenic variants in cilia-CHD genes, Our data shows that variants in ciliary genes contribute to a subtype-specific complex genetic model of CHD and that network analysis provides the biological context for these complex genetic underpinnings. We also identified variants in several cilia genes in the DS+AVSD group, which have previously not been reported in CHD. The results of the present study align with what has previously been shown regarding the association of ciliary genes with CHD and add information on new candidates, providing another group of genes to investigate for involvement in the complex genetics of CHD.

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Abstract ID: 178

Tamoxifen positively regulates the expression of cell adhesion and apoptotic genes in ER/PR-positive Breast Cancer

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Background/ Introduction: Breast cancer is a significant cause of mortality among women globally, with ER/PR-positive breast cancer being a significant subtype. Tamoxifen, a selective estrogen receptor modulator (SERM), is widely used in treating ER-positive breast cancer. Despite its efficacy, the detailed molecular mechanisms underlying tamoxifen's action, particularly on specific genes involved in cell adhesion and apoptosis, require further exploration. This study aimed to investigate the impact of tamoxifen on the expression of cell adhesion gene CDH1 (E-cadherin) and apoptotic gene FAS in MCF-7 breast cancer cell lines to understand its role in cancer cell proliferation and metastasis inhibition.

Objectives: To Evaluate the impact of tamoxifen on the expression of cell adhesion gene CDH1 (E-cadherin) and apoptotic gene FAS in MCF-7 breast cancer cell lines to understand its role in cancer cell proliferation and metastasis inhibition.

Materials and Methods: MCF-7 cell lines were cultured and treated with varying concentrations (10 µM, 20 µM, 30 µM) of tamoxifen for 24 and 48 hours. Cell morphology was monitored, and total RNA was extracted post-treatment. Gene expression analysis for CDH1 and FAS was conducted using quantitative PCR (qPCR). Statistical significance was assessed using a t-test; $p < 0.05$ was considered significant.

Results and Conclusions: Tamoxifen treatment significantly increased FAS gene expression at a 10 µM concentration after 48 hours, suggesting enhanced apoptotic signalling. Similarly, CDH1 expression was upregulated under the same conditions, indicating improved cell adhesion and reduced metastatic potential. No significant changes were observed at higher concentrations or shorter exposure times.

The study demonstrates that tamoxifen positively regulates key genes involved in apoptosis and cell adhesion, highlighting its potential to inhibit cancer cell growth and prevent metastasis. These findings suggest that optimal dosing and timing of tamoxifen administration could enhance therapeutic outcomes in ER/PR-positive breast cancer patients. Further mechanistic studies are needed to evaluate the condition.

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Abstract ID: 179

Bioinformatics Strategies for Decoding the Long-Read Sequencing for Genomic Imprinting Disorders

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Background/ Introduction: Genomic imprinting is a phenomenon where genes are expressed in a parent-of-origin-specific manner. Abnormalities in these genes lead to disorders of genomic imprinting that has clinical manifestation affecting growth, development, and metabolism. Genomic imprinting involves epigenetic modification like DNA methylation that regulate gene expression without altering the DNA sequence. Imprinting disorders often arise from deletion/duplication, uniparental disomy, imprinting center defects, or mutations in imprinted genes involving sequential testing thus complicating their diagnosis.

Objectives: This study aims to develop a comprehensive bioinformatics pipeline optimized for long-read sequencing data from the Oxford Nanopore PromethION P2 Solo platform. The primary objectives are to identify and characterize genetic variants, including SNPs, indels, and structural variants, profile DNA methylation patterns in imprinted regions in addition to relevant pharmacogenomic markers. The pipeline seeks to enhance the accuracy and efficiency of analyzing imprinted regions of human genome.

Materials and Methods: The pipeline was developed using a combination of advanced bioinformatics tools tailored for long-read sequencing data. Initial steps included base calling with Dorado for high-accuracy read generation, followed by quality control using FastQC. Reads were aligned to the human genome reference (hg38) using minimap2, with sorting and indexing performed in samtools. Variant calling involved PEPPER-Margin-DeepVariant for SNPs and indels and NanoSV for structural variants, while WhatsHap was used for phasing variants to assess parent-of-origin effects. Methylation calling was carried out using Megalodon to capture methylation status at CpG sites. CNVs were analyzed using CNVpytor, and homozygosity analysis was conducted using H3M2.Methylation and structural variant data were visualized using IGV for interpretation.

Results and Conclusions: The pipeline successfully identified a spectrum of genetic variants, including SNPs, indels, and structural variations, in imprinted regions of interest. Methylation analysis provided profiles that highlighted DMRs associated with known imprinting disorders. Phased variants revealed

patterns of parent-specific allelic expression, which is critical in understanding the inheritance and manifestation of these conditions.

Conclusion: This bioinformatics pipeline, designed for long-read sequencing data, provides a comprehensive approach to analyzing imprinting disorders by integrating genetic and epigenetic information. The pipeline's ability to phase variants, detect structural variations, and assess methylation patterns offers a robust framework for understanding complex imprinting mechanisms. The results highlight the potential for long-read sequencing to enhance diagnosis and personalized treatment strategies for patients with imprinting disorders. This can be also applied to conditions with genetic heterogeneity to accelerate diagnosis and overcome the need of utilising different techniques for molecular diagnosis.

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Abstract ID: 181

Reversing Glioblastoma's Epigenetic Defense: The Therapeutic Potential of NPTX2 in Tumor-Neuron Interactions

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Background/ Introduction: Background: Glioblastoma multiforme (GBM) is the most aggressive form of brain tumor with unfavorable treatment outcomes. The tumor complexity is enriched by interactions of the surrounding neuronal tissue, which creates a microenvironment that nurtures tumor development and resistance to therapies. Recent studies have identified synaptic-like communications between GBM and neurons that contribute to tumor growth and invasion. Neuronal pentraxin 2 (NPTX2) contributes to synaptic plasticity by modulating receptor trafficking and synaptic activity. Unlike the integral role of NPTX2 in neuronal function, involvement in glioma biology is unexplored.

Objectives: Objective: The study highlights the potential of NPTX2 as a novel target in GBM therapy, with a special emphasis on its role in tumor-neuron interactions and how GBM cells hijack these to promote the disease.

Materials and Methods: Methods: A systematic literature search was done between 2000 and 2024 through databases such as PubMed and Google Scholar. The literature search was performed using specific keywords like "NPTX2," "glioblastoma," "synaptic mimicry," "epigenetic regulation," and "microRNA." We investigated NPTX2 gene expression patterns and methylation signatures and their relation with other molecular markers in GBM disease progression, such as PTEN and NF-κB. We are currently performing bioinformatics analyses to validate NPTX2's therapeutic role in GBM, with findings anticipated to support targeted treatment strategies.

Results and Conclusions: Results: This review highlights that, due to promoter hypermethylation, NPTX2 is inactivated in GBM. There is evidence that re-expression of NPTX2, due to reversal of its methylation status, will result in the induction of apoptosis and inhibits cell proliferation, with possible restoration of sensitivity to chemotherapy. Importantly, microRNA targeting NPTX2, and its regulatory network PTEN and NF-κB, plays a critical role in regulating important GBM cell behaviors.

Conclusion: NPTX2 is a promising yet underexplored in GBM therapy, where its role in synaptic mimicry and epigenetic regulation can be targeted for therapeutic intervention in combination with microRNA-based strategies. Disrupting the interactions between the tumors and neurons would prevent GBM progression, invasiveness, and restore sensitivity. Therefore, NPTX2 can be a potential target for personalized therapy against GBM by reversing chemoresistance and improve outcomes.

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Abstract ID: 182

Implications of EXTL2-Driven CSPG Accumulation in CNS for Multiple Sclerosis Progression

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Background/ Introduction: Background: Multiple sclerosis (MS) is a chronic inflammatory central nervous system (CNS) disease-characterized by neurodegeneration, demyelination, and an inflammation. Glycosyltransferase exostosin-like 2, or EXTL2, is involved in the synthesis of Glycosaminoglycans (GAGs), that plays a role in proteoglycan formation. However, EXTL2 is not directly implicated in the pathology of MS, it regulates chondroitin sulfate proteoglycans (CSPGs), significant for construction of ECM components of the CNS that involves in neuronal repair mechanisms such as axonal regeneration and remyelination, thereby deteriorating the pathology of MS. Based on genetic variation near the EXTL2 locus that correlates with increased susceptibility to MS, a role for EXTL2 likely exists in regulating disease progression.

Objectives: This study investigates the role of EXTL2 in CSPG biosynthesis and its effects on MS pathology with the ultimate goal of finding regulatory pathways and molecular targets that may be used to inform therapeutic strategies to limit accumulation of CSPG, thereby supporting neuronal repair in MS.

Materials and Methods: Methods: The study highlights the expression profiles and regulatory networks associated with EXTL2 in the MS-affected CNS tissues. Pathway mapping and structural analysis would be conducted in order to find a basis of interaction that EXTL2 has in the pathway of CSPG biosynthesis. This provides an overview of the pharmacologic options to modify the levels of CSPG, including the inhibitors of CSPG accumulation and its applications in MS therapy approaches.

Results and Conclusions: Results: It is expected that the study will clarify the regulatory role of EXTL2 in CSPG biosynthesis and its pathological implications in MS. Specific pathways involved in the action of EXTL2 leading to CSPG accumulation are expected to be implicated, along with key interactions within those pathways. This indicates molecular targets that could be modulated in a downward direction to reduce CSPG levels, facilitating neuronal repair and provide a basis for treatment strategy in MS.

Conclusion: The pathway-focused approaches on the discovery of therapeutic targets in MS are pretty crucial. Through demonstration of regulatory mechanisms of EXTL2 in CSPG production can be unfolded, this study aims to point the way to therapeutic strategies hence halting the amassing of CSPG by evoking improvement in neuronal repair and recovery of the disease.

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Abstract ID: 183

Theranostic Role of microRNA in cardiovascular diseases

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Background/ Introduction: Background: Cardiovascular diseases, including myocardial infarction (MI) and heart failure (HF), present significant health challenges worldwide. Recently, microRNAs (miRNAs) have emerged as promising theranostic agents due to their ability to regulate gene expression in processes central to cardiac disease progression, such as apoptosis, fibrosis, and cellular remodeling. This review examines the potential of miR-1, miR-133, miR-208, miR-499, and miR-21, along with miR-132 and its inhibitor, CDR132L—the first drug of its kind to pass early clinical trials—are discussed as advancements in miRNA-targeted therapy.

Objectives: Objectives: The aim of this review is to evaluate the diagnostic and therapeutic implications of selected miRNAs in MI and HF. Additionally, it seeks to examine how miRNAs contribute to early

detection, prognosis, and treatment in these conditions, presenting current findings and future therapeutic possibilities.

Materials and Methods: Methods: A comprehensive literature search was conducted using databases including PubMed and Google Scholar. Keywords used were "miRNA," "myocardial infarction," "heart failure," "therapeutics," and "diagnostics." Inclusion criteria were studies addressing the role of miR-1, miR-133, miR-208, miR-499, miR-21, and miR-132 in MI or HF and published in peer-reviewed journals. Exclusion criteria eliminated studies not directly relevant to these miRNAs or those lacking clinical or experimental data.

Results and Conclusions: Results: The review identified miR-1 and miR-208 as highly specific diagnostic markers in acute MI, while miR-499 and miR-21 show potential in HF diagnosis and prognosis due to their involvement in myocardial remodeling and apoptosis. Therapeutically, miR-133 replacement therapy has demonstrated efficacy in improving cardiomyocyte survival and post-MI repair. The miR-132 inhibitor, CDR132L has demonstrated safety and potential efficacy in early clinical trials, indicating its ability to improve cardiac function in HF patients.

Conclusion: This study supports the theranostic capabilities of miRNAs in MI and HF, with a particular focus on CDR132L as a significant advancement in miRNA therapeutics. Ongoing research into the long-term safety and efficacy of these agents is essential for their integration into cardiovascular treatment protocols, advancing personalized medicine approaches in cardiology.

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Abstract ID: 184

Critical Role of Comprehensive Genetic Analysis in Risky Pregnancy: A Case Study

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Background/ Introduction: This case involves a non-consanguineous couple planning their second pregnancy following a first pregnancy complicated by severe intrauterine growth restriction (IUGR) and fetal distress. The neonate was clinically suspected of having Microcephalic Osteodysplastic Primordial Dwarfism type II (MOPD-II), presenting with overlapping phenotypic features of microcephaly, Seckel syndrome, and skeletal dysplasia complicating the diagnostic landscape. Comprehensive genetic analysis was instrumental in confirming the diagnosis and providing critical insights to guide informed decision-making for the subsequent pregnancy.

Objectives: The aim was to identify the genetic condition and its underlying cause based on the proband's clinical phenotype, highlighting the role of advanced genetic analysis in assessing complex pregnancy risks and supporting informed clinical decision-making in subsequent pregnancies.

Materials and Methods: A series of genetic analyses were performed, including: • Whole exome sequencing (WES) for the proband to identify potential genetic causes. •Carrier screening for both parents via clinical exome sequencing to detect heterozygous pathogenic or VOUS variants. •Spinal muscular atrophy (SMA) screening in parents to rule out additional risks. •Prenatal genetic testing in the second pregnancy through chorionic villous sampling (CVS), targeting PCNT and HSPG2 genes.

Results and Conclusions: WES of the proband identified a homozygous likely pathogenic variant (c.9535dup) in the PCNT gene, consistent with a diagnosis of (MOPD-II). Both parents were confirmed as heterozygous carriers of this PCNT variant. Additionally, the mother was found to carry a variant of

uncertain significance (VOUS) in the HSPG2 gene, while the father carried a polymorphic variant in the same gene.

Prenatal testing for the second pregnancy revealed that the fetus was a carrier of the PCNT variant and compound heterozygous for the HSPG2 variants inherited from the parents. Subsequently, the VOUS variant in HSPG2 was reclassified as benign as per ACMG guidelines, significantly reducing the genetic risk associated with this pregnancy. Furthermore, Ultrasound scans throughout gestation indicated normal fetal development. The integration of genetic testing data along with ultrasound findings confirmed a low genetic risk for the fetus. This case highlights the indispensable role of genetic analysis in refining the risk assessment for complex pregnancies. The precise identification of pathogenic variants, VOUS, and polymorphisms provided crucial insights into the genetic risk profile of the fetus. By leveraging comprehensive genetic data, the clinical team was able to make evidence-based recommendations, resulting in a successful pregnancy outcome. This case underscores the importance of advanced genetic technologies in managing high-risk pregnancies with precision and accuracy.

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Abstract ID: 185

Investigating the link between therapeutic efficacy of lithium and mitochondrial parameters

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Background/ Introduction: Bipolar disorder (BD), marked by severe mood swings, poses significant challenges to affected individuals worldwide. Although lithium is the primary treatment for BD, its effectiveness varies among individuals with some responding well and others not. However, the molecular mechanisms behind its clinical efficacy remain poorly understood.

Moreover, previous studies suggest lithium's potential modulation of mitochondrial function through unknown molecular pathways. Understanding the molecular basis for these effects holds promise for unraveling its therapeutic effects in bipolar disorder and may provide novel insights into the development of more targeted treatment strategies. Thus, integrating mitochondrial studies and investigating the molecular mechanisms of lithium represents a promising approach to advancing our comprehension and treatment of bipolar disorder.

Objectives: Our objective here is to explore the effects of lithium exposure on mitochondrial health in neural lineage cells.

Materials and Methods: Neural precursors (NPCs) and neurons were subjected to varying lithium concentrations (0.5mM to 5mM) in vitro, and subsequent effects on mitochondrial membrane potential and oxygen consumption rate were assessed using flow cytometry and seahorse flux analyzer respectively.

Results and Conclusions: Exposure to lithium concentrations ranging from 0.5mM to 5mM over a 5-day period led to a concentration-dependent decrease in neural precursor cell viability only at concentrations above 2.5mM. Recognizing the interconnectedness of cell viability and mitochondrial function, we investigated the impact of lithium on mitochondrial membrane potential (MMP) and oxygen consumption rate (OCR) across varying concentrations, including clinically toxic (2.5-5mM) and non-toxic (0.5-1.25mM) levels. Notably, lower, non-toxic lithium concentrations did not change MMP & OCR while the

toxic concentrations showed marked decrease in MMP & OCR in control NPCs suggesting a potential link between lithium-induced alterations in mitochondrial bioenergetics and its influence on cell viability.

Our study demonstrates the concentration-dependent effects of lithium on neural precursor cell viability and mitochondrial health. Further, ongoing study involves exploring lithium's effects on stressed/ Bipolar derived patient cells.

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Abstract ID: 186

Spinocerebellar Ataxia-27B caused by repeat expansion in FGF14 gene in the Indian population

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Background/ Introduction: Spinocerebellar ataxias (SCAs) are a heterogeneous group of progressive neurological disorders characterized by impaired coordination of limb, eye movements and dysarthria. They could be caused by pathogenic expansions of short tandem repeats, or deleterious point mutations. Ataxia due to an intronic GAA repeat expansion in FGF14 (SCA 27B) has recently been identified. This is a late onset autosomal dominant neurodegenerative disorder characterized by the onset of gait and appendicular ataxia in adulthood, usually around the age of 55.

Objectives: SCA 27B due to an intronic GAA repeat expansion in fibroblast growth factor 14 (FGF14) seems to occur in many populations. To screen Indian families with movement disorders for pathogenic GAA repeat expansions in the FGF14 gene.

Materials and Methods: 526 ataxia patients who are genetically tested negative for SCA1,2,3 and 12 were selected and underwent genotyping for the allelic distribution of the repeat locus by means of long-range PCR and capillary electrophoresis along with 130 healthy controls. Samples with GAA≥250 in the gene locus were then validated with repeat expansion PCR and Sanger Sequencing to confirm the pathogenic repeat expansion in FGF14 gene. The clinical evaluation of SCA27B-positive patients was documented.

Results and Conclusions: In the healthy controls, 82% chromosomes (213 of 260) carried (GAA)≤25 and the 8 repeats/allele were the most frequent. (GAA)191 repeats were the highest recorded expansion seen in a single individual. In the patient group, 17 patients (3.2%) were suspected to have a pathogenic expansion of GAA≥250 based on electrophoresis. Further, the Sanger sequencing of the control and patient alleles validated the presence of GAA repeat in the FGF14 gene along with 3` TP-PCR. Based on the peak distribution from capillary electrophoresis, patients with GAA repeats are classified into normal (GAA≤100), intermediate (GAA100-249) and pathogenic (GAA≥250). The positive patients were from diverse locations within India. Tremor was a presenting symptom in 5 patients, and gait ataxia was observed in many patients. Slow saccades were also frequently seen, but a downbeat nystagmus was not universal. In all the patients with more than 100 GAA repeats found to show multiple peaks pattern in the expanded allele suggesting multiple repeat sizes in an individual. Approximately 3% of tested ataxia patients had a pathogenic GAA expansion, with an average age of 45 years. This finding supports including SCA27B in routine genetic screening for ataxia in India, given its widespread occurrence. Small

molecule drugs like 4-aminopyridine may offer therapeutic benefits, although further research is needed. We also found intermediate alleles in 3.6% of cases. Investigating the genetic and molecular aspects of this mutation could deepen our understanding of unstable repeats and their link to neuropsychiatric disorders.

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Abstract ID: 187

Prevalence and patterns of chromosomal aneuploidy in a tertiary care center for Neurodevelopmental disorders.

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Background/ Introduction: Neurodevelopmental disorders, which include intellectual disabilities, developmental delays, communication disorders, autism spectrum disorders, ADHD, learning disabilities, and motor disorders have significant genetic contributions such as single gene defects/copy number variations and chromosomal abnormalities. Detecting neurodevelopmental disorders caused by chromosomal abnormalities is crucial for healthcare providers to modify treatment and intervention strategies. Early diagnosis improves quality of life and developmental outcomes by enabling prompt medical, educational, and therapeutic measures. Also, Genetic diagnosis can provide a new criterion for the classification of Neurodevelopmental disorders. Understanding of the particular chromosomal defect can also help in genetic counseling which minimizes the probability of recurrence of this defect in subsequent pregnancies.

Objectives: • To determine the incidence rate of aneuploidy in the patient population. • To identify the most common type of aneuploidy diagnosed in the tertiary care center. •To examine the clinical characteristics associated with different types of aneuploidies. •To evaluate the accuracy of karyotyping test in identifying genetic abnormalities.

Materials and Methods: Peripheral blood was collected and Karyotyping was done on chromosome preparations from peripheral blood lymphocytes. Clinical characteristics and family history details were also evaluated

Results and Conclusions: According to this study, 17.1% of the analyzed samples had chromosomal aneuploidies. The most frequent aneuploidy found is Down syndrome, which accounts for 11.4% of all samples and 66.7% of aneuploid cases. This aligns with statistics from the general population, which shows that the most common chromosomal defect is Down syndrome. Additionally, Klinefelter and Turner syndromes were noted. The identification and understanding of aneuploidy trends in a tertiary care center through karyotyping can provide valuable insights for further research and potential interventions to improve patient outcomes. Our study demonstrates that the karyotyping approach is a reliable, efficient and cost-effective way to identify chromosomal abnormalities. These findings support karyotyping's role as a fundamental diagnostic method and give doctors insightful information about patients' genetic state and can also provide implications for further molecular biology and genetic research aimed at elucidating the pathogenesis of neurodevelopmental disorders.

Abstract ID: 188

Fusion-Gene Detection Techniques: A Strategic Double-Win Assessment with Open-Source Ph+ ALL and Ph-like ALL RNASeq cohort

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Background/ Introduction: Fusion transcripts, resulting from genetic alterations or RNA splicing errors, are valuable biomarkers for cancer diagnosis, subtyping, and treatment. RNA-sequencing (RNA-seq) is the gold standard for identifying these aberrant transcripts. While numerous algorithms exist for detecting fusion transcripts from paired-end RNA-seq data, their performance remains underexplored, especially in the context of specific cancer types. This study leverages publicly available RNA-seq datasets of Ph+ and Ph-like ALL to evaluate the performance of selected fusion detection tools. By focusing on the BCR-ABL1 fusion, a key differentiator between these two subtypes, we aim to identify a highly accurate and efficient tool. This approach not only provides a benchmark for fusion detection tools but also offers a potential method for distinguishing Ph+ and Ph-like ALL.

Objectives: Tool Selection: Identify suitable fusion detection tools based on existing benchmark studies.
Data Acquisition: Retrieve publicly available RNA-seq datasets of Ph+ and Ph-like ALL.
Tool Evaluation: Apply the selected tools to the datasets to assess their performance.

Materials and Methods: A comprehensive literature review was conducted to identify suitable fusion detection tools. Tools with suboptimal performance in previous benchmarking studies were excluded. Subsequently, RNA-seq datasets of Ph+ and Ph-like ALL were retrieved from the ENA repository. The selected tools were then applied to these datasets to assess their performance and computational efficiency.

Results and Conclusions: Based on benchmarking studies, STAR-Fusion, Arriba, FusionCatcher and Cicero were identified as potential fusion detection tools. However, due to their significantly longer processing times on a test dataset, FusionCatcher and Cicero were excluded from further analysis. A total of 91 Ph+ ALL and 49 Ph-like ALL samples were retrieved from the ENA repository. While both STAR-Fusion and Arriba produced accurate fusion detection results, Arriba exhibited significantly faster processing times. Our study concludes that Arriba is a highly efficient and reliable tool for detecting fusion transcripts, particularly in the context of distinguishing between Ph+ and Ph-like ALL.

