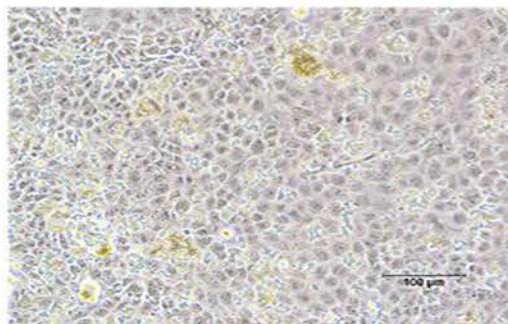
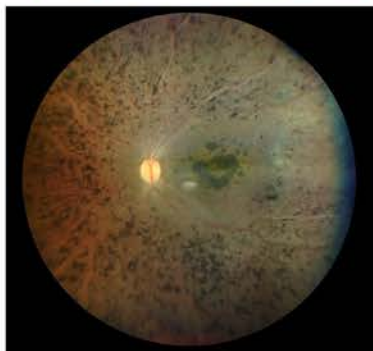
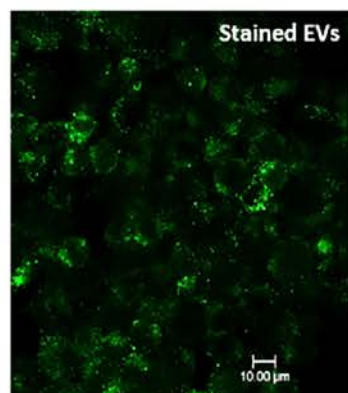
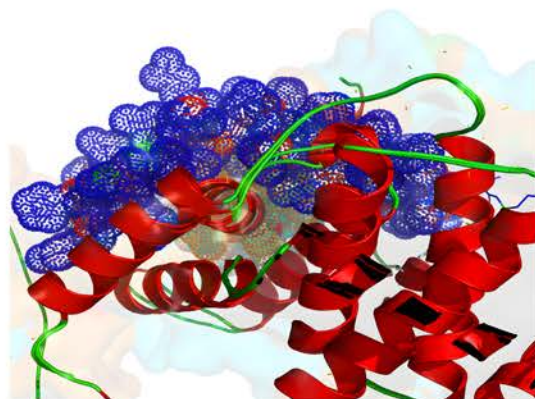


RESEARCH IN OPHTHALMIC SCIENCES

Aravind Medical Research Foundation



Annual Report
2025 - 2026

ARAVIND MEDICAL RESEARCH FOUNDATION

Aravind Medical Research Foundation is recognised
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MISSION

*To eliminate needless blindness by
providing evidence through research
and evolving methods to translate
existing evidence and knowledge
into effective action.*

RESEARCH IN OPHTHALMIC SCIENCES

Dr. G. Venkataswamy Eye Research Institute

Annual Report 2025 - 2026

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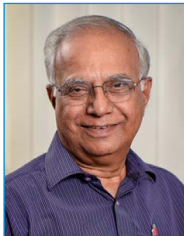


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FOREWORD



It is heartening to see the continuing strides that AMRF is taking as the years roll by. Newer partnerships are being created; newer areas of interests continue to be explored. However, the thrust towards performing relevant research in a responsible manner resides deeply embedded within the DNA of the organization. The advantage of having the clinicians work in close proximity to the basic science researchers goes an extra step in strengthening the above mentioned resolve. AMRF is committed to performing research in a right, relevant and responsible manner and is one of the integral pillar of Aravind Eye Care System, aiding in the mission of AECS in reducing needless blindness.

We look to the future with optimism coupled with a missionary zeal and look forward to further harnessing and integrating the strengths of the various components of AECS, namely Aravind Eye Hospitals, Aurolab and LAICO and create a globally acclaimed ophthalmic research ecosystem.

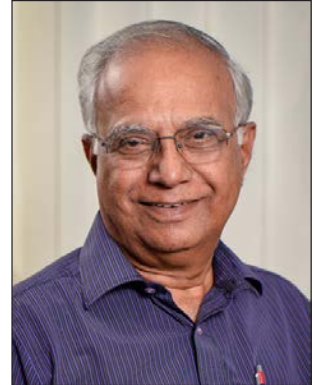
*Dr. N. Venkatesh Prajna
President, AMRF*

INTRODUCTION

Research programs, which received major funding in the previous year are progressing well as can be seen from the report in the relevant sections of the annual report. Some of the research programs now are in the stage of clinical practice. The Juvenile open angle glaucoma study involving families with the history of JOAG is now used for genetic counselling of young children who carry the mutation in their genome. Another study focussing on the clinical grade exosomes will be in the clinical testing stage soon. Predicative markers for the keratitis disease treatment outcome is now explored from another angle using the host microRNAs as signatures in addition to the proteins. New model system to study ocular cancers, started this year, is expected to give us new leads. As in the previous year students and faculty participated in several National and International conferences.

The support of the Government funding agencies and donors helped us immensely to achieve our goals. I take this opportunity to thank all our donors and well-wishers for their continued support for our research activities.

Prof. K. Dharmalingam
Director - Research



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MOLECULAR GENETICS

The genetic diversity underlying retinal dystrophies and optic neuropathies makes them ideal candidates for analysis using next-generation sequencing (NGS). By integrating high-throughput NGS with detailed clinical phenotyping, the department aims to bridge the gap between genetic discovery and clinical translation. Although these conditions have historically lacked effective therapeutic options, this genomic approach establishes a strong foundation for precision medicine in vision science. The laboratory is currently focused on elucidating the genetic architecture of rare neurodegenerative disorders affecting the human visual system, with particular emphasis on conditions such as Leber's Hereditary Optic Neuropathy (LHON), Leber's Congenital Amaurosis (LCA), and Primary Open Angle Glaucoma (POAG). The research program further seeks to establish a comprehensive regional gene registry, creating a strategic framework for future clinical trials. In addition, it aims to enhance the understanding of ethnic-specific genetic variation, improve diagnostic yield, and contribute to the development of population-relevant precision medicine approaches in ophthalmic genetics.

Investigation of nuclear genes involvement in a Mitochondrial Disorder: Leber's Hereditary Optic Neuropathy

Investigators : Dr. P. Sundaresan
Dr. S. Mahesh Kumar
Funding Agency : SERB – CRG
(CRG/2022/000926)

Introduction

Leber hereditary optic neuropathy (LHON) is classically associated with maternally inherited mitochondrial DNA mutations; however, autosomal recessive variants in nuclear genes involved in mitochondrial function can be associated with clinical symptoms of LHON. In this study, next-generation sequencing of seventy LHON probands identified biallelic variants in nuclear genes encoding electron

transport chain (ETC) complex I subunits, including NDUFV1, NDUF57, and NDUF3. These genes are essential for NADH oxidation and proton translocation during OXPHOS. The structural effects of these specific point mutations in these proteins remain poorly understood in LHON patients. Therefore, Molecular Dynamics Simulations (MDS) were employed to assess their impact on protein stability and conformational behaviour. Specifically, the effects of the R386C (NDUFV1), P144L (NDUF57), and P50T (NDUF3) variants were evaluated to elucidate their potential pathogenic mechanisms.

Results

MDS were performed to evaluate the impact of point mutations on protein conformational stability. Wild-type and mutant proteins were simulated for 10 ns in triplicate. Structural stability and flexibility



were assessed using Root Mean Square Deviation (RMSD) and Root Mean Square Fluctuation (RMSF). The wild-type proteins reached stable conformations with average RMSD values of 2.3 Å for NDUFV1, 2.7 Å for NDUFS7, and 0.6 Å for NDUFA3. In contrast, all mutant proteins exhibited RMSD values that differed from those of the wild type. Notably, the R386C variant in NDUFV1 and the P144L variant in NDUFS7 showed the greatest deviations from their respective wild-type structures beginning at approximately 2 ns and persisting until the end of the simulation. In comparison, the P50T variant in NDUFA3 demonstrated minimal deviation during the initial phase of the simulation. These differences in RMSD profiles indicate mutation-induced alterations in protein stability and highlight the structural impact of the substituted amino acids on the overall protein architecture. To further characterise residue-level dynamics, RMSF plots of the C α atoms were generated for all simulated structures. Pronounced fluctuations were observed in regions encompassing residues 5470 in NDUFV1, corresponding to the R386C variant, and residues 1375 in NDUFS7, associated with the P144L variant. In contrast, only mild fluctuations were detected in residue 765 of NDUFA3, irrespective of the P50T substitution (Figure 1). These alterations are likely to influence NADH oxidation and disrupt electron transfer to quinone, thereby potentially impairing OXPHOS..

Conclusion

MDS analysis revealed persistent structural deviations in the NDUFV1, NDUFA3, and NDUFS7 proteins, indicating mutation-induced conformational instability under in silico physiological conditions.

Overall, this study provides important insights into the structural and functional consequences of missense mutations in nuclear genes associated with LHON..

Decoding the unknown genetic etiology to ameliorate the molecular diagnosis of Leber's Congenital Amaurosis

Investigators : Dr. P. Sundaresan,
Dr. S. Senthil Kumari &
Dr. Rupa Anjanamurthy

Junior Research Fellow : S. Shiva Sankari

Funding Agency : Indian Council of Medical Research (ICMR) - Grant-in-aid Scheme (R.11015/10/2023-GIA/HR)

Introduction

Leber Congenital Amaurosis (LCA) is a severe inherited retinal dystrophy and a major cause of childhood blindness. With the advent of gene therapy as a promising treatment, accurate molecular diagnosis is crucial for effective management and intervention. Therefore, this study focuses on the clinical and genetic characterisation of CRB1-associated LCA cases in a South Indian cohort.

Results

Whole-Exome Sequencing (WES) of 77 clinically diagnosed LCA patients identified pathogenic LCA-associated variants in 44 patients (57%). Among the 29 LCA candidate genes, novel variants were

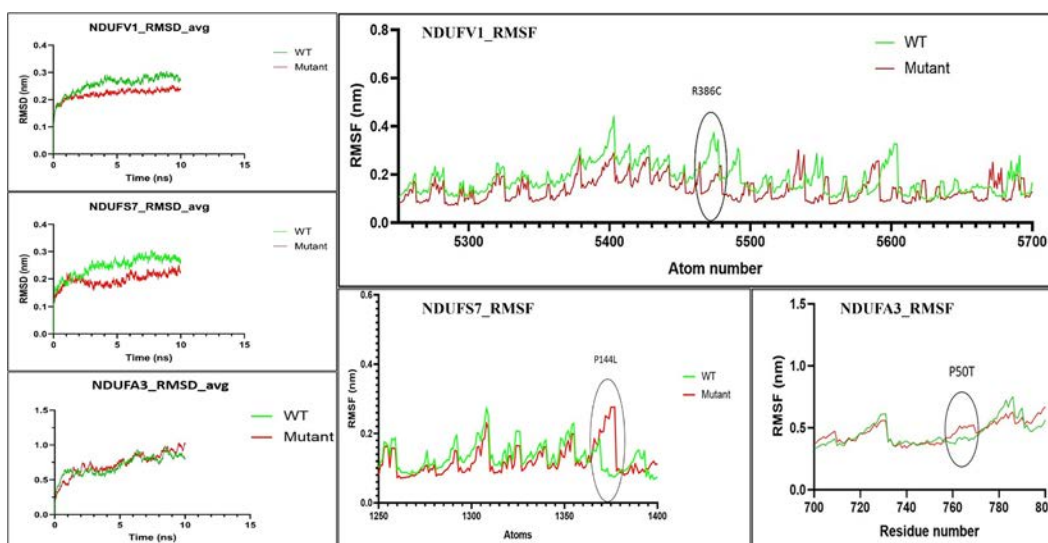


Figure1: RMSD and RMSF profiles of wild-type proteins compared with their corresponding mutant variants

detected in 15 genes of 20 patients. The most frequently affected gene in this cohort was *CRB1* ($n = 9$), followed by *GUCY2D* ($n = 7$). In addition, 15 patients (20%) harboured pathogenic variants in genes typically associated with other Inherited Retinal Diseases (IRDs), including syndromic retinal disorders. Detailed genotype–phenotype correlation was performed for patients with *CRB1* variants. Fundus examination revealed macular scarring and pigmentary retinopathy, while Optical Coherence Tomography (OCT) demonstrated a thickened, immature retina with loss of normal lamination in all *CRB1* mutant patients (Figure 2). Visual acuity assessment showed that 56% of these patients were blind, whereas the remaining 44% exhibited

severe visual impairment. Structural consequences of the identified *CRB1* variants were evaluated using homology modelling in UCSF Chimera, which demonstrated premature protein truncation resulting from nonsense and frameshift mutations, likely disrupting its normal structural integrity and functional role in retinal development and maintenance.

Conclusion

This study provides insight to understand the genetic profile and mutation spectrum of candidate genes of LCA. The clinical and mutational signatures of the *CRB1* gene were demonstrated, providing a comprehensive update of *CRB1* and its association with LCA.

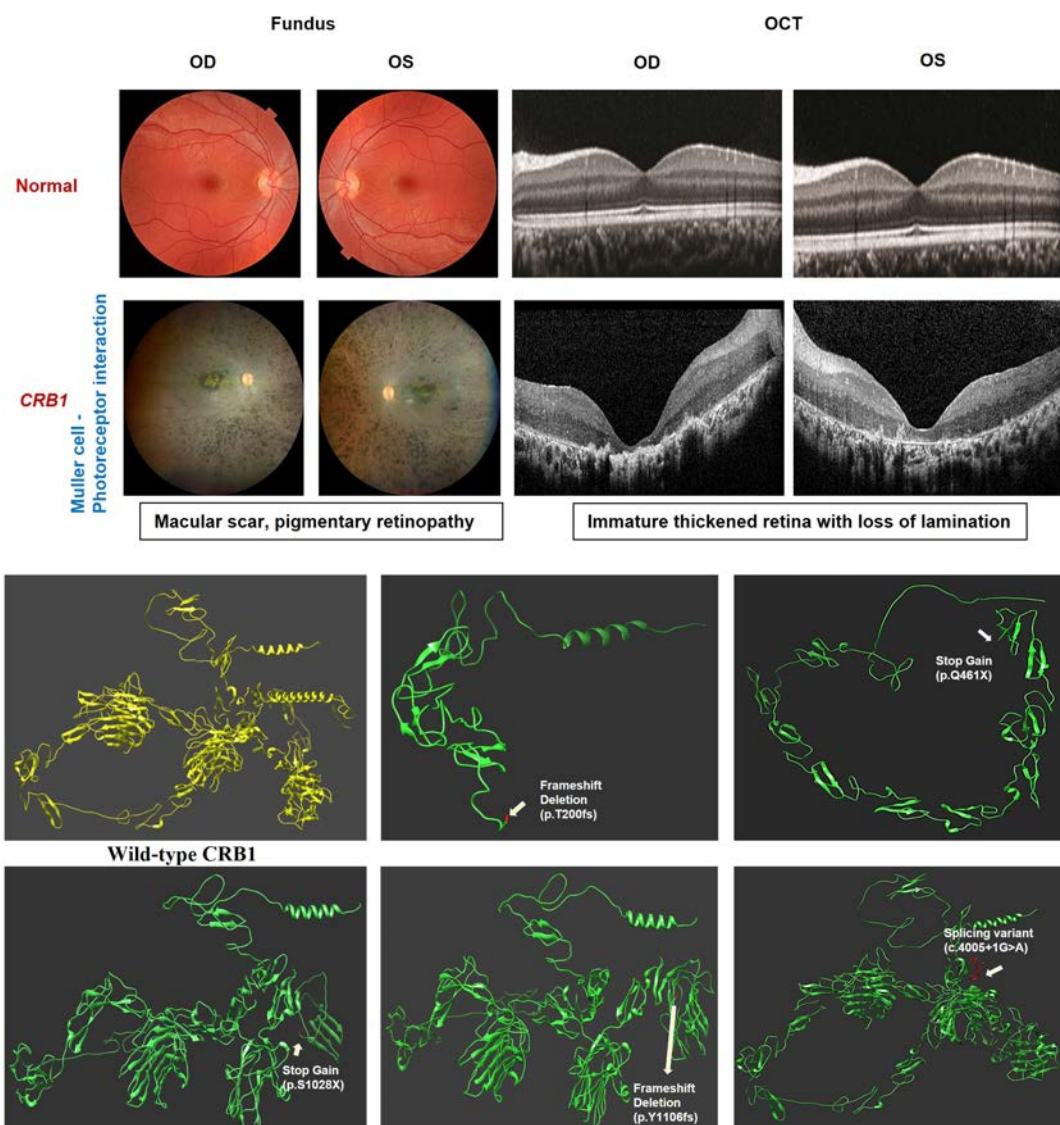


Figure 2: Genotype–phenotype correlation in a *CRB1* patient and in-silico structural prediction of the mutant *CRB1* protein

Mitochondrial association with transcriptomic signatures for trabecular meshwork damage in glaucoma: A tissue-specific omics approach

Investigators : Dr. P. Sundaresan
Dr. S.R. Krishnadas
Collaborators : Dr. Sirisha Senthil
Dr. Rani Pallavi
Dr. Anshuman Verma
Project Technical Support-III : S. Hari Vignesh
Funding Agency : ICMR-Intermediate Grant
(ICMR/03/220/IGP-2024)

Introduction

Glaucoma is a chronic optic neuropathy that leads to irreversible blindness through the degeneration of retinal ganglion cells, optic nerve damage, and Trabecular Meshwork (TM) dysfunction. While elevated Intraocular Pressure (IOP) remains the primary modifiable risk factor, disease progression in patients with normal or controlled IOP indicates that additional underlying mechanisms contribute to disease severity. The TM, which regulates aqueous humour outflow, is highly susceptible to oxidative stress and mitochondrial dysfunction. Mitochondrial DNA (mtDNA) variations have been increasingly linked to impaired cellular energy metabolism and elevated reactive oxygen species (ROS), both of which contribute to Retinal Ganglion Cell (RGC) degeneration. This study employs Whole Mitochondrial Genome Sequencing (WMGS) and single-cell RNA sequencing (scRNA-seq) to explore how mtDNA variations influence TM cell function. Furthermore, this research aims to identify mtDNA variants in TM and blood samples from patients and cadaveric donor eyes to compare the single-

cell transcriptomes of TM tissue in age-matched Normal Tension Glaucoma (NTG) and High-Tension Glaucoma (HTG) patients against cadaveric controls.

Results

A total of 250 study subjects were recruited for this study; 150 patients each from the Primary Open-Angle Glaucoma (POAG-75) and Pseudoexfoliation Glaucoma (PXFG-75) groups, and 100 samples from healthy controls, were collected. DNA was isolated from peripheral blood using the salting-out method, ensuring high-quality genomic material for sequencing.

Whole Mitochondrial Genome Sequencing (WMGS) has been successfully completed for 25 POAG, 12 PXFG, and 5 control samples. The remaining samples are currently in the screening process. Comparative analysis identified five nonsynonymous mtDNA variants unique to POAG patients MT-ATP, MT-CYB, MT-ND6, MT-ND2, and MT-CO3, affecting essential mitochondrial respiratory chain complexes (Figure 3). Table 1 summarises the variants, including zygosity and MitImpact IDs. Notably, four mutations were homoplasmic, while one (MT-CYB) was heteroplasmic. These genetic alterations are linked to oxidative stress and impaired energy metabolism, suggesting mitochondrial dysfunction as a major contributor to retinal ganglion cell (RGC) degeneration.

Conclusion

This study has successfully identified a preliminary set of mitochondrial variants that likely drive the pathophysiology of POAG through respiratory chain disruption and oxidative stress. To build upon these findings, Whole Mitochondrial Genome Sequencing (WMGS) will be completed for the remaining POAG, PXFG, and control cohorts to ensure a statistically

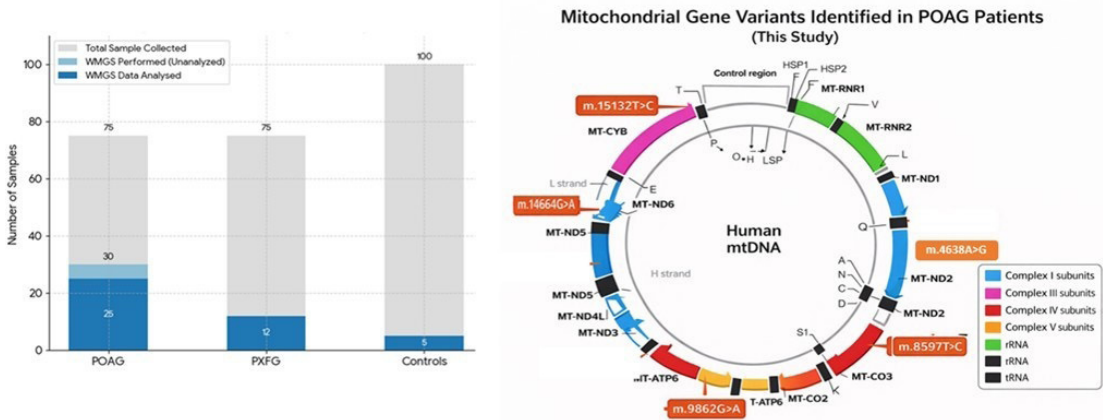


Figure 3: Sample recruitment and mitochondrial genetic spectrum in POAG

Table 1: Nonsynonymous changes identified in POAG patients

S.No	Sample ID	Age/ Sex	Gene	mtDNA Variant	Amino acid Change	Zygoty	Haplo group	MitImpact ID
1	POAG-08	67/M	MT-ATP	m.8597T>C	p.Ile24Thr	Homoplasmy	M42b	ID- MI.153
2	POAG-10	72/M	MTCYB	m.15132T>C	p.Met129Thr	Heteroplasmy	U2b	ID- MI.9152
3	POAG-16	81/M	MT-ND6	m.14664G>A	p.Ala4Thr	Homoplasmy	M39b	ID- MI.24169
4	POAG-23	61/M	MT-CO3	m.9804G>A	p.Ala200Thr	Homoplasmy	M35b	ID- MI.7905
5	POAG-25	65/M	MT-ND2	m.4638A>G	p.Ile57Val	Homoplasmy	M36	ID-MI.13108

robust dataset, with all identified variants undergoing validation via Sanger sequencing. These genomic results will then be correlated with single-cell RNA sequencing (scRNA-seq) data to map specific mitochondrial somatic mutations to differential clustered gene expression within the trabecular meshwork (TM). By linking these variations to cell-specific transcriptomic signatures, this research aims to clarify the mechanisms of IOP-independent TM damage and uncover novel therapeutic targets for treating progressive glaucoma.

Molecular genetics in ocular diseases: Laneway for Personalised medicine especially Leber's Congenital Amaurosis (LCA) and Leber's Hereditary Optic Neuropathy (LHON)

Investigator : Dr. P. Sundaresan,
ICMR – Emeritus Scientist
Funding Agency : Indian Council of Medical
Research (ICMR) (HRD/Head/
IES/2024/7)

Introduction

LCA is a genetically heterogeneous inherited retinal dystrophy that leads to severe visual impairment or blindness in early childhood. LHON is a mitochondrial disorder predominantly associated with dysfunction of the mitochondrial electron transport chain complex I, resulting in impaired oxidative phosphorylation. Consequently, pathogenic variants in mitochondrial or nuclear-encoded genes involved in mitochondrial function and dynamics can give rise to hereditary optic neuropathies. Although curative options remain limited, successful gene therapy has been developed for some of the LCA subtypes and LHON, underscoring the importance of precise molecular diagnosis. This study aims to investigate the clinical and molecular genetic landscape of LCA and LHON to enhance diagnostic accuracy, inform personalised clinical management, and explore the potential for gene-based therapeutic interventions in the near future.

Results

A total of 15 unrelated patients clinically diagnosed with LCA and 20 patients with LHON were recruited

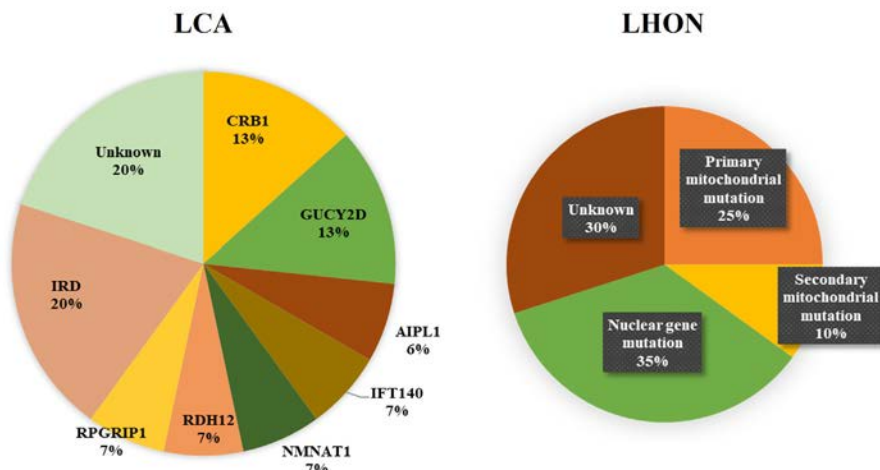


Figure 4: Mutation spectrum of LCA and LHON patients

from the paediatric ophthalmology and neuro-ophthalmology clinic, respectively. Comprehensive whole-exome sequencing (WES) identified nine causative genetic variants in LCA-associated genes among the 15 LCA patients. Consanguinity was reported in five of these cases. The highest gene-specific prevalence was observed for *CRB1* and *GUCY2D* (13.3% each), followed by *AIPL1*, *IFT140*, *NMNAT1*, *RDH12*, and *RPGRIP1* (6.7% each). Using WES, a molecular diagnosis was achieved in nearly 80% of LCA cases, highlighting its effectiveness in variant detection (Figure 4).

In the LHON cohort, whole mitochondrial DNA (mtDNA) sequencing revealed pathogenic mtDNA variants in 35% of probands, including primary mtDNA mutations in 25% and secondary mtDNA mutations in 10% of patients. Additionally, WES identified pathogenic or likely pathogenic variants in nuclear-encoded genes (*NDUFA12*, *DNAJC30*, *MECR*, *POLG*, *KIF1A*, and *OPA1*) in seven

individuals, all of them implicated in mitochondrial function and complex I deficiency. These findings underscore the contribution of both mitochondrial and nuclear genomic alterations, highlighting the critical role of nuclear–mitochondrial interactions in the pathogenesis of LHON.

Conclusion

In summary, this study underscores the crucial role of genetic factors in the pathogenesis of LCA and LHON. The findings highlight the importance of comprehensive genetic assessments, including whole genome sequencing and genome-wide association studies, as essential tools for diagnosing and managing patients who do not possess mutations in the known gene list. These insights may provide the way for novel therapeutic strategies targeting the underlying molecular mechanisms of these vision-threatening inherited retinal and optic neuropathy disorders

GENETICS OF OCULAR TUMORS

Ocular tumours include a wide range of tumours affecting the different layers of the eye. Similar to other cancer, some ethnic and age groups are prone to develop ocular tumours. Retinoblastoma is more common among the paediatric age group whereas ocular lymphoma and melanoma occur in older age groups. Apart from the demographic factors, some of these ocular tumours have a genetic predisposition and hence require a comprehensive screening procedure that involves clinical, pathological and genetic investigations in order to develop methods of early diagnosis, better prognosis and improved treatment. Our multimodal research integrates all these components and focuses on upgrading the livelihood of the patients through restoration of the globe and vision as well. Comprehensive genetic screening of the family members improves our understanding of the disease inheritance pattern and enables the follow-up of at-risk individuals.

Genetic Testing of Retinoblastoma

Investigators : Dr. A. Vanniarajan
Dr. Usha Kim
Genetic Analyst : K. Murugan
Funding Agency : Aravind Eye Care System,
Madurai

Introduction

Retinoblastoma (RB) is a common retinal tumour that mostly affects children under 5 years with rare exceptions. Clinically, it presents as unilateral or bilateral disease, with leukocoria as the most common sign. Undiagnosed and untreated retinoblastoma can be fatal and hence genetic testing plays an important role in early diagnosis and appropriate treatment.