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Abstract ID: 191

Genetic Counseling and Ethical Considerations in PGT-M for Adult-Onset Hereditary Cancer Syndromes: A Case Report on PALB2 Mutation Prevention

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Background/ Introduction: Carrier screening programs can be an effective tool to eliminate the risk of inheritable genetic conditions. Preimplantation Genetic Testing for Monogenic Disorders (PGT-M) allows couples who are known carriers of genetic mutations to reduce the risk of passing on heritable conditions, including adult-onset cancers. This case report describes a non-consanguineous couple who chose carrier screening before their assisted reproduction cycle. Mutations in the PALB2 gene

significantly increase susceptibility to breast and ovarian cancers, presenting unique challenges when making reproductive decisions.

While PGT-M enables the selection of embryos without specific mutations, it raises complex ethical considerations, particularly in cases involving adult-onset conditions. These include the implications for reproductive autonomy, and the moral dilemmas surrounding the selection of embryos based on future health risks. Additionally, psychological and social factors, such as parental guilt, anxiety over reproductive choices, and societal perceptions of genetic testing, must be carefully considered. This report presents a case where genetic counseling and ethical discussions were central to guiding a couple's decision to undergo PGT-M to prevent PALB2 mutation transmission.

Objectives: This case report evaluates the use of PGT-M to prevent PALB2 mutation transmission and discusses the genetic counseling and ethical considerations relevant to adult-onset hereditary cancer predispositions.

Materials and Methods: A 37-year-old couple with a history of reproductive complications, including a previous pregnancy affected by severe congenital anomalies, sought genetic counseling. Carrier screening was advised to rule out recurrence risk in future pregnancies. The male partner was incidentally identified as a carrier of the PALB2 c.2411_2412delCT mutation. Following a thorough discussion of the potential cancer risks, genetic counseling provided insights into the implications of PGT-M and the ethics of embryo selection. The couple underwent IVF, with PGT-M performed on three embryos to detect the PALB2 mutation, followed by aneuploidy screening to increase implantation success.

Results and Conclusions: Of the three biopsied embryos, two were free of the PALB2 mutation and suitable for implantation. The couple, supported by comprehensive counseling, made an informed decision to proceed with implantation. The counseling process considered psychosocial impacts, ethical concerns, and medical benefits, emphasizing a tailored approach.

PGT-M provides couples with actionable options to prevent hereditary cancer risk but requires careful genetic counseling to address ethical issues surrounding adult-onset disease prevention. These issues include the balance between reproductive autonomy and societal implications, the morality of selecting embryos based on future risks, and the emotional impact on families. For families with known genetic cancer predispositions, genetic counseling, and ethical guidance are crucial in PGT-M decision-making. This case report emphasizes the importance of a multidisciplinary approach, involving reproductive medicine and ethics, to address these challenges holistically and optimize patient care.

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Abstract ID: 192

Shared Genetics of Blood Pressure, Smoking and Neurological Disorders in Cross-Ancestry, European and South Asian Populations

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Background/ Introduction: Neurological disorders (ND) such as stroke, dementia, and multiple sclerosis are associated with increasing global morbidity and mortality burden. Both genetic and vascular risk factors (VRF) such as blood pressure traits and smoking are associated with increased risk of ND. However, the shared genetic etiology between these VRFs and NDs is unclear and has been explored less.

Objectives: Investigation of shared genetics between blood pressure traits, smoking and neurological disorders.

Materials and Methods: We analyzed genome-wide shared genetic architecture between blood pressure traits (systolic blood pressure (SBP), diastolic blood pressure (DBP), pulse pressure (PP)), smoking, cerebrovascular diseases (n=10) and neurodegenerative diseases (n=6) in different population groups. Genome-wide association (GWAS) analysis was performed by using PLINK-2 and REGEINE. METAL is implemented for genome-wide meta-analysis for these phenotypes. We used the cross-trait linkage disequilibrium score regression method to estimate genome-wide genetic correlation. PLACO and COLOC methods are used to investigate genome-wide pleiotropy and shared causal genetic loci respectively. Gene-set enrichment analysis is performed to investigate shared genes and biological pathways. The association of genetic risk scores of blood pressure traits with ND was investigated in South Asians from UKB and Indian datasets.

Results and Conclusions: We detected ATXN2, ICA1L, CDK6, HTRA1 and NBEAL1 as highly pleiotropic signals between VRF (SBP, Smoking) and ND. We also detected shared genetic factors which are present in specific phenotypic pairs. Genetic risk scores of blood pressure traits derived from large-scale multi-ethnic studies show an association with ND (SBP-GRS & Ischemic Stroke: beta=0.0057, p=0.0098) in South Asian ancestry samples including Indians.

This is the first study, that shows that genetic factors linked to blood pressure traits are also associated with smoking addiction and these shared genetic factors are also involved in ND of both the central and peripheral nervous system. Understanding shared genetic factors between VRF and ND provides deeper insight into shared biological mechanisms between complex phenotypes and holds potential for novel drug target discovery.

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Abstract ID: 193

Effect of Genetic Polymorphisms on treatment outcomes in Lupus Nephritis patients

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Background/ Introduction: Lupus Nephritis (LN) is a severe renal complication of systemic lupus erythematosus (SLE), often requiring immunosuppressive treatment with cyclophosphamide or mycophenolate mofetil (MMF). However, responses vary, with some patients experiencing significant efficacy and tolerable side effects, while others face limited benefits and severe adverse reactions. Emerging evidence suggests that certain genetic markers (CYP2C19 gene, CYP2B6 gene, etc) may predict better responses and reduced toxicity to these drugs, indicating a potential for genetic-guided therapy in LN.

Objectives: To evaluate the relationship between specific genetic markers and treatment outcomes, including efficacy and adverse effect profiles, in LN patients undergoing cyclophosphamide or MMF therapy.

Materials and Methods: The study will recruit 80 SLE patients with LN and 80 SLE patients without LN initiating treatment with cyclophosphamide or MMF. Genotyping will be conducted using PCR-RFLP, Sanger sequencing or whole exome sequencing to identify markers linked to altered drug response. Patients will be monitored to assess treatment efficacy (complete/partial remission), safety (frequency and

severity of adverse effects), quality of life, renal function and cognitive function. Data analysis will focus on assessing associations between genetic polymorphisms and clinical outcomes.

Results and Conclusions: The study aims to elucidate the role of genetic markers as predictive biomarkers for LN treatment response, which could revolutionize LN management by enabling personalized, genetic-guided therapy. Findings of the study will help in optimizing drug efficacy and minimizing toxicity, which could lead to enhanced patient outcomes and provide a foundation for pharmacogenomics in autoimmune disease treatment.

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Abstract ID: 195

Uncovering Novel NOBOX Variants: Genetic basis of Premature Ovarian Insufficiency in three Indian Families.

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Background/ Introduction: Menstrual irregularities and hypergonadotropic hypogonadism are associated with premature ovarian insufficiency (POI), a clinical condition with a variety of symptoms. A significant contributor to female infertility, POI affects up to 0.01% of women before the age of 20 and 1% of women before the age of 40. Most of the time, the cause of POI is unknown, while it may be iatrogenic, auto-immune, or hereditary. Idiopathic cases must be investigated for a genetic cause, especially when there is a family history of POI. Growing evidence points to a genetic basis being the primary cause of POF in most cases. NOBOX gene mutations have been implicated in POI, but genetic studies in Indian population are limited.

Objectives: Investigate the genetic basis of premature ovarian insufficiency in three Indian families.

Materials and Methods: The families were recruited three cases with primary amenorrhea with absent ovaries after taking informed consent. Exome sequencing was performed and analysed using in-house pipeline based on GATK. The variants were classified as per ACMG guidelines. Sanger validation and segregation analysis was performed for identified variant in the family. qPCR was performed in family with loss of function homozygous NOBOX variant.

Results and Conclusions: Exome sequencing revealed two homozygous loss of function variants and one heterozygous missenses variant in NOBOX gene. NOBOX:c.1322delG is a reported variant in heterozygous state, however our patient had the variant in homozygous state. We have also identified an novel homozygous NOBOX:c.535delC. Function analysis of RNA by qPCR from the family indicated compensated amount of normal NOBOX RNA in asymptomatic mother and severely reduced quantity in Proband and affected sibling. We have also identified novel missense variant NOBOX:c.48G>C, where in-silico characterization is ongoing.

NOBOX, a homeobox gene that encodes a transcription factor that is only expressed by granulosa and oocyte cells in the ovary. The NOBOX protein contains a homeodomain regulate the expression of a series of genes playing key roles in the process of folliculogenesis. NOBOX variants have been identified at the heterozygous state in various cohorts. Population database gnomAD have many LOF variants of NOBOX

in population in low frequency but no homozygous variants. Our observation also suggests that haploinsufficiency caused by LOF variants of NOBOX in could be tolerated in asymptomatic patients as reported in mice models. Our findings improves the mutational spectrum of NOBOX and expand its contribution to the development of POI.

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Abstract ID: 196

Gene expression profile of B-cell subtypes in Granulomatosis with Polyangiitis

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Background/ Introduction: Granulomatosis with polyangiitis is a rare autoimmune systemic small-vessel vasculitis which is characterized by necrotizing granulomatous lesions and vasculitis of majorly the respiratory tracts and kidney. The aetiology of this disease is ambiguous but there is sufficient evidence for the involvement of environmental triggers and genetic susceptibility. B cells have a prominent role in GPA pathogenesis since they produce ANCAs and inflammatory cytokines, although very few studies are present that fully explain their role in GPA pathogenesis.

Objectives: This study aims to elucidate the contribution of memory B cells, naïve B cells, plasmablasts, and transitional stage B cells in GPA pathogenesis by identifying subtype-specific gene expression profiles and leveraging network analysis to highlight critical pathways

Materials and Methods: Transcriptomic data from 7 GPA patients and 4 healthy controls from a North Indian cohort, along with B-cell specific single-cell RNA sequencing data (Project PRJEB27270, ENA), were analyzed. Gene expression profiling was conducted to identify DEGs with thresholds set at $p \leq 0.05$ and log2FC > 1.2 or < -1.2. Enrichment and network analyses were performed to elucidate pathways and protein interactions associated with B-cell subtypes.

Results and Conclusions: A total of 9 significant DEGs were identified, namely RPL13AP5, RPL17P6, MTND1P23, METTL7A, CTSS, LMNTD2, OR2T34, MTCO3P12, POLR2L, with RPL13AP5 observed as a common marker across memory B cells, naïve B cells, transitional stage B cells, and plasmablasts. Enriched pathways included Innate immunity, Neutrophil degranulation, Antigen Processing and Presentation and Basement membrane organization. Protein interaction analysis highlighted networks related to immune regulation and cellular adhesion, indicating potential roles in GPA pathogenesis.

These 9 differentially expressed genes were enriched in pathways involved in innate immunity and neutrophil degranulation, which are critical in GPA pathogenesis. Due to its involvement in several pathways that were previously linked to GPA and other autoimmune illnesses, CTSS stood out among these. Given its function in antigen processing, CTSS may promote the creation of more ANCAs and the development of granulomas. Its potential as a cross-disease therapeutic target is further highlighted by its enrichment in thyroid-related pathways. The 9 novel differentially expressed genes can prove to be effective therapeutic and diagnostic biomarkers for GPA.

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Abstract ID: 197

Mutation spectrum of ALMS1 gene in Alstrom syndrome

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Background/ Introduction: Introduction: Alstrom syndrome (ALMS) (OMIM # 203800) is a rare autosomal recessive ciliopathy that affects multiple organ systems and is caused by biallelic ALMS1 gene mutation. It is primarily characterised by retinal degeneration, sensorineural hearing loss, cardiomyopathy, obesity, and various endocrine complications; like Type 2 diabetes mellitus, hyperlipidaemia, hyperinsulinemia. Additionally, renal, hepatic and pulmonary complications, and fibrosis in multiple organs are reported. While developmental delay is common, cognitive function is typically normal. Vascular like lesions and while matter atrophy in the brain have been recently reported. ALMS is progressive, but the onset of symptoms, phenotype spectrum, and severity, can show inter and intra familial variability. There are a few clinical and genetic case reports from India.

Objectives: Objectives: The aim of our study was to understand the genotype spectrum of ALMS1 mutations in patients from our database (2016-2023) and analyse the phenotype spectrum and variability.

Materials and Methods: Materials and Methods: A cohort received for genetic diagnostic testing along with clinical details and informed consent were subjected to clinical exome or whole exome sequencing. The data generated was analysed using in-house bioinformatics pipeline to identify the disease causative variant.

Results and Conclusions: Results and Conclusions: A total of 75 cases with biallelic variants in ALMS1 gene was reported. Out of which 47 were syndromic (24 pathogenic (P) (51%), 8 likely pathogenic (LP) (17%) and 15 (31%) variants of uncertain significance (VUS)), while 28 non-syndromic cases had 6 P (21%), 4 LP (14%) and 18 VUS (64%) variants. However, 21/30 non-syndromic cases were <10 years and hence other phenotypes might present later. Thirteen cases had single heterozygous variant in the ALMS1 gene (1 P, 12 VUS), 10 were syndromic, and 3 were non-syndromic cases under 10 years of age. These cases could have deep intronic or structural variants not detected by this assay. The main presenting clinical indications in this cohort was cardiac, retinal, renal diseases and obesity. A detailed analysis of the phenotype spectrum and variants reported in this cohort could reveal any genotype-phenotype correlation.

Acknowledgement: We acknowledge all the clinicians who have referred the cases for genetic testing.

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Abstract ID: 198

Insights into Parkinson's Disease-specific α-Synuclein's nuclear physiological role

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Background/ Introduction: α-Synuclein (αSyn) plays a central role in the development of Parkinson's disease (PD) and other synucleinopathies. Despite several decades of research into its cellular roles, its exact nuclear physiological function remains undefined. Interestingly, higher levels of nuclear αSyn are observed under pathological conditions; however, only the αSyn aggregating propensity is widely studied, leaving a significant gap in our understanding of the pathophysiology of nuclear αSyn.

Objectives: In our previous work, we studied the interactions of αSyn with dsDNA and individual histones, where we demonstrated that αSyn-histone interactions are strong and highly specific. This led to our hypothesis that αSyn nuclear role is driven by αSyn-histone interactions. We, therefore, aim to uncover the nuclear physiological role of αSyn and its function in chromatin regulation.

Materials and Methods: We integrated biochemical, biophysical, structural (X-ray crystallography), and cellular approaches in the current study.

Results and Conclusions: We provide the first structural and molecular-level insights into the physiological function of nuclear αSyn, shedding light on its role in chromatin regulation. Through extensive biochemical and biophysical experiments, we showed the interactions of αSyn with the assembled H2a-H2b dimer and (H3-H4)2 tetramer. We further validated our findings with structural and cellular data.

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Abstract ID: 199

Antisense Oligonucleotides Mediated Rescue of ATM Mis-splicing

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Background/ Introduction: Ataxia telangiectasia (A-T) is a progressive neurodegenerative disorder resulting from pathogenic variations in Ataxia telangiectasia mutated (ATM) gene, which encodes a nuclear protein kinase with a critical role in double-strand DNA damage response. Disease-causing variations in ATM are spread across all exons and, result in little or no protein. Variations affecting splicing constitute about a third of all ATM mutations and have a variable effect on protein expression. These mutations are likely candidates for splice-switching antisense oligonucleotides (ASO) therapeutics. Using in silico, ex vivo, and in vitro approaches for functional classification, four ATM variants from Indian A-T patients were selected in the present study, to rescue mis-splicing using (ASO). We designed and developed a set of ASOs that successfully rescued mis-splicing in patient-derived cell lines harbouring the selected ATM variants, resulting in increased levels of correctly spliced ATM transcript. The selected ASOs were further validated to assess their potential to rescue ATM cellular function in addition to splicing.

Objectives: 1. To identify a subset of pathogenic ATM variants that lead to a type of mis-splicing that might be rescued by an ASO.

2.To develop and validate selected ASO's ability to rescue ATM cellular function in addition to splicing.

Materials and Methods: Molecular diagnosis in a cohort of 54 Indian A-T families, identified 46 different disease-causing ATM variants. Of these 46 variants, 21.7% (10/46) were classified as mis-splicing events based on in silico predictions. Four of these variants, c.1898+1G>A(Intron12), c.3153+5G>A(Intron21), c.5178-2A>G(Intron34), and one exonic missense variant c.7307G>A (exon

49), which is recurrently found in our North Indian A-T cohort were identified as ASO-amenable. ASO screens were conducted for these variants using patient lymphoblastoid cells and minigene assays to identify ASOs capable of correcting mis-splicing. To determine if ASOs could rescue ATM cellular function in addition to mis-splicing, we analysed their ability to restore full-length ATM as well as response to radiation-induced damage as measured by phosphorylation of ATM S1981.

Results and Conclusions: The selected four ATM variants were predicted to impair the donor and acceptor splice sites according to Human Splice Finder (HSF) and Splice AI. Transcript analysis from minigene and patient-derived LCLs of 3 variants c.1898+1G>A (Intron12), c.3153+5G>A (Intron21) c.5178-2A>G (Intron34), showed complete skipping of exons 12, 21, and 34 respectively. The variant c.7307G>A (exon49) showed partial skipping of exon 49 with a deletion of 37 nucleotides. ASOs designed to target either the novel splice donor sites in the exons or predicted splice regulatory elements were effective in restoring normal splicing as demonstrated by minigene assay or in patient LCLs in a dose-dependent manner. Selected ASOs were also able to induce a significant amount of full-length ATM and cellular response to radiation as measured by autophosphorylation of ATM-S1981.

Our data underscore the importance of analysing and interpreting the molecular mechanism of mis-splicing enable to successfully design and develop ASOs for restoring normal ATM splicing and cellular function.

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Abstract ID: 200

IL-2/IL-4 stimulation enhances the diagnostic yield of chromosomal abnormalities in Multiple Myeloma

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Background/ Introduction: Multiple myeloma is a genetically complex, heterogeneous clonal plasma cell neoplasm where cytogenetic abnormalities significantly influence the disease prognosis and risk stratification. Several studies have proved that Interphase FISH consistently detects >90% of primary as well as secondary cryptic aberrations even in lowest 5-10% of abnormal plasma cells. Though conventional karyotyping provides an overview of entire genome (all 46 chromosomes, numerical and structural aberrations≤5Mb) abnormal karyotypes were identified only in about 20-50% of multiple myeloma cases, more often in an advanced stage or in a highly proliferative form of the disease with >15% of abnormal plasma cells. Poor mitotic index (inability to get metaphases)/compromised analysis (very few metaphases) due to the limited proliferation of plasma cells in Invitro culture is a limitation in conventional cytogenetics. The present report is to evaluate culture outcomes, frequency of chromosomal abnormalities detected upon IL-2/ IL-4 stimulation of plasma cells and compare the outcomes of conventional karyotyping vs interphase FISH in multiple myeloma.

Objectives: To assess the diagnostic yield of cytogenetic abnormalities and effectiveness of IL-2/IL-4 stimulation on conventional karyotyping in multiple myeloma.

Materials and Methods: Conventional karyotyping was done in 26 multiple myeloma patients. The bone marrow cultures were stimulated independently using B-cell mitogens Interleukin-2 (400ng/3-5 days) and Interleukin-4(200ng/5 days) for karyotyping. plasma cells were purified by using CD138 coated magnetic

beads and Interphase FISH was performed on isolated plasma cells using specific probes. The karyotype and FISH were analysed and interpreted as per ISCN 2020 guidelines.

Results and Conclusions: Results: Out of 26 cases stimulated by IL-2/IL-4 all the cultures were successful with analysable metaphases.42% of cases were detected to have chromosomal abnormalities by karyotyping (11/26). The incidence is high compared to previous studies.31% of cases(8/26) had complex karyotype(>3 abnormalities).Interphase FISH on purified plasma cells detected abnormalities in 80% of cases(21/26) is in consensus with available literature. The proportion of plasma cells in the bone marrow was high in patients with chromosomal abnormalities (median 37%) than in patients without abnormalities (median 9%). However, in all the abnormal cases karyotype detected additional chromosomal abnormalities than FISH.

Conclusion: IL-2/IL-4 stimulation enhanced the diagnostic yield by detecting additional complex chromosomal abnormalities compared to FISH which influenced better patient management in multiple myeloma.

Abstract ID: 201

The Impact of Aging on Synaptic Redox Homeostasis: A Study of Antioxidant Capacity, Oxidative Stress, and the Protective Effects of Emodin

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Background/ Introduction: Aging significantly disrupts synaptic redox homeostasis, a delicate balance critical for neuronal function. This imbalance, marked by increased oxidative stress, reduced antioxidant defenses, and heightened oxidative damage, is a precursor to age-associated neurodegenerative disorders. Synaptosomes, representing functional nerve endings, provide a valuable model to investigate oxidative stress at the synaptic level. Emodin, a bioactive anthraquinone, has shown potential in countering oxidative damage due to its antioxidant properties. However, its therapeutic effects on synaptic redox homeostasis remain largely unexplored. This study focuses on assessing age-related changes in synaptosomal redox parameters and evaluating the protective role of Emodin in mitigating oxidative stress.

Objectives: 1. To assess antioxidant capacity using ABTS and FRAP assays in synaptosomes isolated from young and old male Wistar rats, 2. To measure oxidative species by performing ROS assays, 3. To evaluate oxidative damage through protein thiol, protein carbonyl, and lipid peroxidation assays, and 4. To examine the therapeutic potential of Emodin on the above parameters.

Materials and Methods: . ABTS Assay:
• Reagents: ABTS powder, potassium persulfate, PBS, distilled water, synaptosomes, ascorbic acid (standard), ELISA plate reader.
• Procedure: A stable ABTS radical solution (7 mM ABTS with 2.45 mM potassium persulfate) was prepared and incubated in the dark for 12-16 hours at room temperature. Synaptosome samples (200 µg protein) were mixed with the radical solution, and absorbance was measured at 734 nm after 6 minutes. Antioxidant activity was calculated as a percentage decrease in absorbance. Emodin (20 µL) treatment was performed for 30 minutes before analysis.
2. FRAP Assay:

• Reagents: FRAP reagent (acetate buffer, TPTZ, FeCl3.6H2O), ascorbic acid standards, crude synaptosomes.
• Procedure: FRAP reagent was mixed with synaptosome samples or ascorbic acid standards and incubated for 30 minutes. Ferric reducing power was measured at 593 nm. Emodin treatment was conducted similarly.
3. Protein Thiol Oxidation (DTNB Assay):
• Reagents: DTNB, Tris buffer, SDS, TCA, synaptosomes.
• Procedure: Protein samples were precipitated with TCA, washed, and solubilized. DTNB was added to supernatant and resuspended pellet. Absorbance was measured at 412 nm, and thiol content was calculated.
4. Protein Carbonyl Formation (DNPH Assay):
• Reagents: DNPH, PBS, TCA, acetone, urea.
• Procedure: Crude synaptosome samples were reacted with DNPH, precipitated with TCA, and washed with acetone. Protein carbonyls were dissolved in urea and measured at 370 nm.
5. Lipid Peroxidation (TBARS Assay):
• Reagents: TBA-TCA-HCl reagent, PBS, crude synaptosomes.
• Procedure: Crude synaptosome samples were reacted with the TBA reagent, heated, and centrifuged. TBARS levels were quantified at 535 nm.
For each assay, Emodin treatment was applied, and effects on redox parameters were measured.

Results: 1. Antioxidant Capacity: Old synaptosomes exhibited significantly lower antioxidant capacity (ABTS and FRAP assays) compared to young synaptosomes. Emodin treatment improved antioxidant capacity in old synaptosomes. 2.Oxidative Species: ROS levels were elevated in old synaptosomes, but Emodin treatment significantly reduced these levels. 3.Oxidative Damage: Old synaptosomes showed higher protein thiol oxidation, protein carbonyl formation, and lipid peroxidation, indicative of oxidative damage. Emodin treatment reduced these markers, restoring redox balance. 4. Comparative Analysis: Young synaptosomes demonstrated superior redox homeostasis, but Emodin effectively mitigated oxidative stress and damage in old synaptosomes.

Conclusions: This study demonstrates that aging exacerbates oxidative stress and diminishes antioxidant defenses in synaptic systems. Emodin treatment effectively restores redox homeostasis by reducing oxidative species, enhancing antioxidant capacity, and mitigating oxidative damage. These findings highlight Emodin's therapeutic potential as a neuroprotective agent against age-related oxidative stress. Further studies are warranted to explore its mechanisms and applications in neurodegenerative disease models.

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Abstract ID: 203

Genetic variants in bilateral cleft lip and cleft palate

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Background/ Introduction: Unilateral cleft lip is a common malformation. In children with cleft lip and/or cleft palate, genetic basis can be copy number variants or single nucleotide variants. Isolated cleft palate is more likely to have syndromic cause.

Objectives: To analyse the molecular basis of bilateral cleft lip with or without cleft palate seen in a speciality clinic of tertiary care centre.

Materials and Methods: The patients were enrolled from the Genetic Clinic and Genetic ward of a medical Institute. Informed consent was taken for enrollment and genetic testing as per ICMR guidelines, as part of research study. Next generation sequencing (NGS) testing was performed with pre-test genetic counseling. Ethical clearance was taken from Institute Ethics Committee.

Results and Conclusions: During a 2-year period, 37 cases of cleft palate (CLP)and 13 cases of cleft lip with or without cleft palate (CL-CLP) were observed in a speciality clinic referred for evaluation and counseling. 3 patients had bilateral cleft lip. The presentation included dysmorphism, short stature, and digital anomalies. Whole exome sequencing identified pathogenic variants in these 3 children with bilateral cleft lip and poor growth; which included TFAP2A, TP63, and CHD7 genes separately. Thus, rare genetic disorders were identified in bilateral CL and NGS is preferred testing for bilateral cases. The detailed presentation, implications of diagnosis of multiple malformation syndromes in patients with CL-CLP and importance of genetic counseling is discussed.

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Abstract ID: 204

Whole Exome Sequencing for Fetal Structural Anomalies: Expanding the Diagnostic Yield in Prenatal Care Royal hospital experiences

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Background/ Introduction: Prenatal ultrasound scans can identify fetal structural anomalies, but the underlying genetic cause often remains elusive. Whole exome sequencing (WES) is a powerful tool for identifying genetic variants associated with disease.

Objectives: This study aimed to evaluate the yield of WES in fetuses with structural anomalies and assess its role in prenatal diagnosis

Materials and Methods: We conducted a retrospective cohort study reviewing the electronic medical records of 88 fetuses with structural anomalies identified by prenatal ultrasound scan at the Royal Hospital Muscat between 2017 till 2023. WES was performed on all fetuses.

Results and Conclusions: A molecular diagnosis was obtained in 64 fetuses (73%) while no diagnosis was reached in 24 (27%). The highest diagnostic yield was observed in fetuses with multisystemic involvement. Among the positive results, 44 (69%) were pathogenic variants, and 20 (31%) were variants of uncertain significance (VUS). Notably, we identified AARS2: c.2027 A>C, p.(Gln676Pro) in five unrelated cases, suggesting a potential founder mutation associated with hydrops fetalis. Interestingly, 7% of the positive results had an autosomal dominant inheritance pattern, while 93% followed an autosomal recessive inheritance pattern

Conclusion: WES demonstrated a significant diagnostic yield in fetuses with structural anomalies, particularly those with multisystemic involvement. Our findings support the expanding role of WES as a valuable tool for prenatal diagnosis, potentially alongside conventional genetic testing approaches. The

identification of a potential founder mutation (AARS2: c.2027 A>C, p.(Gln676Pro)) associated with hydrops fetalis warrants further investigation.

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Abstract ID: 205

Making Informed Choices: The Role of Preventive Genetics in Reproductive Planning- A case study of Preimplantation Genetic Testing for Monogenic disorders (PGT-M)

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Background/ Introduction: Asparagine synthetase deficiency (ASD) is a rare autosomal recessive disorder with an estimated 20 cases reported worldwide. It is characterized by neurological impairment due to impaired synthesis of asparagine, an essential amino acid. Preimplantation genetic testing for monogenic disorders (PGT-M) is an important advancement in reproductive medicine and genetic counseling. PGT-M is particularly important for couples who are at risk of passing on inheritable single-gene conditions.

Objectives: This case report details the application of Preimplantation Genetic Testing for Monogenic Disorders (PGT-M) in a couple with a history of asparagine synthetase deficiency. PGT-M enables couples to select embryos that do not carry the mutation, significantly reducing the risk of passing on a hereditary disorder to their children.

Materials and Methods: A third-degree consanguineous couple with a history of two neonatal deaths due to fetal anomalies, were referred for Genetic Counselling. Carrier screening by clinical exome sequencing in couple revealed both partners were heterozygous carriers for a likely pathogenic mutation, c.788C>T in ASNS gene associated with Asparagine synthetase deficiency. Given the autosomal recessive nature of the disorder, the couple was concerned about the recurrence risk and they desire to have a healthy child. Following comprehensive genetic counseling and testing, the couple opted for Preimplantation Genetic Testing for Monogenic disorders (PGT-M) in embryos derived through Assisted Reproductive Technology to identify the likely pathogenic variant in the ASNS gene and prevent Asparagine synthetase deficiency in their next child.

Results and Conclusions: Preimplantation Genetic Testing for Monogenic disorder in conjunction with Preimplantation Genetic Testing for Aneuploidy (PGT-A) was performed in four embryos, amongst which one was identified as a Euploid carrier embryo. Transfer of the genetically healthy embryo lead to a successful implantation and subsequent live birth of a healthy infant.

This case underscores the efficacy of PGT-M in preventing the transmission of genetic disorders and highlights its role in reproductive decision-making for at-risk couples. The implementation of PGT-M not only aids in the avoidance of ASD but also provides valuable insights into the ethical considerations and emotional impacts on families navigating genetic conditions. Further studies are necessary to evaluate long-term outcomes and psychosocial implications for families utilizing PGT-M for rare genetic disorders. Genetic counseling (pre-test and post-test) will be of central importance as it is one of the most effective means to bring awareness on the possibilities of preventing common genetic disorders by educating/making the patient aware of their genetic risks and available reproductive options which enable the couple to take informed decisions.

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Abstract ID: 206

Clinical and genetic profile of patients with Niemann Pick disease

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Background/ Introduction: Niemann-Pick type C (NPC) is a very rare autosomal recessive lysosomal storage disorder caused by mutations in the NPC1 or NPC2 genes. This leads to impaired lipid transport and abnormal accumulation of unesterified cholesterol in the brain and other tissues, resulting in organ system damage and manifesting with primary neurodegeneration. Cohort studies of genetically-established NPC from the Indian population are limited.

Objectives: To describe the clinical and genetic profile of patients with NPC.

Materials and Methods: In the present study, patients with clinically suspected NPC seen in a single Neurology unit from 2015 to 2024 were included. Their clinical, radiological, electrophysiological and genetic data were documented systematically in a predesigned proforma.

Results and Conclusions: The cohort comprised four patients (M:F 1:2) from Bihar (n=1), Tamil Nadu (n=1), Andhra Pradesh (n=1) and West Bengal (n=2). Their age at evaluation ranged from 16 to 35 years. Two patients were children (age<18 years). Age at onset ranged from 10 to 29 years, of which one patient had onset in adulthood. All were born to non-consanguineous parents. A family history of similar illness was present in one patient. All patients presented with cognitive decline and extrapyramidal features. Other clinical features included altered behaviour (n=3) and seizures (n=2). Extrapyramidal features included dystonia in all and parkinsonism in two. Behavioural symptoms included depressed mood, abnormal laughter and apathy. Seizure semiology included generalized tonic-clonic (n=2) and complex partial seizures (n=1). Supranuclear vertical gaze palsy and impaired vertical saccades and pursuits were noted in all. Evoked potentials to visual, auditory and somatosensory stimuli were normal in all. Brain MRI revealed diffuse atrophy. Three patients had splenomegaly. Exome sequencing revealed homozygous pathogenic variations in NPC1 (c.2050C>T) in patient 1, and two heterozygous pathogenic variations in NPC1 (c.302T>G) and c.3704T>C) in patient 2. Patient 3 had a single heterozygous variant in NPC1 (c.3736_3737del). Patient 4 had a homozygous pathogenic variation in NPC2 (c.358C>A). Very few studies have established the clinical and genetic profile of patients with Niemann-Pick disease. Clinical phenotyping and genetic correlation are essential for precise diagnosis and genetic counselling, and these patients may be candidates for upcoming therapies.

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Abstract ID: 207

SEC23B Dysfunction in Congenital Dyserythropoietic Anemia Type II: Insights from Molecular and Gene Expression Studies

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Background/ Introduction: Congenital Dyserythropoietic Anemia (CDA) encompasses a group of rare, inherited, and heterogeneous disorders marked by ineffective erythropoiesis. CDA type II is the most

prevalent form, attributable to mutations in the SEC23B gene. Historically, the diagnosis of CDA has relied on distinct bone marrow abnormalities, including erythroid hyperplasia and the presence of binucleated or multinucleated erythroblasts. However, these morphological features are not unique to CDA, as they are also observed in conditions such as iron deficiency anemia and stress erythropoiesis in preterm infants. Therefore, precise molecular characterization is necessary for an accurate diagnosis.

Objectives: To undertake the molecular characterization of CDA type II in the Indian population and to assess the impact of identified genetic variants on SEC23B gene expression.

Materials and Methods: Patients suspected of having CDA were subjected to molecular characterization using a targeted next-generation sequencing (t-NGS) panel developed in-house. Libraries for sequencing were prepared with the Twist Library Preparation Kit, and raw sequencing data was processed through custom bioinformatics pipelines adhering to GATK best practices. Variants were annotated using the wANNOVAR platform, and their pathogenicity was evaluated using computational tools such as SIFT, PolyPhen-2, MutPred, and MutationTaster. Structural impacts of variants on the SEC23B protein were assessed using DynaMut and Swiss PDB Viewer. To functionally validate these variants, quantitative real-time PCR (qRT-PCR) was employed to analyze SEC23B mRNA levels in patients diagnosed with CDA type II.

Results and Conclusions: A cohort of 28 patients with CDA type II underwent comprehensive molecular and gene expression analyses. Among them, 20 patients harbored a homozygous missense variant, one presented with a homozygous splice site variant, four exhibited compound heterozygous missense variants, and three demonstrated compound heterozygosity involving one missense and one nonsense variant. 3 novel variants were identified. qRT-PCR analysis revealed a marked reduction in SEC23B mRNA levels, with expression decreased by 40–60% compared to healthy controls.

This study represents the first detailed exploration of clinical phenotypes, molecular characterization, and functional consequences of SEC23B gene mutations causing CDA type II in the Indian population. It significantly expands the molecular understanding of CDA type II and highlights the impact of SEC23B variants on gene expression, thereby advancing diagnostic precision for this rare disorder.

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Abstract ID: 208

Biallelic Variants in CCN2 Underlie an Autosomal Recessive Kyphomelic Dysplasia

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Background/ Introduction: Kyphomelic dysplasia is a rare heterogenous group of skeletal dysplasia, characterized by bowing of the limbs, severely affecting femora with distinct facial features. The term "kypho" is derived from ancient Greek word "kyphos," meaning "bent", while "melia" refers to "limb". The term 'kyphomelia' was first used to describe a skeletal dysplasia by MacLean in 1983. Despite its first description nearly four decades ago, the precise molecular basis of this condition remained elusive. Moreover, it was conventionally considered an autosomal recessive condition. Recently, heterozygous de novo variants in KIF5B have been found to be associated with kyphomelic dysplasia, while several bent bone dysplasias do not have a known genetic basis.

Cellular communication network factor 2 (CCN2) is a matricellular protein spanning 349 amino acids, with five exons. It belongs to cysteine rich CCN protein family crucial for proper skeletal growth and development. In vitro studies on CCN2 demonstrated that it promotes DNA synthesis in chondrocytes. Previous investigations on Ccn2 deficient mice showed broader vertebrae, shortened and kinked sterna, along with bending in the radius, ulna, tibia, and fibula. Additionally, they exhibit craniofacial abnormalities including a distorted ethmoid bone, a domed cranial vault, shortened mandibles and secondary cleft palate.

Owing to the high conservation of fundamental signalling pathways and cellular processes involved in skeletal development from fish to humans, zebrafish serve as valuable models for studying skeletal disorders. The function of ccn2a in early skeletal development in zebrafish has not yet been studied.

Zebrafish serve as valuable models for studying skeletal disorders due to the high conservation of fundamental signaling pathways and cellular processes involved in skeletal development. However, the role of ccn2a in early skeletal development in zebrafish has not yet been studied.

We studied three probands from two consanguineous families with bowed long bones, and identified biallelic CCN2 variants segregating recessively. Using zebrafish models, we examined the impact of CCN2 loss of function.

Objectives: The objective of the study was to evaluate multiple affected individuals with kyphomelic dysplasia and to investigate the consequences of CCN2 loss of function in vivo using zebrafish models.

Materials and Methods: Three individuals with kyphomelic dysplasia from two unrelated families of different ethnicities were recruited. Clinical, radiological, and medical history were documented, with written informed consent obtained from participants and their families. The study was approved by the institutional ethics committees.

Exome sequencing was performed for all the three affected individuals with kyphomelic dysplasia. Filtered variants were further analyzed using several in silico pathogenicity prediction tools to assess their potential impact. Conservation analysis was performed using the Clustal Omega tool. The allele frequency of the identified rare variant was estimated from gnomAD and our in-house data of 3188 exomes. Sanger sequencing was performed to validate and segregate identified candidate variants in the proband and their family members. The variants were described according to Human Genome Variation Society (HGVS) nomenclature, with NCBI reference sequences (NM_001901.4, NP_001892.2). Both variants were submitted to the Leiden Open Variation Database (LOVD) database (variant ID: 0000972076; 0000972075). Disulfide bridge between cysteine 148 and a nearby cysteine residue was visualized using PyMOL (protein modelling tool).

CRISPR/Cas9 mediated gene editing was used for generation of ccn2a F0 knockout in zebrafish. Alcian blue staining was performed. Benchling software was used to choose target region of ccn2a. Single guide RNAs (sgRNAs) were synthesized, and phenotypic characterization of the ccn2a F0 knockout zebrafish

was performed. Quantitative real-time qPCR analysis was done for investigating gene expressions of ccn2a, rac1a, rhoAa, col2a1a, sp7, runx2a and gapdh using Ct method (ΔΔCt).

Results and Conclusions: We ascertained two unrelated consanguineous families with kyphomelic dysplasia. They had six affected offsprings and we performed a detailed clinical evaluation, skeletal survey, and exome sequencing in three probands. All the probands had short stature, facial dysmorphism, cleft palate, and micro-retrognathia. Radiographs revealed kyphomelic femora, bowing of long bones (tibia, fibula, radius and ulna), radial head dislocations, scoliosis, mild platyspondyly and irregularities of the knee epiphyses and metaphyses.

Their clinical and radiological manifestations were consistent with what were previously reported in kyphomelic dysplasia. However, the phenotypic manifestations in family 1 were milder compared to those in family 2.

In Family 1, a shared biallelic missense variant, c.443G>A; p.(Cys148Tyr), located in exon 3 of the CCN2, was identified through duo exome sequencing. The variants are present in heterozygous state in the parents and absent in the unaffected sibling. This variant is absent in gnomAD (V3.1.2) and our in-house data of 3188 exomes. In silico mutagenesis analysis revealed a disulfide bridge formed between cysteine residue at position 148 and the nearby cysteine at position 166, is disrupted due to the substitution. Multiple sequence analysis performed revealed conservation of the cysteine residue across several vertebrate species. In family 2, we identified a homozygous frameshift variant, c.779_786del; p.(Pro260LeufsTer7) that was absent from the public and in-house datasets, located in exon 5 of CCN2. Sanger sequencing confirmed the heterozygous status of the parents.

A significant decrease in ccn2a mRNA was observed in the crispants as compared to the NT and WT controls, thus confirming ccn2a editing. The ccn2a crispants showed abnormal body curvature and bent tail suggesting defects in early skeletal development. The ccn2a crispants showed substantial defects in cartilage formation in the craniofacial region as seen by Alcian blue staining at 6.5 days post-fertilization. They had underdeveloped ceratohyal arches, bent or missing ceratobranchial arches and misshapen Meckel cartilage. The levels of established skeletal marker genes such as col2a1a (chondrocyte marker), rac1a and rhoAa (palatogenesis markers), sp7 and runx2a (osteoblast markers) in ccn2a crispants showed a significant decline, as is expected from the phenotype. The crispants also showed poor survival beyond seven days post-fertilization and few survived to adulthood. These adult crispants (F0 KO) showed defects in mineralization and bone structure in specific locations with known endochondral ossification such as missing structures in the tail region and abnormal trunk curvature. The level of knockdown of ccn2a in these adults was confirmed by measuring the mRNA level from trunk tissue and was found to be significantly reduced.

Our observations in humans and zebrafish combined with previously described skeletal phenotype of Ccn2 knock out mice, confirm that biallelic loss of function variants in CCN2 result in an autosomal recessive kyphomelic dysplasia. However, investigation of additional patients and cellular studies are necessary to establish the gene-disease relationship.

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Abstract ID: 209

TLR4 Pathway Hyperactivation in Chronic Suppurative Otitis Media: A Potential Therapeutic Target

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Background/ Introduction: Chronic Suppurative Otitis Media (CSOM) is an uncontrolled inflammation in the middle ear due to bacterial infections. The Toll-Like Receptor 4 (TLR4) pathway is activated by Lipopolysaccharide (LPS) released from gram-negative bacterial cell walls. Currently, there is limited information about the factors responsible for overactivation. One possible factor could be the hypersensitivity of the TLR4 pathway in CSOM patients

Objectives: • To quantify the effect of LPS on TLR4 gene expression levels in the PBMCs of CSOM patients and healthy controls.
• To quantify the effect of LPS on NFkB gene expression levels in the PBMCs of CSOM patients and healthy controls.
• To assess the effect of LPS on TNFα cytokine in the PBMCs of CSOM patients and healthy controls

Materials and Methods: • Case-control study with 63 participants (CSOM patients and healthy controls)
• Peripheral blood mononuclear cells (PBMCs) were cultured for 4 hours with LPS
• TLR4 and Nuclear Factor kappa B (NFkB) gene expression was measured using qPCR
• Tumor Necrosis Factor α (TNFα) cytokine levels were measured using ELISA

Results and Conclusions: Results:
• LPS-induced fold change in TLR4 gene expression was higher in CSOM patients (2.8 vs. 1.6, $p < 0.001$)
• LPS-induced fold change in NFkB gene expression was higher in CSOM patients (3.8 vs. 1.4, $p < 0.001$)
• LPS-induced fold change in TNFα production was higher in CSOM patients (3.2 vs. 1.1, $p < 0.001$)
Conclusion:
• The results indicate hypersensitivity of the TLR4 pathway in CSOM patients, suggesting a potential therapeutic target for the treatment of CSOM.
• Future studies may need to explore the TLR4 pathway for developing drugs that can mitigate the inflammatory damage in CSOM.

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Abstract ID: 210

Genetic Insights into Epidermolysis Bullosa: A Cohort Study Highlighting COL7A1, LAMA3, and LAMB3 Mutations

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Background/ Introduction: Epidermolysis Bullosa (EB) is a group of rare genetic disorders characterized by extreme skin fragility, where even minor trauma can lead to blister formation. Among its subtypes, Dystrophic Epidermolysis Bullosa (DEB) is particularly notable, arising from mutations in the COL7A1 gene. This highlights the critical role of genetic counseling in helping affected families understand the condition's inheritance patterns, assess recurrence risks, and make informed decisions about future family planning.

Objectives: This study analyzes genetic mutations in a cohort of 9 EB patients, focusing on COL7A1 mutations and the impact of genetic counselling for affected families.

Materials and Methods: Genetic testing via Next Generation Sequencing (NGS) panel and Sanger sequencing for mutations associated with EB. Genetic Counselling sessions covered inheritance patterns, carrier risks, and prenatal options.

Results:

Of 9 patients, 5 had COL7A1 mutations (4 pathogenic, 1 uncertain), and the remaining 4 had LAMA3 and LAMB3 mutations (3 pathogenic, 1 likely pathogenic), suggesting junctional EB. Genetic Counselling clarified recurrence risks (25%) and provided options for prenatal and preimplantation testing.

Conclusion: This study highlights the genetic diversity in EB, with a predominance of COL7A1 mutations in DEB. Genetic counseling was crucial for family support, reproductive planning, and understanding genetic risks. Early diagnosis and counseling improved decision-making and quality of life for affected families, with some opting for prenatal genetic testing.

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Abstract ID: 211

Identification of potential risk genes in congenital heart disease through whole exome sequencing

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Background/ Introduction: Congenital heart disease (CHD) is one of the common birth defects characterized by the abnormal structure and function of the heart. The prevalence of CHD is ~8-11 cases per 1000 live births and ventricular septal defect (VSD) is the common type of CHD accounts for 30 to 40% of cases in India. The potential risk genes involved in the manifestation of VSD specific CHD still needs to be established.

Objectives: This study aims to identify the genes using WES and characterizing the gene variations and their pathogenicity in the manifestation of the CHD.

Materials and Methods: The genes reported in VSD were identified through data mining using CHD databases, CHD-RF-KB, CHDbase, and PubMed medical subject headings. Whole exome sequencing was performed for 24 CHD subjects including VSD, VSD-associated atrial septal defects (ASD), and tetralogy of fallot (ToF). The functional effects of variations were assessed using prediction tools and STRING protein-protein interaction network was established to identify the interactions among the proteins.

Results and Conclusions: The findings are: 1) Data mining yielded 50 genes and of these, 34 were common in our 24 CHD subjects with new variations; 2) Of the 34 genes, DLC1 and MTRR recurrently occurred in 18 CHD subjects; 3) The variation, c.524C>T in MTRR was recurrently occurred in nine CHD subjects; 4) A total of 72 nonsynonymous variations were found in 32 genes, of which four variations in GATA4 and one variation in TBX20 showed high pathogenicity; 5) The protein-protein interaction network showed high interactions among transcription factors. The TNNT2, MYL7, and ZFPM2 were the newly identified proteins in the network. This study identified 50 potential risk genes and new variations with high pathogenicity. These can be considered as potential markers to create a gene panel for the diagnosis of CHD and treatment. The implication of these findings will be discussed.

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Abstract ID: 212

Prevalence of deleterious Common and Rare variants in Autism-associated genes in Indian cohort of Autism Spectrum Disorders

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Background/ Introduction: Autism spectrum disorder (ASD) is a complex neurodevelopmental disorder that is behaviorally defined by challenges in social skills, restricted repetitive behaviors, and communication deficits. Considering limited genetic research on ASD in India, we are uncertain of whether its architecture is similar or dissimilar to what is observed globally.

Objectives: Using the Simons Foundation Autism Research Initiative (SFARI)- a well-curated database of ASD-candidate we aimed to investigate the mutation burden in SFARI genes among Indian ASD subjects and study the impact of common and rare variant stratification across populations.

Materials and Methods: Whole Exome Sequencing (WES) was performed for 25 Indian ASD subjects. We included all variations identified in SFARI genes irrespective of their Minor Allele Frequencies (MAFs) and compared them across global and Indian populations. 'NonSFARI' genes were analyzed to identify additional genes with neurodevelopmental functions using multidimensional bioinformatics approaches.