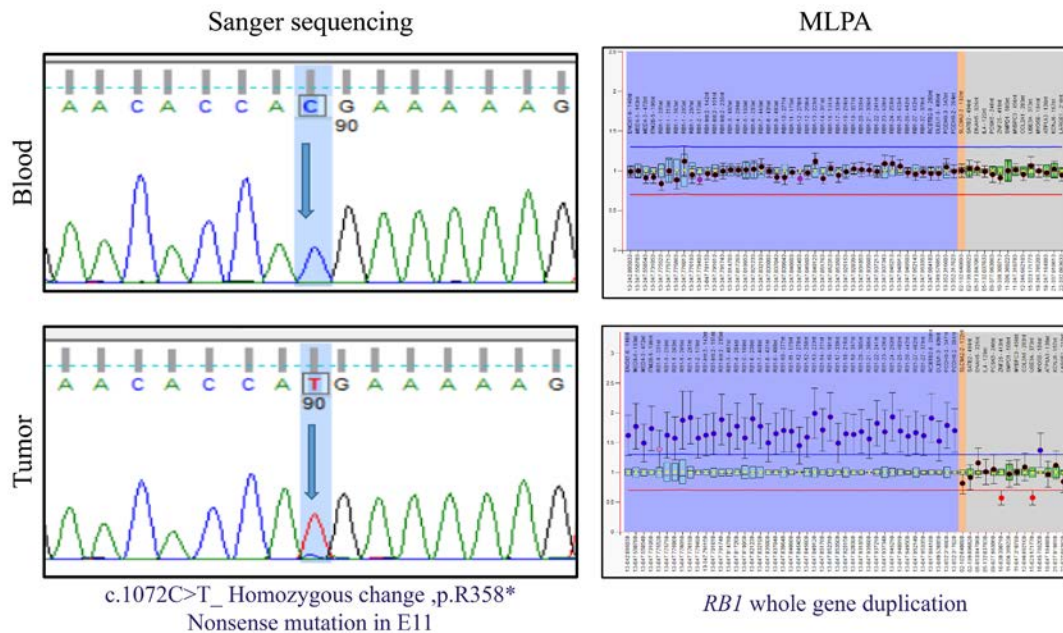
Results

RB1 genetic testing was performed in 29 patients including 20 unilateral and 9 bilateral RB during 2025. Among them, 15 were children diagnosed with RB in this year, while 14 were follow-up patients. Except for one bilateral patient, all the others harboured germline mutations. Among the unilateral patients, only one of them had somatic mutation identified from the tumour. None of the blood samples of unilateral patients had any RB1 alterations.

From the tumour sample, a homozygous nonsense mutation in exon 11 was detected by Sanger sequencing and whole gene duplication was revealed by Multiplex ligation dependent probe amplification (Figure 1). This is a unique case with all the three alleles carrying the mutation. The blood sample of the patient was normal without any of these changes, clearly indicating them as somatic alterations.



Figure 1: Sanger sequencing analysis showing homozygous nonsense mutation in exon 11 and MLPA showing whole-gene duplication in the tumour sample.



Conclusion

Genetic analysis of retinoblastoma patients identified new variants of RB1 that are distinct in our cohort. Genetic counselling was provided to the family members of the patients that helped in disease management.

Elucidating the role of cancer stem cells in chemoresistant retinoblastoma and their therapeutic implications

Investigators : Dr. A. Vanniarajan, Dr. Usha Kim & Dr. Shanthy

Research Scholar : R. Sethu Nagarajan

Funding Agency : ICMR – SRF (Fellowship)

Introduction

Retinoblastoma (RB) is a tumour of good prognosis with timely diagnosis and appropriate treatment. Chemotherapy is a mainstay in the RB treatment. However, some tumours do not respond to the chemotherapy due to multiple factors. Among them, Cancer Stem Cells (CSC) play a pivotal role. Our current study focuses on understanding the role of CSCs and the molecular factors regulating the stemness in chemoresistant RB.

Results

Clinically confirmed chemoresistant RB tumours (n=3), selected based on their treatment responsiveness, along with control neural retina samples (n=3) were used for transcriptome profiling. Differential gene expression analysis revealed 1,440 upregulated and 6,828 downregulated genes at an adjusted p-value < 0.05 and a fold-change cut-off of ± 2 (Figure 2a). Pathway enrichment indicated upregulation of metabolic and signalling pathways, including PI3K–AKT, MAPK, Hippo signalling, and cell cycle regulation. Downregulated genes were primarily associated with olfactory transduction, drug metabolism, complement and coagulation (Figure 2b). Genes associated with the top ten upregulated pathways (163 genes) were selected. StemChecker tool identified 26 stemness-associated genes. Among these, genes reported in two or more stemness databases were prioritised and shown in the figure (Figure 2c).

Conclusion

Transcriptome analysis revealed stemness-associated genes involved in RB chemoresistance, and the identified genes require further validation.

a) Volcano plot

Upregulated: 1442 | Downregulated: 608

b) Key upregulated pathways

Pathway	Gene ratio
Ribosome	~0.65
Cell cycle	~0.15
VEGF signaling pathway	~0.12
Hippo signaling pathway	~0.10
MAPK signaling pathway	~0.08
Pathways in cancer	~0.07
P13K-Akt signaling pathway	~0.06
Metabolic pathways	~0.05

c) Stemness associated genes

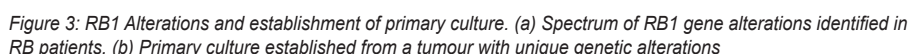
d) Key downregulated pathways

Pathway	Gene ratio
Olfactory transduction	~0.55
Renin-angiotensin system	~0.35
Drug metabolism-cytochrome P450	~0.25
Complement and coagulation cascades	~0.22
Hematopoietic cell lineage	~0.20
Neutrophil extracellular trap formation	~0.18
Neuroactive ligand-receptor interaction	~0.18
Protein digestion and absorption	~0.18

Funding Agency : ICMR

RB is an autosomal dominant paediatric retinal cancer caused by biallelic inactivation of the RB1 gene. This study focuses on a therapeutic approach combining HDR-mediated CRISPR/Cas9 gene editing

RB1 mutation status was evaluated in twenty-nine patients during 2025 and identified a wide spectrum of variants. One tumour sample was used for establishing the primary culture using an optimised protocol (Figure 3). Mutation status of the cultured cells was validated by Sanger sequencing and MLPA with the corresponding tumour and blood samples. An effective guide RNA for CRISPR-mediated gene editing targeting exons 18 and 20 was synthesised. Further, HDR templates for knock-in-mediated mutation repair was designed to enable precise correction of the target variants.



Conclusion

Primary culture and subsequent genetic characterisation of the cells confirmed the genotype similar to the tumour. The efficiency of the designed CRISPR-mediated gene editing system needs to be tested.

Distinct Genetic Aberrations in Bilateral Orbital Mantle Cell Lymphoma

Investigators : Dr. A. Vanniarajan, Dr. Usha Kim & Dr. R. Shanthi
Research Scholar : K. Saraswathi
Funding Agencies : Aravind Eye Foundation (AEF) & Lady Tata Memorial Trust (Fellowship)

Introduction

Intertumoral heterogeneity has a profound impact on disease progression and prognosis of ocular tumours. Such heterogeneity contributes to variable clinical outcomes and poses challenges for accurate prognostication in ocular adnexal mantle cell lymphoma (OA-MCL). Here, we describe a rare case of primary bilateral OA-MCL exhibiting pronounced intertumoral genetic heterogeneity.

Results

A 53-year-old male presented with bilateral orbital swelling. Computed tomography revealed tumour in both orbits (Figure 4) and Immunohistochemistry confirmed the diagnosis of bilateral MCL. WES from both tumours revealed distinct genetic profiles. ATM and KMT2C mutations were detected only in right eye (LT18_R) but not in left eye (LT18_L). ATM deletion was detected in both the tumours; however, the 3q25-29 gain was observed in LT18_L, whereas 1p36 gain was noted in LT18_R (Table 1). Further, variants of KMT2D, PDK2, and ROS1 were different between the two tumours. These alterations underscore intertumoral genomic heterogeneity in the same individual.

Conclusion

This unique case highlights the importance of deciphering genetic variability in ocular MCL to understand tumour evolution and its implications for therapeutic resistance and disease management.

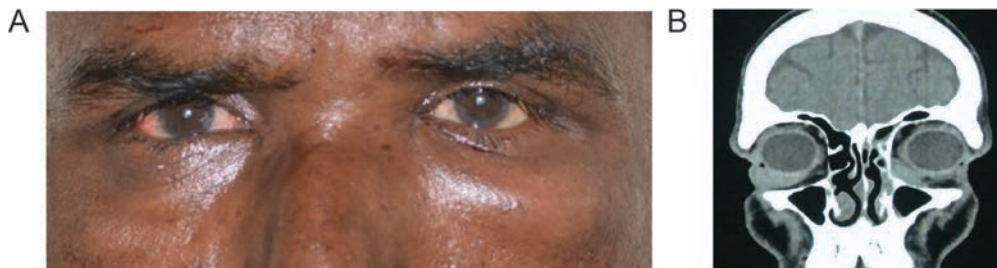


Figure 4: Clinical and Histological Features: (A) Patient with bilateral orbital mass. (B) CT image showing tumour lesions in both eyes engulfing the left nasolacrimal duct and right inferior quadrant encompassing inferior rectus and inferior oblique muscles and extending into pre-septal compartments.

Table 1: Somatic point mutations and CNVs in LT18_L (Black font) and LT18_R (Red font) tumours

Gene	Type of Mutation	c.DNA change	Amino acid change	Mutation ID	ACMG Evidence
ATM	Stop gain	c.C7220A	p.S2407X	rs1555122149	Pathogenic
KMT2C	Missense	c.G9354A	p.M3118I	rs2129110814	Uncertain Significance
KMT2D	Missense	c.A14444G	p.Y4815C	rs778135894	Pathogenic
KMT2D	Missense	c.C3737T	p.T1246M	rs112921115	Benign Strong
PDK2	Stop gain	c.C751T	p.Q251X	rs1441300054	Likely Pathogenic
PDK2	Missense	c.A884G	p.Y295C	rs199817074	Pathogenic Supporting
ROS1	Missense	c.T424C	p.W142R	rs373796868	Pathogenic Supporting
ROS1	Missense	c.G1094C	p.G365A	rs61743088	Pathogenic Supporting

Gene	Length of Structural Variant	Type of Alteration	Chromosomal Loci
ATM	-4598361	Deletion	11q22.3
ATM	-2100	Deletion	11q22.3
BCL6	2322448	Duplication	3q27.3
CDK11A	36151	Duplication	1p36.33

Integrated genomic and transcriptomic analysis of ocular adnexal B cell lymphoma

Investigators : Dr. A. Vanniarajan, Dr. Usha Kim & Dr. R. Shanthi
Research Scholar : K. Saraswathi
Funding Agency : Aravind Eye Foundation (AEF) & Lady Tata Memorial Trust (Fellowship)

Introduction

Ocular Adnexal B-cell Lymphoma (OABL) represents a heterogeneous group of non-Hodgkin lymphomas, with limited prognostic/therapeutic options, particularly in recurrent cases. To elucidate the OABL pathogenesis, we performed integration analysis of whole-exome and transcriptome sequencing data to identify the key altered pathways.

Results

Potentially relevant mutations and copy number variations were assessed for their impact on gene expression across thirteen samples with RNAseq and WES data. We observed significant and consistent

dysregulation of lymphoma associated genes at the transcriptomic level, including TNFAIP3, TBL1XR1, EP300, KMT2D, and MYD88, when compared to controls. Additionally, numerous cancer consensus genes involved in key oncogenic pathways such as B cell signalling, NF- κ B, PI3K, apoptosis, EMT, and KRAS showed significant alterations at both the genomic and transcriptomic levels. Notably, genes including KMT2D, PLEC, BAP1, CLCN2, and FLT4 were overexpressed in recurrent samples compared to primary tumours, indicating their possible role in therapy unresponsiveness (Figure 5).

Conclusion

The integrative multi-omics analysis has identified novel genetic alterations in OABL, supported by corroborative evidence from gene expression profiling. Further validation through functional studies and analysis in larger cohorts is essential. These findings establish a foundation for improved prognostic insights and may aid in identifying potential therapeutic targets to enhance treatment outcomes in OABL.

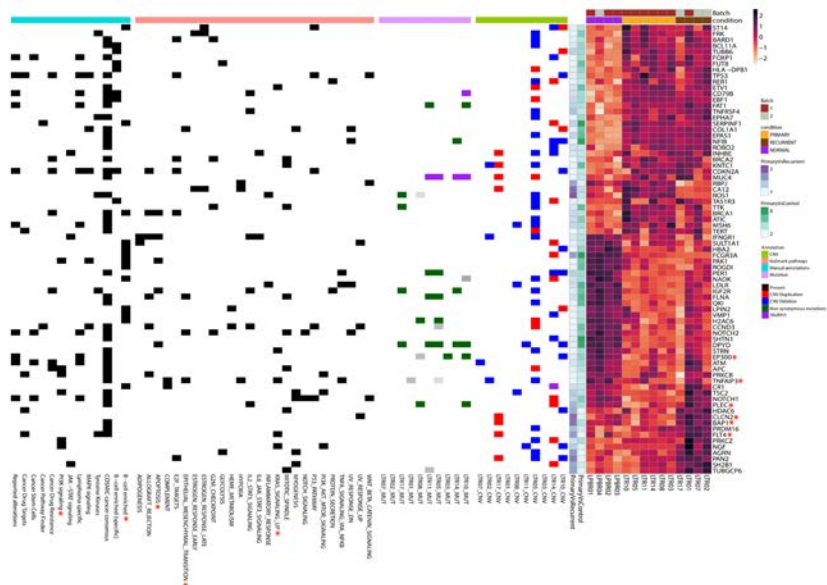


Figure 5: Integration analysis revealing genetic alterations in cancer related pathways. Significantly affected genes with their genetic alterations and pathways (*Genes related to Ocular B cell lymphoma and disease recurrence)

Understanding the basic biology of adult ocular stem cells is significant to develop better stem cell-based therapies for various ocular conditions. This year, the focus of research was on molecular mechanisms regulating the tissue-resident stem cells of human trabecular meshwork, lens epithelium and retinal pigment epithelium. Spatial transcriptomics of the human trabecular meshwork, and retinal pigment epithelium; meta-data analysis of published single-cell transcriptomics data for lens epithelium has enabled confirmation of the location of stem cells and identification of the molecular factors as well as the signaling pathways regulating these stem cells. A new project in developing a cell-free therapy with exosomes for glaucoma has been initiated.

Molecular regulation of adult human lens epithelial stem cells: Changes with age-related cataract

Investigator : Dr. Gowri Priya
Chidambaranathan
Co-Investigator : Dr. Madhu Shekhar
Research Scholar : Thushmitha P
Funding Agency : ICMR - Senior Research
Fellowship

Introduction including Background

The human lens consists of the lens capsule, epithelial cells, and fibre cells. Lens epithelial cells continuously proliferate and differentiate into lens fibre cells throughout life. Yes-Associated Protein (YAP) is a transcriptional co-activator that shuttles between the cytoplasm (inactive, phosphorylated) and nucleus (active, unphosphorylated), where it regulates gene expression through TEAD transcription factors. Conditional YAP knockout in mice leads to cataract formation, highlighting its

association with cataract development. Previous study from the department reported nuclear YAP and SOX2 double-positive cells in the central and equatorial regions of normal lens epithelium, with reduced nuclear and increased cytoplasmic YAP in cataractous lenses. In this study, Western blotting was performed to analyse key YAP pathway proteins (YAP, pYAP, MST2, and LATS). Additionally, publicly available single-nucleus RNA-sequencing data from Van Zyl et al., (2022) were analysed to investigate the molecular regulation of lens epithelial stem cells.

Results:

Alteration in the expression of YAP signalling proteins in human cataractous lens epithelium

pYAP/YAP expression was found to be higher in cataractous lens epithelium. This further validated our previous immunostaining data that the presence of more active (nuclear) YAP in normal lens epithelium. In contrast, the YAP signalling genes, including MST2 and LATS, were identified to be decreased in



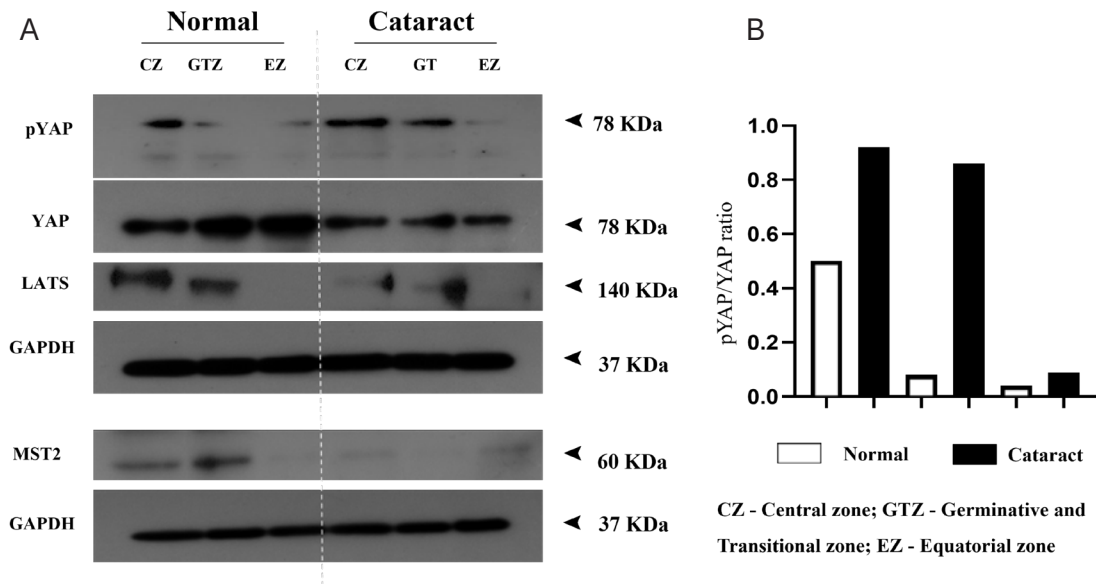


Figure 1. Western blot analysis of proteins involved in the YAP signalling pathway (1A). Increased total YAP and decreased pYAP in normal lenses confirmed the presence of more active YAP in normal condition compared to cataractous lens epithelium. The expression of YAP signalling genes (MST2, LATS) was identified to be reduced in cataractous lens epithelium. GAPDH was used as a loading control. Bar diagram showing differential expression of pYAP and YAP in normal and cataractous lens epithelium (1B).

cataractous lens epithelium compared to normal lens epithelium (Figure 1A and 1B).

Identification of human lens epithelium cell types

A total of eight clusters were identified, including four epithelial-like clusters (cluster 1, 5, 3, 0), three fibre-like clusters (cluster 2, 4, 6), and one stem cell-like cluster (cluster 1). Cluster 1 was identified as a stem cell cluster based on the expression of stemness-associated genes – HGF, HES5, and CCND1). Moreover, KEGG analysis revealed that stemness-associated pathways, including PI3-AKT, Wnt, and

MAPK, were upregulated in cluster 1, representing epithelial stem-like cells. Clusters 2, 4, and 6 were identified as fibre like cluster based on the expression of early and late elongation markers - β -crystallin and γ -crystallin (Figure 2A and 2B).

To assess cellular differentiation states, CytoTRACE analysis was employed to infer relative developmental potential across clusters. Higher stemness scores were observed in cluster 4, representing fibre stem-like cells, suggesting greater transcriptional diversity (Figure 2C).

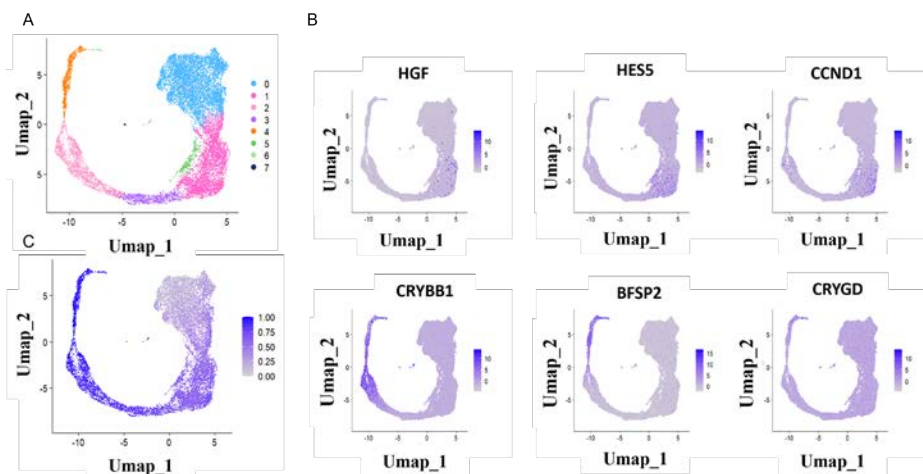


Figure 2: UMAP visualisation of 8 clusters identified in the anterior human lens epithelium (2A). Feature plots showing the expression of representative genes in the lens epithelium and fibre (2B). UMAP visualisation with cytoTRACE scores (2C)

Conclusion:

Western blot confirmed the reduction of active YAP in human cataractous lens epithelium. In addition, single-cell data analysis provided a refined view of lens cellular heterogeneity and highlighted transcriptional genes associated with stem-like properties, contributing to a better understanding of lens biology.

Molecular characterisation of human retinal pigment epithelial stem cells and their role in age related macular degeneration

Investigator : Dr. Gowri Priya Chidambaranathan
Co-Investigators : Dr. D. Bharanidharan, Dr. Kim Ramasamy & Dr. Siddharth Narendran
Research Scholar : Kanthimathi R
Funding Agencies : Indian Council of Medical Research (April to December 2025); CSIR – Senior Research Fellowship (December 2025 to till date)

Introduction including Background

Retinal Pigment Epithelium (RPE) is important for the functioning of photoreceptors and its degeneration leads to age-related macular degeneration, a leading cause of blindness in the elderly population. Previous studies from this laboratory identified the presence of stem cells (SCs) in the peripheral region of human RPE based on their functional characteristics and demonstrated a reduction in their number with ageing. However, specific biomarkers to confirm the exact location of these RPESCs in native tissues are still not known. This study aims to confirm the location of SCs in native human RPE using spatial transcriptomics.

Results

Spatial transcriptomic clustering of retinal tissue sections:

Analysis of the spatial transcriptomic data (Figure 1) identified transcriptionally distinct clusters distributed across the retinal layers. Each cluster was differentiated based on their characteristic marker genes of retinal cell types. The cluster corresponding to the RPE exhibited distinct higher expression of RPE65, RGR, DCT, and OPTC, consistent with functional visual cycle activity and pigment-related processes. Clusters representing rod photoreceptors were defined by expression of RHO, SAG, PDE6G, and RCVRN, whereas ganglion cell clusters showed strong enrichment of SNCG, NEFL, and NEFM. Additionally, clusters with fibroblast-like features displayed expression of COL1A2, DCN, and PRELP, represented transcriptional regulation associated with extracellular matrix organisation. Overall, these gene expression profiles validate the molecular distinction of RPE and other retinal cell populations within the spatial landscape.

Regional variation in gene expression across the RPE:

To analyse region-specific differences in gene expression, differential gene expression analysis was carried out comparing the peripheral RPE with the combined central and equatorial RPE. The volcano plot (Figure 2) highlighted a distinct set of genes significantly enriched in the peripheral RPE. Several of these upregulated genes were associated with stem-like properties: TFPI2, a reported marker of stem-like cells in fetal RPE, showed increased expression in the peripheral RPE. In addition, higher expression of ALDH1A3, along with regulators of BMP and Wnt signalling such as NOG and SFRP1/4, suggested activation of pathways involved in stem cell maintenance. Conversely, genes linked to more differentiated RPE states were comparatively reduced

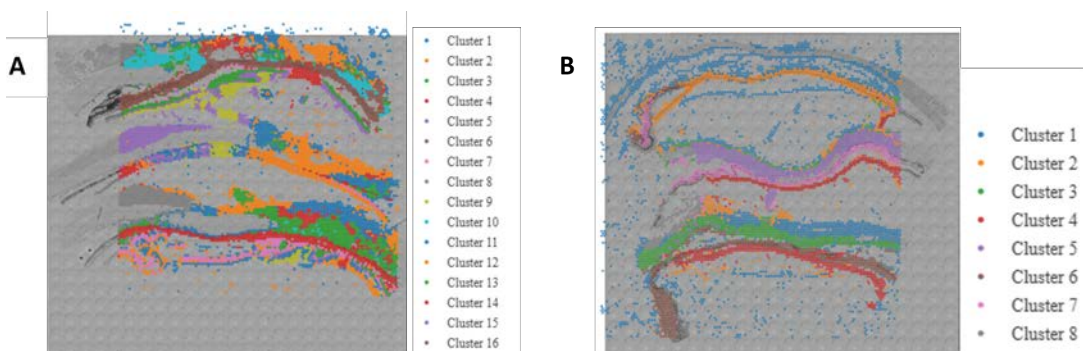


Figure 1: Distribution of spatially resolved transcriptomic clusters within the retina. (A) Sample 1 exhibited sixteen distinct molecular clusters and (B) Sample 2 displayed eight major clusters, reflecting region- and layer-specific gene expression patterns.

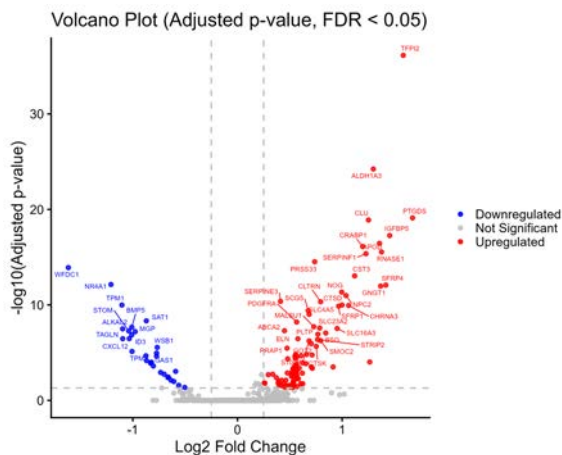


Figure 2: Volcano plot depicting differential gene expression in the peripheral RPE relative to the central and equatorial regions. Genes are displayed based on $\log_2$ fold change and $-\log_{10}$ adjusted p-value. Significantly upregulated genes in the peripheral RPE ($FDR < 0.05$; $\log_2 FC > 0.25$) are shown in red, while significantly downregulated genes ($FDR < 0.05$; $\log_2 FC < -0.25$) are shown in blue.

in the peripheral RPE. Together, these findings indicated a region-specific transcriptional profile consistent with a stem cell-enriched niche in the peripheral RPE.

Conclusion:

This study demonstrated transcriptional heterogeneity across the human RPE and identified enrichment of stemness-associated genes in the peripheral region, supporting the presence of a stem cell-enriched niche. Further studies are essential to identify RPESCs specific markers and the associated signalling pathways.

Molecular regulators associated with the maintenance of human trabecular meshwork stem cells in relation to their reduction in ageing and glaucoma

Investigator : Dr. Gowri Priya Chidambaranathan
 Co-Investigators : Dr. S. R. Krishnadas & Dr. D. Bharanidharan
 Research Scholar : Sneha Nair
 Funding Agencies : Science and Engineering Research Board; LTMT- Junior Research Fellowship

Introduction including Background

Previous studies from this laboratory demonstrated reduced TM cellularity associated with loss of Trabecular Meshwork Stem Cells (TMSCs) with ageing and a more pronounced decline in glaucomatous donor eyes compared to age-matched controls. The present study focuses on elucidating the molecular regulators governing TMSC maintenance and their dysregulation in glaucomatous conditions. Earlier, the miRNA profiling of the Filtering (F) and Non-Filtering (NF) regions of TM was carried out to identify the miRNAs regulating TMSCs using Nanostring. This year the miRNA changes in glaucomatous condition was elucidated. In addition, GeoMx digital spatial transcriptomic analysis was carried out to identify the transcription factors associated with stemness regulation in normal and glaucomatous TM.

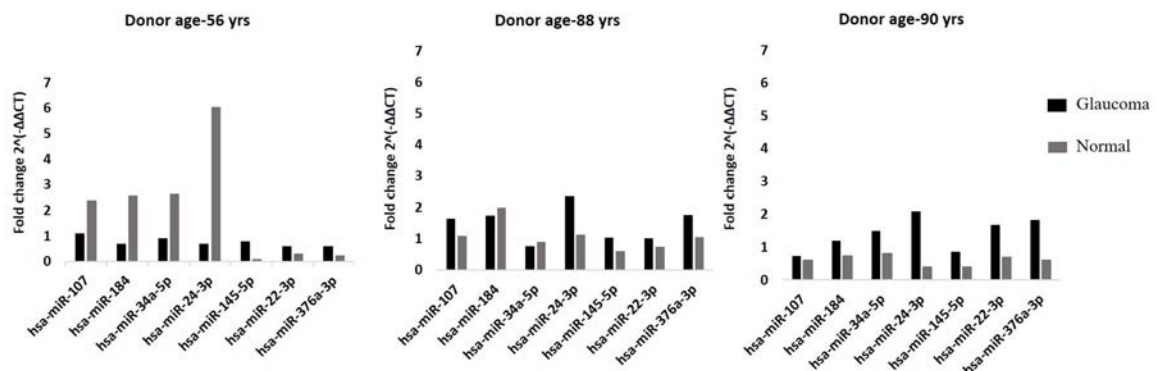
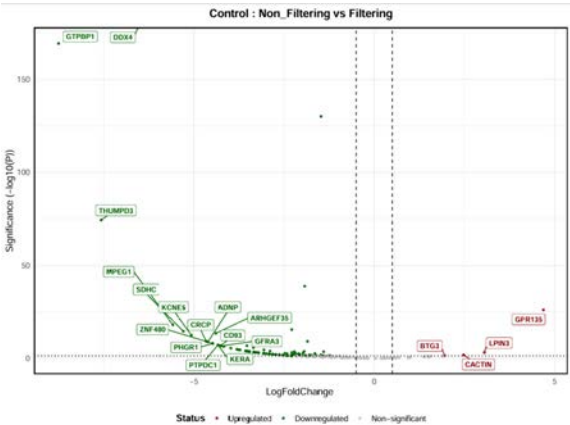


Figure 1: Differential expression of seven selected miRNAs in TM tissues from glaucomatous donors (black bars) compared to age-matched normal controls (grey bars) analysed by quantitative real-time PCR. Each bar represents the mean fold change from technical replicates. Expression was normalised to an internal control (U6) miRNA.

Figure 2. Volcano plot of differentially expressed genes between NF and F regions of human TM. The x-axis corresponds to the $\log_2$ fold change, and the y-axis corresponds to the $-\log_{10}$ adjusted p-value. Significantly upregulated genes in NF regions are shown in red, significantly downregulated genes in green, and non-significant genes in grey.



Results

Expression of differentially regulated miRNAs in Glaucomatous TM

Differential miRNA expression analysis using nSolver identified 26 upregulated and three downregulated miRNAs in the NF region of the TM with a fold change $> \pm 1.2$ and $p < 0.05$ of normal donor TM. In all the three glaucomatous donor tissues, the expression of the down-regulated miRNAs-miR-376a-3p, miR-22-3p and miR-145-5p were upregulated. Conversely, upregulated miRNAs including miR-107, miR-184, miR-34a-5p, and miR-24-3p, exhibited donor-specific variability, with all being downregulated in the 58-year-old glaucomatous donor. In the 88-year-old donor, miR-184 and miR-34a-5p were downregulated, whereas miR-107 and miR-24-3p were upregulated, and in the 90-year-old glaucomatous donor, miR-107

was downregulated while miR-24-3p, miR-184, and miR-34a-5p were upregulated (Figure 1).

Digital spatial transcriptomics identified differential gene expression between NF and F TM

GeoMx digital spatial transcriptomic analysis identified a total of 180 significantly differentially expressed genes between the NF and F region ($p < 0.05$). Of these, four genes were upregulated in the NF, including GPR135, LPIN3, CACTIN, and BTG2, while the remaining genes were downregulated (Figure 2). The most significantly downregulated genes included DDX4, GTPBP1, PTBP1, SUPT7L, MPEG1, SDHC, ARHGEF35, KCNE5, CRCP, ADAM15, and ADNP. Importantly, the consistent expression of established TM markers- CHI3L1, ANKRD1, MGP, AQP1, and MYOC confirmed the identity of the profiled regions and validated the spatial transcriptomics dataset.

Pathway analysis of differentially expressed genes in NF TM

KEGG pathway enrichment analysis using Shiny GO identified multiple significantly enriched pathways. Twenty pathways containing more than 20 target genes and meeting the significance threshold ($p < 0.05$) were selected for further analysis. Among these, MAPK, PI3K–AKT and Ras signalling pathways that are functionally associated with stemness were enriched (Figure 3). In addition, pathways involved in focal adhesion and regulation of actin cytoskeleton, were also significantly enriched.

Conclusion

The expression profile of miRNAs was altered in glaucomatous donor TM compared to controls. Spatial transcriptomic profiling identified genes highly expressed in the NF region of TM, where the stem

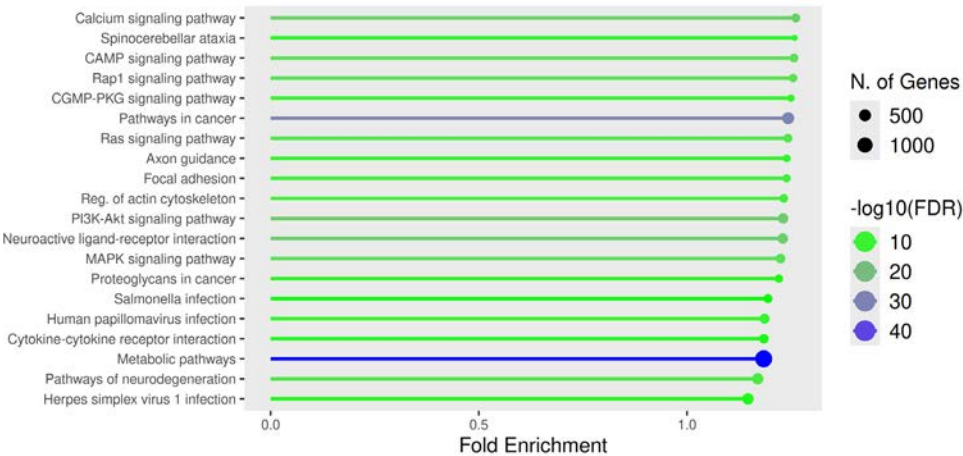


Figure 3: Shiny GO pathway enrichment analysis showing 20 significantly enriched pathways with more than 20 target genes

cells are located. Identification of genes that regulate the stem cells maintenance in the NF region and the changes in their expression in the glaucomatous condition will aid in developing better cell-free therapy for glaucoma wherein there is a TM stem cell and total cell loss.

Adult stem cell derived exosomes: A cell-free therapy for glaucoma

Investigator : Dr. Gowri Priya
Chidambaranathan
Co- Investigators : Prof. K. Dharmalingam,
Dr. S.R. Krishnadas, & Prof.
Nirmal Jayabalan
Post-Doctoral
Fellow : Dr. K. Balachander
Funding Agency : ICMR - Investigator Initiated
Projects-Intermediate Grant

Introduction including background:

TM drains the aqueous humour from the anterior chamber of the eye and thus regulates the IOP homeostasis. Previous studies from this laboratory demonstrated a drastic loss of TM cells and significant reduction in TMSCs in glaucomatous donor eyes indicating the need for a cell-based therapy for glaucoma. Studies using TMSC derived exosomes in vitro revealed their enhanced wound healing and anti-oxidant potential compared to TM

cell derived exosomes. In continuation, this study aims to develop clinical grade TMSC and Adipose Derived Stem Cell (ADSC) exosomes and evaluate their efficacy in restoring normal IOP in in vitro and in vivo steroid induced glaucoma models.

Results:

Establishment of TMSC Cultures under Xeno-free Conditions

Human TM cells were isolated from donor tissue and were cultured in standard Stem Cell Growth Medium (SCGM) and a commercial xeno-free medium STEMIn1™, while ADSCs were cultured in STEMIn1™. Phase-contrast microscopy showed that all cultures exhibited a typical mesenchymal morphology with uniform, elongated, spindle-shaped fibroblast-like cells (Figure 1).

Phenotypic Characterisation of Cultured Cells

Immunostaining was performed to assess stem cell marker expression in TMSCs cultured under standard and xeno-free conditions. Consistent with Iswarya et al. (2024), putative stem cell markers ABCG2 and p75 were minimal/absent by Passage 3 in both media, indicating loss of the naive stem cell phenotype upon in vitro expansion (Table 1). Despite this, TMSCs retained a MSCs phenotype (Figure 2). At Passage 3, cells cultured in STEMIn1™ expressed MSC markers CD90 (74%), CD105 (78%), and CD73



Figure 1: Representative phase-contrast images of human TMSCs cultured in SCGM (A) and xeno-free STEMIn1™ medium (B), and ADSCs cultured in STEMIn1™ medium at Passage 3 (C).

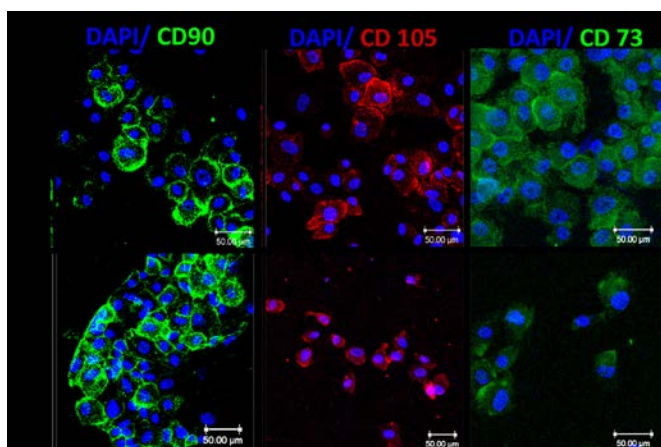


Figure 2. Representative confocal images of TM cells cultured in SCGM and STEMIn1™ medium stained for MSC markers CD90 (FITC-green), CD105 (Alexa 633-red) and CD73 (FITC-green) with nuclear counterstain-DAPI (blue).

Table 1. Comparison of stem cell marker expression in TMSCs cultured in SCGM and STEMIn1™

Markers	Percentage of positive cells					
	Passage 3		Passage 4		Passage 5	
	SCGM	STEMIn1	SCGM	STEMIn1	SCGM	STEMIn1
ABCG2	0	0	0	0	0	0
P75	2	4	0	0	0	0
CD90	72	74	65	67	53	56
CD105	76	78	67	65	49	53
CD73	75	79	66	63	54	56

Table 2. Stem cell marker expression of cultured ADSCs

Markers	Passage 3	Passage 4	Passage 5
CD90	95%	93%	91%
CD105	93%	91%	87%

(79%), comparable to those cultured in SCGM, both media exhibited gradual marker decline by Passage 5, suggesting reduced stemness with prolonged culture (Table 1).

Phagocytic property of cultured TMSCs: The cells cultured in both SCGM and STEMIn1™ medium effectively internalised FITC-labelled zymosan particles, confirming preserved TM-specific phagocytic activity.

Characterisation of ADSCs: ADSCs cultured in xeno-free STEMIn1™ medium showed higher MSC marker expression. The expression of CD90 and CD105 was about 90% from passage 3 to passage 5 (Table 2).

Conclusion

A clinically relevant, xeno-free culture system for expanding TMSCs and ADSCs was established supporting their suitability for downstream cell-free therapeutic applications.

PROTEOMICS

The Proteomics Department employs multi-omics approaches integrated with advanced cell and molecular biology techniques and robust computational analyses to investigate major ocular diseases contributing significantly to vision loss in the Indian population, including diabetic retinopathy, fungal keratitis, keratoconus, pterygium, glaucoma and age-related macular degeneration. The department explores disease mechanisms, identify biomarkers, and develop therapeutic interventions for various ocular diseases. The department also started to study the inter-relationship between chronic metabolic conditions in diabetes and glaucoma through a major grant from ICMR. In addition to the existing mass spectrometers at the proteomics facility, the newly added state-of-the-art equipment, named O-link Proteome analyzer and a flow cytometer to strengthen the department's research activities. The department continues to study the role of extracellular vesicles (EVs) in ocular diseases, utilizing ultracentrifugation and Nanoparticle Tracking Analysis (NTA) housed in the Exosome innovation centre. The department aims to translate the insights gained from the research into diagnostic and therapeutic strategies to enhance disease management and improve patient outcomes.

Characterisation of the secretome and secretome - derived Extracellular Vesicles from *Aspergillus flavus* saprophyte and clinical isolates.

Investigators : Prof. K. Dharmalingam
Research Fellows : G. Hariharan &
K.S. Vedha Varshini

Introduction and Background

Extracellular vesicles (EVs) of Fungi are nanostructured vesicles, surrounded by intact lipid bilayer and are involved intercellular communication, virulence, pathogenesis and environmental adaptation etc. The cargo of the EVs includes proteins, lipids, and nucleic acids, which facilitate the host-pathogen interactions. Understanding the composition and functional role of the secretome

proteins and EVs from the saprophyte and clinical isolates grown under different culture conditions will provide valuable insights into their functional capability and perhaps their potential diagnostic utility.

The current study focuses on the proteomics of the secretome and the EVs from *A.flavus* saprophyte (ATCC26) and clinical isolate (MTCC 13369) grown in solid and liquid culture conditions in different media

Summary of Previous Work:

The growth rate of the clinical isolates is more sensitive to Congo red (CR) compared to saprophyte. CR treatment markedly altered protein cargo of the EVs isolated from the drug treated cultures. Pathway and gene ontology enrichment analyses of the EV proteins demonstrated that CR treatment altered the pathways related to cell wall organisation and polysaccharide catabolism. These observations



highlighted the differential stress response of clinical isolates compared to saprophytes.

Proteome Profiling of Secretome and EVs derived from the secretome of *A. flavus* isolates.

Cultures were propagated on wheat bran as described previously. Secreted proteins including the EVs were collected and filtered and then used for EVs isolation using ultracentrifugation. Secretome and EV proteins were identified using Orbitrap Mass Spectrometer.

Comparison of the proteome profile (fig 1.) of the secretome of Saprophyte and the clinical isolate showed that about 50% of the proteins are common between the two cultures. Ideally the EV cargo also should be included in the identified proteins of the unfractionated secretome, if the experimental set up is ideal. Therefore, we focused on the EV proteins from the EVs from the secretome of both cultures further.

1. Gene Ontology-Based Comparative Functional Profiling of *Aspergillus flavus* EVs and Secretome:

The distribution of the proteins in the secretome and the EV fraction of the secretome is shown below.

Gene ontology (GO) analysis was performed using the FunRich software to classify identified proteins under biological process, molecular function, and cellular component categories. Total number of proteins identified on EV is ATCC 26- 1034 (29 was recognised using uniprot and 24 was annotated) MTCC 13369 – 1318 (35 was recognised using uniprot and 28 was annotated). In secretome ATCC 26- 1167 (44 was recognised and 42 was annotated) MTCC 13369 – 1048 (42 was recognised and 39

was annotated. These annotated protein/Genes were used for the Gene ontology analysis. Proteins in the EV fractions were predominantly associated with translational processes, ribosomal organisation, carbohydrate metabolism, cellular stress response and amino acid biosynthesis pathways whereas the total secretome showed enrichment in cell wall modification, carbohydrate catabolism, secondary metabolite biosynthesis, extracellular and metabolic pathways.

Under molecular function, the EV proteomes were characterised by enrichment of catalytic, structural, and RNA-binding activities, consistent with ribosomal and enzymatic proteins. Secretome by contrast, displayed stronger representation of hydrolase, oxidoreductase, and transferase activities, reflecting diverse extracellular enzymatic roles.

In terms of cellular component, EV-associated proteins were primarily mapped to cytosolic and ribosomal compartments, including the cytosolic small ribosomal subunit (up to 30% enrichment), cytosol, and Golgi apparatus, indicating the intracellular origin of many vesicular constituents. In contrast, the secretome proteins were predominantly have proteins of the cell wall, plasma membrane, and extracellular regions, consistent with their secretory nature.

Overall, FunRich-based GO categorisation revealed distinct functional segregation between the vesicle-specific and total secreted protein repertoires of both *A. flavus* isolates. The EV fractions were enriched in intracellular and translational components, while the secretome were dominated by extracellular enzymatic and metabolic proteins, reflecting two complementary aspects of the fungal extracellular proteome.

Comparison of EV proteome and the secretome proteome of Saprophytic and clinical isolate culture.

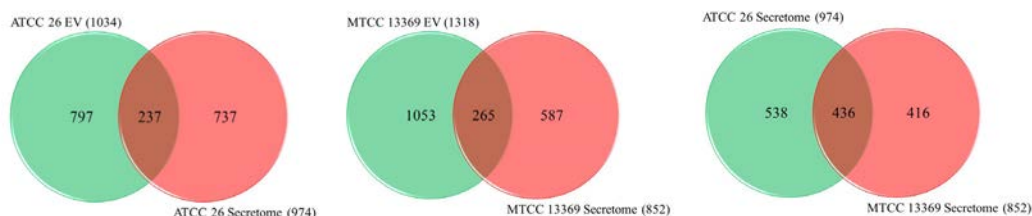


Fig. 1: Venn diagrams showing shared and unique proteins between *A. flavus* ATCC 26 and MTCC 13369 EV and secretome fractions.

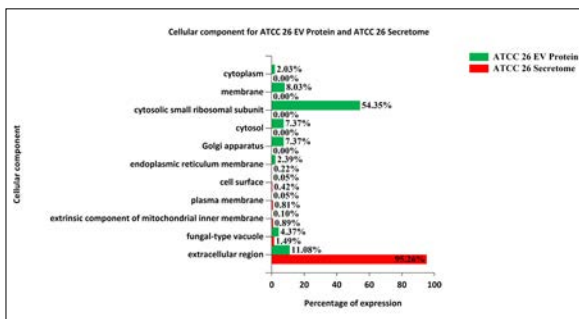
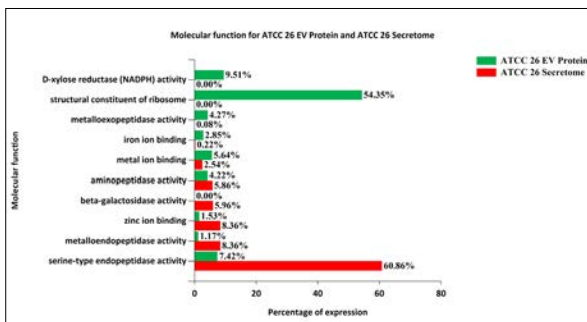
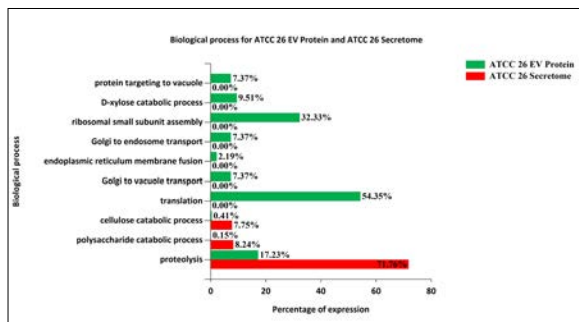


Fig. 2: GO analysis of *Aspergillus flavus* ATCC 26 EV and secretome proteins showing enrichment under biological process, molecular function, and cellular component categories. EV proteins (green) were mainly ribosomal and translational, while secretome proteins (red) were enriched in proteolysis and extracellular components.

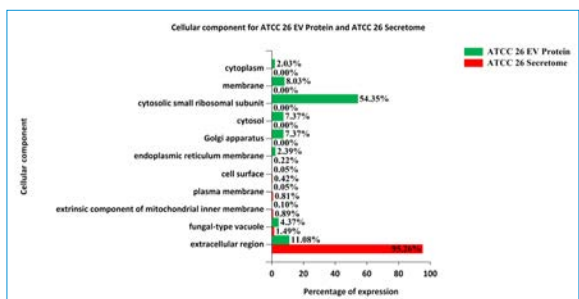
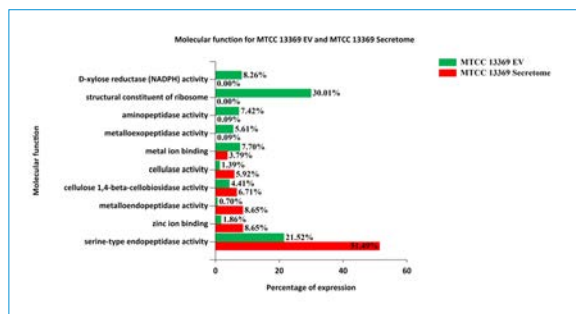
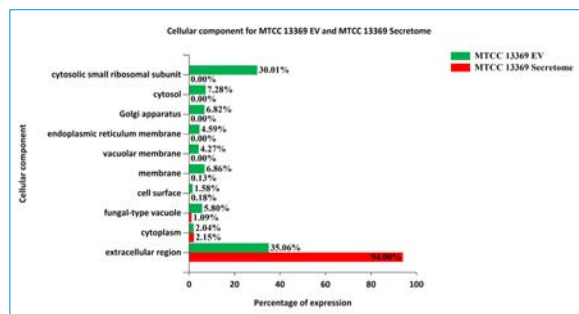


Fig. 3: GO analysis of *Aspergillus flavus* MTCC 13369 EV and secretome proteins showing enrichment under biological process, molecular function, and cellular component categories. EV proteins (green) were mainly ribosomal and translational, while secretome proteins (red) were enriched in proteolysis and extracellular components.

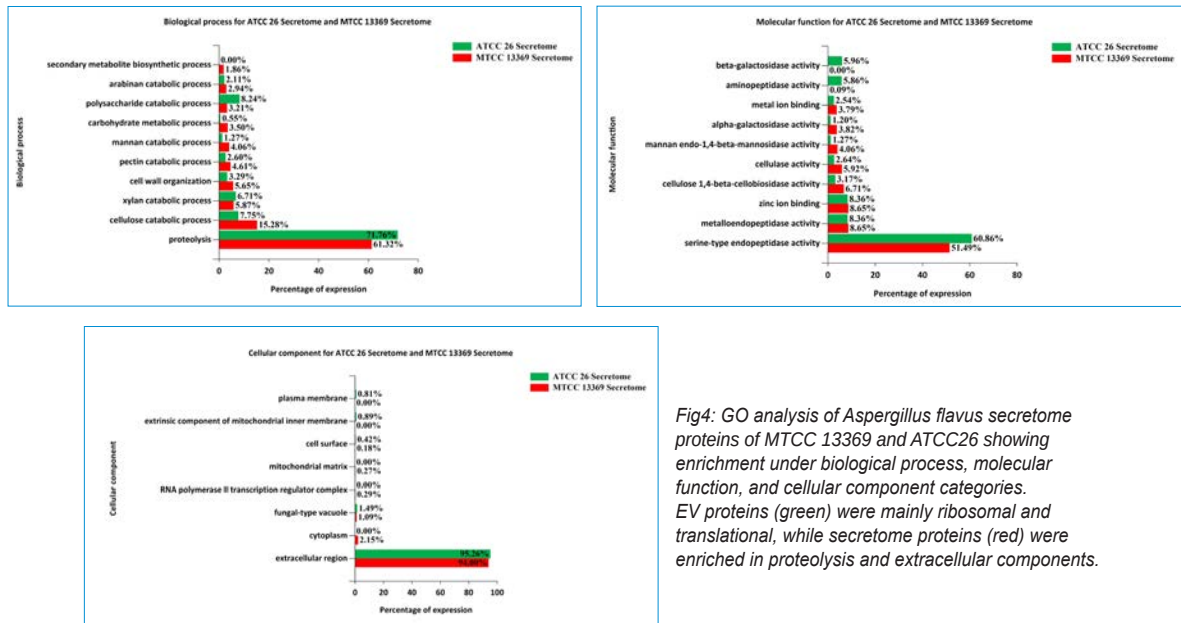


Fig4: GO analysis of *Aspergillus flavus* secretome proteins of MTCC 13369 and ATCC26 showing enrichment under biological process, molecular function, and cellular component categories. EV proteins (green) were mainly ribosomal and translational, while secretome proteins (red) were enriched in proteolysis and extracellular components.

Conclusion:

Extracellular vesicles showed enrichment of some specific pathways compared to the secretome. The functional implication of this enrichment indicates that the exosome cargo has the ability to affect the interaction and outcome specifically. The ability of the EVs to enter and alter the host cells indicate that enriched pathways are of significance in host pathogen interaction.

Functional characterisation and identification of the cargo of *A.flavus* EVs

Investigator : Prof. K. Dharmalingam
Research Fellow : Dr. K. Lavanya

Introduction and background

During ocular surface fungal infection, EVs first encounter the corneal epithelium, the outermost layer of the cornea; therefore, Human Corneal Epithelial (HCET) cells were used as an in-vitro model to study

fungal EV–host cell interactions. For fungal EVs to exert biological effects, either they must be efficiently internalised and deliver functional cargo intracellularly or bind to appropriate receptors to activate the recipient cells. Dil-based membrane labelling was used to visualise EV uptake by HCE.

Results

Fungal EVs are internalised by HCET cells.

Fungal EVs labelled with the lipophilic membrane dye Dil showed clear uptake by HCET cells following 24 hours of co-culture at a Multiplicity of Infection (MOI) of 5000. Confocal microscopy revealed distinct intracellular punctate red fluorescence distributed within the cytoplasm, indicating internalisation of intact EVs rather than surface association. Uptake was visualised after fixation, with DAPI counterstaining confirming cellular localisation. These observations demonstrate that fungal EVs are efficiently internalised by HCET cells through active uptake mechanisms.

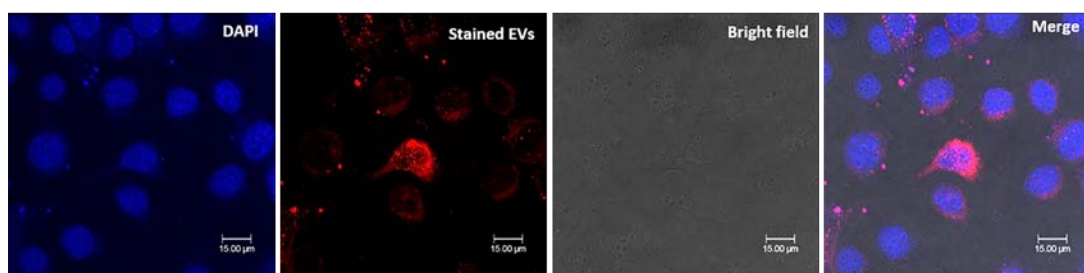


Fig 1. Dil-based uptake of fungal extracellular vesicles by HCET cells. Representative confocal microscopy images showing uptake of Dil-labelled fungal EVs by HCET cells after 24 h of co-culture at a multiplicity of infection (MOI) of 5000. EV membranes were labelled with Dil (red). Cell nuclei were counterstained with DAPI (blue). Cells were fixed prior to imaging. Scale bar: 15 µm.

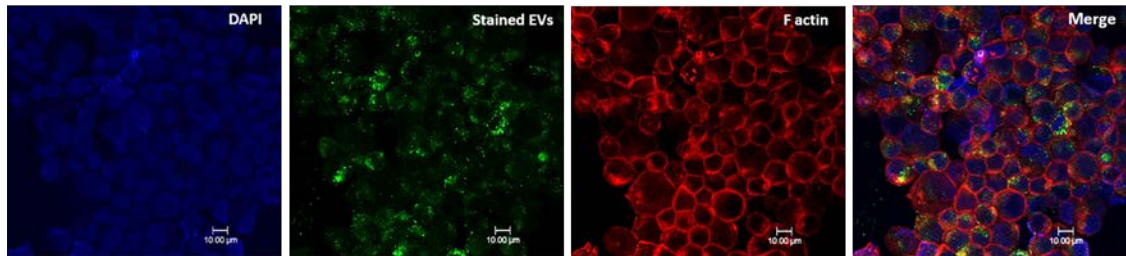


Fig 2. Functional uptake of fungal EV cargo assessed by Calcein-AM assay. Representative confocal images of HCET cells incubated for 24 h with Calcein-AM-labelled fungal EVs at an MOI of 5000. Actin cytoskeleton was stained with phalloidin (red), and nuclei were stained with DAPI (blue). Cells were imaged without fixation. Scale bar: 10 µm

Functional characterisation of *A.flavus* EVs after the uptake by HCET

EV integrity and functional cargo retention after uptake, was demonstrated using fungal EVs labelled with Calcein-AM prior to incubation with HCET cells. Confocal microscopy showed intracellular green fluorescence, indicating uptake of intact EVs containing esterase, with F-actin co-staining confirming intracellular localisation.