Results and Conclusions: Only 15% of SFARI genes carried 241 common and rare deleterious variations in our ASD cohort. The most frequent variants identified were Protein Truncating Variants (PTVs) (69.7%). The SFARI Score-1 genes had more missense variants than PTVs hinting at their evolutionarily conserved nature. Seven PTVs in six genes, SON, KMT2C, EXOC6B, RBFOX1, CAPN12 and KIZ were found to be recurrently altered in multiple ASD individuals. About 32% of 241 deleterious variations identified were common (MAF ≥0.05) across populations. Asian population had significantly more rare PTVs than global populations. Functional analysis identified 101 nonSFARI genes with neurodevelopmental functions, of which six genes were found altered in unrelated ASD subjects globally

which could be the new potential ASD-candidate genes. Thereby, our study provides evidence that Indian ASD subjects are more burdened by PTVs. Common variants levy additional genetic-risk necessitating further validation in larger cohorts. Using population specific human genome reference is important to obtain insights into population-specific risks for a disease.

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Abstract ID: 213

A study on analyzing the DNA methylation patterns, its roles in Down syndrome and congenital heart disease- A Review Article

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Background/ Introduction: Down syndrome (DS), caused by trisomy 21, is frequently associated with congenital heart disease (CHD), observed in up to 63% of individuals with DS. Emerging evidence indicates a pivotal role of epigenetic modifications, specifically DNA methylation, in the etiology of CHD among individuals with DS. Understanding these epigenetic patterns can elucidate the mechanisms driving CHD and potentially guide therapeutic approaches

Objectives: This review aims to explore the role of DNA methylation in CHD pathogenesis in individuals with DS, focusing on hypomethylation and hypermethylation patterns, their impact on gene expression, and the involvement of key cardiac development genes. Additionally, the review examines population-specific methylation trends, including findings in Indian cohorts, to highlight gaps and future research opportunities.

Materials and Methods: A systematic review of the existing literature was conducted, encompassing studies on epigenetics, DNA methylation, and CHD in DS populations. Data on differentially methylated regions (DMRs), global methylation patterns, and gene-specific methylation changes were analyzed with an emphasis on their biological relevance to cardiac development and congenital anomalies.

Results and Conclusions: Findings reveal global hypomethylation and hypermethylation patterns in DS individuals, with notable epigenetic changes in cardiac development genes such as GATA4, NKX2-5, and TBX5. Specific DMRs associated with cardiac muscle function and metabolic pathways were identified, with significant variations observed between DS individuals with and without CHD. Indian population studies indicate methylation alterations in genes like MTHFR and CRELD1, linking them to CHD phenotypes. However, regional variations in methylation patterns necessitate further localized research.

Conclusions: DNA methylation plays a crucial role in CHD development among DS individuals by modulating gene expression and epigenetic memory. Population-specific studies, particularly in underrepresented regions like India, are critical to understanding environmental and genetic interactions shaping epigenetic landscapes. This knowledge could inform novel diagnostic markers and therapeutic strategies, improving outcomes for individuals with DS and CHD.

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Abstract ID: 214

Taste Sensitivity and Obesity Risk in Indian Adults: The Impact of Genetic Variations in Sweet and Fat Taste Receptors

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Background/ Introduction: Chronic diseases like obesity and diabetes are closely linked to dietary intake of highly palatable sugary and fatty foods. Individuals with obesity may respond differently to diets due to abnormalities in taste perception. Specific single nucleotide polymorphisms in taste receptors, especially those for sweet and fat, have been linked to altered dietary behaviour and obesity development. Despite these associations, limited research has investigated the relationship between T1R2/T1R3 polymorphisms, and fat taste perception among Indian population. Hence, the study aims to explore these relationships in Indian individuals with varying BMIs.

Objectives: The study aimed to investigate the relationship between genetic polymorphism in sweet and fat taste receptors and taste sensitivity; the relationship between genetic polymorphism and anthropometric and biochemical parameters; impact of genetic variations on taste sensitivity and their potential role in obesity risk among Indian adults with varying BMI.

Materials and Methods: Healthy participants were recruited according to specified inclusion and exclusion criteria. Anthropometric measurements were recorded, and blood samples were obtained for genotyping and serum parameter analysis. TaqMan Probe-based RT-PCR was employed to determine the polymorphisms among T1R2, T1R3, and CD36 genes. The general labelled magnitude scale (gLMS) was used to assess the taste sensitivity for Linoleic acid and Sucrose.

Results and Conclusions: Participants with polymorphisms in CD36, T1R2, and T1R3 were associated with significantly higher BMIs, waist circumferences, triglyceride levels and Triglyceride-Glucose Index after adjusting for age and gender. Individuals with obesity showed lower sensitivity than healthy adults, as indicated by higher detection thresholds for sucrose and linoleic acid (LA). Moreover, a positive correlation ($r = 0.5299$, $p < 0.0001$) was observed between LA and sucrose thresholds. Hence, the fact that people with polymorphic alleles in CD36, T1R2, and T1R3 had higher BMI, and lower taste intensity ratings highlights the interplay between genetic and physiological factors in the development of obesity and taste perception.

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Abstract ID: 215

Prevention and treatment strategies using preimplantation genetic testing (PGT) for thalassemia and other hematological disorders: A Jaslok Hospital experience.

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Background/ Introduction: The prevalence of hematological disorders like beta thalassemia is high in India. Preimplantation Genetic Testing (PGT) is an additional step during in vitro fertilization (IVF) to

select unaffected embryos from fertile carriers of familial monogenic disorders (PGT-M). This avoids the trauma of termination of multiple affected pregnancies. PGT can help families cure their affected children by selecting HLA matched unaffected embryos for a future bone-marrow transplant from the young 'Savior sibling'.

Objectives: To offer IVF-PGT for couples carriers for hematological disorders to select unaffected euploid embryo to get disease free own child or HLA matched Savior Sib.

Materials and Methods: PGT-M for hematological disorders was carried out in 29 couples (19 beta-thalassemia including 4 for HLA matching, 4 sickle cell anemia, 2 beta-thalassemia/sickle cell anemia and 1 each of alpha/beta thalassemia, hemochromatosis, MTHFR and G6PD with MTHFR). In IVF cycle, trophectoderm cells biopsied from blastocysts were subjected to aneuploidy screening by PGT-A followed by PGT-M for pathogenic variants. Unaffected euploid embryos were selected for implantation in 19/29(65%) couples. Prenatal diagnosis reconfirmed PGT results. A single HLA matched unaffected euploid embryo was used for implantation in 1/4 beta-thalassemia-HLA matching cases. For alpha/beta thalassemia (husband homozygous for alpha triplication and wife carrier for beta-thalassemia) PGT-M for beta-thalassemia was carried out.

Results and Conclusions: Pregnancy was achieved in 16/19(84%) couples: 4(25%) ongoing, 11(69%) full term deliveries of unaffected healthy children (8 cases of beta-thalassemia, 2 sickle cell anemia, 1 MTHFR) while a twin pregnancy miscarried (G6PD with MTHFR). One of the births was a HLA matched brother – a savior sib who helped cure his elder beta-thalassemia affected sister with a successful bone marrow transplant.

At our centre, we could successfully help in prevention of hematological disorders using PGT with full term unaffected healthy live births and cure of a beta-thalassemia affected child with an HLA matched sibling.

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Abstract ID: 216

Uncovering Genetic Mutations in Male Infertility: Insights from Whole Exome Sequencing

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Background/ Introduction: Male infertility is a complex condition with diverse etiologies, where genetic factors play a critical role. Whole Exome Sequencing has emerged as a powerful tool to identify mutations that contribute to infertility by analyzing exonic regions across the genome.Understanding these genetic underpinnings can aid in personalized reproductive counseling and informed therapeutic approaches.

Objectives: This study investigated the genetic basis of male infertility through WES to identify pathogenic mutations linked to spermatogenic failure, embryo maturation arrest, and other reproductive anomalies. The findings are intended to support clinical decisions for managing male infertility.

Materials and Methods: A cohort of 50 male patients diagnosed with infertility was selected for the study. Each participant underwent WES after Genetic counseling to detect mutations in key genes associated with reproductive function. Variants were assessed for pathogenicity based on established clinical criteria, with a focus on identifying variants likely to impact fertility.

Results and Conclusions: Out of 50 patients, mutations were identified in 18 patients related to specific phenotypes: Spermatogenic failure: FSIP2, CFAP58, CFAP43, and TTC21A, Embryo maturation arrest:

PADI6 and ZP1, Hypogonadotropic hypogonadism: ANOS1, Congenital bilateral absence of vas deferens: SLC26A8, Autoinflammation with arthritis and dyskeratosis: NLRP1, Ciliary dyskinesia: DNAH17, DNAH10, and CCDC40. Among the detected variants, 8 were classified as likely pathogenic, while 10 were variants of uncertain significance (VUS). These findings indicate a strong genetic contribution to infertility in the cohort, with WES revealing rare and potentially actionable mutations.

Whole Exome Sequencing (WES) achieved a diagnostic yield of 36% in this cohort, identifying mutations in 18 out of 50 patients with male infertility. The identification of specific mutations informs targeted genetic counseling, offering patients valuable insights into their reproductive options, including preimplantation genetic testing for monogenic disorders (PGT-M) and the use of donor sperm when appropriate. These findings underscore the importance of WES in supporting informed decision-making and optimizing reproductive outcomes for patients.

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Abstract ID: 217

Diagnostic Efficacy of Next Generation Sequencing in Pediatric Neurological Disorders

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Background/ Introduction: Genetics plays an important role in the aetiology of majority of neurological disorders in children. Yield depends on the type of genetic testing and the type of the disorder studied. Broadly yield is around 30 to 40% for various paediatric neurological disorders like neurodevelopmental disorders, epilepsy, neuromuscular disorders and around 20-25% for dystonia. Genetic testing helps in accurate and early identification, appropriate disease modifying treatment, and antenatal and peri-conceptional counselling.

Objectives: To determine the diagnostic efficacy of next generation sequencing (NGS) in a cohort of pediatric patients suspected with neurological disorders.

Materials and Methods: Retrospective chart review of pediatric patients who presented from the period of June 2017 to October 2022, with a clinically suspected neurogenetic disorder and have undergone genetic testing were recruited. Cases were subcategorized into genetic-positive group comprising those with pathogenic/likely-pathogenic, and genetic-negative group.

Results and Conclusions: We identified 420 patients (males: 256,59.5%), with median age of 5 (IQR 2 to 10 years). The clinical spectrum comprised neurodevelopmental (n=89, 21.2%), neuromuscular disorders (n=45,10.7%), epilepsy syndromes (n=146,34.7%), movement disorders (n=34,8%), autism spectrum disorders (n=37,8.6%), mitochondrial cytopathies (n=27,7.9%), neurocutaneous syndromes (n=8,5.1%), neurometabolic (n=37,8.8%), pediatric stroke, (n=2, 0.4%) and unclassified (n=3,0.7%). Next-generation sequencing revealed pathogenic/likely-pathogenic variants in 203 cases (yield 48%), VUS in 154 (n=36.3%), of which matching clinical phenotypes were identified in 115 (26.7%) cases, while 62 (n=14.4%) cases were genetic-negative. Genetic yield was significantly higher in the subcategories of neurodevelopmental disorders (p=0.016), progressive myoclonic epilepsy spectrum (p=0.017), while ataxia (p=0.095), and neuro-regression (p=0.064) showed a trend towards significance. Combining the phenotypically matching VUS could enhance the yield further to 55.4%.

Conclusions: The current study represents one of the largest paediatric neurogenetic disorders across the globe. It essentially highlights the overall utility of NGS across the entire spectrum of neurogenetic disorders in the pediatric population, including each of the individual subspecialities of neurology, and the common genes implicated in this part of the globe.

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Abstract ID: 218

Exploring the Phenotypic spectrum in Prader Willi Syndrome patients along with genotypic analysis.

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Background/ Introduction: Prader-Willi Syndrome (PWS) is genetic imprinting disorder caused by loss of function of SNRPN gene on paternal chromosome 15 (15q11-q13), occurring 1 in 10,000–30,000 live births. It results from paternal deletion (70–75%), uniparental disomy (UPD) (20–25%), or other imprinting defects (1–3%). PWS is characterized by hypotonia, feeding difficulties, hyperphagia leading to obesity, short stature, behavioral and learning difficulties, obsessive-compulsive traits, growth hormone (GH) deficiency and hypogonadism. Early initiation of GH therapy, nutritional and behavioral counseling, weight control, management of hyperphagia combined with pulmonary and skeletal surveillance is the recommended care for PWS.

Objectives: To characterize phenotypic spectrum in PWS and analyze correlation with underlying genotypic abnormalities.

Materials and Methods: Study involved 9 PWS patients (aged 10 months to 15 years, 3 females and 6 males) confirmed priorly through MS-MLPA. Structured clinical proforma was utilized to collect information regarding phenotypic characters, clinical history, and medications. Descriptive analysis was performed to evaluate spectrum of phenotypes and their correlation with genotypes. DNA extracted from blood of four individuals was further analyzed using long read sequencing to identify methylation patterns, extent of deletion and alterations within PWS 15q11-13 region.

Results and Conclusions: Study included 4 UPD and 5 deletions PWS individuals. Anthropometric data revealed consistent trends of short stature and obesity, with obesity being more pronounced in older patients. Distinct phenotypic differences were noted between genetic subtypes: UPD patients exhibited higher prevalence of psychiatric manifestations, including possessiveness and food-related tantrums, while those with deletion predominantly demonstrated severe obesity. Perinatal histories frequently reported reduced fetal movements, all patients exhibited hypotonia and feeding difficulties in infancy. We are further analyzing long-read sequencing data of four PWS individuals (2 UPD and 2 deletion) to observe mutation spectrum along with methylation to determine genotypic association with phenotypic severity which is a novel approach in characterizing PWS to facilitate personalized genetic counseling and management.

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Abstract ID: 219

Functional annotations of transcriptomic heterogeneity in ischemic stroke: Evidence from bioinformatic meta-analyses

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Background/ Introduction: Stroke ranks top five leading causes of fatality and disability in India. Early diagnosis and treatment are crucial for improving patient outcomes in Ischemic Stroke (IS). CT scan-based diagnosis is not only expensive and infrastructurally intensive but also exposes the patient to ionizing radiation. Identification of common pathomarkers and pathopathways from transcriptomics datasets across populations will enable scan-free diagnosis and novel biotherapies.

Objectives: 1) To detect shared pathomarkers across transcriptomic datasets from GEO which significantly modulated in blood and tissue of IS patients. 2) To demarcate the pathopathways (biological processes, molecular functions, and cellular components) over or under-represented in IS.

Materials and Methods: Transcriptomic datasets of ischemic stroke were extracted methodically from the GEO database. Datasets were case-control mRNA expression data by microarray or RNA-Seq from blood or tissue samples were included and datasets with control sample size n<4 and of noncoding RNA were excluded. Selected datasets underwent a multimodule meta-analytic workflow comprising quality control, expression profiling of pooled data and functional enrichment using Bioconductor packages in R 4.4.2. Differentially Expressed Genes (DEG) between IS and control subjects were compared. Genes with $P < 0.05$ and $|\log2FC| \geq 1$ were considered significant DEGs and visualized using volcano plots. Functional enrichment analysis was used to identify the cellular components, molecular functions, and biological processes represented in IS.

Results and Conclusions: Four datasets were finally screened out from 199 GEO DataSeries (GSEs): one tissue array dataset (GSE162955), two blood microarray datasets (GSE22255 + GSE16561) and a blood RNA sequencing dataset (GSE158312). Detailed meta-analysis results will be presented during the conference.

Identified pathomarkers and pathopathways could facilitate developing drug targets for IS. Findings of functional enrichment provide valuable insights into the underlying pathopathways of IS. Experimental validation of the variability of top-regulated pathomarkers in cell and animal model systems of ischemic stroke can unlock their translational potential.

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Abstract ID: 220

Rare variant in the NID1 gene causing Occipital encephalocele: Case report

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Background/ Introduction: Encephalocele is a neural tube defect which is a sac-like projection of the brain and its membranes that opens through the skull. This happens due to improper closure of the neural tube during the fetal development. Occipital encephalocele is the most common type of encephalocele,

accounting for approximately 75% of cases. The NID1 is one among the gene associated with autosomal dominant Dandy-Walker malformation and occipital encephalocele (Vanda McNiven et al., 2019. Sofie Dietvorst et al., 2023, Benjamin W Darbro et al, 2013). The NID1 gene which codes for nidogen 1 (entactin) protein is an essential glycoprotein that plays a vital role in the structure and stability of the basement membranes [Uwe Töpfer., 2024].

Objectives: To identify a rare variant causing occipital encephalocele in prenatal sample by exome sequencing.

Materials and Methods: The whole exome sequencing which focuses on most of the coding regions. DNA extracted from product of conception was used to perform targeted gene capture using a custom capture kit. The libraries were sequenced to mean depth of >80-100X on Illumina sequencing platform. Here in we discuss a case of a POC whose antenatal scan showed occipital encephalocele, prominent cisterna magna and inferior vermis hypoplasia. We have identified a heterozygous nonsense variation of uncertain significance in the NID1 gene [c.1930C>T; p.Gln644Ter]. There is limited literature evidence supporting loss of function in this gene can cause disease.

Results and Conclusions: The occipital encephalocele can be detected in the early stages of fetal development by neuroimaging techniques and through advanced genetic testing, such as whole exome sequencing and timely surgical intervention will overcome the challenges associated with the encephalocele. The genetic mutation in the NID1 seems to follow an autosomal dominant inheritance pattern, which is suggested by the presence of multiple affected individuals in the family. Further segregation studies in the affected and unaffected family member would be helpful to confirm the exact inheritance pattern and to understand better the full spectrum of genetic risk.

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Abstract ID: 221

Unravelling Early-Onset Polycystic Kidney Disease: The Role of Bi-Allelic PKD1 Variants in Atypical Presentations

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Background/ Introduction: PKD1 gene located on chromosome 16 encodes for polycystin-1 protein which form complex with polycystin-2 protein encoded by another gene PKD2 to form heteromeric calcium-permeable ion channel and regulates calcium permeable cation channels and intracellular calcium homeostasis. Heterozygous mutations in PKD1 gene are known to cause polycystic kidney disease 1 (ADPKD1). ADPKD1 is a late-onset (usual onset in middle age) multisystem disorder characterized by renal cysts, liver cysts, and intracranial aneurysm, acute and chronic pain and nephrolithiasis. However, very early onset polycystic kidney disease (PKD) are reported in literature with bi-allelic hypomorphic variants in PKD1 gene.

Objectives: To investigate the prevalence of early onset polycystic kidney disease with biallelic variants in PKD1 gene over a period of 5 years.

Materials and Methods: Whole exome sequencing or clinical exome sequencing or Polycystic kidney disease panel genes sequencing were employed to investigate genetic cause of early onset PKD patients.

Results and Conclusions: We detected bi-allelic likely causative PKD1 variants in 20 patients with early onset PKD. Among these, 2 were fetus, with homozygous variants, c.6096dup and c.7073T>G. 4 were neonates with homozygous variant [c.2158G>A] and compound heterozygous variants, [chr16:g.(2062497_2062600)_(2117791_2118462)del and c.6928G>A; c.4746G>A and c.8310C>G; c.4579del and c.8549C>T]. One 10 years old child with homozygous variant (c.8302G>A). Our study highlights the presence of bi-allelic variants in the PKD1 gene as a significant genetic cause of very early-onset PKD, distinct from the typical late-onset ADPKD1 associated with heterozygous mutations. Among the cases analyzed over five years, we identified seven patients with early-onset PKD carrying bi-allelic variants, including homozygous and compound heterozygous mutations. These findings expand the phenotypic spectrum of PKD1-associated disorders, emphasizing the need for comprehensive genetic testing in early-onset PKD cases to improve diagnosis and management.

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Abstract ID: 223

Androgen Insensitivity Syndrome (AIS)

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Background/ Introduction: Androgen receptor gene defects male sexual development in 46,XY individuals, leading to androgen insensitivity syndrome (AIS), an X-linked recessive condition that results in a failure of normal masculinization of the external genitalia due to the inability to respond to the male sex hormones (called androgens) due to the functionally impaired androgen receptor (AR). They may have mostly female external sex characteristics or signs of both male and female sexual development and as a result, the affected individual has some or all of the physical traits of a woman, but the genetic makeup of a man. Normal male (46,XY)sex determination relies on the presence of the Y-chromosome, specifically on expression of SRY at the appropriate time and place during gonad development hence, mutations in SRY accounts for sex reversal cases.

Objectives: 1. molecular examination of the underlying genetic factors that could be associated with patients who are diagnosed with the male to female sex reversal.

2.To investigate the role of CAG repeats as genetic background affecting the androgen insensitivity syndrome (AIS) phenotype. To the check the presence of SRY gene through sequencing and Fluorescent in situ hybridization (FISH).

Materials and Methods: samples were collected Various centers. The samples were processed and DNA extracted, the genes of interest were amplified, Sanger sequenced.

Results and Conclusions: In this study, the coding regions of SRY and AR genes were sequenced and analysed in 20 cytogenetically confirmed XY females. A reported mis-sense mutation c.733G>A, in exon 5 of the AR gene was seen in six patients of the same family in both homozygous and heterozygous condition. The SRY gene was also screened to check for any variations in these patients. FISH was performed on all six patients to check for the presence and location of SRY. All the individuals had CAG repeats that were within the normal range.

Abstract ID: 224

Achondroplasia and related Disorders

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Background/ Introduction: Skeletal dysplasias are a heterogeneous group of generalised cartilage and bone disorders. The phenotype of the diseases can range from a mild arthropathy in relatively average stature individuals to severe dwarfism with perinatal mortality. Achondroplasia (ACH) and Hypochondroplasia (HCH) are two common skeletal dysplasias. The abnormalities are however, milder in HCH than in ACH. These disorders are genetically allelic and are associated with recurrent mutations in the FGFR3 gene. Pseudoachondroplasia (PSACH) too, is a disorder of the skeletal system that clinically manifests with disproportionate short stature, lax joints and ligaments, vertebral anomalies and precocious osteoarthritis. PSACH is caused due to mutations in a gene called COMP (Cartilage Oligomeric Matrix Protein). All three disorders (ACH, HCH and PSACH) have an autosomal dominant inheritance pattern.

Objectives: To screen for the most common variants in individuals clinically diagnosed with ACH and HCH. COMP gene screening was carried out for clinically suspected cases of PSACH and those that resulted negative for ACH. Genotype-Phenotype correlation and comparison with previously reported data. Development of an NGS panel for skeletal dysplasias.

Materials and Methods: All the samples were collected at the genetic counselling clinic at Centre for Human Genetics. An elaborate family history in the form of pedigrees, medical history of the patient were obtained. The samples were processed and DNA extracted, the genes of interest were amplified, Sanger sequenced and analysed.

Results and Conclusions: In this study, two hotspot pathogenic variants, one each in ACH (Gly380Arg, which accounted for more than 97% of the cases) and HCH (Asn540Lys) were screened. Screened for mutations in the COMP gene where, a novel mutation (c.1052G>T) and a reported small deletion (c.1120-1122del) were found in two cases. Mutation screening plays a major role in making a definitive diagnosis for clinically suspected genetic disorders.

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Abstract ID: 225

MAX-Mediated Transcriptional Regulation of Pseudouridine(Ψ) Modified RNAs and Interacting RNA-Binding Proteins: Implications in Cancer Regulatory Networks

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Background/ Introduction: Pseudouridine modified mRNAs interact with RBPs and regulate various cellular processes including cell growth, proliferation and death. Ψ is found to be a potential biomarker for several types of cancers. Ψ modulates binding of various RBPs linked to cancer. However, transcriptional regulatory mechanisms coordinating these processes are still poorly understood.