Conclusion

The HCET cells demonstrate the ability to internalise fungal EVs, and further studies are in progress to evaluate the effects of these EVs on recipient cells.

Characterisation of Cell-Derived Nanovesicles from Human Corneal Epithelial Cells incubated with Fungal Extracellular Vesicles and Spores

Investigator : Prof. K. Dharmalingam
Research Fellow : Karthik Alagarsamy

Introduction and Background

Cell-Derived Nanovesicles (CDNs) are membrane-bound vesicles typically ranging from 30–200 nm in size and closely resemble exosomes in terms of composition, structure, and biological function. CDNs are artificially prepared as described before. The objective of this study was to compare the characteristics of the CDNs from the HCET infected with fungal EVs.

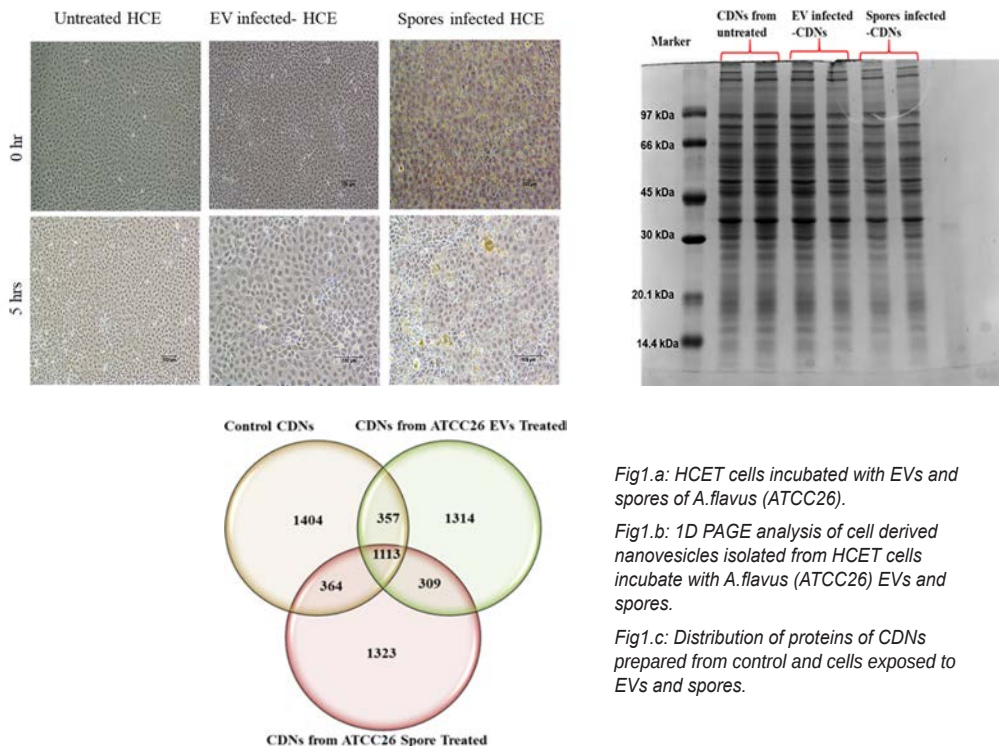


Fig1.a: HCET cells incubated with EVs and spores of *A.flavus* (ATCC26).

Fig1.b: 1D PAGE analysis of cell derived nanovesicles isolated from HCET cells incubate with *A.flavus* (ATCC26) EVs and spores.

Fig1.c: Distribution of proteins of CDNs prepared from control and cells exposed to EVs and spores.

Results:

HCET cells were cultured and used for infection with fungal spores or extracellular vesicles derived from ATCC-26 as described before. Cells were incubated with fungal spores or EVs for up to 5 hours. Microscopic examination revealed clustering of fungal spores on the cell surface after 5 hours of incubation (Fig 1a). CDNs were generated using repeated freeze–thaw cycles as described previously.

Comparative analysis showed no major differences in the overall protein profiles among control, and experimental samples in 1D PAGE analysis (Fig 1b).

CDNs obtained after size-exclusion chromatography showed a size range of 50 – 350 nm in NTA analysis with an average concentration of 1.38×10^9 particles produced by 1×10^7 cells.

CDN proteins resolved on SDS-PAGE gels were analysed using a shotgun proteomics approach. More than 3000 proteins were identified from all three sample and comparative analysis was performed to identify common and unique proteins across control, EV-infected, and spore-infected CDN samples (Fig 1c). Further analysis is in progress.

Conclusion:

The interaction of EVs and spores with HCET cells led to distinct pattern of proteins in the CDNs.

Repurposing statins as anti-fibrotic agents targeting the TGF-β–ECM–Myofibroblast axis in pterygium

Investigator : Dr. Daipayan Banerjee
Co-Investigators : Dr. Tejaswi Prasad,
Dr. N. Venkatesh Prajna &
Prof. K. Dharmalingam
Research scholar : Mathan L
Funding Agency : SERB-SRG

Introduction:

Pterygium is a common ocular surface disorder characterised by a fibrovascular growth onto the cornea, driven by chronic inflammation, oxidative stress, and extracellular matrix (ECM) remodelling. Although surgical excision remains the standard of care, increasing attention is being directed toward molecular mechanisms that could enable pharmacological modulation of disease progression.

Recent studies have identified several candidate therapeutics targeting fibrotic, oxidative, and inflammatory pathways. Kisspeptin inhibits chemokine (C-X-C motif) ligand signalling to suppress pterygial cell proliferation (1), while

Nintedanib—an FDA-approved tyrosine kinase inhibitor for pulmonary fibrosis—modulates the miR-23b-3p/TGFB2 axis to block epithelial–mesenchymal transition (EMT) (2). Celestrol activates the SIRT1/PGC-1α/Nrf2 pathway to mitigate oxidative injury (3), MERTK inhibition reduces fibroblast proliferation (4), and Teprotumumab—an FDA-approved IGF-1 receptor antagonist—decreases fibroblast viability (5). These findings underscore the therapeutic potential of repurposing drugs with established safety profiles for ocular fibrosis.

Our transcriptomic and proteomic profiling of pterygium and control conjunctiva revealed heightened activation of the TGF-β–mediated ECM–myofibroblast signalling axis, a core driver of fibrotic remodelling. Prior reports support this pathway as a pharmacologic target: Cyclosporine A inhibits TGF-β2–induced myofibroblast differentiation (6), and Bromfenac attenuates TGF-β1–driven fibrosis (7). Given their pleiotropic properties, statins—widely used HMG-CoA reductase inhibitors—offer a promising drug-repurposing opportunity for pterygium. Beyond lipid lowering, statins exert anti-inflammatory, anti-oxidative, and anti-fibrotic effects, including suppression of TGF-β–induced myofibroblast activation (8). Compared with other experimental agents, statins offer the advantage of an established systemic safety profile, multi-targeted anti-fibrotic activity, and ready clinical translatability through drug repurposing. This study investigates the potential of statins to modulate the TGF-β–ECM–myofibroblast axis in human conjunctival fibroblasts and pterygium fibroblast, establishing a mechanistic rationale for their therapeutic application in pterygium.

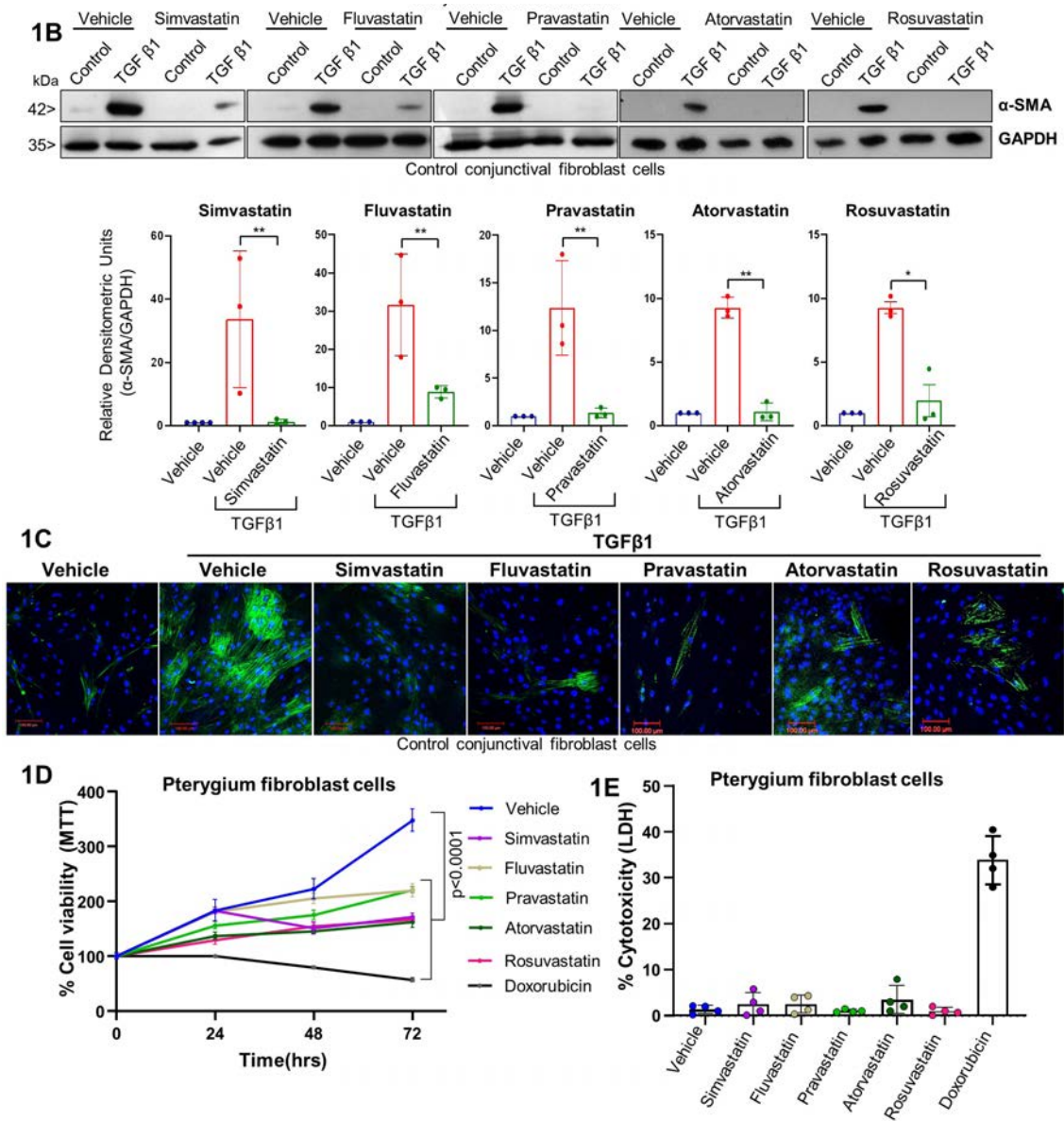
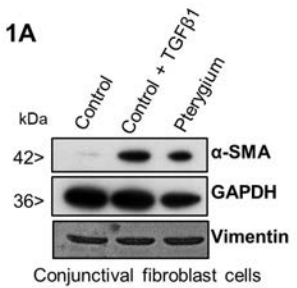
Results:

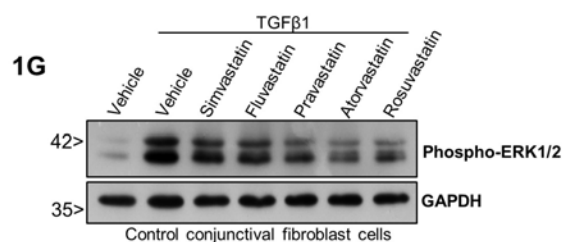
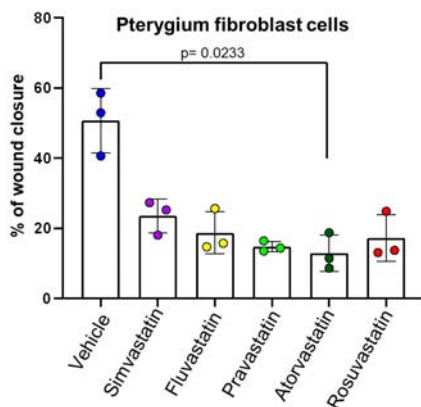
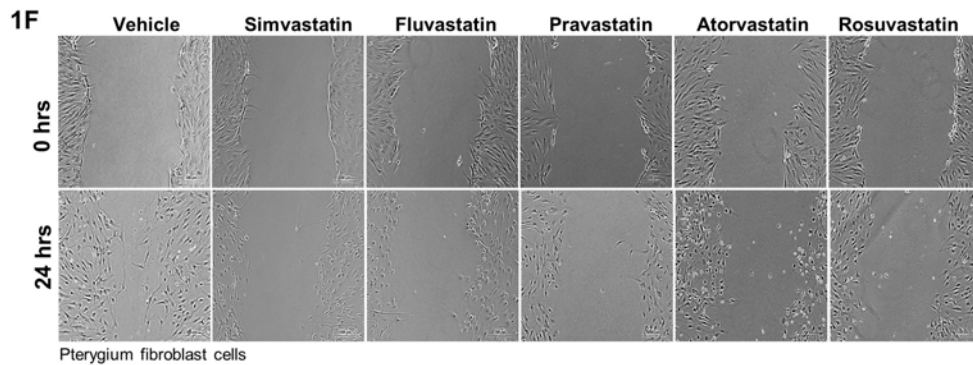
Primary human conjunctival fibroblasts (HCFs) and pterygium fibroblasts (HPFs) were cultured under standard conditions. For induction of myofibroblast differentiation, cells were stimulated with recombinant human TGF-β1 (2 ng/mL) for 48 h. To evaluate the effects of statins, cells were pre-treated for 2 h with simvastatin (1 μM), fluvastatin (1 μM), pravastatin (10 μM), atorvastatin (10 μM), or rosuvastatin (10 μM) prior to TGF-β1 exposure. Protein expression of α-smooth muscle actin (α-SMA) and phosphoERK1/2 was analysed by immunoblotting using GAPDH as a loading control. Immunofluorescence staining was performed to visualise α-SMA expression in statin-treated and control fibroblasts. Cell viability and proliferative capacity were determined using the MTT assay after 72 h of treatment, whereas cytotoxicity was assessed by quantifying lactate dehydrogenase (LDH) release after 48 h. Fibroblast migration was evaluated using a scratch wound-healing assay, in

which confluent monolayers were linearly scratched and wound closure imaged at 0 h and 24 h. For signalling analyses, cells were pre-treated with statins for 2 h followed by TGF- β 1 stimulation (2 ng/mL) for 1 h, and Iuvasta-ERK1/2 levels were examined by Western blot.

Comparative analyses confirmed that TGF- β 1 treatment induced α -SMA expression in control conjunctival fibroblasts to levels comparable with those observed in untreated pterygium fibroblasts (Fig. 1A). Treatment with simvastatin, fluvastatin, pravastatin, atorvastatin, and rosuvastatin significantly reduced α -SMA expression, as demonstrated by immunoblotting and immunofluorescence (Fig. 1B and 1C). Consistent with this, statin-treated fibroblasts displayed reduced proliferative activity as assessed

by MTT assay, without evidence of cytotoxicity based on LDH release, in contrast to doxorubicin-treated controls which exhibited both proliferation loss and cell death (Fig. 1D-E). Additionally, scratch wound assays revealed a marked decrease in fibroblast migration following statin treatment (Fig. 1F). Mechanistically, statin exposure attenuated TGF- β 1-induced phosphorylation of ERK1/2, suggesting suppression of downstream pro-fibrotic signalling (Fig. 1G).





Conclusion:

Collectively, these findings demonstrate that statins effectively suppress TGF-β1-induced myofibroblast differentiation, proliferation, and migration in human conjunctival fibroblasts through attenuation of ERK1/2-mediated pro-fibrotic signalling. These results identify statins as potent modulators of the TGF-β–ECM–myofibroblast axis, supporting their potential repurposing as anti-fibrotic therapeutics for pterygium.

Proteomic profiling of cell-derived nanovesicles reveals fibroblast activation signatures in pterygium and TGF-β1–stimulated conjunctival fibroblasts

Investigator : Dr. Daipayan Banerjee
 Co-Investigators : Dr. Tejaswi Prasad,
 Dr. N. Venkatesh Prajna &
 Prof. K. Dharmalingam
 Research Scholar : Mathan L
 Funding Agency : SERB-SRG

Introduction:

Cell-derived nanovesicles (CDNs) serve as critical mediators of intercellular communication, carrying proteins that mirror the physiological and activation

state of their parent cells. In fibroblasts, these vesicles can reflect dynamic changes in extracellular matrix remodelling, stress responses, and signalling activity. Profiling the proteomic content of fibroblast-derived CDNs provides insight into the cellular pathways that are activated or suppressed during phenotypic transitions such as myofibroblast differentiation. This study was designed to define the protein composition and signalling pathways represented in CDNs secreted by conjunctival fibroblasts under different activation states. Comparative proteomic analysis was performed between CDNs derived from primary pterygium fibroblasts and those from healthy conjunctival fibroblasts, as well as from TGF-β1–stimulated fibroblasts, to identify shared and distinct molecular features of fibroblast activation. This approach enables the identification of CDN-associated proteins and pathways that may mediate extracellular matrix remodelling, oxidative stress adaptation, and altered vesicle-mediated signalling characteristic of fibroblast activation.

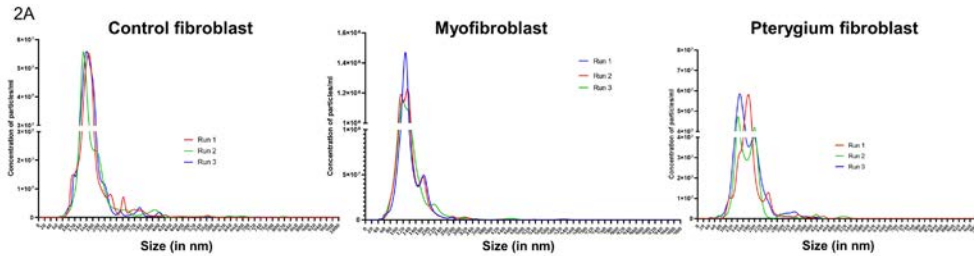
Results:

CDN isolation and characterisation: Primary fibroblast cultures were established from excised pterygium tissue (during pterygium excision surgery) and from normal conjunctiva obtained during small-incision cataract surgery (SICS) (N=3 patients in each

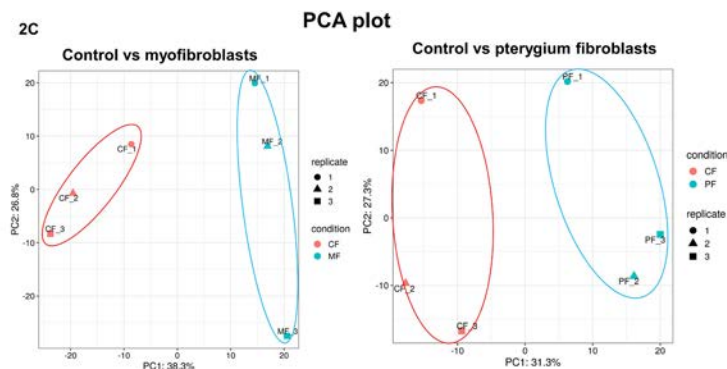
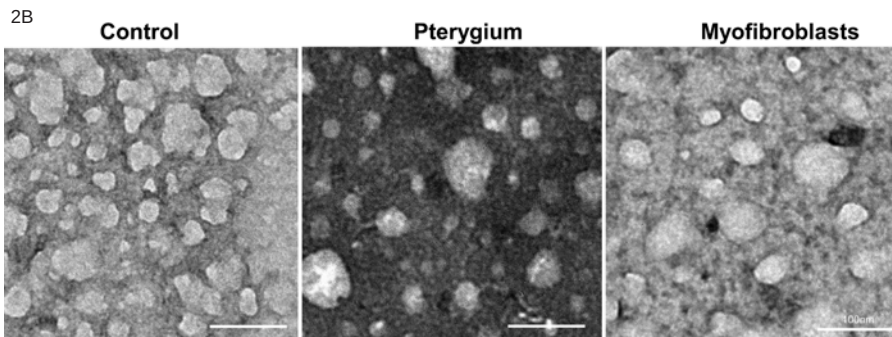
group). To generate CDNs, the cell suspension was subjected to repeated freeze–thaw cycles to induce mechanical membrane disruption. The nanovesicle-containing supernatant was subsequently processed by serial membrane extrusion through polycarbonate membranes of defined pore sizes (e.g., 10 μm , 8 μm , 400 nm followed by 200 nm) using a mini-extruder system to obtain uniformly sized CDNs. Nanoparticle Tracking Analysis (NTA) and Transmission Electron Microscopy (TEM) confirmed size distribution and vesicular morphology of CDNs (Figure 2A, 2B).

Proteomic analysis:

CDN proteins were extracted, digested with trypsin, and analysed using shotgun liquid chromatography–tandem mass spectrometry (LC–MS/MS). Three independent patient-derived samples were analysed per group. Only proteins consistently detected in all three biological replicates were retained for downstream analyses. Principal Component Analysis (PCA) demonstrated clear clustering of samples within each group (Figure 2C).



	Conjunctival fibroblast	Myofibroblast	Pterygium fibroblast
Mean(nm)	171.8	148.0	196.3
Mode(nm)	166.9	143.6	162.0
D10(nm)	190.1	176.3	129.3
D50(nm)	191.4	190.0	176.8
D90(nm)	265.3	170.8	239.5
CDN concentration/million cells	4.2×10^8	4.88×10^8	3.04×10^8



Shotgun proteomic analysis identified 979 proteins in CDNs derived from healthy conjunctival fibroblasts (control group), 968 proteins in CDNs derived from pterygium fibroblasts, and 776 proteins in CDNs derived from TGF- β 1-treated fibroblasts (myofibroblasts). Among these, 676 proteins were common between the healthy and pterygium groups and were selected for comparative quantitative analysis. Using a threshold of $|\log_2 \text{fold change}| > 0.5$, 123 proteins were significantly upregulated and 37 proteins were downregulated in CDNs from pterygium fibroblasts compared with controls. Comparative analysis between the healthy and myofibroblast groups revealed 574 commonly identified proteins, among which 141 were upregulated and 74 were downregulated in CDNs derived from TGF- β 1-treated fibroblasts.

Comparison of CDNs derived from pterygium and healthy conjunctival fibroblasts:

Proteomic analysis of CDNs derived from pterygium fibroblasts revealed a distinct molecular signature consistent with persistent fibroblast activation, oxidative stress adaptation, and secretory remodelling. The upregulated proteome was dominated by proteins involved in extracellular matrix (ECM) organisation, cytoskeletal remodelling, and vesicular trafficking, including SPARC, Calponin-3 (CNN3), Tropomyosin- α 3 (TPM3), Paxillin (PXN), and Cysteine- and glycine-rich protein-1 (CSRP1), which collectively reflect enhanced contractility and focal adhesion signalling. Elevated levels of Annexins (A3, A5, A11), Rab5C, VPS26A, and TRIM25 indicate increased vesicle biogenesis and exosomal secretion, while enrichment of ER chaperones (FKBP9, FKBP10, ERP29) and proteasome regulators (PSMB10, PSME1, UBA7) suggests heightened proteostatic control. Metabolic and antioxidant enzymes such as LDHA, GSTP1, PRDX6, GLRX3, and ALDH1A3 highlight redox homeostasis and metabolic resilience, features typical of long-term oxidative stress adaptation. Conversely, the downregulated proteome comprised proteins associated with translation, RNA processing, and nuclear maintenance, including ribosomal subunits (RPL3, RPL4, RPLP1), translation factors (EIF4E, EIF5B, EIF6), RNA-binding proteins (FUS, PRMT1), and nuclear transport mediators (KPNA4, KPNA5, IPO11), indicating reduced biosynthetic and nuclear renewal capacity. The decline in DNA-repair enzyme PRKDC, mitochondrial aconitase (ACO1), and heat-shock protein HSP105 further points to suppressed nuclear and metabolic activity, balancing the energetic demands of chronic activation. Collectively, these alterations define a fibrotic, stress-tolerant vesicular proteome, wherein pterygium fibroblasts downregulate general biosynthetic pathways while

selectively enriching CDNs with ECM, redox, and signalling cargo—hallmarks of a chronic, secretory fibroblast phenotype central to pterygium pathogenesis.

Comparison of CDNs derived from TGF- β 1-treated and healthy conjunctival fibroblasts:

Proteomic profiling of CDNs from TGF- β 1-stimulated conjunctival fibroblasts revealed a coordinated reprogramming of the vesicular proteome reflecting fibroblast activation, cytoskeletal remodelling, and metabolic adaptation. Upregulated proteins prominently included cytoskeletal and contractile regulators such as Transgelin (TAGLN), Calponin-3 (CNN3), Tropomyosins (TPM3, TPM4), Cofilin-2 (CFL2), Myosin regulatory light chain-9 (MYL9), and Actin-related proteins (ARPC1B, ARPC2, ACTR2), indicative of actin filament reorganisation and a transition toward a myofibroblast phenotype. Concurrent elevation of SPARC, Fibrillin-3, and Annexins (A2, A5) denotes enhanced ECM deposition and mechanotransductive signalling. Metabolic enzymes including LDHA, ALDOA, PGK1, G6PI, and NNMT reflected a glycolytic and redox-adaptive metabolic shift, while upregulation of proteasomal subunits (PSMA2, PSMB5, PSMB6) and 14-3-3 family proteins (YWHAЕ, YWHAG, YWHAZ) highlighted increased proteostasis and stress-response activity. Conversely, downregulated proteins comprised ribosomal subunits (RPL3, RPL4, RPL22, RPL36, RPS23), tRNA synthetases, and RNA helicases (DDX6, DHX9), consistent with suppressed translation and RNA metabolism. Reduced expression of nuclear transport factors (IPO7, XPO-T, UBC9, SAE2) and DNA-repair enzymes (PRKDC, XRCC5) indicates downscaling of nuclear maintenance processes, while decreased metabolic enzymes (ACO1, TKT, LTA4H) points to metabolic reprioritisation. Together, these shifts suggest that TGF- β 1 drives fibroblast CDNs toward a selective, stress-resilient secretory phenotype, emphasising ECM and signalling cargo at the expense of global biosynthetic and housekeeping functions—mirroring the early molecular hallmarks of fibrotic myofibroblast activation.

Conclusion:

The present study provides the first proteomic characterisation of cell-derived nanovesicles (CDNs) from conjunctival fibroblasts under basal and activated states, revealing a coordinated shift toward extracellular matrix remodelling, oxidative stress adaptation, and vesicular signalling under TGF- β 1 stimulation and in pterygium-derived cells. Future work should focus on functional validation and mechanistic dissection of these findings to establish

how specific vesicular proteins influence recipient cell behaviour and contribute to fibroblast–epithelial or fibroblast–vascular communication in the conjunctival microenvironment.

In –vitro analysis of wild type and mutant myocilin functions in HEK 293T cells and conjunctival fibroblast cells

Investigators : Dr. O.G. Ramprasad,
Dr. A. Vanniarajan,
Dr. D. Bharanidharan &
Prof. K. Dharmalingam
Funding Agency : Aravind Eye Hospital

Introduction including background:

Myocilin protein is expressed in many ocular tissues, including the trabecular meshwork, and was revealed to be the trabecular meshwork glucocorticoid-inducible response protein. The trabecular meshwork regulates intraocular pressure, and mutations in MYOC have been identified as the cause of hereditary juvenile-onset open-angle glaucoma (JOAG). In a recent study from the lab (Kader et al., 2024), myocilin gene mutation N480K (c.1440C>G) was detected in 20 members of a family with some having JOAG and some having primary open angle glaucoma. It is known that mutations in coding region of myocilin gene causes misfolding of the protein which leads to intracellular sequestration and resultant cellular cytotoxicity (Scelsi et al., 2023). Wild type myocilin is usually secreted by the cells to carry out its physiological functions. While the physiological roles of other myocilin mutations are known, the cellular role of N480K mutation remains

unknown. Hence our objective was to compare N480K expression with respect to wild type myocilin gene and characterisation of the protein complex in HEK293T and fibroblast cell line. We also wanted to analyse the secretory and other biochemical properties of N480K mutant in comparison to wild type myocilin in fibroblast cells.

Results and Conclusion:

In a preliminary study, wild type myocilin construct (gift of Prof. Raquel Lieberman, USA) was transfected into HEK293T cell line and primary conjunctival fibroblasts derived from conjunctiva of patients undergoing cataract surgery. Transfection efficiency was very high in HEK293T cells (75%-80%) but was very low in primary conjunctival fibroblasts (10%-12%) as assessed by transfecting a GFP expressing plasmid.

HEK293T cells transfected with wild type myocilin (55-60 kDa), expressed the protein in secreted (cell culture medium) and insoluble fractions (membranous components, cellular and protein aggregates). No expression was seen in soluble fraction (cell lysate) (Fig. 1). Extra myocilin bands were seen in the secreted fraction of transfected cells (around 35 and 25 kDa). Control, untransfected cells showed no expression of myocilin in any of the fractions.

Primary conjunctival fibroblast cells did not secrete myocilin in the culture medium after transfection, but most of the transfected myocilin was present in the soluble fraction and in insoluble fraction (Fig.2). In conclusion, HEK293T cells recapitulated the secretory functions of wild type myocilin while conjunctival fibroblasts did not. In future studies, trabecular meshwork fibroblast cells will be used to study the functions of wild type and N480K mutant myocilin.

Fig :1

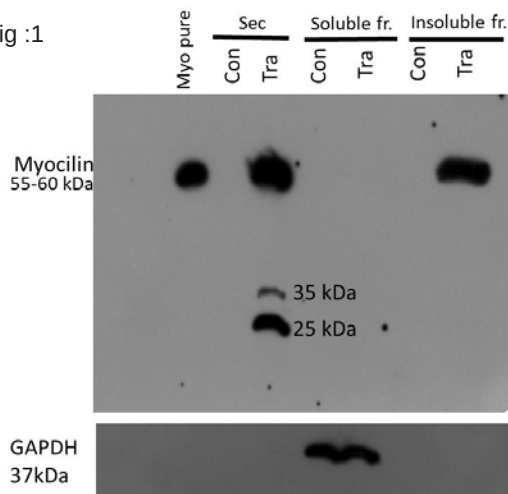
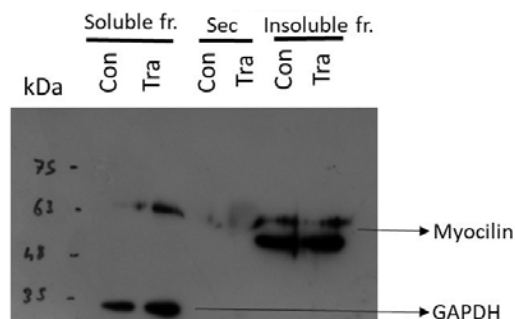


Fig :2



WT myocilin expression in HEK293T fractions (Fig. 1) and in primary conjunctival fibroblast fractions (Fig. 2). Myo pure is a pure protein for positive control. Sec stands for secreted fraction. GAPDH was used as a loading control for soluble fraction. Con represents untransfected control cells while Tra represents transfected cells.

OCULAR PHARMACOLOGY

Glaucoma is the second leading cause of blindness worldwide. Elevated IOP is the only modifiable risk factor and its medical management mainly relies on the topical application of anti-glaucoma medications which act either by reducing the aqueous humour formation (Adrenergic agonist, Beta blockers and carbonic anhydrase inhibitors etc.) or by increasing the uveoscleral outflow (prostaglandin derivatives). The surgical treatment of glaucoma lowers IOP either by decreasing aqueous inflow or by increasing aqueous outflow.

Trabeculectomy is the traditional incisional glaucoma filtration surgery in which aqueous outflow is enhanced either by augmenting the existing outflow pathways or by creating artificial outflow pathways into the sub-conjunctival or sub-tenon's space. The success of the trabeculectomy greatly depends upon the prevention of post-surgical scarring over the bleb with the use of antimetabolites, 5FU (5-fluorouracil) and MMC (Mitomycin C). Despite of this, about 10% of the patients fail surgery leading to vision-threatening complications. With the use of MMC can cause some complications such as hypotony maculopathy and a leaky bleb that is susceptible to endophthalmitis results in surgical failure. Therefore, there is a pressing need to develop/identify anti-fibrotic agents to preserve the filtering bleb and maintain good IOP control.

Ocular Pharmacology team is working on investigating the IOP lowering property and anti-fibrotic properties of human relaxin (RLX2) on TGF β 2-induced elevated IOP ex vivo model of glaucoma using Human Organ Cultured Anterior Segment (HOCAS).

Evaluation of IOP lowering property and Anti-fibrotic property of Relaxin on TGF β 2-induced Elevated IOP Ex vivo model of glaucoma using Human Organ Cultured Anterior Segment (HOCAS)

Investigator : Dr. S. Senthilkumari
Co-Investigator : Dr. R. Sharmila
Junior Research : Lakshmi Devi Priya & Mohana
Fellows : Jeya Priya
Funding Agency : Science & Engineering Research Board - Core Research Grant [2023-26]

Introduction

Relaxin is a heterodimeric peptide hormone with known vaso-dilatory and anti-fibrotic activity. This hormone was first identified by Frederick Hisaw in guinea pig model of pregnancy and parturition. In our previous study in identifying the differentially expressed genes between steroid responsive and non-responsive human cadaveric eyes in perfusion culture, we found that relaxin 1 gene was significantly down-regulated in steroid responder eyes as compared to non-responder eyes and its expression was evident in 7d group as compared to 16h steroid-treated HTM cells. However, the role of relaxin and its



non-allelic gene variants in human aqueous outflow facility and IOP regulation is not clearly understood.

Considering the distribution of relaxin and its receptors in human aqueous outflow pathway tissue, it is hypothesised that relaxin2 may have an active role in the regulation of outflow facility and IOP in human eye. Currently, the establishment of TGF β 2-induced elevated IOP ex vivo model of glaucoma has been established successfully. Further the demonstration of IOP lowering property of Relaxin2 on elevated IOP induced by TGF β 2 in an ex vivo model is in progress. We believe that relaxin 2 with proven IOP lowering and anti-fibrotic properties will be more beneficial in the management of glaucoma.

Results

Effect of Relaxin on Elevated IOP Induced by TGF β 2 Treatment using Human Organ Cultured Anterior Segment (HOCAS).

Human Donor Eyes: Post-mortem human eyes were obtained from the Rotary Aravind International Eye Bank, Aravind Eye Hospital, Madurai. All eyes were examined under the dissecting microscope for gross ocular pathological changes, and eyes without such changes were used for the experiments. The

presence or absence of glaucomatous changes in the study eyes was confirmed by histo-pathological analysis of the posterior segments. The study protocol was approved by the standing Human Ethics committee of our institute [RES2023022BAS]

HOCAS was established using paired human donor eyes as described previously by us and others (Ashwinbalaji et al., 2019; Haribalaganesh et al., 2022). After baseline (BL) equilibration (~2-3 days), anterior segments were exchanged with 5 ml of hrTGF β 2 (3ng/mL) at 200 μ l/minute as described by Gottenka et al., 2004 whereas, the contralateral anterior segment received the plain medium. Then the flow rate is resumed to 2.5 μ l/min and continued for 7 d. AH outflow facility (OF) (μ l per minute/mm Hg) was calculated as the ratio between the inflow rate (μ l/ minute) and the measured IOP (mm Hg). The OF was calculated every hour as the average of 6 values recorded every 10 minutes, beginning 4h before the drug infusion and continuing for the duration of the culture. The average OF value between 3–4h before hrTGF β 2 infusion was taken as baseline.

After TGF β 2 treatment for 7d, one eye received recombinant human relaxin (RLN2) (500ng/mL) whereas, the contralateral eye received the plain

Effect of RLN2 on the Expression of Fibrotic Markers by Western blotting

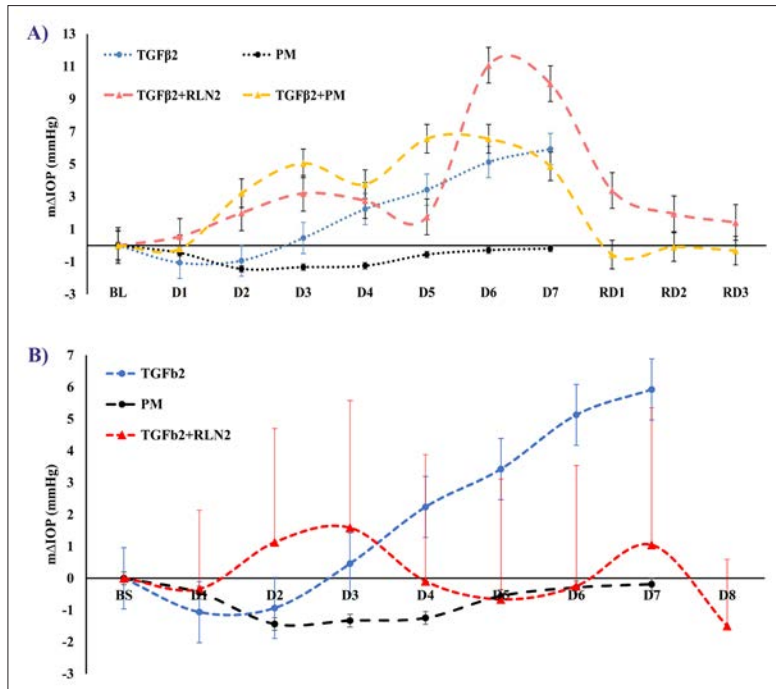
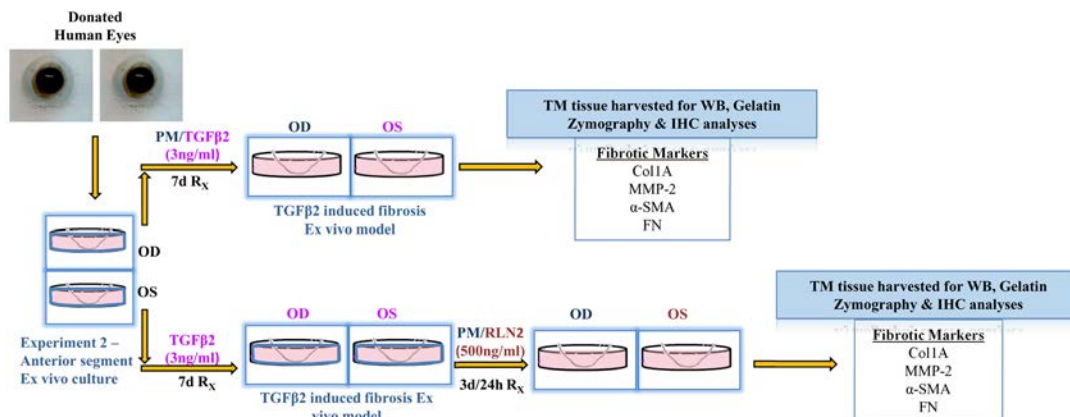


Figure showing the effect of Relaxin2 (RLX2) on TGF β 2-induced Elevated IOP

Note: TGF β 2-treatment reduced the outflow facility by 14%. The presence of RLN2 showed a marginal reduction in IOP in both 24h/3d post RLN2 treatment. A) TGF β 2 Treatment (7d) + RLN2 (3d); B) TGF β 2 Treatment (7d) + RLN2 (24h)



Study design showing the effect of RLN2 on the expression of fibrotic markers using anterior segment ex vivo culture model system.

There was a significant reduction in the fibrotic marker with 24h post RLN2 treatment

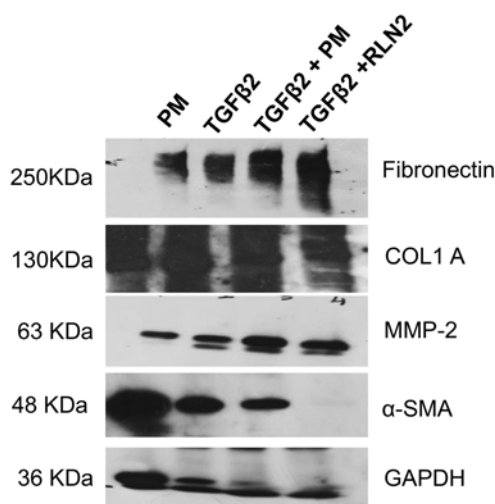


Figure Showing the Effect of RLN2 on Fibrotic Markers by Western blotting.

Note: The presence of RLN2 significantly reduced α -SMA expression and increased MMP2 expression as compared to TGF β 2 treated eyes. There is no significant change in the expression of COL-1A in 24h post RLN2 treated eyes as compared to TGF β 2 treated eyes.

For westernblot analysis, the TM tissues were isolated after the anterior segment ex vivo model with TGF β 2 +/-RLN2 treatment. Medium as vehicle control for another 3d/24h. At the conclusion of the experiments, the eyes were fixed by perfusion with 7 ml of 4% paraformaldehyde for assessing the morphology of aqueous outflow tissues and IHC analysis.

Conclusion

TGF- β 2 treatment reduced the outflow facility (OF) by 14 %. The reduction in OF was substantiated with the increased expression of fibrotic markers. No significant change in outflow facility was observed with RLN2 treatment. However, the presence of RLN2 alleviated the fibrosis induced by TGF- β 2 treatment.

Prospective study analysing the role of Cytokine- Mediated IOP reduction in POAG /OHTN patients receiving Therapeutic Ultrasound for Glaucoma (TUG) Treatment

Investigators : Dr. Sharmila Rajendrababu, Dr. Senthilkumari Srinivasan & Prof. K. Dharmalingam

Collaborators : Dr. Donald Schwartz, President and Founder, EyeSonix, Los Angeles, USA, & Eric Schultz, CEO, CTO, EyeSonix, Los Angeles, USA

Project Assistant : Fazil H.

Funding Agency : EyeSonix, USA

Introduction

Glaucoma accounts for 5.5% of total blindness caused in India is expected to affect 27.8 million more people of the Asian population by 2040. In consideration of alternative treatments for glaucoma and OHT, therapeutic ultrasound for glaucoma (TUG) which utilises a low power, low-frequency, focused ultrasound device can be used in controlling the IOP. Earlier studies have reported the beneficial effects of TUG on triggering cascade of cytokines which in turn leads to the reduce in IOP through mild inflammatory effect casing effective drainage of aqueous humour through TM. Therefore, the present study is proposed to investigate the mechanism & levels of cytokines involved (IL-1 α , IL-1 β , IL-6, TNF α , MMP-3, MMP-9 and TGF β) in reducing IOP after TUG treatment in glaucoma patients undergoing cataract surgery

(Phacoemulsification). This study will provide the insight into the key role of cytokines in reducing IOP.

The inclusion criteria for this study includes patients with POAG or OHT with significant cataract with age > 18 years, IOP more than 21 mmHg and less than 30 mmHg, not on any anti-glaucoma medications (AGM), disc showing features of mild to moderate glaucoma (<0.7 CDR) and VFI not worse than 60 % will included in this study after giving their written informed consent. The study protocol was carried in accordance to the tenets of Declaration of Helsinki.

This study included POAG/OHT patients receiving TULIP prior to cataract surgery (n = 4), compared with age-matched cataract controls without glaucoma (n = 5). Blood, aqueous humour and tear samples were analysed for cytokines (CNTF, IL-1 β , IL-6, MMP-3 and TNF- α) via multiplex Luminex assays and mTOR by ELISA.

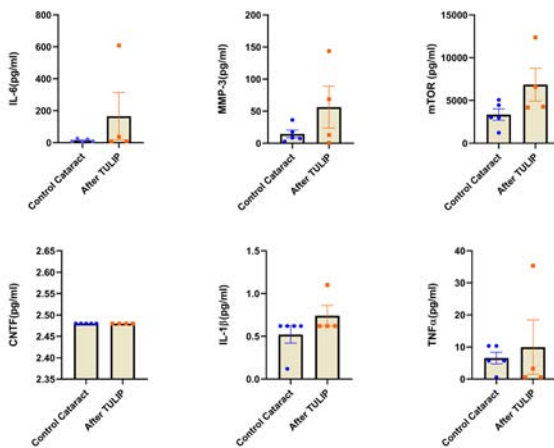


Figure showing the levels of Cytokines in Aqueous Humour of POAG patients who has received TUG treatment.

Note: There was increased levels of IL6, MMP-3 and mTOR in aqueous humour of POAG patient after TUG treatment and the levels of other cytokines such as CNTF, IL-1 β and TNF α were not significantly altered.

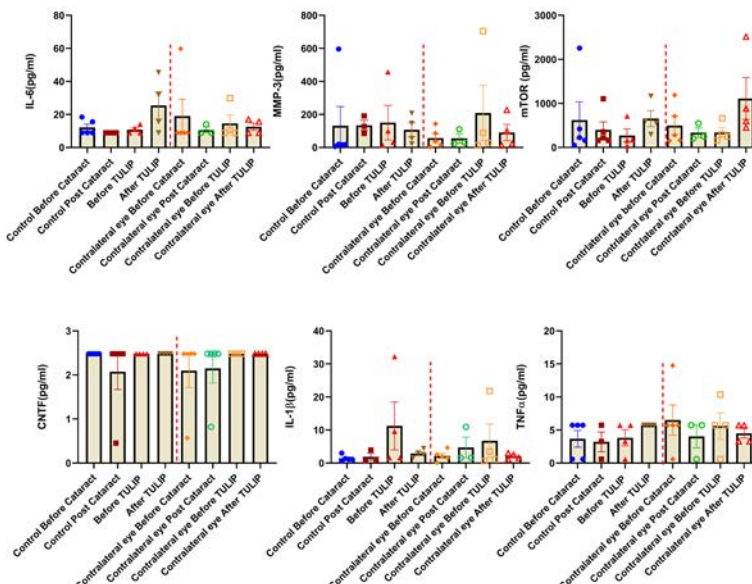


Figure showing the levels of Cytokines in tear samples collected from the study participants at different time intervals

Note: Increased levels of IL-6 and mTOR were observed in tear samples following TUG treatment collected from the study eye and other levels of cytokines were not altered significantly.

Results

In aqueous humour, there was an increased levels of IL-6 (11-fold), MMP-3 (3.8-fold) and mTOR levels (2-fold) whereas in tear samples, the same trend was seen with IL-6 (2.3 fold) and mTOR (2 fold) levels.

Conclusion

Increased levels of IL-6, MMP-3 and mTOR in tear and aqueous humour of those patients who received TULIP treatment. No elevation of IL-1 β or TNF- α

or CNTF was observed following TULIP treatment. There was no significant change was observed in plasma samples collected from those patients. Therefore, TUG is a safe, non-invasive therapy that lowers IOP and may act through both local and bilateral neural-cytokine pathways. These findings support TUG as a promising primary or adjunctive therapy for glaucoma.

The Bioinformatics Department develops and applies computational methods for the analysis of next-generation sequencing and imaging data in ophthalmic research. Its major activities include variant discovery from whole-genome/exome sequencing, transcriptomic and alternative splicing analyses, ocular microbiome profiling, and deep learning-based OCT image analytics. By integrating genomics, transcriptomics, and machine learning, the department supports biomarker discovery, disease stratification, and translational applications in eye diseases.

Identification and analysis of alternative transcripts in retinoblastoma progression

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Dr. A.Vanniarajan &
Dr. Usha Kim.
Funding : ICMR-SRF
Research Scholar : Mohamed Hameed Aslam A

Background

Alternative splicing is markedly altered in retinoblastoma, with thousands of genes exhibiting differential splicing compared with the normal retina (Yang et al., 2020). It may contribute to RB progression alongside the RB1 pathway, influencing cell cycle, DNA repair, metabolism, and apoptosis (Moore et al., 2010; Yang et al., 2020). Despite recognition of its importance, few studies validate these splicing events experimentally (Z. Liu & Rabadan, 2021). Our study aims to bridge computational detection and experimental confirmation of splicing events using RT-qPCR and bioinformatics modelling, providing evidence of functional effects at molecular and structural levels (P.

Wang et al., 2005; W. Zhang et al., 2024). We focus on four events, exon skipping of CCNB1, ENO2, and LUC7L, and intron retention of CDK5RAP3, that represent key biological functions and were chosen for their significance and feasibility. The findings may help improve diagnosis, prognosis, and therapy in retinoblastoma and related cancers (Murphy et al., 2022).

Results and conclusion

RT-qPCR analysis successfully detected and quantified all four predicted alternative splicing events in retinoblastoma samples using junction-specific primers, which ensured selective amplification of splice variants without detecting canonical transcripts. Relative expression levels were determined using the $2(-\Delta\Delta Ct)$ method, normalised to B2M expression.

Among the four variants, the CCNB1 exon-skipped isoform showed a markedly higher expression in retinoblastoma tissues compared with normal retina ($p = 0.01$, fold change = 6.02). The $-\Delta Ct$ values for tumours ranged from -3 to $+5$, while controls showed a narrower range (-5 to -1), indicating strong upregulation in tumour samples. In contrast, the



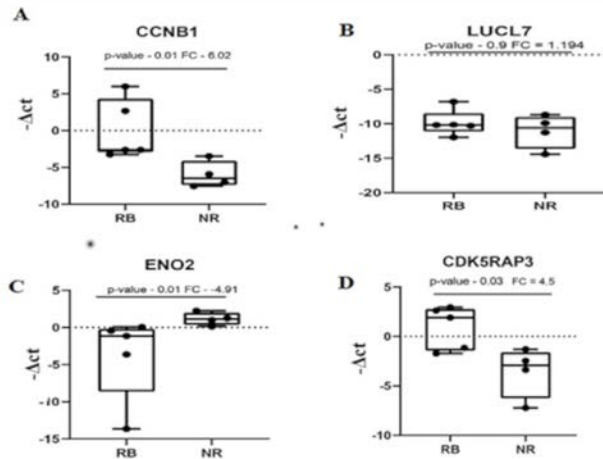


Figure 1: Relative Expression ($-\Delta Ct$) of alternative splice variants in retinoblastoma (RB) versus normal retina (NR) tissues. (A) CCNB1 exon skipping: $p = 0.01$, $FC = 6.02$. (B) LUC7L exon skipping: $p = 0.9$, $FC = 1.194$. (C) ENO2 exon skipping: $p = 0.01$, $FC = 4.91$. (D) CDK5RAP3 intron retention: $p = 0.03$, $FC = 4.5$. Box plots show median, interquartile range, and individual data points.

exon-skipped variant of ENO2 exhibited significantly lower expression in retinoblastoma ($p = 0.01$, fold change = -4.91), with $-\Delta Ct$ values from -13 to -4 compared with -2 to $+2$ in normal retina, reflecting substantial downregulation in tumour tissue.

The LUC7L exon-skipped variant showed no significant difference between retinoblastoma and normal retina ($p = 0.90$, fold change = 1.19). Both tissues displayed consistently low expression, with $-\Delta Ct$ values ranging from -12 to -8 in retinoblastoma and -16 to -10 in normal retina. This suggests that, although LUC7L undergoes alternative splicing in retinoblastoma, the event is not tumour-enriched. The CDK5RAP3 intron-retained variant (RI.6), however, exhibited significantly higher expression in retinoblastoma compared with controls ($p = 0.03$, fold change = 4.5), with $-\Delta Ct$ values ranging from -1 to $+3$ in tumours versus -8 to -2 in normal retina, indicating robust intron retention.

The specificity achieved through junction-targeted primer design validated the detection of genuine splice variants and excluded interference from wild-type transcripts (Govindkumar et al., 2022). Furthermore, the non-significant finding in LUC7L provides an essential internal control by distinguishing tissue-specific splicing events from those genuinely associated with tumour progression (Y. Zhang et al., 2021).

Computational analysis showed that splicing events alter protein domains, affecting function and localisation (J. Xu & Zhang, 2010). The exon-skipped CCNB1 variant had a moderate structural change (RMSD $1.87 \text{ \AA}$; TM-score 0.8), damaging the nuclear export signal domain (Figure 2). This disrupts recognition for nuclear export, impairing nucleo-cytoplasmic transport and CCNB1 localisation, leading to cell-cycle dysregulation essential for proper mitosis (Toyoshima et al., 1998; Hagting et al., 1998).

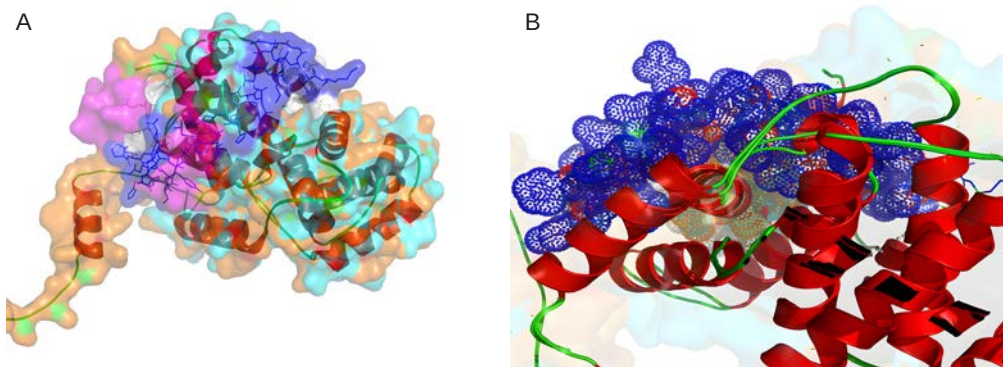


Figure 2: Structural comparison of CCNB1 reference and exon-skipped proteins. (A) Superimposed structures of reference CCNB1 (cyan surface) and exon-skipped model (orange surface). The blue region indicates the exon-skipped sequence, magenta marks the altered segment in the spliced protein, and white highlights flanking regions. Helices are coloured red, loops are green, and sheets are yellow. (B) Nuclear export signal (NES) domain visualised with blue dotted representation; regions marked by blue dots illustrate NES, which is partially absent in the exon-skipped protein.

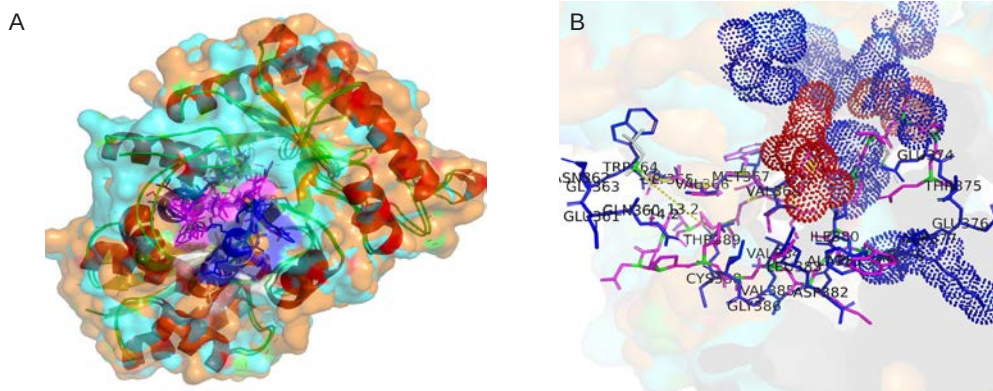


Figure 3 Structural mapping of the ENO2 exon skipping event. (A) Superimposed structures of reference ENO2 (cyan surface) and exon-skipped model (orange surface). The blue region highlights the exon-skipped segment, magenta depicts altered residues within the spliced protein, and red, green, and yellow indicate helices, loops, and sheets, respectively. (B) Active site (red dots) and substrate binding site (blue dots) are shown. Blue sticks with residue labels denote amino acids from the exon-skipped region that directly affect the active site and substrate binding site.

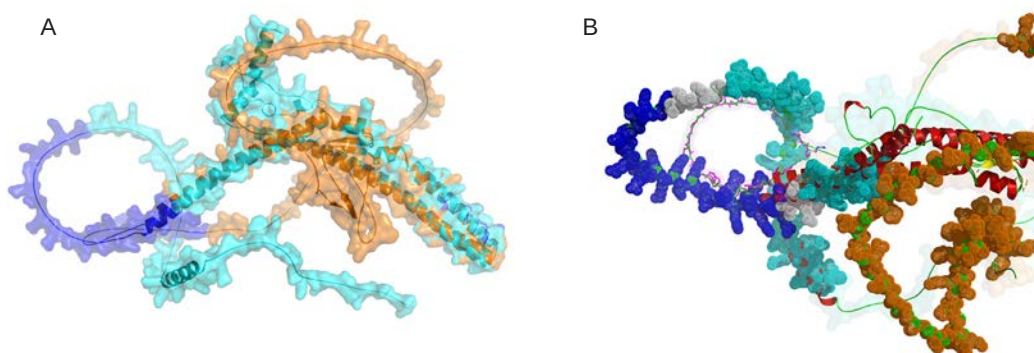


Figure 4: Structural comparison of LUC7L reference and exon-skipped proteins. (A) Superimposed structures of reference LUC7L (cyan surface) and exon-skipped model (orange surface), with the skipped region shown in blue. (B) Dotted blue regions indicate the RS-rich domain that is skipped in the exon-skipped LUC7L protein, highlighting the impact of exon skipping on domain architecture.