Objectives: To investigate the transcriptional regulatory mechanisms connecting pseudouridine-modified mRNAs, RBPs, and cancer hallmarks, focusing on the role of MAX in oncogenesis.

Materials and Methods: We obtained the details of pseudouridine-modified sites on mRNA from available literatures and details of RBPs binding to it from ENCODE database. We used iRegulon to identify transcription factors regulating these mRNAs and RBPs. We performed functional enrichment analysis and assessed their implication in cancer.

Results and Conclusions: Our analysis identified MAX (MYC associated factor X) as a key oncogenic transcription factor involving regulation of both Ψ modified mRNAs and RBPs. It regulates oncogenic splicing factors like SRSF1, cancer associated helicase like DDX3X, DDX55 and protooncogenes like EXOSC5. MAX also regulates critical metabolism regulators like NOLC1 and ATP5E which play essential role in cancer cell metabolism and energy homeostasis. MAX mediated regulation of tumor suppressor PCBP1 and protooncogene IGF2BP3 indicates its dual regulatory influence in oncogenesis. This MAX-centered transcription regulatory network shows pseudouridine mediated cancer regulation through regulation of RBPs and RNA metabolism.This study highlights how Ψ mediated processes are linked to cancer hallmarks by modulating RBP binding. This underscores therapeutic potential of targeting MAX mediated regulatory mechanism in cancer.

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Abstract ID: 226

Hereditary spastic paraplegia due to hyperhomocysteinemia: A potentially modifiable genetic disorder

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Background/ Introduction: Hereditary spastic paraplegias (HSP) are genetically diverse disorders that cause length-dependent degeneration of the corticospinal tracts leading to progressive spasticity and weakness affecting predominantly the lower limbs. Remethylation defects leading to the accumulation of homocysteine (Hcy) are potentially treatable causes of HSP. Data on the course and outcome of these patients are limited.

Objectives: To describe the profile of patients with hyperhomocysteinemia presenting with HSP phenotype.

Materials and Methods: This retrospective study included patients with HSP phenotype and markedly elevated serum Hcy levels (> 100 µmol/L) seen in a single neurological unit between 2012 and 2024 (n=34, M:F 25:9, mean age=26.53±9.8 years). Clinical exome sequencing was performed.

Results and Conclusions: Results:

Age at onset ranged from one to 34 years. Nine patients were born to consanguineous parents. Clinical features included marfanoid habitus (n=11), cognitive decline (n=10), seizures (n=8), impaired vision (n=5), lens dislocation (n=3) and leg vein/cerebral venous thrombosis (n=3). Mean Hcy levels were 184.84±60.5 µmol/L. Nerve conduction studies showed axonopathy in six patients, visual evoked potentials were abnormal in 12. Brain MRI showed peri-ventricular hyperintensities in a majority of patients. Exome sequencing (n=21) showed MTHFR mutations in 14 (homozygous=9, compound heterozygous=5). The most common variant was c.459C>G, detected in seven patients. Other variants included c.642G>C (n=2) and c.374T>C, c.1860C>G, c.584C>T, c.456C>G, c.665C>T, c.1060C>T, c.1591_1593delAAG and c.386C>T in one patient each. Three of these patients had additional mutations in SPG7, ALDH18A1, SETX, and COL4A1. Three siblings from a single family had heterozygous

mutations in MTRR (c.147A>G) and CBS (c.1080C>T). Four patients had mutations in genes unrelated to the homocysteine biosynthetic pathway (ATP13A2, TTC37, PMP22, MARS, and HNRNPDL). All patients received vitamin B12, folic acid, riboflavin and pyridoxine, in addition to symptomatic treatment for spasticity, seizures and prothrombotic states. At follow-up (n=21), 13 patients improved partially, while two worsened due to non-compliance with medications.

Conclusion: This is one of the largest cohorts of HSP due to hyperhomocysteinemia. Identifying hyperhomocysteinemia has important implications for genetic counselling and therapeutic interventions to overcome metabolic defects and prevent cardiovascular complications.

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Abstract ID: 227

Generation of Long-Lived Lymphoblastoid Cell Lines from Patients with Chromosomal Abnormalities Using Epstein - Barr virus

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Background/ Introduction: Cell immortalization refers to cells that grow in vitro induced by or under the influence of external factors and whose growth cycle is different from that of normal cells. The trend of senescence in these cells is avoided in order to have high proliferation and long passages .B cells are the principal targets of Epstein - Barr virus (EBV) infection, primarily due to their expression of CD21, the major receptor for the virus. The EBV is a ubiquitous transforming herpes virus derived from the marmoset cell line B95-8, that infects and immortalizes EBV receptor positive B lymphocytes. This results in polyclonal activation of B lymphocytes in vitro and in vivo leads to Lymphoblastoid cell lines (LCL).

Objectives: To establish long-live, continuously proliferating B cell lines in order to retain their chromosomal abnormalities for future research by using EBV.

Materials and Methods: Heparinized peripheral blood samples are processed by density gradient centrifugation of diluted blood layered over a density gradient medium like Histopaque. This yields Peripheral Blood Mononuclear Cells (PBMCs). The PBMCs are cultured and exposed to EBV. The culture is supplemented with additives like ITSS, which supports the expression of EBNA1, EBNA2, and LMP1 proteins, facilitating the immortalization of B cells. To further support B cell proliferation, cyclosporine A is used to suppress EBV-specific cytotoxic T lymphocytes. Successful transformation is indicated by the appearance of pseudopodia, clump formation, and an increase in cell count. Once transformed, the cell lines were cryopreserved for long-term propagation.

Results and Conclusions: A panel of well-characterized LCLs derived from patients with rare chromosomal abnormalities retains the chromosomal abnormalities even after a decade of revival. This makes them useful for studying the long-term effects of such disorders. These LCLs also provide a lasting resource for future research on the genetic and immunological aspects of chromosomal abnormalities.

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mBand FISH in characterizing derivative X - Chromosome in 2 patients with Turner Syndrome

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Background/ Introduction: Turner syndrome is a genetic condition that exclusively affects females and results from the complete or partial loss of one of the X chromosomes. The typical manifestations of Turner syndrome include short stature, ovarian insufficiency, and a webbed neck, along with other features including skeletal abnormalities, heart defects, and kidney issues, resulting in a diverse array of clinical signs.

Objectives: 1. Karyotyping to confirm the genotype in 2 patients with Turner Syndrome, and 2.To characterize the derivative X- chromosome using mBand FISH.

Materials and Methods: Two patients with a Turner syndrome phenotype were referred for conventional cytogenetic testing. Heparinized blood was cultured in RPMI-1640 medium. Cells were arrested at metaphase stage with colcemid and harvested. Slides were prepared and stained using GTG banding technique. mBand FISH (multi-colour banding), an advanced cytogenetic method, was applied to identify the chromosomal rearrangement. In mBand technique, five fluorophores generate a characteristic multi-color banding pattern by quantifying fluorescence intensity ratios all along the chromosomes. Each band has a unique ratio, producing a band-specific pseudo-color, enabling accurate analysis of complex inter- and intra-chromosomal aberrations.

Results and Conclusions: The karyotype findings for case 1 showed mos 45,X[5]/46,X,der(X)[15], and for case 2, mos 45,X[12]/46,X,der(X)[3]. mBand FISH results revealed the breakpoints and structural events of the derivative chromosome, where case 1 displayed mos 45,X[5]/46,X,der(X)t(X;X)(pter—>q13::q13–>pter)[15] and case 2 exhibited mos 45,X[12]/46,X,der(X)t(X;X)(pter—>q22::p11–>pter)[3]. The condition involves partial X chromosome loss or mosaicism with characteristic Turner karyotype (45,X), giving rise to a variant of Turner syndrome. Differences in gene dosage contribute to the variation in patient phenotypes. Case 2, with a greater number of monosomy cell lines than der(X) cell lines, presents with more diverse phenotypic features, while in case 1, where monosomy cell lines are fewer than der(X) cell lines, shows fewer variant phenotypic traits. The clinical management approach includes hormone therapy, cardiological surveillance, and psychosocial care.

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Abstract ID: 230

Recurrent pregnancy loss

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Background/ Introduction: Recurrent pregnancy loss (RPL) is defined as the loss of two or more pregnancies before 20 weeks of gestation. RPL is broadly-classified as primary, secondary, and tertiary RPL. Primary RPL is experiencing multiple pregnancy losses before 24 weeks whereas secondary RPL is a pregnancy loss that occurs after one or more live births. Tertiary RPL is the pregnancy loss interspersed between live births/normal pregnancies.

RPL has a multifactorial etiology, with both genetic and non-genetic causes. Among them, parental Chromosomal Abnormalities (CA) are present in approximately 4% of the couples facing RPL and are an important causes of early miscarriages.

Objectives: This study explored the role and prevalence of CAs in RPL and examined the implications of cytogenetic causes on genetic counselling and reproductive decision-making.

Materials and Methods: This study includes 145 individuals. For chromosome analysis, peripheral blood was cultured, harvested, and dropped onto slides, Chromosomes were then stained using G-banding technique. C-banding and NOR staining were applied in cases of normal chromosomal variations.

Results and Conclusions: Of the 145 couples studied, CA was identified in 52 individuals. The prevalence of different CAs was as follows: Structural abnormalities were seen in 31 individuals, numerical abnormality was seen in 1 female and polymorphisms were seen in 8 individuals. The most common CA was reciprocal translocations, seen in 12 individuals.

This study is crucial, as understanding the significance of CA in RPL has proven valuable in assessing pregnancy outcomes for couples experiencing RPL. Therefore, our study emphasizes that cytogenetic analysis should be a standard examination in all such cases. Genetic counselling has a major implication in guiding couples with RPL regarding the recurrence risk based on the type of chromosomal rearrangement and the chromosome involved and aids in assisting couples with suitable reproductive options.

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Abstract ID: 231

Use Of Primed In-Situ Labelling (Prins) As A Molecular Cytogenetic Technique In Genome Research And Molecular Diagnostics

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Background/ Introduction: Primed in-situ labelling is a molecular cytogenetic technique that combines PCR with the localization of specific DNA sequences on chromosomes using oligonucleotide primers targeted to the specific regions of interest. Although PRINS has been used in genomic research as a molecular cytogenetic tool, it has not been adapted to diagnostics. PRINS can serve as a potential diagnostic tool for the identification of numerical abnormalities in lymphocytes, amniocytes, preimplantation embryos and in the analysis of structural aberrations, such as translocations, marker chromosomes, and ring chromosomes. It is highly sensitive and involves cytogenetic localization of chromosome specific unique and repetitive sequences, which is based on sequence specific annealing of primer, followed by chain elongation by Taq DNA polymerase. Incorporation of directly or indirectly labelled nucleotides during the reaction helps in the visualization of region of interest.

Objectives: The current study aimed at validating PRINS as a potential diagnostic tool and as a faster, cost-effective alternative to FISH in routine diagnostic procedures. The aim of the study was to create a panel of primers for the detection of autosomal and sex chromosome aneuploidies, detection of marker chromosomes and commonly observed aneuploidies in cancers.

Materials and Methods: PRINS was standardized for the detection of α satellite regions of various chromosomes and telomeric regions. PRINS was also standardized for the detection of more than one chromosome/ region on the same slide using combined reactions. The primers were validated using FISH

probes and by testing on abnormal patient samples. One of the newer approaches involved primer designing and standardization of the PRINS reaction for detection of rDNA repeats on the p arms of acrocentric chromosomes which has not been demonstrated in humans before.

Results and Conclusions: The technique has proved to be useful in research and in routine chromosomal screening as an efficient molecular cytogenetic technique for in-situ nucleic acid detection.

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Abstract ID: 232

Characterizing the impact Runs of Homozygosity on psychiatric disorder risk in Indian population using Whole-Exome Sequencing(WES)

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Background/ Introduction: Regions of homozygosity (ROH) are genomic stretches with identical alleles from both parents, often resulting from consanguinity, inbreeding, or population bottlenecks. Analyzing ROH helps identify recessive variants linked to disease. This study examines the impact of ROH and inbreeding on schizophrenia and bipolar disorder in the Indian population using WES.

Objectives: To examine the impact of ROH and inbreeding in schizophrenia and bipolar disorder in Indian population using WES.

Materials and Methods: We performed WES for n = 182 individuals using Illumina HiSeq sequencing platform.The samples were collected as part of family genomic study, and comprised of patients with bipolar disorder(BD, n = 77),schizophrenia(SZ, n = 58) in addition to healthy controls(HC,n = 47).After standard quality control,alignment was performed to reference genome hg19 using BWA v0.7.18, and variants were called using GATK HaplotypeCaller v4.5.0.The ROH segments were called from genomic variant calling format file(n = 316497 variants) using Bcftools/ROH v1.17,with default parameters, and minimum length of ROH was set as ≥1Mb.

Results and Conclusions: A greater frequency of longer runs of homozygosity was observed in families with affected individuals compared to control groups. Within the subgroups of cases,SZ exhibited a higher frequency of ROH(fROH = 0.0279) and mean ROH length(80.4 Mb) compared to BD and HC.Additionally,two different overlapping ROH segments measuring between 1-2 Mb in length were detected on chromosome 4 and 16 on SZ and BD with greater prevalence that was absent in the HC cohort.The burden analysis showed significant number of genes associated with SZ(n = 141 p.val < 0.05) within the ROH regions.

In populations with high endogamy and consanguinity, studying ROH offers insights into genetic risks for SCZ and BD. Genes in ROH may reveal the genetic architecture of these disorders, while gene set enrichment and functional analyses could uncover pathways burdened with recessive risk alleles.

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Abstract ID: 233

Prevalence of Chromosomal abnormality of couples with Primary Infertility.

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Background/ Introduction: Infertility is a growing global health disability affecting 2.6% - 10.2% of the population in India and 10% -15 % worldwide. "Primary Infertility is defined as the inability to conceive after 1 year of unprotected intercourse of reasonable frequency". It can affect both men and women and has a multifactorial etiology including genetic factor. The role of chromosomal abnormalities as a cause of infertility is becoming increasingly recognized with advances in diagnostics techniques and treatment modality. This study aims to assess the proportion of chromosomal abnormalities in couples with primary infertility.

Objectives: To evaluate the prevalence of Chromosomal abnormalities among couples with Primary infertility.

Materials and Methods: After approval from the scientific and ethics committee, 192 participants were recruited for the study. The blood samples were subjected to chromosomal analysis by G banding technique of karyotyping.

Results and Conclusions: Among the 192 study participants, 5.7% (11) participants exhibited chromosomal abnormalities. Of these 3.6% (7) had Structural chromosomal abnormality, 1%(2) had Numerical chromosomal abnormality, 0.5% (1) had structural and numerical chromosomal abnormality, 0.5% (1) had sex reversal. The prevalence of (5.7%) chromosomal abnormality in participants with primary infertility was significantly higher than reported in previous studies. Additionally, the male participant had a higher prevalence than females, highlighting the importance of genetic screening in infertility evaluations.

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Abstract ID: 235

A Case study of HLA DQ gene mutation in relation to Celiac Disease

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Background/ Introduction: The primary storage protein found in wheat grains is gluten. Gliadin and glutenin are the two primary components of gluten, which is a complex mixture of hundreds of related but different proteins. An adverse reaction to the protein gluten, which is present in wheat, barley, and rye, is a hallmark of celiac disease (CD), an autoimmune condition. This disorder, which mostly affects people who are genetically predisposed, causes inflammation and damage to the small intestine, which impairs food absorption. Fatigue and sleep issues have been associated with gluten sensitivity, especially in the context of CD. The prevalence of CD is approximately 1% globally, with symptoms ranging from gastrointestinal issues to non-gastrointestinal manifestations. There are numerous genes known to be involved in the pathophysiology, and the Human Leukocyte Antigen (HLA)is believed to play a critical role. Nearly all affected patients carry the HLA-DQA1 and HLA-DQB1 genes, which encode the HLA-DQ2 and HLA-DQ8 molecules and are the primary determinants of hereditary susceptibility. Certain variants of the HLA-DQA1 and HLA-DQB1 genes increase the risk of celiac disease. These genes provide

instructions for making proteins that play a critical role in the immune system. The human leukocyte antigen (HLA) complex is a gene family that includes the HLA-DQA1 and HLA-DQB1 genes. Certain HLA-DQA1 and HLA-DQB1 variants are present in nearly all celiac disease patients, and they appear to increase the risk of an inappropriate immune response to gliadin.

Objectives: An 8-year-old male child who presented with fatigue, sleep disturbances, and white spots on his skin was referred to us for genetic testing for food allergies. He underwent Global microarray testing to identify any food allergies. We identified an intronic variant in the HLA DQ gene.

Materials and Methods: Blood was collected from the patient and sent for the Global Screening Array (GSA) test on the Illumina platform. We analyzed the HLADQ gene.

Results and Conclusions: We have identified an intronic variant in the HLA DQ gene. We recommended Anti - gluten diet for him. After 2 weeks of following an anti-gluten diet reduced symptoms were noted, his sleeping quality was increased and also white spots were reduced.

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Abstract ID: 236

A meta-approach to predicting the impact of missense of mutations and case studies of mucopolysaccharidosis (MPS)

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Background/ Introduction: Missense mutations occur due to a single nucleotide change in the genome that leads to the substitution of one amino acid for another in a protein. These alterations can significantly affect the protein's structure and function by disrupting critical non-covalent interactions essential for its activity. Such changes may result in protein misfolding, instability, or aggregation, often associated with diseases, including neurodegenerative disorders. Several bioinformatics tools have been developed to predict the impact of missense mutations, evaluating parameters like sequence conservation, changes in local hydrophilicity, structural implications on the protein fold, and alterations of key interactions at the protein's active site. However, these tools provide only a partial view of the mutational effects due to their specific strengths and limitations.

Objectives: To develop a comprehensive meta-approach that integrates various predictive methods into a unified platform for a holistic analysis of missense mutations.

Materials and Methods: We developed MutXplor (http://bts.ibab.ac.in/mutXplor.php), a bioinformatics tool that integrates online and offline mutation prediction tools. MutXplor uses web scraping techniques to gather data from multiple online tools and incorporates offline tools via a custom script, eliminating the need for users to individually run multiple applications. This system generates an all-encompassing mutational report that can be used to develop datasets for machine-learning models. The resulting data, including features such as protein physicochemical properties and sequence conservation at the mutation site, were applied to build a predictive model for phenotypic outcomes in mucopolysaccharidosis patients. The predictive model is hosted on Meta-Predictor (http://bts.ibab.ac.in/predictor.php).

Results and Conclusions: MutXplor streamlines mutation analysis by generating comprehensive mutational reports that can be for used for machine learning model for phenotype prediction. Meta-

Predictor leverages these reports to enhance genotype-phenotype predictions, especially for mucopolysaccharidosis. This approach demonstrates the potential of integrating multiple bioinformatics tools for a more robust and precise mutation impact analysis, advancing both basic research and clinical applications.

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Abstract ID: 237

BMAL1 Underexpression and Sleep disturbances in Autism Spectrum Disorder: Exploring Circadian and Biological correlates

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Background/ Introduction: Sleep disturbances are commonly reported in Autism Spectrum Disorder (ASD) with a prevalence of 40-80% compared to typically developing children. Sleep, immune and circadian dysfunction are interlinked and probably play a crucial role in ASD pathophysiology.

Objectives: 1. To assess sleep abnormalities and quantify the levels of expressions of inflammatory, circadian & sleep homeostatic pathway-related genes in ASD children and compare with Healthy Controls (HC). 2.To correlate the gene expression levels with sleep dysfunction in ASD & its severity.

Materials and Methods: A prospective, case-control study was conducted in Departments of Neurology, Child and Adolescent Psychiatry and Human Genetics, NIMHANS, Bengaluru between June 2022- May 2024 after obtaining ethical approval. Sleep disturbances were screened using Children's Sleep Habits Questionnaire (CSHQ). Plasma samples from 57 ASD children and 56 healthy controls aged 2-10 years were collected with informed consent. Gene expression of 5 genes (Circadian-BMAL1 and CLOCK, Sleep homeostasis-Orexin, Inflammatory-STAT3 and IL6) was quantified using qPCR and ddPCR(IL-6). Statistical analysis was done by biostatistics department.

Results and Conclusions:

Results: This study found sleep dysfunction in 66.7% of ASD children and 14.3% of healthy controls ($p < 0.001$). BMAL1 gene showed significant underexpression in ASD children ($p = 0.02$), with BMAL1 correlating positively with CLOCK ($p = 0.046$) and Orexin ($p = 0.022$). Orexin also positively correlated with CLOCK ($p = 0.006$) and IL-6($p < 0.001$). No significant differences were found for CLOCK, Orexin, IL-6 and STAT3 gene expressions, nor correlations between gene expressions and sleep dysfunction in either group.

Discussion: Sleep dysfunction was significantly more prevalent in ASD children. Differences in BMAL1 expression and its correlations with CLOCK and Orexin suggest these genes might influence sleep disturbances in ASD, though no direct link to sleep dysfunction was found.

Conclusion: BMAL1 gene has a significant role in ASD's molecular mechanisms and its correlation with other genes implies the potential therapeutic targets for sleep disturbances in ASD.

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Abstract ID: 238

Dietary Interventions in Classical Phenylketonuria with an indigenously manufactured metabolic diet

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Background/ Introduction: Classical Phenylketonuria (PKU) OMIM #261600 is caused by bi-allelic mutations in PAH gene on chromosome 12q. Affected new-borns appear normal at birth but soon develop progressive neurological problems, seizures and depigmentation of skin and hair without treatment. Early introduction of a Phenylalanine (PA) restricted diet can prevent most of the complications. We describe our experience in treating PKU with a low-cost indigenously manufactured metabolic diet.

Objectives: To assess and follow up children affected with PKU on dietary management.

Materials and Methods: 60 affected children were diagnosed with classical and variant PKU from a southern Indian centre over ten years (2013 – 2023). 22 affected with classical PKU diagnosed between 9days – 5years age have been receiving regular dietary therapy and metabolic review. Metabolic diet by Metanutrition PKU (Pristine Organics, Bengaluru) was commenced along with diet calculated by a specialist metabolic dietician (2/3rd of protein requirement through formula and 1/3rd through naturally occurring proteins, divided into 4 – 6 daily feeds. Regular monitoring for compliance and PA monitoring continues. PA values at diagnosis, at 6m and 1y after starting therapy were documented (Table 1).

Results and Conclusions: 22 children (9 male, 13 female) from 12 families were included. Parental consanguinity was present in 10/12 of families. Genetic studies confirmed PAH mutations in all. Therapy was begun in two within the first month with reduction and near normal blood PA at one-year. The others showed variably reduced values (Table 1). Clinical and behavioural improvement was noted in most with 15/22 able to attend mainstream school in later years. The indigenously manufactured PKU diets available at a fraction of the cost of imported products appear promising with lowering of blood PA values and clinical improvement. Metanutrition products are the only Indian diets to have secured FSSAI approval.

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Abstract ID: 239

Clinico-genetic profile of inborn errors of cobalamin metabolism: a series of 11 cases from a tertiary care hospital

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Background/ Introduction: Disorders of intracellular cobalamin metabolism, or cobalaminopathies, have a variable phenotype and age of onset that are influenced by the severity and location within the pathway of the defect. The common varieties are methylmalonic acidemia, cblC type, and homocystinuria. Dysfunction of any step of cobalamin metabolism causes elevated levels of homocysteine, methionine or methylmalonic acid in serum/ urine leading to their toxic accumulation and diverse clinical manifestations.

Objectives: To describe the various clinical features, imaging and biochemical abnormalities in a cohort of patients with defects in cobalamin metabolism.