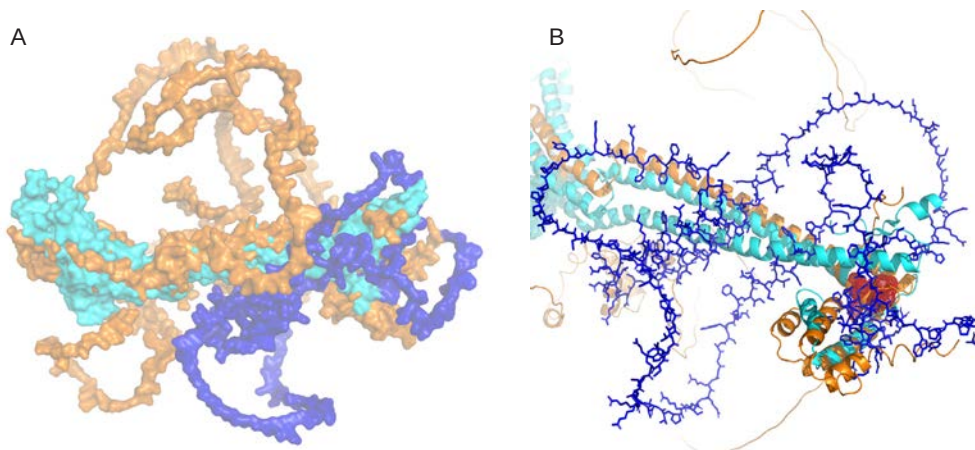


Figure 5: Structural comparison of CDK5RAP3 reference and intron-retained isoforms. (A) Superimposed structures of reference CDK5RAP3 protein (cyan) and intron-retained model (orange), with the retained intronic region highlighted in blue. (B) The LXXLL motif residues are represented as dots, visualising their spatial distribution and showing how the motif is affected by intron retention within the protein structure.

Mislocalisation could contribute to cancer. The ENO2 variant was conserved (RMSD 1.23 Å; TM-score 0.88) but showed disruption at the active site, affecting substrate binding and catalysis (Figures 3), potentially altering glycolytic function to support rapid proliferation in retinoblastoma (L. Gao et al., 2023). The LUC7L variant lacking the arginine-serine-rich domain had significant structural divergence (RMSD 2.22 Å; TM-score 0.48), losing interactions crucial for splicing regulation (Figures 4). This may act as a dominant negative and disrupt normal splicing. Retained intronic sequences in CDK5RAP3 affected the LXXLL motif (TM-score 0.42; RMSD 5.18 Å), essential for protein interactions regulating cell-cycle checkpoints (Figures 5). Disruption could impair CDK5 interaction, promoting abnormal cell cycle and reduced control. In conclusion, this study experimentally validated tumour-enriched alternative splicing events in CCNB1, ENO2, and CDK5RAP3 and demonstrated their associated structural and functional protein alterations. The findings support a contributory role of aberrant splicing in retinoblastoma progression and highlight its potential as a source of diagnostic and prognostic biomarkers.

A comprehensive POAG database

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Prof. K. Dharmalingam &
Dr. A. Vanniarajan
Research Fellow : Dr. R. Santhosh
Funding Agency : ICMR – CAR

Background

Primary open-angle glaucoma (POAG) is the most common subtype of glaucoma. POAG is distinctly described as a multifactorial optic neuropathy that is irreversible, chronic, and progressive with a characteristic of optic nerve fibres losses acquired.

Such loss develops in the presence of open anterior chamber angles, characteristic visual field abnormalities, and intraocular pressure that is too high for the continued health of the eye. Because of the silent nature of glaucoma, patients usually do not present with any symptoms or visual complaints until late in the disease course, particularly with POAG. There is significant attention should be given to the patient's clinical history: ocular history, medical history, current medications etc. So, this is an essential to develop a database of POAG patients, The POAG patients' data are clustered as the patients with and without diabetes mellitus (DM), strict exclusion of the patients who has diabetic retinopathy (DR). In this clinical feature extraction categorised into two major parameters (i) Systemic parameters and (ii) Ophthalmic parameters, where, (i) Systemic parameters are patient having diabetes, hypertension, depression, blood pressure, and in-addition to the personal details of the patient as age, gender, ethnicity, and family history of the glaucoma. (ii) Ophthalmic parameters are Baseline Intraocular Pressure (IOP), Central Corneal Thickness (CCT), Follow-up duration and intervals and the medication intake of the patient.

Results and Conclusion

The POAG database was created using CGI and MySQL. Currently, the database contains the 207 patient's information were collected from the clinical department of the Aravind Eye Hospital, Madurai. The collected data were clustered by diabetes mellitus (DM) and the database pipeline has been designed by the clustered information and displayed in the figure 2.1 and 2.2. Furthermore, the collected information is classified into family history and respective DM and non-DM patients. This assists to store, concurrency, security & integrity and mainly to identify the data relationship can be obtained using machine learning.

OCULAR MICROBIOLOGY

The department investigates the molecular and immunological basis of vision-threatening ocular infections and inflammatory diseases. Current work integrates corneal transcriptomics and miRNA profiling to define pathogen-specific host immune signatures in fungal and bacterial keratitis and identify prognostic biomarkers. The lab also explores extracellular vesicle-mediated miRNA transfer as a strategy to modulate multidrug-resistant *Pseudomonas aeruginosa* virulence and biofilm formation. In parallel, tear-based immune profiling and MIF-centred network analyses are being developed to stratify and prognosticate patients with uveitis and other ocular inflammatory conditions, enabling precision diagnostics and targeted immunomodulatory approaches.

Role of human corneal miRNAs in the onset and severity of fungal keratitis

Investigators : Dr. D. Bharanidharan (PI),
Dr. K. Dharmalingam,
Dr. Venkatesh Prajna &
Dr. Lalitha Prajna
Research Scholar : Shreya Dinesh
Funding Agency : ICMR

Purpose

Identification of differentially expressed genes involved in common and pathogen-specific host immune responses, for the understanding of the host response in fungal keratitis.

Background

Fungal keratitis, predominantly caused by *Fusarium spp.* and *Aspergillus flavus*, is a major cause of ocular morbidity in tropical regions and is frequently associated with poor treatment outcomes despite antifungal therapy. In this study, we identified

dysregulated miRNAs that plays an important role in disease severity and pathogenesis. Growing evidence suggests that these pathogens trigger distinct host immune responses, which may critically influence disease progression and therapeutic efficacy. An unbiased transcriptome analysis was performed to investigate common and pathogen-specific host mechanisms in fungal keratitis, *Fusarium spp.* and *A. flavus* infected corneal samples to identify differentially expressed genes involved in host immune regulation. The RNA seq. was performed for *A. flavus* and *Fusarium spp.* infected samples and DEGs that were common to *A. flavus* and *Fusarium spp.* infection were identified. Enrichment analysis was performed separately in each group. Based on relevant pathways DEGs common to both infection and the genes that unique to *A. flavus* and *Fusarium spp.* infected samples were selected and validated using RT-qPCR. In addition, selected DEGs identified as targets of the validated miRNAs hsa-miR-19a-3p and hsa-miR-16-5p, and



implicated in TGF- β signalling and mitophagy, were further validated in *Fusarium spp.*-infected samples compared with cadaveric controls to confirm miRNA–mRNA regulatory interactions.

Results and Conclusion

This study identified common and pathogen-specific host immune responses triggered by *Fusarium sp.* and *Aspergillus flavus* in fungal keratitis. Pathway enrichment analysis revealed common immune pathways, such as IL17, TNF, and chemokine signalling, shared by both fungal infections (Figure 1.1). Similarly, Pathogen-specific DEG analysis revealed distinct immune signatures between *Fusarium spp.* and *Aspergillus flavus* keratitis. C3 was uniquely enriched in *Fusarium spp.* keratitis, associated with acute inflammatory response and neutrophil extracellular trap (NET) formation, and was absent in *A. flavus* keratitis, supporting its implication in complement pathway. Additionally, *Fusarium spp.* keratitis showed unique enrichment of IL19 and LRG1, suggesting novel roles in cytokine signalling, immune regulation, and tissue repair. In contrast, *A. flavus* keratitis exhibited pathogen-specific hub DEGs, including TREM2 and APOE, enriched in adaptive and immune response pathways (Figure 1.1).

From the network analysis 6 genes dysregulated in both *A. flavus* and *Fusarium spp.* infected samples 5 genes unique to *Fusarium spp.* and 4 genes unique to *A. flavus* infected samples were validated using RT-qPCR. S100A7, S100A8, S100A9, CXCL8, and IL17A were upregulated in both *Aspergillus flavus* and *Fusarium spp.* keratitis, while IL19, C3, and LRG1 were specifically elevated in *Fusarium spp.* keratitis and TREM2 and APOE in *A. flavus* keratitis. Although IL6, FOS, and EGR1 did not show RT-qPCR concordance, the findings highlight distinct pathogen-specific immune activation profiles (Figure 1.2).

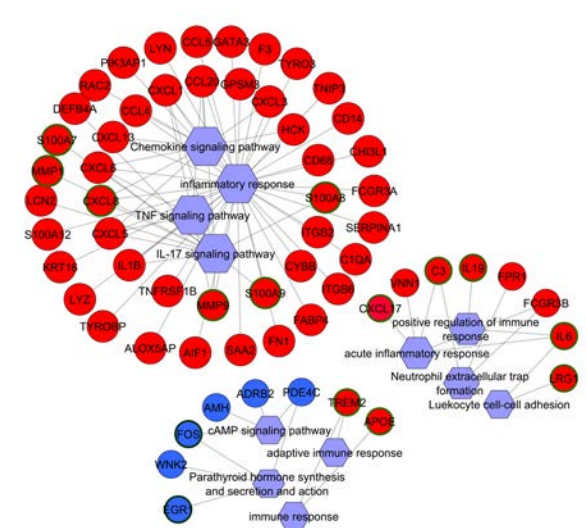


Figure 1.1: Functional network created using selected KEGG pathways and GO-biological processes for dysregulated genes Upregulated and downregulated genes are marked as red and blue respectively. The hub genes are outlined in green in colour. Hexagonal shapes represent the pathways or GO.

Six genes WNT4, GABARAPL1, FMOD, THBS1, PDGFD, and DKK3 were selected for RT-qPCR validation based on their association with significantly dysregulated miRNA expression and relevance of their associated pathways in 10 control and 10 infected corneal tissue samples. All six genes were significantly downregulated in infected tissues, demonstrating consistent inverse miRNA–mRNA expression patterns with their corresponding miRNAs and providing experimental support for the predicted miRNA–target regulatory relationships (Figure 1.3).

This study characterises both shared and pathogen-specific host immune responses in *Fusarium spp.* and *Aspergillus flavus* keratitis, revealing common enrichment of IL17, TNF, and chemokine signalling pathways. *Fusarium spp.* keratitis was distinguished

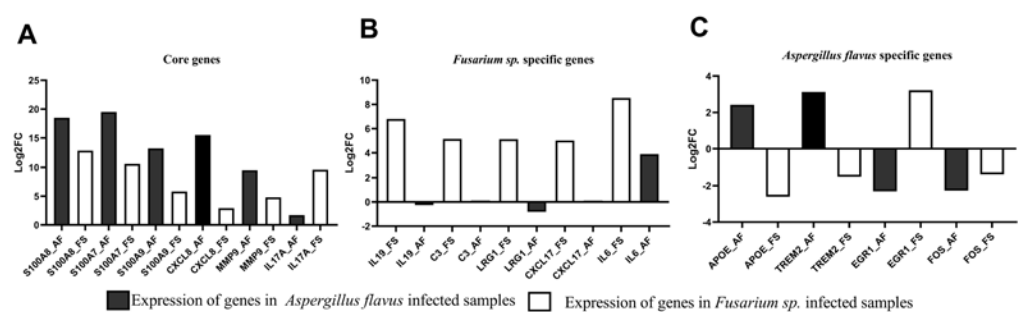


Figure 1.2: RT-qPCR validation of selected genes in Cadaver corneal tissue samples and *Fusarium sp.* and *Aspergillus flavus* infected tissue samples. Plot of log2 fold change (Log2FC) values of A) Common genes B) *Fusarium sp.* specific genes and C) *Aspergillus flavus* specific genes.

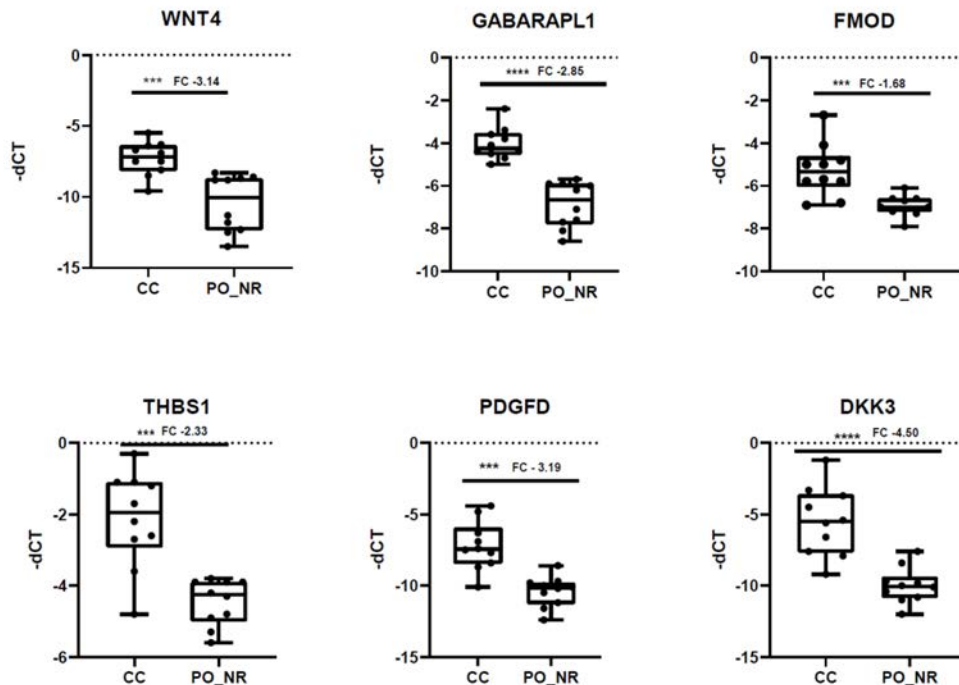


Figure 1.3 - The relative expression of six selected target genes of the validated miRNAs were plotted in corneal tissues compared to cadaver controls for 10 samples Median values are shown by horizontal lines using the Mann-Whitney test. * p-value < 0.05. CC – Cadaver Control, PO-NR – poor outcome tissue - Non-Responder. Figure 1.3 - The relative expression of six selected target genes of the validated miRNAs were plotted in corneal tissues compared to cadaver controls for 10 samples Median values are shown by horizontal lines using the Mann-Whitney test. * p-value < 0.05. CC – Cadaver Control, PO-NR – poor outcome tissue - Non-Responder.

by enrichment of C3, IL6, IL19, and LRG1 in inflammatory and immune-regulatory pathways, whereas *A. flavus* keratitis showed predominant involvement of TREM2- and APOE-associated adaptive immune and cell migration responses. Importantly, validation of miRNA target genes supports a functional role for dysregulated miRNAs in fungal keratitis pathogenesis through modulation of key pathways, including TGF- β signalling and mitophagy.

Altered expression of human corneal miRNAs and its predicted role in *Pseudomonas aeruginosa* keratitis pathogenesis

Investigators : Dr. D. Bharanidharan (PI),
Dr. Venkatesh Prajna, &
Dr. Lalitha Prajna
Research Scholar : R. Praveenkumar
Funding : DBT-JRF

Background

Pseudomonas aeruginosa (PA) keratitis is a severe corneal infection that can rapidly progress

to vision loss without appropriate medical care. Despite effective bacterial clearance by antibiotic administration, the clinical outcomes are compromised due to inflammation-driven corneal ulceration. In our earlier study, we profiled 122 differentially expressed (DE) human corneal miRNAs from ulcerated corneal buttons of PA keratitis patients undergoing Therapeutic Penetrating Keratoplasty (TPK). Now, we have further validated this work by confirming the altered expression of three selected miRNAs namely, miR-184, miR-21-5p and miR-22-3p using RT-qPCR in an additional set of samples. Further, we have predicted the role of these miRNAs by constructing a functional network.

Results and Conclusions

i) Target prediction and shortlisting, pathway enrichment and network analysis

Target prediction of the top sixty abundant differentially expressed (DE) miRNAs identified 6,900 targets (prediction score >0.9; restricting to 3' UTR), commonly predicted by miRWalk and miRDB. Filtering these targets using in-house human corneal transcriptomic data yielded a final set of 5,476 cornea-expressed genes, which were subjected to

pathway enrichment analysis using DAVID. Among the significantly enriched KEGG pathways, the MAPK signalling pathway, neurotrophin signalling pathway, and autophagy pathway were prioritised based on their relevance to corneal pathology. Network was constructed with top DE miRNAs, predicted targets and selected enriched pathways using Cytoscape v 3.10. In our network, miR-21-5p was the most abundant upregulated miRNA, whereas let-7a-5p was the most abundant downregulated miRNA (Figure 2.1). miR-185-5p and let-7a-5p showed the highest number of predicted targets (10 and 7 respectively), whereas the gene which was targeted frequently by most number of miRNAs was NTRK2 (6 miRNAs) (Figure 2.1). From the network analysis, literature support and disease relevance, five miRNAs namely, miR-184, miR-21-5p, miR-22-3p, let-7c-5p and let-7a-5p were chosen for RT-qPCR study.

ii) Altered expression of miRNAs and its implication in PA keratitis corneal ulceration

Expression of miR-184, miR-21-5p, miR-22-3p, let-7a-5p, and let-7c-5p was assessed by RT-qPCR in PA keratitis corneal buttons (n = 10) and cadaveric controls (n = 10), with normalisation to U6 snRNA. miR-22-3p and miR-21-5p were significantly

Figure 2.1: Network illustrating differentially expressed (DE) miRNAs and their predicted target genes. The network was constructed using Cytoscape v10.3. Selected enriched KEGG pathways are represented as circles, DE miRNAs as diamonds, and their predicted target genes as hexagons. The colour gradient from violet to red denotes the fold change ($\log_2FC$) of DE miRNAs, with violet indicating downregulation and red indicating upregulation. The size of each diamond corresponds to the relative abundance ($\log_2CPM$) of the respective DE miRNA.

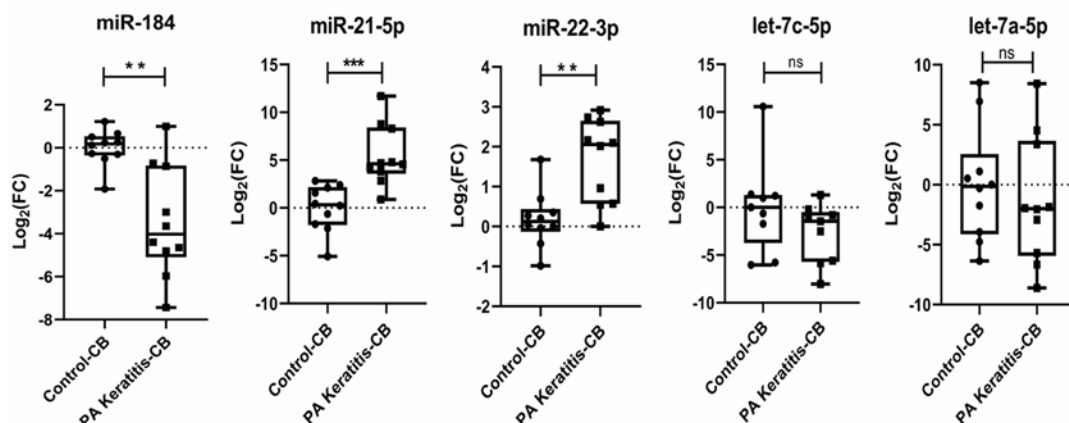


Figure 2.2: Differential expression of miRNAs by RT-qPCR. The Y-axis represents Log₂Foldchange values for each miRNA (miR-184, miR-21-5p, miR-22-3p, let-7c-5p and let-7a-5p), calculated with by qPCR Ct values and normalised to the internal control snRNA-U6. The X-axis represents the sample types: Control Corneal Button (Control-CB) or *Pseudomonas aeruginosa* Keratitis Corneal Button (PA keratitis-CB). Statistical significance was calculated using the Mann–Whitney U test and is indicated by asterisks: $p < 0.05$ (*), $p < 0.01$ (**), and $p < 0.005$ (***) or ns for no-significance.

Collectively, the altered expression of miR-184, miR-21-5p, and miR-22-3p may contribute to corneal ulceration in PA keratitis through disruption of neurotrophin signalling, enhancement of inflammation-associated cell death, and delayed wound healing.

Attenuation of drug-resistance in Ocular Multidrug-Resistant *Pseudomonas aeruginosa* using human corneal miRNA cargo of Extracellular Vesicles

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Sri Sanjana. S.V.

Funding Agency : ICMR

Background

Bacterial keratitis (BK) is a major cause of corneal blindness worldwide, accounting for nearly two million cases of vision loss annually. In India, the incidence of BK is estimated at 700 cases per 100,000 population.

Among the causative pathogens, *Pseudomonas aeruginosa* (PA) has emerged as the most prevalent and clinically aggressive agent in recent years. Clinical data from Aravind Eye Hospital, Madurai serving seven districts of Tamil Nadu highlight a substantial burden of PA keratitis. Between 2018 and 2022, PA constituted approximately 29–32% of culture-positive BK cases (444 of 1,427 patients). This large and demographically diverse patient population provides a robust clinical framework for investigating disease pathogenesis and therapeutic outcomes across varied socio-economic backgrounds. PA keratitis is a rapidly progressive ulcerative infection frequently associated with contact lens wear or ocular trauma. Although prompt treatment with broad-spectrum antibiotics is essential, clinical outcomes are increasingly compromised by the emergence of multidrug-resistant *P. aeruginosa* (MDR-PA). Recent Indian studies report resistance rates of 65–75% to commonly used antibiotics, including fluoroquinolones, cephalosporins, carbapenems, and aminoglycosides. Our surveillance data further indicate that over 20% of PA isolates identified in 2022 were MDR, underscoring a growing therapeutic challenge. Several adjunct therapeutic strategies, including antimicrobial peptides, plant-derived compounds, monoclonal antibodies, corneal cross-linking, and corticosteroids are under evaluation. However, outcomes remain poor, particularly in infections caused by virulent exoU PA strains. Interestingly, our prior genomic analysis identified a patient infected with an extensively drug-resistant exoU strain who demonstrated favourable clinical recovery, suggesting a critical role for host-mediated factors.

Results and conclusion

Pseudomonas aeruginosa infection model was optimised using AO/PI live–dead staining. An MOI of 1:1 was chosen and taken forward for extracellular vesicle isolation. EVs were isolated from conditioned media of control and *Pseudomonas aeruginosa* (PA) infected human corneal epithelial cell cultures. PA infection was performed using the laboratory reference strain PA14 and a clinically isolated multidrug-resistant keratitis strain, BK8.

Isolated EVs were thoroughly characterised to confirm their identity and quality. Nanoparticle tracking analysis (NTA) demonstrated a homogeneous population of vesicles within the expected size range for small EVs, with no significant contamination from cellular debris.

Western blot analysis further confirmed EV enrichment by detecting established EV marker proteins, validating the integrity of the isolated vesicles.

Comparative profiling of EV-associated microRNAs revealed significant upregulation of host-derived miRNAs, particularly let-7a-5p and let-7b-5p, in

EVs released following PA infection compared to uninfected controls. To determine whether these EV-associated miRNAs could be transferred to bacteria, EVs isolated from infected epithelial cells were co-incubated with PA14 and BK8 strains. Uptake studies confirmed the successful delivery of EV-derived miRNAs into both PA strains, demonstrating cross-kingdom transfer of host miRNAs.

RT-qPCR was performed to assess the expression levels of miRNA Let-7a-5p and Let-7b-5p in Control EV's (uninfected HCE-T EV's) and Infected EV's derived from PA14 and BK8. Statistical analysis was performed using one-way ANOVA (****p < 0.0001).

The functional impact of EV-mediated miRNA transfer was assessed using phenotypic assays. Treatment with infection-derived EVs resulted in a significant reduction in biofilm formation in both PA14 and MDR BK8 strains compared to untreated controls. This observation suggests that host-derived EV miRNAs can modulate bacterial biofilm regulatory pathways, a key factor contributing to bacterial persistence and antimicrobial resistance.

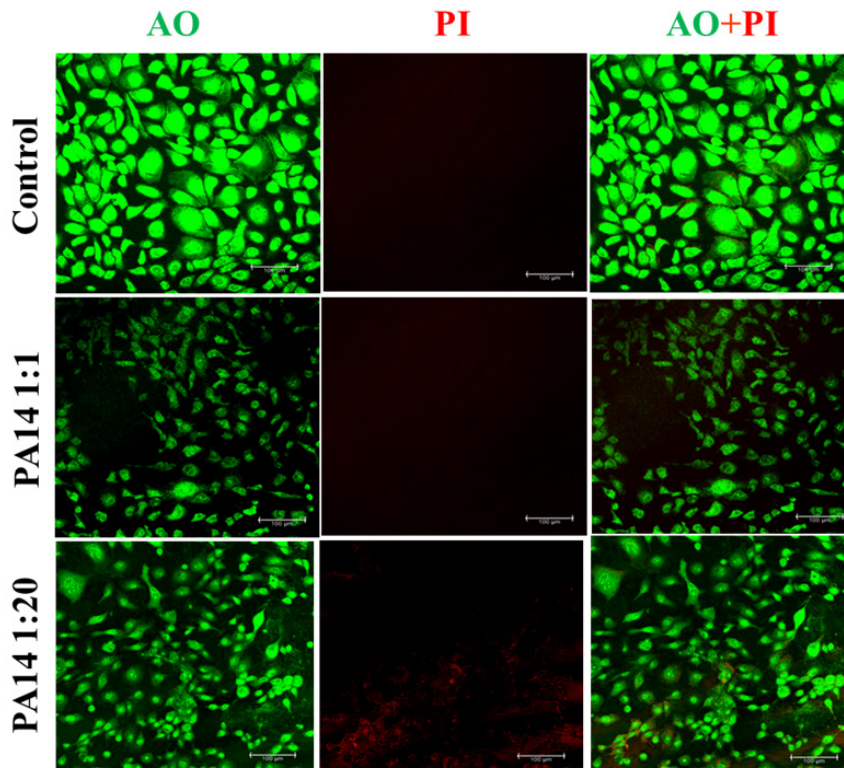


Figure: 3.1 PI/AO staining of HCE-T cells following 6 hours of infection with *Pseudomonas aeruginosa* (PA14) at different multiplicity of infection (MOI).

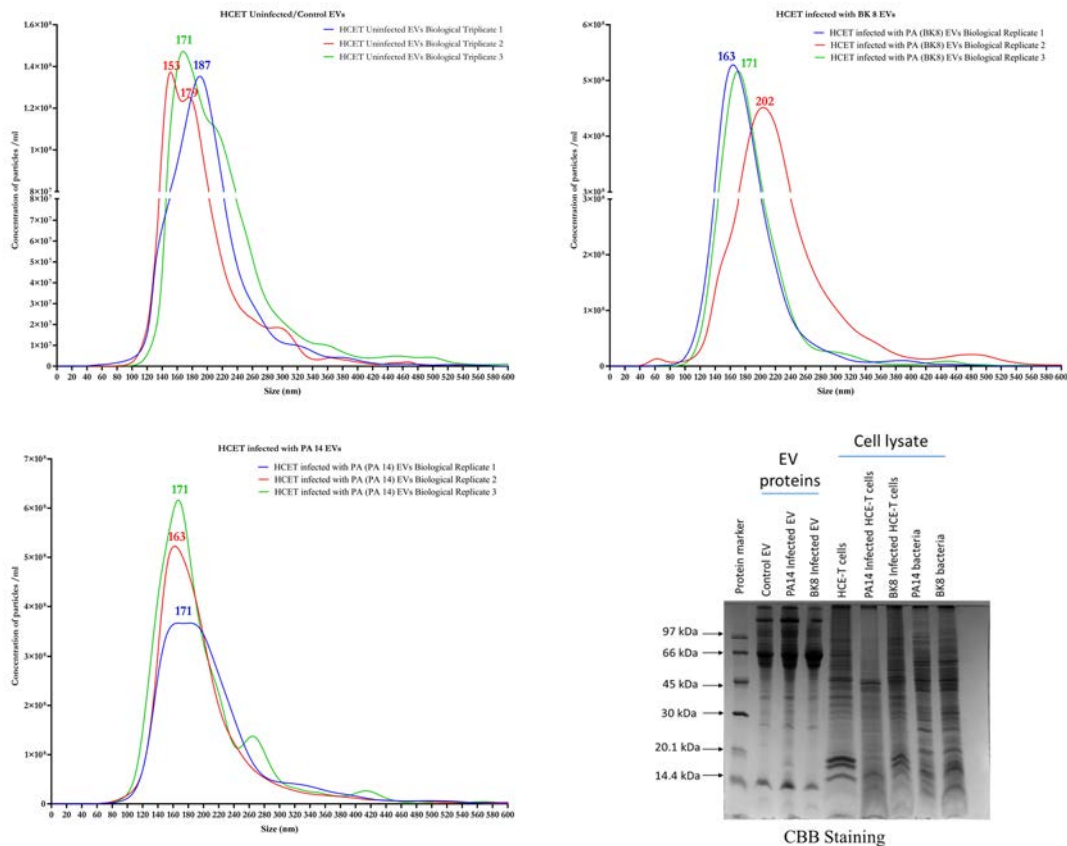


Figure 3.2: NTA analysis of Uninfected EV and PA infected EV. Protein profile of EV, cell lysate and bacterial cell lysate in 12.5% SDS PAGE.

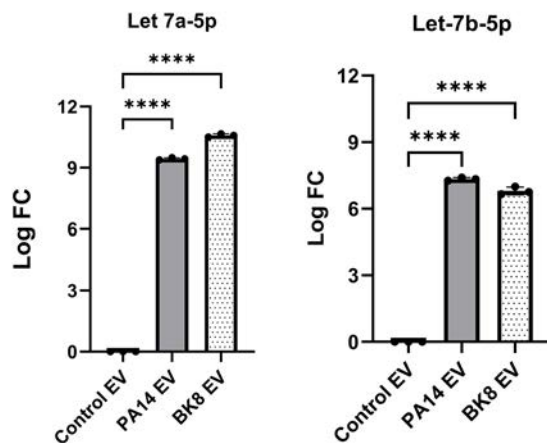


Figure 3.3 Relative expression of miRNA Let-7a-5p and Let-7b-5p in control and *Pseudomonas aeruginosa* infected EVs.

Ocular macrophage migration inhibitory factor-based stratification and prognostication of patients with vision threatening ocular inflammatory conditions

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Co-Principal investigators: Dr. N.V. Prajna, & Dr. Swagata Ghosh

Research Students : Dhanu Prakash M & Anusha Erulandi

Funding Agency : ICMR

Introduction and Background

Public Health Problem: Ocular inflammatory and infectious diseases remain a significant cause of vision morbidity worldwide. In the United States alone, approximately 14.3% of all patients with ocular conditions annually present with inflammatory or infectious diseases. Among these, uveitis—a heterogeneous group of intraocular inflammatory diseases involving the iris, ciliary body, and choroid—accounts for the majority of ocular inflammatory

diseases. Uveitis is estimated to cause 10–20% of blindness in developed countries and up to 25% in developing regions.

Unmet Clinical Need: Despite advances in ocular diagnostics and immunosuppressive therapies, there is no clinically implementable molecular guidance strategy for disease monitoring or risk stratification in uveitis. Differences in ocular inflammatory profiles might account for heterogeneity in disease progression and treatment response in patients. However, there is a substantial gap in our knowledge about how the variability in ocular immune profiles relate to different uveitis types, their progression and treatment response.

Rationale and hypotheses: To address this gap, our work focuses on developing host immune-inflammatory parameter-based approaches for stratification, prognostication, and treatment planning in uveitis and related ocular inflammatory conditions. We hypothesise that non-invasive tear samples can provide reliable insight into intraocular immune activity. By studying immuno-inflammatory mediators such as cytokines, chemokines, enzymes, and regulatory proteins, and more importantly, by analysing their relationships and interaction patterns, we aim to identify correlation-based immune monitoring signatures.

Macrophage Migration Inhibitory Factor (MIF) plays a dichotomous role in host immunity, functioning as either an immune activator or suppressor and influencing a wide range of human diseases. Although increased MIF levels and MIF gene polymorphisms have been previously associated with different uveitis subtypes, there is no patient-based evidence regarding how MIF interacts with immune-augmenting and immune-dampening factors across different ocular inflammatory conditions. We hypothesised that MIF is differentially associated with immune-activating and inhibitory factors in distinct ocular inflammatory diseases.

In order to test our hypothesis, we aim to assess i) MIF and its associated cytokine and enzyme networks in the tear-fluids of different ocular inflammatory diseases (uveitis, fungal keratitis, scleritis etc.); ii) the distribution of MIF gene polymorphisms in our patient groups and how these differ between diseases.

To date, our analyses have encompassed two subtypes of non-infectious uveitis and the ocular surface inflammatory disease scleritis. The fungal keratitis cohort is presently being established and will be examined in forthcoming assays.

Results

Overview of Study Cohorts and Samples: Tear and blood samples were collected from patients at Aravind Eye Hospital, Madurai, representing multiple clinical entities:

- Vogt–Koyanagi–Harada (VKH) disease – autoimmune uveitis (n=28)
- Idiopathic uveitis – inflammation of unknown etiology (n=30)
- Scleritis – ocular surface inflammation (n=30)
- Healthy controls – age-matched individuals (n=20)

The idiopathic uveitis and scleritis patient cohort analysed in this study includes clinically identified cases of non-infectious etiology. Tear samples were collected using Schirmer strip, followed by protein extraction in 1xPBS and were analysed by bead-based multiplex ELISA on a flow-cytometer. Cytokine panels included both low-concentration analytes (e.g., IL-1 β , IL-2, IL-6, IL-10, IL-13, TNF- α , IFN- γ , TGF- β 1, IL-17A) and high-concentration analytes (e.g., MPO, NGAL, MIF, MMP-9, TIMP-1). DNA isolated from peripheral blood was analysed for MIF gene polymorphisms by PCR amplification and Sanger sequencing.

A. Tear Cytokine Profiles and Disease-Specific Alterations:

IL-6 emerged as the only cytokine consistently elevated across all disease groups compared to the controls (Fig 1). IL-17A, IL-13, IL-10, and TGF- β 1 were significantly increased in VKH and scleritis, while IFN- γ elevation was restricted to VKH. IL-2, MIF, NGAL, and MMP-9 showed significant elevation in both VKH and idiopathic uveitis but not in scleritis (Fig 1). Conversely, TIMP-1 was uniquely decreased in scleritis underscoring disease-specific immune alteration patterns (Fig 1). Idiopathic uveitis displayed high intra-group variability in cytokine concentrations, possibly due to its heterogeneous etiology.

B. Diagnostic Discrimination Using AUROC Analysis:

To assess the discriminative potential of these tear factors, Area Under the Receiver Operating Characteristic (AUROC) analysis was performed (Table 1). In VKH, IL-2, IL-17A, IL-13, IL-10, IL-6, and TGF- β 1 showed high AUC values, demonstrating their ability to distinguish VKH from healthy individuals with strong sensitivity and specificity. In scleritis, only IL-6 retained significant discriminative potential. Logistic regression modelling identified specific multi-factor signatures (Fig 2). While IL-13 and IL-10 combined best differentiated VKH from

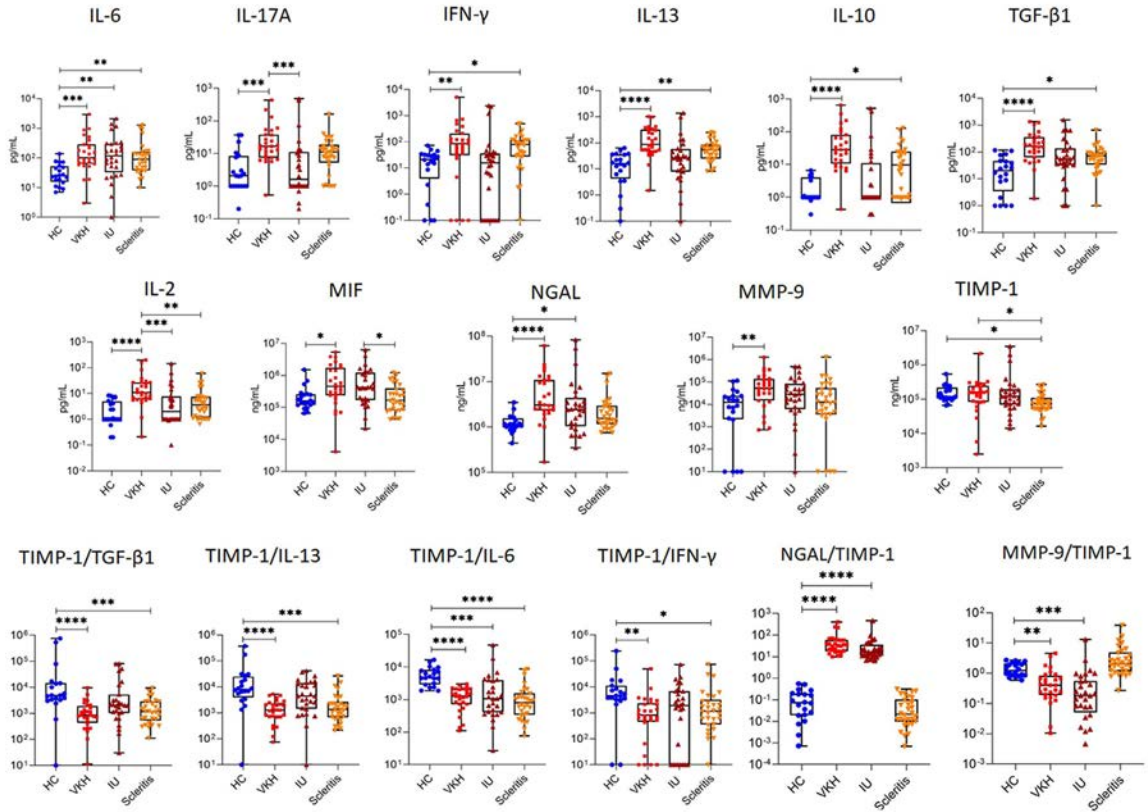


Figure 1: Comparison of absolute concentrations and relative abundance of factors between disease groups and controls. HC-Healthy controls, VKH- Vogt-Koyanagi-Harada disease, IU- Idiopathic uveitis.

controls, a combination of IL-6, IL-13, NGAL, and TIMP-1 perfectly discriminated idiopathic uveitis (AUC = 1.0). IL-6, IL-13, and TIMP-1 together effectively distinguished scleritis from controls. Interestingly, MIF alone, though with low AUC, was the only marker that could differentiate scleritis (ocular surface) from uveitis (intraocular), emphasising its potential as a unifying immune modulator across disease boundaries.

C.Relative abundance of tear factors:

We examined the relative abundance of all pairs of factors to identify ratios most effective in distinguishing disease groups from controls, as well as among disease subtypes. The TIMP-1/IL-6 ratio was significantly reduced across all disease groups (Fig 1). Ratios of TIMP-1 to TGF-β1, IL-13, and IFN-γ were markedly lower in VKH and scleritis (Fig 1).

Groups	Factors	Cut off concentration	Sensitivity %	Specificity %
HC vs VKH	IL-2	>5.300 pg/mL	88.89	75
	IL-17A	>11.04 pg/mL	70.37	85
	IL-13	>41.12 pg/mL	85.19	90
	IL-10	>5.821 pg/mL	96.30	95
HC vs Scleritis	TIMP-1	<93747 ng/mL	73.33	85
	IL-13	>26.68 pg/mL	83.33	70

Groups	Ratios	Cut off criterion	Sensitivity	Specificity
HC vs VKH		>5.253	100	100
HC vs IU	NGAL/TIMP-1	>3.491	100	100
Uveitis vs Scleritis		<3.365	100	100
HC vs VKH	MMP-9/TIMP-1	<7.041	75	95
HC vs IU		<0.8043	82.14	85

Table 1. Table showing the results of the AUROC analysis for both the absolute concentrations and relative abundances of factors. The upper section presents the cut-off concentrations, while the lower section shows the cut-off proportions used to distinguish between disease groups and controls, or among disease subtypes, along with their corresponding sensitivity and specificity values.

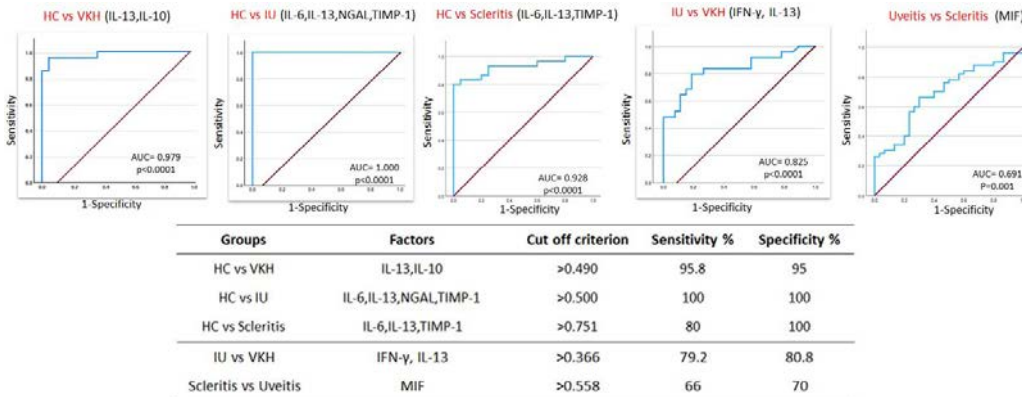


Figure 2: Results of the Logistic regression analysis comparing different study cohorts. Factor or specific combinations of factors, their cut off criterion along with their specificity and sensitivity values are shown to distinguish the respective disease groups and controls or among disease subtypes.

Interestingly, levels of NGAL with respect to TIMP-1 significantly differed between uveitis subtypes and scleritis, being higher in VKH and idiopathic uveitis but lower in scleritis (Fig 1). Notably, while the ratios of TIMP-1 to IL-6, IL-13, or TGF-β showed increased AUC values and significance in distinguishing scleritis from healthy controls compared to individual factors (data not shown), the NGAL/TIMP-1 ratio separated scleritis from each uveitis subtype with a perfect AUC of 1.0 and 100% sensitivity and specificity (Table 1).

D. Spearman's rank Correlation analyses:

Correlation matrices provided further insight into immune network alterations in the disease groups compared to the healthy controls (Fig. 3). Significant correlations were observed between IL-2, MIF, MMP-9, MPO, NGAL, and TIMP-1, reflecting homeostatic immune readiness. VKH exhibited a dense correlation network, indicating synchronised innate and adaptive activation characteristic of autoimmune inflammation. Idiopathic uveitis and scleritis showed

distinct disruptions in these correlations, highlighting disease-specific immunopathogenic dynamics. A notable observation was the context-dependent relationship between MIF and IL-2. In healthy tears, moderate MIF–IL-2 correlation likely represents balanced crosstalk between innate and T-cell–derived immune activity. In VKH, this correlation strengthens, reflecting MIF's potential role in potentiating T-cell IL-2 production and amplifying inflammation. In scleritis, the correlation disappears, suggesting loss of MIF–IL-2 coordination and a shift toward Th2-dominant cytokine patterns (high IL-10, IL-13, low IFN-γ, IL-17A). Moreover, MIF's and IL-2's correlation with ECM modifying MMP-9 and TIMP-1 and stress response factors-MPO and NGAL were also found to be strengthened in VKH and completely disrupted in scleritis. These findings support a model where MIF–IL-2 interactions maintain ocular immune homeostasis, regulate ECM modulation and stress response and deviations from this relationship mark pathological or disease-specific immune alterations.

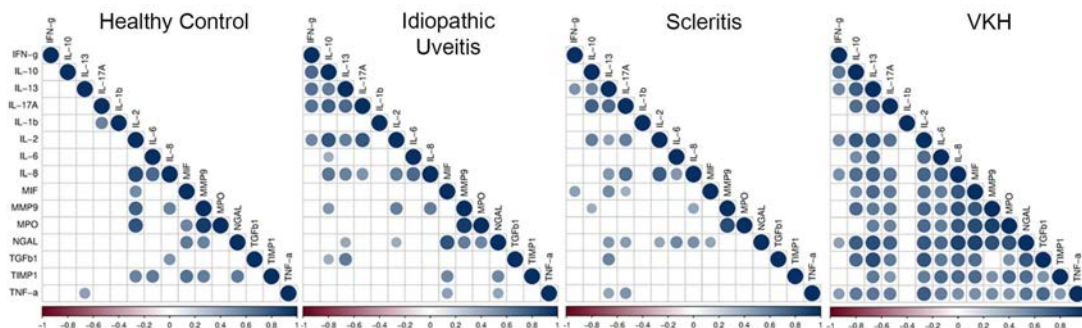


Figure 3: Spearman's rank correlation matrix of tear factors for the healthy controls and different disease groups showing distinct disease specific patterns.

E.MIF Gene Polymorphisms and Disease Associations:

Genetic analysis focused on three MIF polymorphisms- rs755622 (-173 G>C, promoter region), rs2096525 (+254 T>C, first intron), rs5844572 (-794 CATT₅₋₈, short tandem repeat in promoter) (Fig 4). rs755622 (-173 G>C) variant carriage (GC or CC genotypes) was higher in VKH (50%) and idiopathic uveitis (42.9%) than in scleritis (7.2%). rs2096525 (+254 T>C) variant carriage was 60% in VKH, 28.6% in idiopathic uveitis, and 14.3% in scleritis. Fisher's exact test confirmed significantly higher variant frequencies in VKH and idiopathic uveitis. Longer CATT repeats (rs5844572) appeared more frequent in VKH, although statistical significance was limited by cohort size. These data align with previous reports linking MIF gene polymorphisms with uveitis susceptibility. In addition, these data indicate that the SNP alleles might associate with MIF-coordinated inflammation in VKH while the wild type allele correlates with uncoupling of MIF from the ocular immune network in scleritis.

Conclusion

Our study demonstrates that tear samples serve as an effective, non-invasive biofluid for immunological profiling in ocular inflammatory diseases. Tear

immuno-inflammatory factors reflect disease-specific immune alterations, distinguishing between intraocular and ocular surface inflammatory conditions as well as between disease subtypes. Differences are evident not only in absolute cytokine levels but also in relative abundance and correlation structures which are consistent with the expected differences in underlying immune mechanisms, thus validating the tear-fluid factors for carrying functionally relevant information. The MIF-IL-2 relationship emerges as a potential marker of ocular immune balance, with possible utility in monitoring treatment response in ocular inflammatory and infectious diseases. MIF gene polymorphisms are differentially distributed in uveitis and scleritis, supporting MIF's integral role in immune response variability in ocular inflammatory diseases.

Future directions include expanding the cohort to include infectious uveitis and fungal keratitis, enabling cross-comparisons between infectious and non-infectious inflammatory paradigms. Thus, this project aims towards precision ocular immunology, leveraging non-invasive tear-based biomarkers to inform patient-specific diagnosis, risk assessment, and therapeutic strategies.

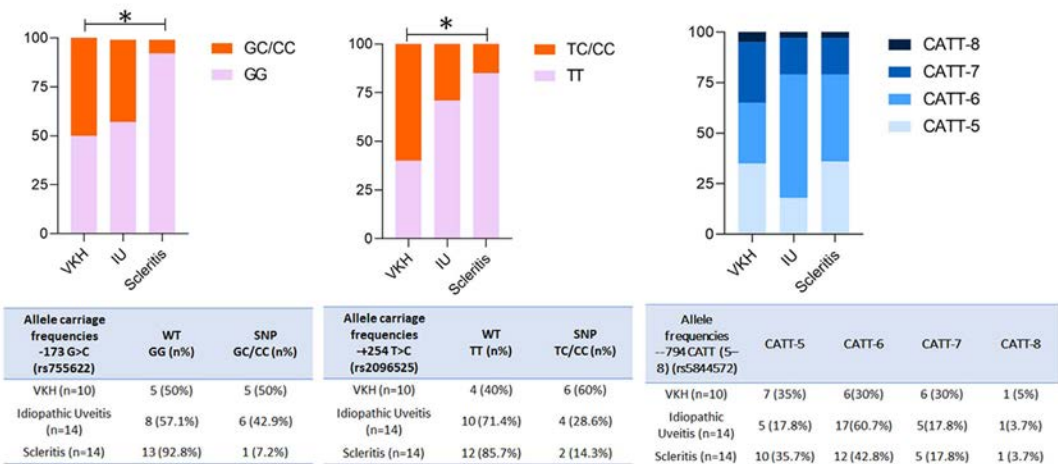


Figure 4: Graphs and tables representing difference in SNP allele and STR distribution in different disease groups

REGIONAL RESEARCH CENTRES (RRC)

Close interactions of clinicians with the scientists are essential for resolving the societally relevant problems. Hence, the regional research centres were established at Pondicherry, Coimbatore and Tirunelveli, equipped with instruments needed for the basic analysis. Advanced analyses were carried out in AMRF to derive meaningful conclusions. Clinician Scientists at RRCs identified the clinical problems that are more prevalent in the region or unique to the region. Larger families with Glaucoma were identified by the clinicians and DNA isolation was done at RRCs followed by genetic analysis of those families at AMRF. Cornea specialists identified the higher prevalence of Keratoconus among Down syndrome patients and their clinical and molecular characterisations were carried out. Viral keratitis was another important problem taken up for improving the treatment procedures. RRCs play an important role in translational research for improving the patient care.

RRC, COIMBATORE

The Coimbatore Regional Research Centre of AMRF integrates basic science, translational research, and clinical studies in ophthalmology. The centre was officially inaugurated on 27 August 2025. Research activities are organised into five core areas that together support the translation of scientific findings from laboratory studies to clinical applications. Basic and discovery research is undertaken through the Aravind Centre for Excellence for Genetic Interventions in Sight (AEGIS), Aravind Centre for Disparities in Ocular Diseases and Health Equity (ADORE), and the Aravind Centre for Regeneration and Innovation in Sight Engineering (ARISE). Translational research and clinical evaluation are carried out through Aravind Drug Innovation, Repurposing, Evaluation and Clinical Translation (A- DIRECT) and the Aravind Centre for Clinical Trials (ACT). Through this integrated structure, RRC aims to ensure that basic scientific findings are systematically evaluated and translated into clinically relevant interventions.

AEGIS- Project 1

Multicentric diagnostic and therapeutic evaluation of a rapid molecular platform for fungal endophthalmitis, and metagenomic sequencing to characterise the microbial spectrum in endophthalmitis

Principal Investigator : Dr. Siddharth Narendran
Co-Investigators : Dr. R. Kim, Dr. Lalitha
Prajna, Dr. V.R. Saravanan,
Dr. Karthik Srinivasan, Dr.
Bharanidharan Devarajan, &
Dr. Jaishree Pandian
Project students : Balaji, Brindha,
Seethalakshmi, & Niveditha
Funding Agency : ICMR Intermediate Grant

Background

Fungal endophthalmitis is a severe intraocular infection associated with poor visual outcomes and high rates of globe loss. Conventional diagnostics (smear and culture) can have low sensitivity and long turnaround times, leading to delayed or inappropriate therapy. This gap is especially important in tropical and resource-limited settings, where fungal infections

are more frequent. There is therefore a clear need for rapid, sensitive, and clinically deployable molecular diagnostics that can support early antifungal intervention.

Preliminary results

This project builds on RID-MyC (Rapid Identification of Mycoses using CRISPR), a CRISPR-Cas12a-based assay for rapid fungal detection in intraocular samples.

Key findings from preliminary multicentric evaluation (133 intraocular samples from 117 patients):

- Higher detection yield than conventional methods (positivity: RID-MyC 17.29%, pan-fungal PCR 12.78%, culture 1.50%).
- Rapid turnaround (mean: 48 minutes).
- Strong diagnostic performance: sensitivity 88.24% (CI 63.56–98.54) and specificity 93.10% (CI 86.86–96.98), improving after discrepancy analysis to 90.48% sensitivity and 96.43% specificity.
- Improved resistance to inhibitors in ocular fluids with markedly higher analytical sensitivity than PCR (example: *Aspergillus flavus* LoD in aqueous humor: RID-MyC 165 aM vs PCR 165 fM).

Key objectives (next phase)

- Multicentric clinical validation of RID-MyC against standard microbiological methods.
- Microbial spectrum characterisation using targeted and unbiased sequencing, with emphasis on culture-negative cases.
- Evaluation of the clinical impact of RID-MyC-guided therapy through a randomised controlled trial.

Expected outcomes and translational relevance

The project aims to establish a rapid molecular diagnostic workflow that can be integrated into clinical practice to support timely antifungal therapy and improved outcomes. Findings are expected to inform diagnostic algorithms and evidence-based management of fungal endophthalmitis, with potential applicability to other invasive fungal infections.

AEGIS - Project 2

ARIDE-CAP: Strengthening Clinical, Molecular, and Computational Research Capacity for Inherited Retinal Diseases in India (Aravind Registry for Inherited Diseases of the Eye – Capacity Building Program)

Principal Investigator	: Dr. R. Kim
Co-Principal Investigators	: Dr. P. Sundaresan, Dr. D. Bharanidharan, & Dr. Siddharth Narendran
Key Collaborators	: Prof. Fred Chen (Lions Eye Institute, Australia) & Prof. Nikolas Pontikos (University College London / Moorfields Eye Hospital, UK)
Student Project Lead	: Jayashree
Funding Agencies	: GKD Charity Trust, MAK Controls (Private), Velux Stiftung (Ophthalmology in LMICs).

Background:

Inherited retinal diseases are an important cause of irreversible blindness. India has limited large-scale, longitudinal registries linked to genomics, and Indian populations remain under-represented in global datasets. A structured registry that combines standardised phenotyping, imaging, genetics, and

analytics is needed to support accurate diagnosis, variant interpretation, and future trial readiness and therapy access.

Results and conclusion (pilot feasibility, Jan–Jun 2025)

- Registry registered in Clinical Trials Registry–India (CTRI/2024/07/071686) with governance/SOPs and high completion of standardised multimodal imaging (~98%).
- 1,439 screened; 693 enrolled (491 probands, 202 relatives); ~98% uptake among referred probands.
- Diagnostic profile among affected participants: retinitis pigmentosa 56%, cone–rod dystrophy 10%, Stargardt disease 7%, macular dystrophy 4%, atypical RP 4%, syndromic RP 4%, rod–cone dystrophy 3% (others smaller).
- First 108 sequenced: 188 variant calls across 90 genes; ACMG classification: 24 pathogenic, 36 likely pathogenic, 128 variants of uncertain significance; commonly represented genes included USH2A, EYS, ADGRV1, ABCA4, with recurrent alleles already evident.

Future directions

- Expand to a multicentre network enrolling 15,000 patients across five Aravind centres with longitudinal follow-up.
- WES of 1,000 index cases, using eyeVarP adapted for South Asian allele spectra and founder variants; generate an India-specific gene/allele atlas and India-optimised diagnostic panel specification.
- Build integrated data infrastructure using FHIR / OMOP / DICOM standards and evaluate image-based gene prediction (Eye2Gene) in Indian imaging datasets.

ADORE - Project 1

Ethnolinguistic differences in diabetic retinopathy within a shared geographic setting in South India (The Gudalur Diabetic Retinopathy Study)

Principal Investigator	: Dr. Siddharth Narendran
Co-Investigators	: Dr. Sujay Jaju & Dr. Jaishree Pandian
Project students	: Niraja & Vijayakrishna
Funding Agency	: Conducted through community outreach screening & institutional research activity.