Materials and Methods: We retrospectively collected data on genetically confirmed cases of cobalaminopathy in patients under 18 years of age who presented to our centre between 2018 and 2023, including clinical history and examination, biochemical investigations, MRI brain findings, and treatment details.

Results and Conclusions: Eleven cases were included, out of which seven were female. Two cases had a cystathionine beta synthetase (CBS) gene mutation, 7 had methylene tetrahydrofolate reductase (MTHFR) mutation, and 1 each of MMACHC and MMUT. The age at diagnosis ranged from 7 months to 15 years. Nine patients had developmental delay and intellectual disability, four had progressive cognitive decline, and four had seizures. Ophthalmological manifestations like optic atrophy and poor vision were noted in three, and one had ectopia lentis. Two patients had marfanoid habitus, five had dysmorphic facies, and one had kyphoscoliosis. Hypopigmented hair was seen in two, while diffuse osteopenia, in one. Seven cases showed diffuse cerebral atrophy and confluent T2/FLAIR white matter hyperintensities, one had multiphasic infarcts with bilateral ICA stenosis and one had normal MRI Brain. Serum homocysteine was elevated in all, methionine was elevated in two cases, and one case showed elevated methylmalonic acid (MMA).

Conclusion: Defects of cobalamin metabolism are rare metabolic disorders with a wide spectrum of clinical manifestations which if identified early, are potentially treatable.

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Abstract ID: 240

Structural and Functional modification of Long Noncoding RNAs induced by Single Nucleotide Polymorphism

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Background/ Introduction: Long noncoding RNAs (lncRNAs) are the category of regulatory noncoding which are widespread throughout the genome. From recent research, they are found to be the key element for the functioning of cellular transcriptomes. They execute most of the function by interacting with RNA Binding Proteins (RBPs). Genome wide association studies (GWAS) has reported that noncoding regions of the genome include a large number of the single nucleotide polymorphism (SNP), a mutational phenomenon where a single nucleotide is replaced by another nucleotide at specific genomic position. The noncoding RNAs also emerged from the noncoding region of the genome. So, transcription of lncRNA containing SNP may lead to various detrimental diseases. This makes the SNP study crucial to decipher the possible mechanism behind the association among SNP, lncRNA, and RBP.

Objectives: In our study, the impact of SNP was investigated on the lncRNA configuration. Apart from that, SNP-induced RNA-RBP remodelling was also studied.

Materials and Methods: For structural prediction of lncRNAs with or without SNP, softwares like RNA Structure, RNAFold and CentroidFold were employed. RBPs interacted with lncRNAs were obtained from Encyclopedia of RNA Elements (ENCORE) data analysis. MPRDock molecular docking was implemented to study RNA-RBP binding.

Results and Conclusions: From in silico work, SNP-induced structural alterations are found in the lncRNA, which are validated by above mentioned RNA structure prediction softwares. RBPs interacting with lncRNAs were obtained by analyzing the published data acquired from the ENCORE. From

molecular docking, the SNPs are found to be associated with the remodelling of RNA-RBP interaction. This ultimately affects the functions performed by lncRNAs. Therefore, the interconnection among SNP, lncRNA and RBP is essential to understand the underlying cellular mechanism of disease pathogenesis.

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Abstract ID: 241

Interphase Chromosome Profiling (ICP), A Novel cytogenetic technique

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Background/ Introduction: Identifying chromosomal abnormalities is crucial for effective management. Over time, techniques like karyotyping and fluorescent in situ hybridization (FISH) have been developed to detect such abnormalities. However, molecular genetic testing has advanced the field by offering faster, more sophisticated methods, overcoming the limitations of traditional cytogenetic techniques. Interphase chromosome profiling helps us in identifying numerical and structural abnormalities. Using a combination of FISH probes along the length of the chromosome on nuclei allows us to study the entire chromosome. Interphase chromosome profiling probes will have 4 to six hybridization sites along the length of chromosome in an equidistant manner.

Objectives: • Standardization of Interphase Chromosome Profiling (ICP) for normal control samples (PB). • ICP to screen for chromosomal aberrations and validating ICP as a diagnostic method for detection of numerical and structural chromosomal abnormalities.

Materials and Methods: Samples were cultured to attain best slide preparations. Slides can be prepared from directly processed samples or from cultured samples. Cells were cultured using RPMI 1640 medium. Colcemid was added and incubated for one hour and harvested in the metaphase stage. Samples were dropped on slides and hybridised using ICP probes. Post hybridisation wash was carried out the next day, counterstained using DAPI and analysed using fluorescence microscope. Although ICP probes are designed for interphase nuclei, we have attempted ICP on both interphase nuclei and metaphases for the better understanding of banding patterns.

Results and Conclusions: All the samples showed a desired signaling pattern corresponding to the particular chromosome. To better understand the benefits and disadvantages of the test, we have used a combination of possible conventional and molecular cytogenetics techniques. We used karyotyping and ICP on both interphase cells and metaphases. The combination of different techniques proved ICP, as a test is reliable and was concordant with our other results.

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Abstract ID: 242

X-Linked Leigh's Disease in Siblings with PDH Complex Deficiency: A Case Report

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Background/ Introduction: Leigh's disease (LD), one of the most common phenotypes of mitochondrial disorders, is usually autosomal recessively inherited. X-linked inheritance is rarely reported.

Objectives: To describe two siblings with X-linked LD due to a pathogenic variant in PDHA1.

Materials and Methods: Deep phenotyping, which included clinical, biochemical, electrophysiological and radiological characterization, and clinical exome sequencing.

Results and Conclusions: These siblings from West Bengal in India were born to non-consanguineous parents with unremarkable antenatal and perinatal periods. Patient 1 (proband), a four-year-old boy, had global developmental delay, left-hand preference, paroxysmal non-kinesogenic dyskinesia, ataxic gait, falls, ankle contractures, hyporeflexia and extensor plantars. At 22 months of age, he developed fever-triggered encephalopathy, seizures and quadriparesis followed by orofacial dystonia. Patient 2 (older brother), a 13-year-old boy, manifested with mild developmental delay, fever-triggered encephalopathy, orofacial dystonia and quadriparesis. Examination revealed pes cavus, hammer toes, ankle contractures and hyporeflexia. Elevated plasma lactate, creatinine kinase and alanine levels, sensorimotor axonal polyneuropathy on nerve conduction studies, and bilaterally symmetrical hyperintensities in the globus pallidi on brain MRI were noted in both patients. Patient 2 had additional hyperintensities in the substantia nigra, and dentate nuclei. Exome sequencing revealed a hemizygous missense variant in exon 4 of PDHA1 (c.328C>T, p.Arg110Cys) in both patients. This variant had previously been reported as pathogenic and associated with Pyruvate Dehydrogenase Complex deficiency (PDCD) in the ClinVar database. Muscle biopsy (Patient 2) revealed neurogenic atrophy and reduced mitochondrial respiratory chain complex I activity (24%). Patient 1 showed mild improvement in dystonia, dysarthria, and gait with mitochondrial cocktail at 3 years follow-up.

Conclusion: PDCD primarily impacts carbohydrate metabolism, leading to a complex clinical picture and disproportionate involvement of the central nervous system and LD phenotype. Our case highlights the X-linked inheritance pattern of LD. Further research into targeted therapies, including metabolic and gene-based treatments, is essential to enhance outcomes for individuals with PDCD.

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Abstract ID: 243

VEGFA Polymorphisms in the Pathogenesis of Diabetic Retinopathy: A Systematic Review and Meta-Analysis

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Background/ Introduction: Diabetic retinopathy (DR) is a leading cause of visual impairment, particularly among individuals with long-standing diabetes mellitus. While hyperglycaemia and disease duration are well-established risk factors, genetic predispositions significantly contribute to DR development. DR, one of the most prevalent microvascular complications of diabetes, is characterised by retinal microvasculopathy. Angiogenesis, a hallmark of advanced DR stages, plays a critical role in disease progression, often leading to visual impairment. Among the numerous factors involved in angiogenesis, vascular endothelial growth factor A (VEGFA) has emerged as a key mediator. VEGFA induces pathological vascular permeability, disrupts the blood-retinal barrier, and promotes neovascularisation, all of which contribute to DR progression. Despite extensive research into VEGFA

and its genetic variants, inconsistencies in findings highlight the need for a systematic evaluation of its role in DR susceptibility and pathogenesis.

Objectives: This study aims to systematically evaluate the association between VEGFA single nucleotide polymorphisms (SNPs) and DR susceptibility, while considering population-specific differences. This meta-analysis identifies significant polymorphic variants associated with DR.

Materials and Methods: A systematic review was conducted using all relevant published literature available to date. Data from genetic association studies focusing on VEGFA SNPs relevant to DR were extracted and analysed. Publication bias evaluations were undertaken to ensure robust conclusions.

Results and Conclusions: This study identified rs2010963 and rs833061 as two extensively studied VEGFA polymorphisms located in the upstream region of the VEGFA in relation to diabetic retinopathy. Our meta-analysis shows that the rs833061 polymorphism is significantly associated with DR, suggesting that the C allele increases susceptibility. VEGFA polymorphisms, particularly the upstream region polymorphism, rs833061, play critical roles in DR pathogenesis by modulating VEGFA expression and activity. The findings underscore the importance of genetic variability in the development of DR. These insights could inform personalised approaches to DR prevention and management.

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Abstract ID: 244

Gene-FCCSS: A ressource for investigating genetic factors predisposing to iatrogenic pathologies in childhood cancer survivors

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Background/ Introduction: With the increasing number of childhood cancer survivors, iatrogenic effects are becoming a very important issue. The French Childhood Cancer Survivors Study (FCCSS) focused on the late effects of childhood cancer treatments with the longest follow-up. All patients included in the FCCSS study have a whole-body radiation dose reconstruction and detailed clinical, therapeutic and follow-up data.

Objectives: The Gene-FCCSS project has been set up to sequence and analyse the genetic data from FCCSS. The main objective is to identify genetic susceptibility factors for cancerous and non-cancerous pathologies following treatment for childhood cancers and to propose polygenic risk scores (PRS) for better personalised treatment of childhood cancers.

Materials and Methods: Whole genome sequencing was completed in December 2024, on saliva or blood samples from 2673 FCCSS patients.

Results and Conclusions: Sequencing depth is 30X on average, and greater than 15X for more than 99% of patients. The most common types of cancer were renal tumours (15%), neuroblastoma (12%) and Hodgkin's lymphoma (8%). Of these, 1518 were treated with radiotherapy and 2130 with chemotherapy for their childhood cancer.

Analyses will focus on genetic susceptibility to different treatment side effects, such as the risk of a second primary neoplasm (n=440), grade 3 heart disease (n=240), treated diabetes mellitus (320), severe ototoxicity (n=300), surgically treated cataract (n=60), chronic renal failure (n=70) and stroke (n=110). The final objective of the association analysis will be to propose a PRS for each phenotype tested and/or to validate the PRS already published.

In conclusion, Gene-FCCSS is currently the largest deep European cohort of childhood cancer survivors including constitutional WGS data and focusing on iatrogenic effects.

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Abstract ID: 246

Impact of gene-gene interaction of IFN-γ/ IL12 immune-axis in Progressive Supranuclear Palsy (PSP)

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Background/ Introduction: Progressive Supranuclear Palsy (PSP) is a rapidly progressive primary tauopathy that constitutes the third most common form of neurodegenerative disease. PSP is mainly characterized by vertical gaze palsy, repeated unprovoked falls and mild Dementia. Systemic and neuroinflammation play important roles in PSP. Peripheral levels of interleukin 12 (IL-12) and interferon-gamma (IFN-γ) were associated with cognitive impairments in tauopathy. However, the impact of genetic variations and gene-gene interactions of the elements of the IL-12/IFN-γ axis has not yet been studied in PSP.

Objectives: To investigate the impact of IFN-γ/ IL12 immune-axis by examining the genetic variation of IL12B, IL12RB1, IFNGR1, IFNGR2, and ISG15 genes and interactions amongst these genes in Indian patients with sporadic PSP.

Materials and Methods: A case-control study was conducted on PSP patients of Indian origin using a non-random purposive sampling procedure. PSP patients (n=95) fulfilling the MDS criteria for PSP and healthy controls (n=145) were recruited. A total of 8 single nucleotide polymorphism (SNPs) located within IL12B (rs3212227), IL12RB1 (rs436857 and rs393548), IFNGR1 (rs1327474 and rs2234711), IFNGR2 (rs1059293), and ISG15 (rs15842 and rs1921) genes were selected in this study based on functional significance. Genotyping was performed by TaqMan biallelic discrimination assay. The differences in the frequencies in the genotypes between groups were checked by SNPStats. The linkage disequilibrium analysis was performed in the SHEsisPLUS online version. Gene-gene interactions were analyzed using multi-dimensionality reduction tests (MDR 3.0.2).

Results and Conclusions: The distribution of observed genotypic frequencies of all the SNPs was consistent with the Hardy-Weinberg equilibrium (HWE). The frequency distribution of each genotype of all the SNPs was checked between the groups using five inheritance models (codominant, dominant, recessive, overdominant and log-additive). However, statistically significant differences in the frequencies of genotypes were not detected between PSP and controls. A strong LD was observed between rs436857 and rs393548 (D´=0.96), rs1327474 and rs2234711 (D´=1.0), and rs15842 and rs1921

(D´=0.99). Synergistic epistasis interaction was observed within rs1921 (ISG15) and rs393548 (IL12RB1), rs1921 (ISG15) and rs2234711 (IFNGR1), rs3212227 (IL12B) and rs393548 (IL12RB1).

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Abstract ID: 249

Epistatic interactions of MAPT, STX6, MOBP, and EIF2AK3 genes in Progressive Supranuclear Palsy: New genetic perspectives in the Asian Indian population

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Background/ Introduction: Progressive supranuclear palsy (PSP) is a heterogeneous neurological disorder classified as an atypical Parkinsonian syndrome that significantly affects movement, gait, balance, speech, vision, and cognition. Genes play a crucial role in determining the risk of PSP, with major risk genes such as MAPT, STX6, MOBP, and EIF2AK3 identified across diverse ethnic groups. However, the impact of the interactions among these genes on the risk of PSP remain unexplored.

Objectives: To investigate the role of the gene-gene interactions among MAPT, STX6, MOBP, and EIF2AK3 genes in Indian patients with sporadic PSP.

Materials and Methods: A prospective case-control study was conducted on patients with PSP of Indian origin. PSP patients (n=106) fulfilling the MDS criteria for PSP and healthy controls (n=109) were recruited for the study. A total of 12 single nucleotide polymorphism (SNPs) located within MAPT (rs1467967, rs242557, rs3785883, rs2471738, rs8070723, rs7521, rs12185268, rs62063857), STX6 (rsrs3747957, rs1411478), MOBP (rs1768208), EIF2AK3 (rs7571971) genes were considered for this study based on functional significance. Genotyping was performed using TaqMan biallelic discrimination assay in a QuantStudio 6 Flex Real-Time PCR platform (Applied Biosystem, USA). Gene-gene interactions were analyzed using multi-dimensionality reduction (MDR) (version 3.0.2).

Results and Conclusions: We found significant synergistic interactions within the MAPT gene between i) rs1467967 and rs244557 (1.37%), ii) rs244557 and rs3785883 (1.64%), iii) rs3785883 and rs7521 (1.13%), and iv) rs3785883 and rs2471738 (1.39%) genetic variants. Besides, a strong synergistic gene-gene interaction was observed between rs7521 (MAPT) and rs1768208 (MOBP) (1.05%), and moderate synergistic interactions were found between i) rs1768208 (MOBP) and rs1411478 (STX6) (0.84%), ii) rs1768208 (MOBP) and rs3785883 (MAPT) (0.63%). Interestingly, our analysis revealed that rs7571971 (EIF2AK3) and rs1411478 (STX6) were statistically significant in distinguishing females and males, respectively. The present findings for the first time suggest strong synergistic interactions among MAPT, STX6 and MOBP genes in modulating the risk of PSP.

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Abstract ID: 251

Transformation of Quality of Life of Alzheimer's disease Cases by Yoga Therapy

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Background/ Introduction: Alzheimer's disease (AD) constitutes a formidable challenge, profoundly affecting the fabric of individuals' lives and intricately entwined with disturbances in their quality of life.

Objectives: Our aim is to unravel the impact of a yoga intervention on their overall quality of life with AD and mild cognitive impairment subjects.

Materials and Methods: A case control yoga interventional study was conducted on 30 subjects (male-18 and female-12) were enrolled from the department of neurology and neurocognitive assessments were done in department of Anatomy, AIIMS New Delhi, India. The multifaceted evaluation encompassed the Geriatric Depression Scale (GDS) including the evaluation of 15 questions and the Montreal Cognitive Assessment (MoCA) scale including for overall quality of life assessment with measuring language, memory, attention, visuospatial, naming, delayed recall, abstraction, and orientation for quality-of-life of the AD subjects.

Results and Conclusions: The AD subjects cohort exhibited statistically significant enhancements ($P < 0.001$) in quality-of-life scores (GDS & MoCA) pre and post Yoga intervention. GDS scores of 15 questions experienced a transition from pre-yoga (8.36 ± 2.7) to post-yoga (5.13 ± 3.0). While total MoCA scores ascended from pre yoga (18.65 ± 4.13) to post yoga (25.06 ± 6.3). MoCA scores of individual points pre yoga includes language (1.10 ± 0.2), memory (no points), attention (3.54 ± 1.8), visuospatial (4.0 ± 1.30), naming (2.16 ± 0.4), delayed recall (3.55 ± 0.25), abstraction (0.50 ± 0.0), and orientation (3.80 ± 0.18). MoCA scores of individual points post yoga includes language (2.16 ± 0.6), memory (no points), attention (4.60 ± 1.3), visuospatial (4.90 ± 2.50), naming (2.85 ± 0.75), delayed recall (4.85 ± 0.50), abstraction (1.40 ± 0.20), and orientation (4.30 ± 0.45). This discerning study illuminates the transformative potential of yoga, showcasing significant enhancements in the quality of life of individuals grappling with mild to moderate Alzheimer's disease.

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Abstract ID: 252

Possible reasons and outcomes associated with no-call results on Non-invasive Prenatal Screening

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Background/ Introduction: NIPT is a screening test that utilizes cell-free DNA (cfDNA) to screen for fetal chromosomal aneuploidies. The fetal cfDNA found in the maternal plasma originates from the placental trophoblast and is commonly referred as fetal fraction (FF). FF is a critical parameter used while analysing NIPT results. Occasionally, a result cannot be provided due to inadequate FF or other laboratory-related reasons (post-analytical failures). Various reasons have been known to affect FF such as maternal weight, gestational age and maternal medications (low molecular weight heparin). In cases with low FFs, it has been suggested that there is an increased risk (4.7% - >17%) for fetal aneuploidy and the guidelines (ACMG, 2016) recommend that a second redraw sample may not be advisable, instead diagnostic testing should be offered.

Objectives: To understand the possible reasons for no-call results on NIPT and the possible results on repeat samples.

Materials and Methods: Retrospective analysis was performed on NIPT samples with no call results. The results were recorded and tabulated accordingly.

Results and Conclusions: Overall, 2.1% of NIPT results were reported as no-call on the first sample during the study period. In 74% of cases a repeat sample was sent. Low risk was obtained for 80.9% of samples on second redraw and in about 15.1% of cases a conclusive result could not be provided. About 2% cases, high risk result was obtained on repeat sample. This study demonstrates that a repeat sample can be offered to pregnant women with a no call result on first sample. The decision to submit a repeat sample should be made after thorough evaluation of the background risk while considering the gestational age and maternal weight.

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Abstract ID: 253

Complex Chromosomal Abnormalities: A Single Case with Balanced Translocation, Mosaicism, and Ring Chromosome

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Background/ Introduction: Turner syndrome (TS) is a genetic disorder affecting approximately 1 in 2,500 live female births, typically caused by the complete or partial absence of one X chromosome. It is characterized by short stature, ovarian insufficiency, infertility, elevated gonadotropins (FSH, LH), and low anti-Müllerian hormone (AMH) levels. While classic TS is typically a 45,X karyotype, some individuals exhibit complex chromosomal abnormalities, this case discusses a 27-year-old woman with secondary amenorrhea, absent ovaries, a hypoplastic uterus, and undetectable AMH levels, leading to a genetic evaluation that revealed complex chromosomal rearrangements.

Objectives: This case highlights the importance of genetic testing in TS patients, especially when complex chromosomal abnormalities like mosaicism, translocations, and ring chromosomes are suspected.

Materials and Methods: The patient underwent genetic evaluation for secondary amenorrhea. Karyotyping using GTG banding revealed a mosaic pattern with 45,X and 46,X,r(X) cell lines, along with a translocation between chromosomes 2 and 11. FISH confirmed the X ring chromosome, and CMA showed no additional chromosomal imbalances.

Results and Conclusions:

Genetic analysis confirmed a complex mosaic karyotype: mos45,X, t(2;11)(q37.1;q13.1)/46,X,r(X), t(2;11)(q37.1;q13.1), with 45,X and 46,X,r(X) cell lines, and a translocation between chromosomes 2 and 11. FISH confirmed the X ring chromosome, and CMA revealed no chromosomal imbalances. Imaging studies showed a hypoplastic uterus and absent ovaries. Laboratory tests indicated undetectable AMH and elevated FSH/LH levels, confirming premature ovarian failure and supporting the diagnosis of TS with complex chromosomal rearrangements.

This case highlights the significance of a thorough diagnostic approach in patients with suspected TS, especially when mosaicism and complex chromosomal abnormalities are present. Traditional karyotyping is crucial for identifying structural anomalies, such as ring chromosomes and translocations, that may not be detected by chromosomal microarray. FISH is essential for confirming ring chromosomes. A multidisciplinary approach, combining clinical, genetic, and imaging evaluations, is vital for accurate diagnosis and tailored management of TS and other chromosomal disorders.

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Abstract ID: 255

Origin, Evolution, and Pathophysiological Diversifications of Vertebrate α1-antitrypsin-like Serpins

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Background/ Introduction: The α1-proteinase inhibitors (α1-PIs), originating over 500 million years ago, diversified through gene duplications, playing crucial roles in regulating inflammation, coagulation, and RAAS, reflecting critical vertebrate system evolution.

Objectives: To study the detailed evolutionary origin of α1-proteinase inhibitors focusing on details of functional overlaps and pathophysiological details of these proteinase inhibitors

Materials and Methods: Methods: Using comparative genomics and bioinformatics analyses of α1-proteinase inhibitors aided by ENSEMBL Genome browsers, homology detection tools and MEGA-X.

Results and Conclusions: Results: We found that this locus duplicated into three: one harbored non-inhibitory AGT, while the other expanded by tandem duplications, evolving from a single serpin (lampreys) to three (elephant shark), five (Fugu), seven (birds and frogs), and 11 (human). The ancestral locus harbored α1-antitrypsin-like genes, with AGT transitioning from inhibitory (GTEAKAETVVGIMPI†SMPPT) in lampreys (Lethenteron, Lampetra, Petromyzon) to non-inhibitory in vertebrates like tilapia (GAEPQDPTQEEGVPL†KLSIN), flycatcher, turtle, and human over 500 MY.

We further created three distinct sub-groups within vertebrate group V2 serpins:

(a) V2_I: α1-antitrypsin-like genes arose by tandem duplications and functional diversifications via RCL mutations, including α1-antitrypsin (SERPINA1), α1-antitrypsin-related protein (SERPINA2), antichymotrypsin (SERPINA3), kallistatin (SERPINA4), protein C inhibitor (PAI3/SERPINA5), corticosteroid-binding globulin (SERPINA6), thyroxine-binding globulin (SERPINA7), centerin (SERPINA9), protein Z-dependent proteinase inhibitor (SERPINA10), Vaspin (SERPINA12), and two uncharacterized serpins (SERPINA11, SERPINA13).

(b) V2_II: AGT, the primary carrier of active angiotensin hormones in the renin-angiotensin system regulating fluid homeostasis, blood pressure, and vascular formation.

(c) V2_III: Heparin factor II (HCII), thrombin-inhibitory with glycosaminoglycans, conserved in vertebrates, embedded inside PIK4CA's largest intron.

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Abstract ID: 256

An Integrated Analysis of Genetic Alterations in Breast Cancer Signaling Pathways

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Background/ Introduction: Breast cancer (BC) is the most diagnosed cancer and the leading cause of death among women. Abnormal signaling due to gene mutations in signaling pathways contributes to the progression of normal cells to BC cells. To explore mechanisms associated with BC, large-scale analyses are required to map gene mutations to cellular pathways.

Objectives: To perform an extensive pathway and protein-protein network analysis of BC genes to identify key mutations and signaling pathways involved in BC progression and identify potential therapeutic targets.

Materials and Methods: Mutational data from 18,373 genes across 9,555 BC samples was collected via cBioportal. After filtering genes with mutational counts >40, a list of 1,174 BC genes was generated. Pathway and protein-protein interaction analyses were performed using EnrichR and STRING 11, respectively. Genes were classified into five tiers based on mutational frequencies.

Results and Conclusions: BCtier_I genes (mutational frequencies >5%): 12 genes, including PIK3CA (35.7%), TP53 (34.3%), and others. Pathway analysis revealed top pathways: PI3K-AKT, TP53, NOTCH, HIPPO, and RAS pathways. Mutational hotspots of several BC genes were identified, including shared genes across BC panels (e.g., BRCA1, BRCA2, TP53). These findings emphasize the role of genetic perturbations in BC signaling and protein interaction networks, highlighting their importance in the development of targeted therapeutic strategies.

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Abstract ID: 257

Impact of genetic and epigenetic changes within MAPT gene on the risk of Progressive supranuclear palsy (PSP)

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Background/ Introduction: Progressive supranuclear palsy is a rare and rapidly progressive neurodegenerative disorder. The symptoms include postural instability, falls, ocular motor dysfunction, cognitive decline. The microtubule-associated protein tau (MAPT) gene has emerged as major genetic determinant of PSP. The genetic landscape and DNA methylation changes at MAPT gene locus in Indian population remain unexplored.

Objectives: To investigate the impact of MAPT gene variation and DNA methylation in promoter region of MAPT gene on the risk of PSP in Asian-Indian population.

Materials and Methods: A total of 106 patients, 109 controls were recruited. Six SNPs in MAPT gene (rs1467967, rs242557, rs3785883, rs2471738, rs8070723 and rs7521) were selected. Genotyping was performed using TaqMan biallelic discrimination assays. Sequencing of Exon1, Exon10, and Exon10 flanking intronic regions of the MAPT gene was carried out by Sanger sequencing. Genomic DNA samples were bisulfite-converted and MAPT promoters upstream of Exon0 and between Exon0 to Exon1 were sequenced. Genotype associations were assessed using Pearson's chi-square test. Linkage disequilibrium and haplotype analysis were performed by SHEsisPLUS

Results and Conclusions: Significant differences in the frequencies of AG ($p<0.001$) and GG ($p<0.001$) genotypes of rs1467967 and AA genotype ($p=0.044$) of rs242557 of the MAPT gene were observed between patients and controls. Strong LD was observed among rs2471738, rs1467967 and rs7521 in our data. Risk associations were identified with three MAPT haplotypes (H1c, H1o, and H1b). H1c and H1o sub-haplotypes showed an increased risk, while H1b was linked to a decreased risk of PSP. However, we didn't observe any known mutations in the Exon1, Exon10, and Exon10 flanking intronic regions of the MAPT gene in Indian PSP patients. No significant differences in promoter DNA methylation patterns were observed between patients and controls. This is first Indian study for MAPT observed in European and Caucasian populations. The novel genotypes and haplotypes added further dimension for understanding of MAPT-related risk of PSP.

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Abstract ID: 258

Alterations of gut-immune axis in Guillain Barre Syndrome: A metagenomic study

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Background/ Introduction: Guillain-Barre Syndrome (GBS) is an acute, and immune-mediated-monophasic disease of the peripheral nervous system. Among several risk factors, Campylobacter jejuni is the major trigger for GBS. Various immune cells and molecules have been suggested to drive the pathogenetic pathways of GBS. Notably, gut microbiota has appeared important in preventing colonization of the pathogenic microorganisms as well as in maintaining immune homeostasis. The role of gut microbiota and its cross-talk with immune cells, especially mucosal-associated invariant T (MAIT) cells have not been explored yet in GBS. However, the precise mechanism underlying disease etiology and progression is not properly understood.

Objectives: To evaluate the interaction between gut microbiota and MAIT cells in the immunopathogenesis of GBS.

Materials and Methods: A cross-sectional study design was followed and 28 GBS patients and 20 healthy controls (HC) were recruited. Immunophenotyping of MAIT cells was performed on the BD Lyric platform. Whole genome metagenomic sequencing was performed by Next generation sequencing to assess the gut microbiota profile. Quantification of the expression levels of RORC, PLZF, and T-bet genes by qPCR and measurement of plasma levels of IL-17A, IFN- γ, and TNF-α by multiplex suspension assay was carried out in a subset of study participants (GBS=9, HC=10).

Results and Conclusions: Result: A significantly decreased percentage of circulating MAIT (TCR Va 7.2+ CD161+) cells (p=0.010) and activated MAIT cells(p=0.09) were observed in GBS compared to HC. A significant reduction in the abundance of gut microbiota belonging to the Firmicutes and Actinobacteria phyla was observed in GBS compared to HC.

Conclusion: This study, for the first time, demonstrates a significant role in the altered frequency and function of MAIT cells and gut microbiota in GBS.

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Abstract ID: 259

Polysomy of CEP8 Correlates with Tumor Mitotic Activity in Breast Carcinoma: Analysis of 45 FFPE Sample

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Background/ Introduction: Chromosomal instability, particularly polysomy, is associated with prognosis and treatment response in breast cancer. Polysomy refers to the presence of extra copies of a chromosome, which can influence cancer progression. Chromosome 8 (CEP8) polysomy is of particular interest due to its significant clinical implications in breast cancer, affecting prognosis and therapeutic strategies. However, the molecular mechanisms linking CEP8 polysomy to breast cancer pathology remain poorly understood.

Objectives: 1. To investigate the prevalence of polysomy involving chromosome 8 (CEP8) in various histological subtypes of breast cancer, including invasive ductal carcinoma (IDC) and ductal carcinoma in situ (DCIS) with microinvasion.

2. To analyze the correlation of CEP8 polysomy with mitotic activity of tumor.

Materials and Methods: This study employed retrospective and prospective analyses of formalin-fixed paraffin-embedded (FFPE) breast carcinoma tissues. Tumor sections were prepared for fluorescence in situ hybridization (FISH) using fluorescent-labeled DNA probes specific for c-MYC genes and their centromeric region (CEP8). Tissue samples were deparaffinized, digested enzymatically, and hybridized with the probes. Hybridization signals were visualized and enumerated using a Leica fluorescence microscope by two independent investigators in at least 50 intact tumor cell nuclei per sample. Polysomy was assessed by the average centromere signal count, while MYC amplification was evaluated based on signal ratios (MYC/CEP8 $\geq$ 2.2).

Results and Conclusions: IDC shows a higher prevalence of polysomy (26.6%) compared to Pure DCIS (6.6%) and DCIS with Microinvasion (6.6%). Notably, mitotic activity appeared to correlate with CEP8 polysomy, suggesting its role in aggressive tumor phenotypes. Statistical Analysis: The Fisher's Exact Test is used to evaluate the association between cancer subtype and polysomy. P-value 0.048 statistically significant. There is a statistically significant difference in the prevalence of polysomy across the subtypes.

Conclusion: The findings highlight that CEP8 polysomy is more prevalent in IDC, whereas DCIS predominantly shows disomy. This suggests that polysomy may be linked to tumor progression and aggressiveness, while disomy might characterize early-stage or less aggressive lesions. These insights underscore the potential of CEP8 polysomy as a biomarker for breast cancer prognosis and therapeutic stratification. Mitotic activity corresponds with the polysomy of CEP8.

Abstract ID: 260

Understanding Taste Receptor Polymorphism in Indian Population

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Background/ Introduction: Taste receptors (TASRs) play an important role in evaluating food quality and affecting nutritional value. Given their possible association with non-communicable diseases (NCDs), it is important to investigate these receptors in the Indian population. The genetic basis of taste perception is increasingly recognized for influencing susceptibility to various diseases, as genetic variations can impact dietary choices, metabolism, and disease risk.

In this study, 158 taste genes are manually curated, and their variations across the Indian population are checked by principal component and fixation index (Fst) analysis. The PCA analysis showed the distribution of variations, revealing a significant grouping of taste-related genes across language and geographical groups. Principal component analysis (PCA) demonstrates that both location and language influence genetic diversity, with separate clusters in the Southern/Dravidian and Northeastern/Tibeto-Burman populations. The Fixation Index (Fst) assesses population divergence in terms of genetic diversity. Most regions show minor genetic differences, according to pairwise FST analysis, with the exception of the Northeast. Additionally, a phenotypic study involving 401 individuals was conducted to examine taste perception using Bartovation Taste strips, which classified the population as supertasters, non-tasters, and recessive supertasters based on taste score. In future research, we will do a genotypic analysis of the individuals we study phenotypically and their link with Ayurvedic Prakriti so that we may use Ayurveda's precise, personalized approach to select primary preventative, diagnostic, and therapeutic methods.

Objectives: To analyze the spectrum of variation in taste-related genes in Indian population

Materials and Methods: Gene Selection, Variant Filtering, Data Analysis, Frequency and Fst Calculations, Principal Component Analysis (PCA)

Results and Conclusions: The clustering suggests some genetic similarity within each geographical region, with distinct clusters for the South and Northeast regions.

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Abstract ID: 261

Investigating the genetics of Non-Syndromic Tetralogy of Fallot: Insights from WES and *in silico* analysis of NOTCH1 variants

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Background/ Introduction: Tetralogy of Fallot (TOF) is a common subtype of conotruncal cardiac anomalies and one of the most prevalent forms of congenital heart defects (CHDs). TOF can occur as part of a syndrome or in a non-syndromic form.

Objectives: The primary objective of our study was to identify potential genetic variants associated with non-syndromic TOF through whole exome sequencing (WES) and to evaluate the underlying molecular mechanisms contributing to the development of TOF in the proband.

Materials and Methods: In this study, WES was performed on an 11-year-old female proband of non-consanguineous Indian origin, who presented with non-syndromic TOF. In silico analyses were conducted to evaluate the pathogenicity of the identified variant. Conservation analysis was performed to assess the evolutionary significance, and structure analysis was done to determine the effect of the variant on the protein structure and stability. Segregation analysis was also carried out to confirm the inheritance pattern.

Results and Conclusions: WES revealed a heterozygous missense variant in the NOTCH1 gene. Segregation analysis confirmed that the variant was inherited from the mother, indicating an autosomal dominant inheritance pattern. In silico analysis showed that the variant was highly conserved across multiple species and predicted to be deleterious. Structural analysis suggested that the variant could disrupt the functional domain of the NOTCH1 protein, potentially affecting its binding ability and involvement in critical signaling pathways. This suggests that the abnormalities in vascular development and associated signaling defects may contribute to the pathogenesis of TOF. This study provides valuable insights into the molecular mechanisms underlying this congenital cardiac condition.

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Abstract ID: 262

Genetic Screening in optimizing assisted reproductive technologies (ART) Outcomes in cases of Male Infertility and Recurrent Pregnancy Loss- A case series

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Background/ Introduction: Genetic screening is critical in addressing reproductive difficulties, especially in assisted reproductive technologies. Male Infertility and RPL are major causes of infertility and the current burden in India is around 23% and 7.4% respectively. Chromosomal abnormalities and single-gene diseases are among the underlying genetic etiologies of infertility and recurrent pregnancy loss (RPL). Genetic counseling prior to ART has emerged as an effective strategy for enhancing pregnancy outcomes by identifying potential genetic factors.

Objectives: The purpose of this study is to demonstrate the importance of genetic counseling and testing prior to ART procedures in cases of male infertility and recurrent pregnancy loss, as well as the impact on reproductive decision-making and pregnancy outcomes.

Materials and Methods: Here, we discuss 3 different cases where 2 of the couples presented with recurrent pregnancy loss and 1 of them presented with male infertility. Pre-Test and Post-Test Genetic Counseling were provided to them before their ART procedure as per their requirement.

Results and Conclusions: In all 3 cases, the couples were able to make informed reproductive decisions through personalised counseling, resulting in successful ART cycles and the delivery of healthy babies. This case series demonstrates how genetic counseling can help couples with reproductive difficulties optimise the results of ART. By determining the genetic factors causing infertility or recurrent

miscarriages, clinicians can provide personalised treatment regimens and improve the chances of successful pregnancies. The positive outcomes seen in these cases highlight the value of integrating genetic counseling into the standard of care for couples undergoing ART procedures, particularly those with a history of reproductive difficulties. In conclusion, genetic screening before ART procedures play a crucial role in addressing the underlying genetic factors contributing to male infertility and recurrent pregnancy loss. Genetic counseling enhances ART outcomes by providing couples with personalized reproductive care tailored to their genetic makeup.

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Abstract ID: 901

Differential Expression Of miRNA'S In Coronary Artery Disease.

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Introduction: Coronary artery disease (CAD) contnues to be a leading cause of mortality and morbidity globally, contributing significantly to the burden of cardiovascular diseases (CVD). According to recent statistics from the World Health Organizaton (WHO) and the Global Burden of Disease (GBD) study, CVDs account for approximately 18.6 million deaths annually, representing nearly one-third of global deaths. Ischemic heart diseases, including CAD, are the primary contributors, with 9.1 million deaths recorded in 2019 alone. Risk factors such as high systolic blood pressure, poor dietary habits, and air pollution exacerbate the prevalence of CAD worldwide. Epidemiological studies suggest genetic factors play a significant role, contributing to 40-60% of CAD susceptibility. Recent advancements in molecular biology have identified microRNAs (miRNAs) as crucial players in CAD pathogenesis. MiRNAs are small, non-coding RNAs, approximately 20-24 nucleotides in length, that regulate gene expression post-transcriptionally. Unlike messenger RNAs, miRNAs do not encode proteins but instead negatively regulate their target genes, affecting key biological processes such as cell proliferation, apoptosis, and inflammation. Importantly, miRNAs are tissue-specific, stable in circulation, and easily detectable, making them promising candidates for diagnostic and prognostic biomarkers. In the context of CAD, alterations in miRNA expression profiles have been linked to atherosclerosis, endothelial dysfunction, angiogenesis, and vascular remodelling. Analyzing miRNA's can help establish disease-specific signatures. These signatures can be used for early detection, disease monitoring, or prognosis of a disease. Non-invasive sampling (e.g., blood or urine) for miRNA profiling adds to their clinical utility as markers. By determining disease-specific miRNA profiles and their downstream targets, research can help in stratifying patients based on molecular characteristics, enabling tailored treatments. Hence, this study aims to investigate the association of miRNAs in CAD pathogenesis by identifying differentially expressed miRNAs, analysing their expression in CAD patients and healthy controls, and validating their predicted target genes.

Materials and Methods

Identification of Candidate miRNAs: A comprehensive literature review and database analysis were performed to identify miRNAs associated with CAD. PubMed was ulized to screen studies reporting miRNA expression levels in CAD, while datasets from the Gene Expression Omnibus (GEO) database

were analyzed for experimentally validated miRNA data. GEO2R, an online tool for differential expression analysis, was used to identify differen□ally expressed genes (DEGs) between CAD and control groups, applying thresholds of p-value < 0.05 and log fold change criteria. Overlapping genes across multiple datasets were identified using Venny 1.0, ensuring biological relevance.

Subject Recruitment and Sample Collection: A total of 250 participants (125 CAD patients and 125 healthy controls) were recruited from the Cardiology Department of Sri Ramachandra Medical Centre (SRMC). CAD cases were clinically confirmed, while controls were angiographically verified to have no signs of CAD. Demographic and clinical data, including age, sex, smoking status, HbA1c, lipid profiles, leti ventricular ejection fraction (LVEF), and Syntax scores, were collected to control for confounding factors. Blood samples (3 mL) were collected from all participants, and total RNA was extracted using TRIzol reagent.

miRNA Expression Analysis: Extracted RNA was subjected to poly(A) tailing for miRNA conversion, followed by quantification using TaqMan Advanced MicroRNA Assay kits. Expression levels of eight candidate miRNAs were measured and normalized using RNU44 as an endogenous control. The comparative Ct (cycle threshold) method was employed to analyze relative miRNA expression differences between CAD patients and controls.

Target Gene Prediction and Validation: Target genes of significant miRNAs were predicted using bioinformatic tools, including miRTarBase, TargetScan, miRDB, and HMDD. These tools employ distinct algorithms, such as seed sequence matching and machine learning, to ensure prediction accuracy. Cross-referencing of predictions was performed using Venny 1.0 and the Intervene interface to identify overlapping target genes.

Gene Expression Analysis: Target gene expression was quantified using quantitative real-time PCR (qRT-PCR) with SYBR Green chemistry. RNA was converted to complementary DNA (cDNA) using a high-capacity cDNA reverse transcription kit, and GAPDH was used as an endogenous control for normalization.

Statistical Analysis: Statistical analysis was conducted using SPSS so it ware. Group comparisons were performed using appropriate tests, and a p-value < 0.05 was considered statistically significant.

Results: Four GEO datasets on miRNAs in CAD using blood and its constituents were chosen atier a thorough search for them: GSE28858, GSE49823, GSE59421, and GSE105449. There were 1146, 35, 468, and 461 differently expressed miRNAs in these datasets, respectively. Following filtering and rearrangement of the datasets based on p-value, miRNAs were identified and selected for further analysis. The mean age of CAD patients was 53.6 ± 12.3 years, compared to 49.5 ± 13.6 years in controls. Among the ten miRNAs analyzed, five (miR-196a, miR-499, miR-423, miR-100, and miR-149) showed significant differential expression between cases and controls ($p < 0.05$). Upregulation of miR-499 and miR-423, and downregulation of miR-196a, miR-100, and miR-149, were observed in CAD patients. Bioinformatic analyses identified critical target genes for these miRNAs, including AKT, MTOR, TRAF, IRAK1, ADIPOQ, SIRT1 and ENOS. Expression analysis revealed altered levels of these target genes in CAD patients, supporting their involvement in disease pathogenesis. Functional annotation indicated that these genes regulate inflammation, angiogenesis, vascular remodelling, and other processes integral to CAD.

Conclusion: This study identified five miRNAs (miR-196a, miR-499, miR-423, miR-100, and miR-149) as significantly dysregulated in CAD patients compared to healthy controls, highlighting their relevance in the disease. Bioinformatic analyses revealed that these miRNAs influence key molecular pathways, including vascular inflammation and remodelling, through interactions with target genes. These findings highlight the intricate regulatory networks driving CAD and underscore the utility of miRNA-target interactions in disease diagnosis and therapy. While this study provides valuable insights, further research with larger cohorts and functional studies is essential to confirm these findings and explore their translational potential.

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Abstract ID: 907

Genetic and Clinical Correlations in Cardiomyopathy in Dakshina Kannada.

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Abstract: Cardiomyopathies are genetic heart problems that affect the myocardium, altering the structure and function of the heart muscle. They can cause heart failure or sudden cardiac death and contribute to global cardiac morbidity and mortality. These diseases are classified as hypertrophic cardiomyopathy (HCM), dilated cardiomyopathy (DCM), restrictive cardiomyopathy (RCM), and arrhythmogenic cardiomyopathy (ACM) based on their morpho-functional phenotype. Genetic testing and genetic counselling are crucial tools in diagnosing, managing, and treating patients with cardiomyopathy. In India, there is a lack of understanding of genetic factors responsible for cardiomyopathies, particularly in India. An observational study was designed to study the mutational spectrum in the local population, focusing on 28 genes with the highest causal rate of cardiomyopathies. The study recruited 179 patients, diagnosing 62 with HCM, 102 with DCM, 15 with RCM, and finding no cases of ACM. The study identified three new mutations that might cause HCM, 18 mutations in DCM, and six patients with the RCM phenotype due to hereditary transthyretin cardiac amyloidosis. A genotype-phenotype correlation study was performed to better understand the clinical significance of the mutations, helping clinicians formulate better treatment regimens. The study also identified a novel transthyretin (TTR) mutation, p. Ile93Val, that leads to an RCM phenotype due to cardiac amyloidosis. This research was the first to identify a TTR mutation causing amyloid cardiomyopathy in India. The findings of this exploratory study could be expanded to include additional patients and genes to gain a deeper understanding of the genetic landscape of cardiomyopathy in India.

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Abstract ID: 908

A Novel Frameshift BRCA2 Gene Deleterious Variant in TNBC setting: A possible candidate for Poly (ADP-Ribose) Polymerase Inhibitors

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Background: Triple negative breast cancer (TNBC) is the most difficult form of breast cancer (BC) with limited treatment options. The BRCA2 is a tumor suppressor gene and presence of a pathogenic gene mutation can disrupt its function, leading to an increased risk of breast, ovarian, pancreatic, prostate and other cancers. The current study describes a case of 42-year-old pre-menopausal female with complete bilateral mastectomy with TNBC phenotype.

Methods and Results: Germline, Next Generation Sequencing (NGS) unmasked a novel BRCA2 gene, inDel variant. Cross-platform validation via targeted Sanger sequencing technology, confirmed the NGS data findings. Also, various bioinformatic tools predicted the variant to be "Deleterious/Damaging/Pathogenic" in nature as it resulted in a premature truncated protein.

Conclusion: To our knowledge, the detected BRCA2, exon-10 mutation is a clinically significant gene variant in a high-grade bilateral TNBC setting, being reported from India (a global arena of TNBC cases). The variant has not been reported previously in any major clinical variant databases available. Hence, the detected BRCA2 gene variant has been submitted to ClinVar database for global access and review. Also, the detected BRCA2 gene pathogenic variant makes the patient a likely candidate for targeted PARP inhibitor's in the presented clinical setting. The present study, illuminates the significance of NGS technology in cancer care in the era of "personalized medicine" which brings a novel actionable target to fore.

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Abstract ID: 909

Clinical exome sequencing identifies two homozygous *LOXHD1* variants in two inbred families with pre-lingual hearing loss from South India.

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Background: In recent years, numerous genetic variants have been linked with prelingual hearing loss (HL). Variants in the *LOXHD1* gene (Lipoxygenase homology domain – 1) associated with *DFNB*77, are highly heterogeneous, with different auditory characteristics varying from stable to progressive and mild to profound. To date, 168 *DFNB*77 cases have been recorded worldwide. Variants in the *LOXHD1* gene (MIM#613072) are causally linked to the *DFNB77* locus (Deafness, autosomal recessive 77) (MIM #613079)

The *LOXHD1* gene spans 43 exons and encodes a protein that is 2,211 amino acids long protein with 15 PLAT domains. Variants in *LOXHD1,* particularly in PLAT 10, significantly impede inner hair cell mechanotransduction shortly after birth, despite the hair bundle largely maintaining its structural integrity).

Objectives: To identify the rare/novel variants in deafness causing genes among assortative mating hearing impaired families from India.

Materials and Methods: Forty-one hearing-impaired probands, who were previously excluded for a set of four common deafness-causing genes (*viz., GJB2, GJB6, SLC26A4, CDH23)* from 33 hearing impaired (HI) families, were subjected to clinical exome sequencing (CES) involving 285 genes associated with HL. This was followed by a segregation analysis, of the available members in the family.

Results: We identified two pathogenic *LOXHD1* variants in two unrelated inbred families. One is a novel homozygous pathogenic nonsense variant (c.3999C>A; p.C1333X), while the other is a likely pathogenic missense variant (c.6046G>T; p.E2046K). *In silico* tools such as SIFT, Polyphen-2, Mutation taster, CADD and REVEL scores were used to predict variant pathogenicity. Furthermore, ACMG guidelines specific to HL were applied to finally classify a variant as pathogenic or otherwise. The root specificity of these affected families lies precisely in parental inbreeding, which increases the likelihood of homozygosity of rare pathogenic variants, such as presented in this study. The frequency of *LOXHD1 variants* identified in our study is 4.88 % (2/41). This is the first *LOXHD1* report associated with non-syndromic hearing loss in South Indian families.

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Abstract ID: 910

Role of genetic factors in Thrombosis during pregnancy and puerperium

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Background/ Introduction: Pregnancy itself is an independent risk for thrombosis, and thrombosis poses significant problems in pregnancy, a time when objective diagnosis and prompt treatment are crucial. Though maternal morbidity in India is relatively high (around 4 – 5%), there is limited data available on the risk assessment for thrombosis during pregnancy. One of our previous studies showed that in patients with pregnancy-associated cerebral venous thrombosis (CVT), prothrombotic markers can be multiple and are associated with increased odds of mortality. This study was undertaken based on the hypothesis that there are underlying inherited genetic risk factors, making women more prone to a prothrombotic state during this physiologic high-risk period and thus resulting in thrombosis.

Objectives: Recruiting women with thrombotic event(s) during pregnancy and puerperium period as cases and the ones without but categorized as clinically high-risk (CHR) group based on the clinical questionnaire in their third trimester as controls, and investigating the association between selected single nucleotide polymorphisms (SNPs) with thrombosis in pregnancy and puerperium.

Materials and Methods: Seventy-five patients with thrombotic events were recruited as cases, and 201 pregnant subjects were identified as CHR controls, and they were followed up to the puerperium period. As an individual with genetic mutations/polymorphisms can develop thrombosis when exposed to acquired

(modifiable) risk factors like pregnancy and puerperium, we studied 16 previously reported genetic polymorphisms in the cases and controls using PCR-RFLP approach.

Results and Conclusions: Out of the 16 polymorphisms studied, we found a significant difference in PROC (C6152T, p=0.001), Factor XII (C46T, p=0.049), and Factor XI (p=0.009) between cases and controls. Our results thus confirm that genetic predisposition plays a crucial role in patients with thrombotic events during pregnancy and the puerperium period in the ethnic population we have studied and warrants a larger study to demonstrate the same in other parts of India.

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Abstract ID: 911

Benign breast disease with association of single Nucleotide Polymorphism of Codon 72

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Introduction: Benign breast disease (BBD) arises from various breast tissues like epithelium, stromal adipocytes, or vascular tissue and is considered a physiopathological disorder. It is a public health issue as it significantly increases the risk of breast cancer, the most common neoplasm among women worldwide. Breast cancer progresses through multiple stages, from normal tissue to hyperplasia with and without atypia, carcinoma in situ, invasive carcinoma, and metastasis. Common benign breast lesions such as fibroadenoma and fibrocystic disease, account for 90% of clinical breast disease presentations. Some benign lesions with malignant properties, like loss of growth control, do not invade or metastasize but are called premalignant or precursor lesions, including ADH, ALH, DCIS, and LCIS. Genomic and proteomic analyses help in early detection and susceptibility assessment of malignant conversion, particularly studying TP53 mutation, which are associated with cancer risk modulation through distinct genotype (Arg/Arg, Pro/Pro, Arg/Pro).

Aims & Objective: 1. To detect SNP & susceptibility of cancer in cases & control armed. 2. To determine the variant of genotype & frequency in cases & control armed. 3. To detect biomarker which are responsible for breast cancer in cases & control armed.

Material & Methods: A case-control pilot study in GRMC Gwalior, in collaboration with Cancer Hospital research Institute and Jiwaji University. Triple assessment conformed diagnosis then surgical procedure was done, tissues & blood samples were gathered from benign breast disease &breast cancer patients while only blood samples collected from control armed. Using PCR and RFLP methods, DNA genotypes were determined.

Conclusion: The study found variant genotype, Arg/Arg, Pro/Pro, Arg/Pro, respectively in case & control armed. Arg/Pro variant was common in both group although Pro variant is significantly associated with breast cancer while Arg variant have protected against cancer. Hence current study recommend Arg72 Pro molecule used as a prognostic/diagnostic marker for cases & control armed, screening of non-hereditary cancer & chemo-prophylaxis.

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Abstract ID: 912

Clinical, genetic and structural delineation of *RPL13*-related spondyloepimetaphyseal dysplasia suggest extra-ribosomal functions of eL13

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**These authors jointly supervised this work: Giedre Grigelioniene, Katta M Girisha .*

Introduction: Spondyloepimetaphyseal dysplasia with severe short stature, RPL13-related (SEMD-RPL13) is a rare autosomal dominant skeletal disorder caused by variants in the ribosomal protein gene *RPL13*. It is characterized by disproportionate short stature and skeletal abnormalities, including spondylar and epimetaphyseal changes, primarily affecting the lower limbs. First reported in 2019, SEMD-RPL13 belongs to the broader group of ribosomopathies, disorders linked to defects in ribosome biogenesis or function. Hitherto, only thirteen patients (nine families) have been reported in the literature. Ribosomal proteins like eL13, encoded by RPL13, are critical components of ribosomes but also have extra-ribosomal functions, such as RNA binding and translational regulation. These additional functions suggest potential mechanisms for the specific skeletal abnormalities seen in SEMD-RPL13, which remain poorly understood.

Methods: Clinical and radiographic data were systematically collected from 12 affected individuals of seven unrelated families, alongside health histories and genetic analyses. Whole exome and genome sequencing was performed to identify disease-causing variants in *RPL13*. Structural analysis of the eL13 protein was conducted to assess the impact of the identified variants on its RNA-binding and ribosomal functions. Ethical approvals and informed consents were secured for all subjects prior to data collection.

Results: Clinical evaluations revealed a wide phenotypic spectrum. Affected individuals displayed varying degrees of short stature, ranging from mild to severe, accompanied by delayed carpal ossification, platyspondyly, and epimetaphyseal dysplasia in the lower limbs. Radiographic findings were consistent

across individuals, showing irregular vertebral endplates, coxa vara, and deformities of the hips and knees. Notably, the severity of the skeletal manifestations varied significantly, even within families.
Genetic analysis identified three pathogenic variants in *RPL13* (NM_000977.4), including two novel missense variants, c.548G>A (p.Arg183His) and c.569G>T (p.Arg190Leu), and a previously reported splice-site variant, c.477+1G>A. These variants clustered within the conserved RNA-binding motif of eL13, indicating a mutational hotspot critical for protein function. Segregation analysis highlighted an autosomal dominant inheritance pattern, although cases of gonadal and somatic mosaicism were observed, underscoring the complexity of transmission.
Structural modelling revealed that these variants disrupt eL13's RNA-binding capacity by altering the conserved helix H7. Specific changes included the introduction of steric hindrance, and electrostatic alterations which disrupted key binding interactions with 28S rRNA. These disruptions likely impair eL13's ability to mediate essential molecular functions, such as stabilizing RNA structures or interacting with regulatory factors. The analysis also suggested that impaired extra-ribosomal activities, particularly those involving NF-κB signaling pathways, may underlie the skeletal-specific phenotype observed in SEMD-RPL13.

Conclusion: This study broadens the clinical and genetic understanding of SEMD-RPL13 by identifying novel pathogenic variants and providing detailed phenotypic characterization in seven unrelated families (12 individuals). Additionally, the study highlights the importance of RNA-binding domains in ribosomal proteins for maintaining skeletal integrity. The clustering of pathogenic variants within the RNA-binding motif of eL13 suggests a critical role for this domain in skeletal development. Moreover, the findings support the hypothesis that the extra-ribosomal functions of eL13, including interactions with mRNA encoding skeletal regulatory factors such as NF-κB, are critical to normal bone growth and remodeling. Disruption of these functions due to pathogenic variants may lead to the distinct skeletal abnormalities' characteristic of SEMD-RPL13. Overall, these findings underscore the tissue-specific importance of ribosomal proteins and their extra-ribosomal roles in skeletal development. Further research is needed to explore these mechanisms and potential therapeutic targets.

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Abstract ID: 913

Identification of novel gene variants for Autism Spectral Disorders using Whole Exome Sequencing

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Background: Autism Spectrum Disorder (ASD) is a significant health concern in children globally due to its increasing prevalence, complex phenotype manifesting in a range of neuro-developmental disabilities which persist lifelong. It is mainly characterized by varying degrees of impairment in Communication/

Language, Social interactions and Restrictive repetitive/stereotyped behaviour. The prevalence rate of autism in India is Approx. 1 in 500 or 0.20% and currently more than 10 million are suffering. Etiology of ASD is

complex with multifactorial basis involving factors like maternal-uterine/foetal effects. Early intervention is linked to improved outcomes, and lack of specific screening and diagnostic criteria is an important concern

besides treatment challenges. There is a need to establish or define ASD signature by comprehending critical genes of pathological pathways and their variants for their clinical significance.
Objective: The objective of the study is to identify clinically potential mutations in ASD patients through Whole Exome Sequencing (WES).
Methodology: 30 ASD patients were recruited from KIMS Hospitals and Providence Microbiome Research Centre from whom blood samples were collected and processed for WES at Neuberg Diagnostics.
Results: The results revealed 25 different mutations which were heterozygous and novel. Three mutations were found to X-linked while the rest were autosomal dominant. Two pairs of genes coding common proteins were observed: CACNA1H and CACNA1A; SETD5 and SETD1B. 60% of the genes identified are implicated in Neuronal development. Three nodal protein interactions were identified
Conclusion: The results indicate three critical genes and their mutations need to be screened systematically on a large sample.

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Abstract ID: 914

Understanding the role of IFN-γ/ IL12 immune-axis in Guillian-Barre Syndrome.

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Background: Guillain-Barre syndrome (GBS) is a rare acute, immune-mediated polyradiculoneuropathy and characterized by rapid-onset muscle weakness and paralysis, often triggered by an infection. Post-infectious T cell-driven cytotoxicity specifically mediated by the Th1 cells was reported in GBS. The growing evidences suggest that the genetic background of the host plays an important role in GBS susceptibility. However, the impact of genetic variations of the Th1 cell-driven IFN-γ/ IL12 immune-axis has not yet been explored in GBS.

Objective: To explore the impact of IFN-γ/ IL12 immune-axis by analyzing the genetic variation of *IL12B, IL12RB1, IFNGR1, IFNGR2,* and *ISG15* genes in the patients with GBS.

Method: The study was conducted on patients of Indian origin with GBS (n=130) fulfilling the National Institute of Neurological Diseases and Stroke criteria for GBS and had a Brighton Level of diagnostic

certainty of 1 or 2. Additionally, genetically unrelated healthy controls (n=144) were recruited. A total of 8 single nucleotide polymorphism (SNPs) located within *IL12B* (rs3212227), *IL12RB1* (rs436857 and rs393548), *IFNGR1* (rs1327474 and rs2234711), *IFNGR2* (rs1059293), and *ISG15* (rs15842 and rs1921) genes were assessed in this study by TaqMan biallelic discrimination assay. The differences in genotype

frequencies between groups were assessed using SNPStats. Linkage disequilibrium (LD) and haplotype analysis were conducted Haploview (version 4.2) software.

Results: A significantly decreased frequency of major allele 'G' of rs1921 (*ISG15*) was observed in patients with GBS (p=0.017). Further, significant differences in the frequencies of the GA genotype (p=0.004) of rs1921 (*ISG15*), CC genotype (p=0.039) of rs1059293 (*IFNGR2*) were observed between patients and controls. Strong LD (D′>0.8) was observed in *IL12RB1* (rs436857 and rs393548), *IFNGR1* (rs1327474 and rs2234711) and *ISG15* (rs15842 and rs1921) genes. Additionally, the CA haplotype (Case vs Control:

0.271/0.365, p=0.017) of the *ISG15* gene was associated with a decreased disease risk of GBS. Additionally, the AT haplotype of *IFNGR1* gene showed a trendline association (Case vs Control: 0.118/0.174, p=0.060).

Conclusion: The association of alleles, genotypes, and haplotypes of IL-12/IFN-γ pathway-related genes in GBS patients strengthens the current understanding of the role of host genetic background in the risk of GBS.

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Abstract ID: 915

Investigating the Efficacy of *Medhya rasayana* on Alzheimer Disease induced Mouse Model

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AD is a common neurodegenerative disease, and it levies substantial economic and non-economic burdens. Ayurveda, the Indian traditional medicinal system known to promote "healthy ageing," and the Medhya rasayana described in it aim to improve the brain functions and mitigate various neurodegenerative diseases, including AD. C. asiatica (Mandukaparni) and B. monnieri (Brahmi) are two commonly used Medhya rasayana plants by Ayurveda practitioners for treating various brain-related disorders. However, there is very little evidence showing their functional and molecular effects on alleviating AD. In the present investigation, an attempt was made to understand the molecular changes in AD-induced C57/B6 mice and also the effects of C. asiatica and B. monnieri. In this study, the four-month-old mice were fed with rasayana for two months, and then AD was induced chemically. The behavioral tests were performed to assess the impact of rasayana on cognition in AD-induced mice. The hippocampus samples were subjected to transcriptomics analysis to understand the functional and molecular role of rasayanas. The data indicated the differential expression of genes in AD and also the reversal of expression of a few genes in rasayana-pretreated animals. Thus it may

be opined that these rasayana may potentially enhance brain functions and may be used as adjuvants in the therapeutics of AD.

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Abstract ID: 917

Induction of Sister Chromatid Exchanges (SCEs) and Chromosome aberrations in Chinese hamster ovary cell line (CHO cell line) using medium dose of K- alpha Radiation (X-rays) and Antineoplastic drugs.

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Chinese hamster ovary (CHO Cell line) cell line was obtained from National Facility for Animal Cell & Tissue culture Facility, University of Poona and maintained in our Laboratory using C0 2 incubator at 37 0 C using Minimal Essential Medium and Foetal Calf serum and antibiotics Penicillin and Streptomycin and P H adjusted to 7.0. The Cell cycle kinetics studies i.e First and Second cell cycle includes G 1, S, and G 2 +M stages are standardised using standard protocol and G0 achieved by using serum free medium. \The cells were exposed to medium dose of K-alpha radiation (i.e X-rays) 2.53 Gy, 5.06, 10.12Gy and 15.18Gy Units) at G 1 , S, G 2 +M phases. The cells were harvested after 1 st cell cycle i.e 13 hours and after 2 nd cell cycle i.e 26 hours by injecting colchicines (0.02ml of 0.2%) just before two hours before harvesting the cells for air dried technique standardised in the Laboratory. The induction of Sister Chromatid Exchanges (SCE s) and Sister Chromatid Differentiation (SCD) are achieved by injecting 5-Bromo-2-Deoxy Uri dine (BrdU) hourly which is obtained from Sigma Chemical Company, Mumbai.

The Gene mutations and also Chromosomal aberrations and SCE s studied using medium dose of X-rays (2.53Gy, 5.06Gy, 10.12Gy and 15.18GyUnits) and incombination with antineoplastic drugs such as Actinomycin-D, Benzamide (RNA-inhibitors), Mitomycin –C (DNA inhibitor) and Cycloheximide (Protein inhibitor) among different cell cycle phases i.e G1, S and G2+M phases. The induction of X-ray induced gene mutations are inhibited by Cycloheximide and Actinomycin-D in combination with X-rays, whereas the gene mutations are enhanced by the addition of Mitomycin C with radiation. The additive effect/ inhibitive effect of radiation in combination with antineoplastic drugs are discussed in detail.

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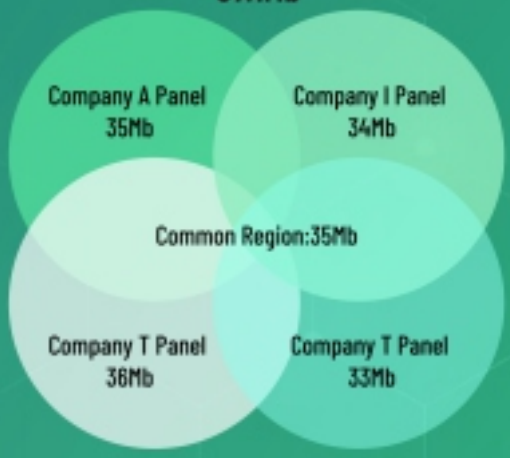

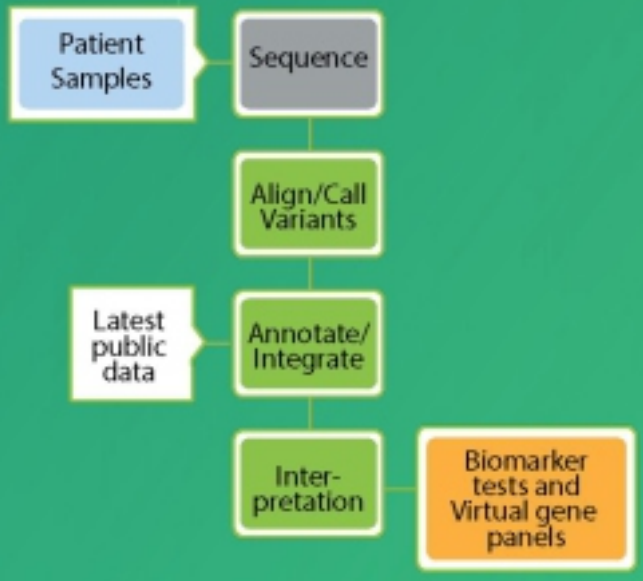

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NextSeq 1K & 2K Systems

NovaSeq 6000 System

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NGS Target Enrichment

Exome & Custom NGS Target Enrichment Solutions

- Exceptional Quality & Performance
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- Greater Flexibility

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Don't Settle for Less in Targeted Sequencing

aws

AWS FOR GENOMICS

Driving Better Patient Outcomes with Genomics Data

Accelerating Genomics Discoveries with AWS

For almost a decade, organizations of all sizes and disciplines have utilized AWS to accelerate the translation of raw sequencing data into actionable insights through scalable, secure, and cost-effective solutions. Alongside the flexibility to choose the right tool for the job, AWS provides the fastest pace of innovation and the most extensive global infrastructure, making modernization and innovation more easily accessible to all.

AWS for Genomics offers a robust portfolio of curated industry tools and purpose-built solutions designed to help genomics organizations:

- Globally access, integrate, and collaborate with genomics and multi-modal datasets
- Reduce compute and long-term data storage costs
- Easily deploy industry tools and leverage high performance computing (HPC) and machine learning (ML) to accelerate data analysis and interpretation
- Elastically scale up and down to meet workflow ebbs and flows and optimize workflow automation
- Adhere to industry and regional security and compliance needs

With AWS, organizations can dedicate more time and resources to science, achieve breakthrough research faster, collaborate globally, and reduce turnaround times and costs, all while adhering to security and compliance standards.

AstraZeneca leverages AWS to run over 51 billion statistical tests in less than 24 hours.

"We rely on the strength of AWS tools as a backbone that allows us to focus on designing genomics-specific algorithms. As researchers' and clinicians' needs change, we can easily deploy new features and versions of our products." - VP of Bioinformatics and Instrument Software, Illumina

"With AWS, (we enable) enterprises worldwide to perform genomic analysis and clinical studies in a secure and compliant environment at a scale not previously possible." - CEO, DNAnexus

Nationwide Children's Hospital leverages AWS to securely process genomics data on a massive scale—while keeping privacy and HIPAA concerns top priorities.

"Previously, we would often find ourselves still downloading the data for a review on the day it was due. But with all the data on AWS, we can perform targeted analysis very quickly with local access to open data sets." – Dr. Divya Tej Sowpati, Scientist, CCMB

Powering Personalized Care with Genomics

Genomics is quickly becoming a critical piece of clinical decision-making. In the past decade, it has become exponentially faster, cheaper, and more efficient. The challenge now is making sense of the data and reducing barriers to widespread access.

Industry leaders around the world rely on AWS and AWS Partner solutions to find meaning in genetic data and to make more personalized, data-driven diagnoses and treatment decisions. Empowering patients with targeted, individualized care can improve health outcomes—and spark the next generation of healthcare delivery.

Whether working in a research environment, a clinical setting, or any combination of the two, AWS can help you transform genomics data into better results—for scientists, providers, executives, and patients alike. Discover how AWS can help your organization reduce time to discovery, gain better control over genomics-related costs, ensure security and regulatory compliance, and collaborate on a global scale.

Scan the code to learn more about AWS for Genomics

DNBelab C Series Single-cell Product

Facilitate Large-scale Research On Single-cell Omics

Automated Library Preparation

MGISP-100
1-8 Samples/Run

MGISP-960
8-96 Samples/Run

High-throughout Sequencing

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1-8 Samples/Run

DNBSEQ-T7
4-40 Samples/Run

Pocket Single-cell Lab

Comprehensive and Efficient

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Quick Analysis Package

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Are you suspecting a **Lysosomal Storage Disorder (LSD)** in your patient?

Patients with the following signs and symptoms* may have a **Lysosomal Storage Disorder...**

Cerezyme® imiglucerase

GAUCHER DISEASE

- Enlarged liver and spleen
- Delayed or stunted growth in children
- Easy bruising and bleeding
- Anemia and thrombocytopenia
- Unexplained bone pains
- Unexplained avascular necrosis of head of femur

ALDURAZYME® (LARONIDASE)

MPS I# DISEASE

- Coarse facial features
- Early onset joint stiffness/ claw-hand deformities/contractures
- Corneal clouding (leading to light sensitivity or impaired vision)
- Recurrent respiratory infections (including sinuses and ears)
- History of recurrent hernia repair in young age

Myozyme™ (alglucosidase alfa)

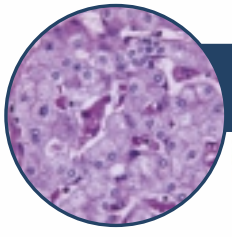
POMPE DISEASE

- "Floppy" appearance in infants or young children
- Unexplained cardiomyopathy
- Progressive respiratory muscle weakness or insufficiency
- Progressive limb-girdle muscle weakness (in late-onset cases)

Fabrazyme® agalsidase beta

FABRY DISEASE

- Severe burning pain in hands and feet
- Intolerance to heat and cold
- Inability (or decreased ability) to sweat
- Red, purple spots on skin (angiokeratomas)
- Evidence of early renal involvement (nephropathy)
- History of stroke in young age



ACID SPHINGOMYELINASE DEFICIENCY (ASMD)

Xenpozyme® (olipudase alfa)

- Enlarged liver and spleen
- Bleeding manifestations
- Skeletal abnormalities and growth delays

#EveryLifeisPrecious

- For the use of a Registered Medical Professional, a Hospital or a Laboratory
- Enzyme Testing Services are available only for the above shown diseases and only through Sanofi supported testing service – Disha.
- * https://www.webmd.com/children/what-are-lysosomal-storage-disorders • Consent/Assent has been taken for patient images displayed
  # Mucopolysaccharidosis I
- For more information, please contact Sanofi Healthcare India Pvt Ltd, Sanofi House, CTS No.117B, L&T Business Park, Saki Vihar Road, Powai, Mumbai-400072, India

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