Background

Marked differences in diabetic retinopathy (DR) prevalence have been reported between Tamil Nadu and Kerala. To reduce confounding from broad geographic and health-system differences, this study compared Tamil- and Malayalam-speaking adults living within the same geographic setting using a community-based screening design.

Results

- N = 295 adults with type 2 diabetes mellitus (Malayalam n = 171; Tamil n = 124).
- Malayalam speakers were older ($P = 0.008$) and had longer diabetes duration ($P < 0.001$); HbA1c was comparable between groups.
- Any DR prevalence was higher in Malayalam speakers (49.7% vs 30.6%; $P = 0.001$), with higher proliferative DR prevalence (12.9% vs 6.5%) and different severity distribution ($P = 0.026$).
- Malayalam mother tongue remained associated with any DR after adjustment (adjusted OR 1.88; 95% CI 1.10–3.22; $P = 0.021$).
- Malayalam speakers reported higher awareness and higher annual eye-examination uptake (both $P < 0.001$).
- Dietary subset (n = 264): energy intake and saturated fat contribution were higher in Malayalam speakers (both $P < 0.001$).

Conclusion:

Even within a shared geographic setting, measurable differences in DR burden persisted between ethnolinguistic groups after adjustment for key clinical factors. These findings support context-specific prevention and counselling strategies alongside continued screening.

ADORE - Project 2

Gender disparity in cataract prevalence: Deciphering the Estrogen–Alpha-Crystallin Axis in cataract progression in women

Principal Investigator : Dr. Siddharth Narendran
Co-Investigators : Dr. Rekha, Dr. Daipayan Banerjee & Dr. Jaishree Pandian
Project Students : Elakkiya Nandhini & Akshaya
Funding Agency : ICMR Small Grant.

Background:

Women experience a disproportionate burden of cataract-related visual loss. This project evaluates disparities across sex, age, and socioeconomic strata using large-scale cataract surgical data and focused clinical studies, with emphasis on the menopausal transition and associated risk pathways.

Results

- Large retrospective dataset (Aravind Eye Care System; 2010–2023): 3,362,256 cataract surgery records (women 1,833,801; men 1,528,455).
- Women underwent cataract surgery at a younger age than men (median 60 vs 64 years).
- Female predominance was most pronounced in midlife: in the 40–49-year age group, women accounted for 9.1% of surgeries compared with 6.5% among men; the female-to-male ratio peaked in the 40–49 age group and remained higher across strata of preoperative visual acuity.
- Internal 10-year analysis (~3.3 million procedures): women aged 31–50 had higher odds of cataract surgery than men (OR 2.66; 95% CI 2.57–2.75; $P < 0.0001$).
- Presenile case–control study (women ≤ 50 years; 86 cases, 254 controls): cases were older (median 45 vs 40 years). Postmenopausal status (25.6% vs 3.5%), regular betel nut use (14.0% vs 4.7%), and lowest socioeconomic quintile (5.8% vs 0.8%) were more frequent among cases; mean age at menopause was lower in cases than controls (42.64 vs 46.89 years).
- Age-adjusted analysis in the case–control cohort showed strongest association with postmenopausal status (age-adjusted OR 5.92; 95% CI 2.27–15.46, vs premenopausal reference).
- Regular betel nut use was associated with higher odds of presenile cataract (age-adjusted OR 2.55; 95% CI 1.01–6.48, vs never).
- Lower socioeconomic class was associated with higher odds (Class IV vs III: age-adjusted OR 2.44; 95% CI 1.34–4.44; Class V vs III: age-adjusted OR 7.15; 95% CI 1.18–43.24).

Conclusion

Across population-scale surgical data and focused clinical studies, women show a consistent pattern of earlier cataract surgery. Signals point toward contributions from the menopausal transition alongside socioeconomic and lifestyle factors. Ongoing work focuses on refining risk estimates, addressing confounding carefully, and translating findings into practical screening and counselling approaches for midlife women.

Topographic Retinal Pigment Epithelium Immunogenic Heterogeneity and Its Implications in Degenerative Diseases

Principal Investigator : Dr. Siddharth Narendran
Co-Investigator : Dr. Jaishree Pandian
Project students : Karvannan & Janani

Background:

Age-related macular degeneration (AMD) is a leading cause of irreversible vision loss in older adults. A defining feature is the selective vulnerability of the macular retinal pigment epithelium (RPE) compared with relatively preserved peripheral RPE. This project aims to characterise regional differences between macular and peripheral RPE, focusing on immunogenic profiles and regulatory networks relevant to degeneration.

Results

- Meta-analysis of single-cell RNA sequencing of regionally defined RPE cells demonstrated marked topographic heterogeneity between macular and peripheral RPE populations.
- Distinct transcriptional signatures were observed, including differences in immunoregulatory genes, differentiation/lineage-associated genes, and cell fate-related programs.
- Macular RPE showed increased variability in immune-related pathways, suggesting a heightened immunogenic state that may contribute to selective vulnerability in degenerative disease contexts.

Future directions

- Mechanistic validation of candidate immunogenic and regulatory factors driving regional RPE identity and vulnerability.
- Assessment of impact on Bruch's membrane integrity, extracellular matrix remodelling, and RPE–Bruch's membrane interactions.
- Network-level analysis of how region-specific factors shape downstream programs under disease-relevant stress.
- Disease-context modelling to evaluate how altered immunogenic states influence RPE dysfunction and degeneration in AMD.

Delineation of the regulatory pathways governing corneal endothelial cell regeneration

Principal Investigator : Dr. Siddharth Narendran
Co-Investigators : Dr. Divya &
Dr. Jaishree Pandian
Project Student : Vijayakrishna

Background

The corneal endothelium maintains corneal transparency through barrier and pump functions. In humans, corneal endothelial cells have limited proliferative capacity in vivo, and cell loss is commonly compensated through migration and enlargement rather than true regeneration. Once endothelial cell density falls below a functional threshold, irreversible corneal edema and vision loss can occur. Emerging evidence suggests latent regenerative potential and regional heterogeneity, but the molecular mechanisms governing quiescence and regenerative competence remain incompletely understood.

Planned work and future directions

- Characterise region-specific or progenitor-like endothelial cell subsets, particularly in the peripheral cornea, that exhibit enhanced regenerative capacity.
- Examine how Descemet's membrane composition, stiffness, and age-related alterations influence endothelial repair and survival.
- Assess how regenerative pathways are altered in pathological conditions such as Fuchs endothelial corneal dystrophy and iridocorneal endothelial syndrome.
- Evaluate pharmacologic modulators (e.g., ROCK inhibitors), gene regulatory targets, and cell-based approaches to enhance endothelial repair in preclinical models with emphasis on clinically feasible interventions.

RRC - MADURAI, PONDICHERRY & COIMBATORE

Screening of family members of Juvenile Open Angle Glaucoma (JOAG) patients for myocilin gene mutations

Investigators : Dr. S.R. Krishnadas, Dr. Indira Durai, Dr. R. Venkatesh, Dr. S. Kavitha, Dr. Swati Upadhyaya, Dr. Ganesh V. Raman, Prof. K. Dharmalingam, Dr. A. Vanniarajan, & Dr. D. Bharanidharan

Research Personnel: V. Saravanan, Sridevi, & Lalitha

Funding Agency : AMRF, AEF

Background

Genetics is now becoming an integral part of the clinical evaluation of glaucoma as there is an increased risk of the disease in individuals with a positive family history. Myocilin gene was the most commonly altered gene among those individuals with multiple members affected in the family and correlating with the severity of the disease. Juvenile Open Angle Glaucoma (JOAG) patients seeking treatment at Aravind Eye Hospitals in Madurai, Pondicherry and Coimbatore were screened for Myocilin gene mutations and risk is predicted based on the mutation pattern in family members.

Results

AEH, Madurai

Among 224 JOAG patients who visited the hospital, we identified 52 probands with a family history of glaucoma. Blood samples were collected, isolated

DNA, and stored for further experiments. For the myocilin gene mutational screening, exon 3 was first screened; if it turned negative, the other two exons were screened. Four families were identified with known pathogenic mutations and four families with novel myocilin mutations.

Known pathogenic myocilin mutations include G367R, N480K, A363T and T377M identified in four families. In addition, one novel heterozygous mutation, Y365S, and one novel homozygous mutation, A363, were detected in one and three families, respectively, which requires further screening of family members and experimental studies to confirm their pathogenicity.

AEH, Pondicherry

From RRC Pondicherry, 58 samples of juvenile-onset open-angle glaucoma (JOAG) were identified. Among these, 24 samples were collected from nine families. Further screening of these nine families revealed that in one family, six members were affected with JOAG or ocular hypertension (OHT) and a pathogenic T377M mutation was detected in the father and son.

AEH, Coimbatore

A total of 42 patients have been recruited for the study from Coimbatore. Genomic DNA was successfully extracted from all 42 samples, and screening of the Myocilin gene was performed. Among eleven families analysed, two families were found to have heterozygous known pathogenic MYOC mutations: P254L and T377M. In one family, ten members with variable phenotypes such as juvenile-onset open-angle glaucoma (JOAG), glaucoma suspect, or normal phenotype were screened, and the pathogenic T377M mutation was detected in eight of these family members (Figure 1).

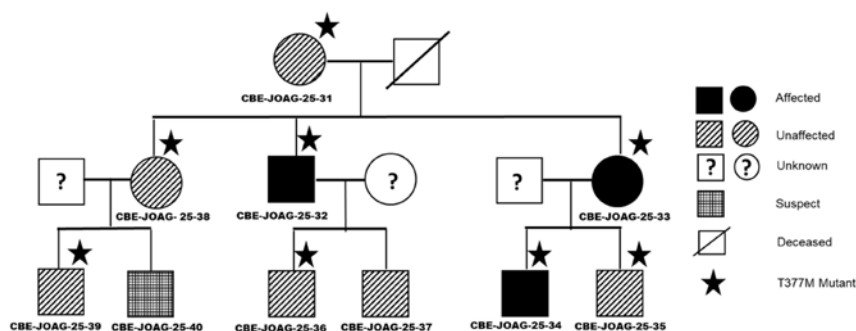


Figure1: A large family with multiple members affected with JOAG. The mutation T377M was observed in both the affected and unaffected individuals from the same family indicating that there may be additional genes that may be causing the disease

Keratoconus severity and tomographic patterns between eyes in individuals with Down syndrome

Investigators : Dr. Anitha Venugopal,
Dr. Meenakshi Ravindran,
Dr. Josephine Christy,
Dr. OG. Ramprasad,
Dr. A. Vanniarajan, &
Prof. K. Dharmalingam

Background:

Keratoconus (KC) is a chronic inflammatory disorder characterised by progressive corneal thinning, irregular astigmatism, and corneal scarring, leading to defective vision. The incidence of KC is higher among patients with associated disorders such as chronic allergic eye disease, atopy, hormonal imbalance and genetic disorders like Down syndrome.¹ Specifically, KC is higher among Down syndrome individuals compared to the general population, varying up to 71%. Hence this project is aimed to evaluate the severity of Keratoconus (KC) associated with tomographic characteristics in Down syndrome (DS) individuals based on Amsler Krumeich classification

Methods:

A cross-sectional study was conducted in 96 DS patients with KC (55 males, 41 females; mean age: 15.7±7.3 years) at AEH, Tirunelveli and Pondicherry. Participants were grouped under four KC stages using Amsler-Krumeich classification with rotating Scheimpflug imaging (Pentacam HR) and slit-lamp biomicroscopy. Key parameters included mean keratometry (K), corneal thickness (CT), astigmatism, ARTmax, higher-order aberrations (HOA) and pachymetry.

Results:

We could identify 75% of the patients in early stages of KC (Stage I ((31), stage II (41) and the tomographic indices such as, ARTmax and ZKmax were helpful in differentiating KC eyes from non-KC eyes. As the severity increases (Stage III and IV), the corneal parameters such as mean CT, mean K, astigmatism, ZKmax, HOA and PE at the thinnest show significant correlation with disease progression and comparative to the results from other geographic locations among DS with KC. Logistic regression further confirms that mean CT, mean K and astigmatism are strongly associated with more severe stages of KC. Conversely, ARTmax, ARC and PRC are negatively associated with severity of KC in DS.

Conclusion:

Tomographic patterns align very well with the severity of the disease. Detection of KC in DS patients at early stages makes the intervention possible as corneal collagen cross-linking (CXL) may still be effective in stage II patients. Our findings emphasise the importance of targeted screening in DS individuals to address unique progression patterns and manage KC at early stages. The severity of the disease will be further analysed and characterised through the molecular studies.

Tear protein profiling for recurrent viral keratitis

Investigators : Dr. Josephine Christy,
Dr. Aditi Parab,
Prof. K. Dharmalingam, &
Dr. A. Vanniarajan

Background:

Approximately 1.5 million new cases are diagnosed annually with corneal opacification and infectious blindness due to viral keratitis. In India, about 10% of corneal infections are due to herpes simplex virus (HSV). HSV-1 and HSV-2, along with Varicella zoster virus (VZV), manifest various forms of keratitis, such as superficial punctate keratitis, dendritic ulcers, stromal keratitis, disciform keratitis, endothelitis. This study is aimed to profile the proteomic alterations in tears of HSV patient presenting as epithelial, stromal and endothelial keratitis.

Results:

Tear samples were categorised into three groups – (1) Different forms of Keratitis from the infected eye (2) Control from the contralateral eye (3) Healthy control. So far 33 tear samples each from the infected eye and contralateral eye along with 2 samples from healthy controls. Keratitis samples include 12 dendritic; 9 immune stromal; 7 disciform and 5 keratouveitis. Initial proteomic analysis showed distinct profiles in tears of infected eye compared to contralateral eye.

Conclusion:

The profile alterations in the infected keratitis are distinct and further analysis will identify the changes in the profile among the different groups of viral keratitis.

ICMR - CENTRE FOR ADVANCED RESEARCH (CAR) IN GLAUCOMA

Early detection of open angle glaucoma in diabetes patients with increasing age and family history of glaucoma

This year the Indian Council of Medical Research, Govt of India, sanctioned the Centre for Advanced Research (CAR) in Glaucoma to AMRF. This is a prestigious program for a period of Five years. In this ambitious project we will be using the combined effort of multiple investigators to develop and deploy a prediction tool to identify this serious vision threatening disease well ahead of the development of the earliest disease related changes of Glaucoma among clinically undiagnosed individuals with Diabetes. The ultimate objective is using multi omics including clinical imageomics for the development of this novel prediction tool.

Investigators : Dr. S.R. Krishnadas (PI),
Dr. K. Dharmalingam,
Dr. A. Vanniarajan &
Dr. D. Bharanidharan
Project Fellows : Dr. K. Lavanya, Dr. R. Santhosh,
R. Kaviya, T. Saravanakumar,
V. Ramya, K. Petchimuthu,
T. Maheswari
Funding Agency : ICMR-CAR

Introduction:

Glaucoma is the leading cause of irreversible blindness, with an estimated prevalence of about 4% in India (Rewri, 2023). The prevalence of glaucoma among diabetic individuals above 40 years in India ranges from 4.9% to 20% (Bhutia et al., 2018), which is notably higher than the estimated prevalence. Hence, early detection of glaucoma in this targeted group of diabetic patients can help prevent glaucoma progression. We propose early detection of Open Angle Glaucoma (OAG), the most common type in the targeted risk group of diabetes patients with increasing age and family history of glaucoma. This study uniquely integrates multi layers of evidence—clinical, imageomics, genomic, transcriptomic and proteomic biomarkers—to develop a comprehensive predictive model for early diagnosis.

Genomic analysis of POAG patients (T. Saravanakumar)

Background:

Genetics plays an integral role in the identification of the individuals at risk of glaucoma. Increasing age and a positive family history are indicators of POAG. Participants aged 40 years and above with a confirmed diagnosis of POAG are recruited with

complete clinical investigations including intraocular pressure (IOP) measurement, visual field (VF) testing, retinal nerve fibre layer (RNFL) assessment, and cup-to-disc ratio analysis. Blood samples from the probands and family members were collected and isolated the DNA.

Results:

Five families fulfilling the criteria have been recruited and carefully evaluated. In one representative family, the proband and all of his siblings are affected by POAG. Inherited from the previous generations. Additionally, the next generation, including the proband's niece and nephew, have also been diagnosed. This pattern of affected individuals across multiple generations strongly suggests a familial aggregation consistent with autosomal dominant inheritance.

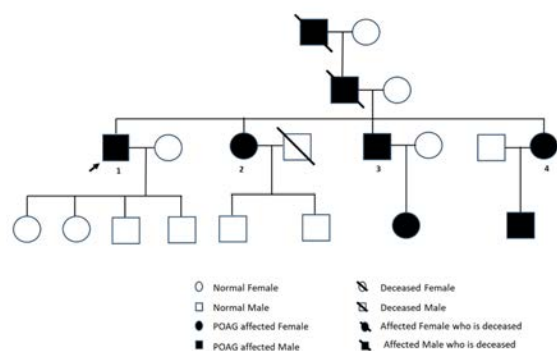


Figure 1: A family with multiple affected POAG members across 4 generations

Conclusion:

Families with several members affected with POAG were identified and they will be further characterised by whole genome analysis to identify the causative variants.

Proteome profiling of aqueous humour from diabetic POAG patients (R. Kaviya)

Background:

The study includes five diabetic patient groups: (i) DM without primary open-angle glaucoma (POAG), (ii) DM with ocular hypertension (OHT), (iii) DM with mild POAG, (iv) DM with moderate POAG, and (v) DM with severe POAG. The experimental strategy comprises a discovery phase using MS/MS-based proteomic analysis, followed by a pilot phase using the Olink assay platform, and a final validation phase employing multiplex ELISA for confirmation of candidate biomarkers.

	Cohorts	DM POAG					Total
		No	OHT	Mild	Moderate	Severe	
No. of samples	AH	0	1	6	13	16	36
	Plasma	0	7	30	47	62	146

Table1: Samples collected for the study

Results:

Aqueous humour samples are collected intraoperatively from patients undergoing glaucoma or cataract surgery at Aravind Eye Hospital, Madurai. Diabetic POAG cases and diabetic non-glaucoma patients are being recruited prospectively based on surgical eligibility, while age-matched diabetic cataract patients serve as controls. Comprehensive clinical data, including intraocular pressure, visual field parameters, and systemic comorbidities, are being documented alongside sample collection. All samples are immediately stored at -80°C following collection to ensure preservation of analyte integrity.

In parallel, standardisation and optimisation of mass spectrometry-based proteomic profiling are being carried out using control aqueous humour samples. To begin with, we investigated whether there are differences in the proteomic profiles between male and female subjects in both control and patient groups. As an initial step, the protein profiles of aqueous humour from control cataract male and female patients were analysed. Mass spectrometric analysis was performed following 1D gel profiling of pooled aqueous humour samples from male and female control cataract groups. The analysis revealed substantial differences in the proteomic profiles between males and females, with the presence of both common and unique proteins. Further analysis

of the top 100 identified proteins showed that 25 proteins were unique to either male or female samples, while 75 proteins were common between the two groups.

Conclusion:

Preliminary proteomic analysis of aqueous humour indicates distinct gender-specific differences in protein profiles, underscoring the importance of standardised mass spectrometry workflows and demographic stratification prior to large-scale biomarker discovery in diabetic POAG.

Extraction of image features OCT images using deep learning method (K. Petchimuthu)

Background:

Optical coherence tomography (OCT) has become a cornerstone tool for glaucoma assessment, as it enables sensitive evaluation of inner retinal structural changes, particularly in the macular retinal nerve fibre layer (mRNFL) and the ganglion cell-inner plexiform layer (GCIPL). To reliably quantify these subtle layer-specific alterations, deep learning-based retinal layer segmentation is increasingly used to accurately delineate clinically meaningful boundaries (e.g., ILM, RNFL, GCIPL) and compute objective thickness measurements (global and sector-wise), which are established biomarkers of glaucomatous

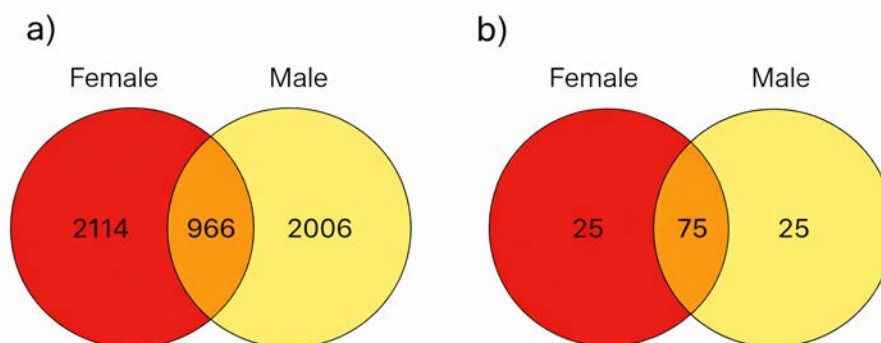


Figure 2. Gender-specific proteomic profiling of aqueous humour.

(a) Venn diagram showing common and unique proteins identified in aqueous humour from male and female control cataract subjects following MS/MS analysis. (b) Distribution of the top 100 proteins shared between male and female samples ranked based on peptide spectrum matches (PSMs), highlighting abundantly detected common proteins across both groups.

neurodegeneration. We used deep learning method which have integrated segmentation free-driven thickness with classification using EfficientNet-B0 backbone pretrained on ImageNet to assess the RNFL thickness (Mariotoni et al., 2020).

Results and Conclusion:

A total of 1,400 OCT images were included in this study. Of these, 1,000 images (500 per class) were used for model training. Images were split at the patient level into training, validation, and testing subsets with a ratio of 1000:200, ensuring that no subject overlap occurred between subsets. All OCT scans were pre-processed using standard procedures, including CLAHE enhancement, normalisation (ImageNet mean and standard deviation), and resizing to 224×224 pixels prior to model training. For RNFL thickness estimation, a segmentation-free deep learning approach was employed using an EfficientNet-B0 backbone pretrained on ImageNet. The model was trained using proxy thickness labels derived from disease class associations (NORMAL: 97µm) with smooth L1 loss as the objective function. Training was accelerated using mixed-precision automatic gradient scaling on an NVIDIA T4 GPU, achieving convergence within 10 epochs. On the held-out validation set, the model achieved a mean absolute error (MAE) of 4.59µm and a Pearson correlation coefficient of $r = 0.8881$ ($p < 0.001$) between predicted and target thickness values, with an R^2 score of 0.7592. For visualisation and clinical interpretation, three complementary methods were employed: (1) thickness heatmaps displaying color-coded RNFL thickness distributions

using a jet colormap (40–150 µm range).⁽²⁾ thickness profile plots showing lateral thickness variation across A-scans with predicted average and normal range indicators (80–110 µm); and (3) 3D surface plots providing topographic representations of RNFL thickness landscapes. Post-hoc gradient-based boundary detection was applied for qualitative validation of thickness predictions, employing Sobel edge detection with CLAHE preprocessing (clip limit = 3.0) and Savitzky-Golay smoothing (window = 41, polynomial order = 2) to identify ILM and RNFL-GCL interfaces. These visualisation techniques enabled comprehensive assessment of RNFL structural integrity and facilitated comparison with clinical thickness norms.

References:

- Thompson AC, Jammal AA, Berchuck SI, Mariotoni EB, Medeiros FA. Assessment of a Segmentation-Free Deep Learning Algorithm for Diagnosing Glaucoma from Optical Coherence Tomography Scans. *JAMA Ophthalmol*. 2020 Apr 1;138(4):333-339. doi: 10.1001/jamaophthalmol.2019.5983. PMID: 32053142; PMCID: PMC7042899.
- Mariotoni, E.B., Jammal, A.A., Urata, C.N. et al. Quantification of Retinal Nerve Fibre Layer Thickness on Optical Coherence Tomography with a Deep Learning Segmentation-Free Approach. *Sci Rep* 10, 402 (2020). <https://doi.org/10.1038/s41598-019-57196-y>

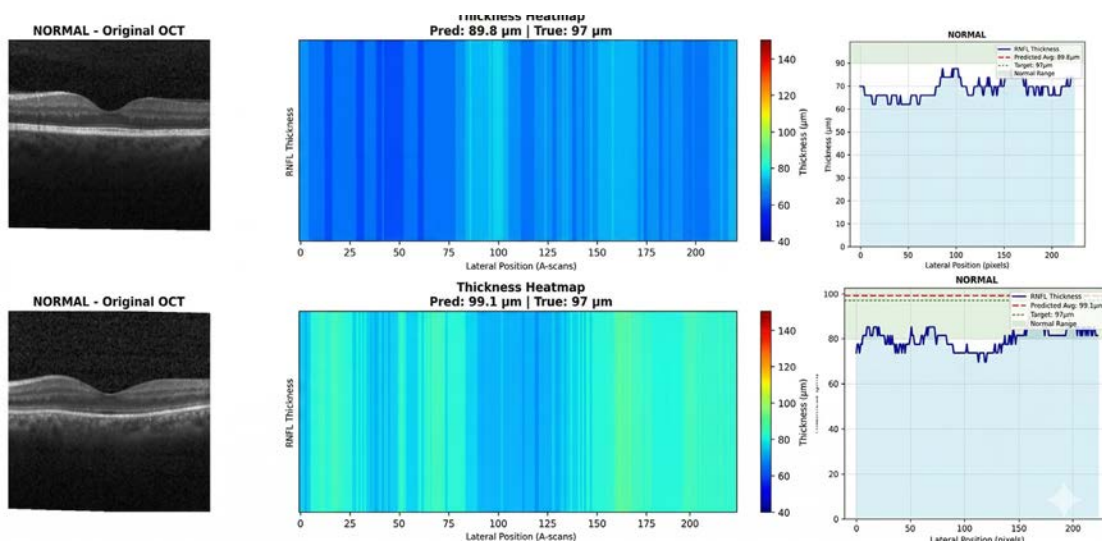


Figure 1: (A) Normal Eye - original OCT Image (B) Heat map of RNFL Thickness. Prediction of 89.8µm and 99.1µm. (C) Prediction profile of RNFL Thickness.

CONFERENCES / MEETINGS

83rd Annual Conference of the All India Ophthalmological Society (AIOS) 2025

New Delhi, 3-6 April 2025

Dr. P. Sundaresan, ICMR Emeritus Scientist participated at the 83rd Annual Conference of the All India Ophthalmological Society (AIOS 2025) held at New Delhi.

Paper presentations:

- Mitochondria and Wound healing (Session: Cells Talk: Innovations in Cell-Based Ophthalmology)
- Translational Genomics of Eye Diseases (Session: Advances in Multi-Omics Approaches to Understanding Eye Diseases)
- A Journey in Glaucoma Genetic Research (Session: Genetics in Ophthalmology: Current Methods of Genetic Analysis and Potential Genetic Treatments)

International Seminar on New Horizons in Plant Science (NHPS-2025)

Kariavattom, 8-10 April 2025

Prof. K. Dharmalingam, Director-Research, AMRF has delivered the invited talk on “Microbiome Science and Applications” during the International Seminar on New Horizons in Plant Science (NHPS-2025)

- “Harnessing Bio-Resources: Opportunities in Prospecting, Sustainable Utilisation and Bioresource-based Entrepreneurship” organised by Department of Botany, Kerala University Business Innovation and Incubation Centre (KUBIIC), University of Kerala.



Dr. S. Senthilkumari at ARVO 2025, USA

Annual Meeting of the Association for Research in Vision and Ophthalmology (ARVO) 2025

USA, 3-8 May 2025

Scientists from AMRF participated in the Annual Meeting of the ARVO 2025, held at Salt Lake City, Utah, USA.

Dr. P. Sundaresan

- Functional Analysis of GUCY2D Mutant: An Intrinsic Cause of Leber's Congenital Amaurosis (Poster presentation)

Dr. Senthilkumari Srinivasan

- Hsa-MiR-483 -3p regulates the Extracellular Matrix Proteins via TGF β 2/SMAD4 Signalling in the Glucocorticoid-responsive Human Trabecular Meshwork Cells – (Paper Presentation Session: “Glaucoma: Aqueous outflow cell biology, fibrosis and wound healing”) Her work was selected for the hot topic of the ARVO 2025.

Dr. P. Sundaresan at AIOS Conference 2025, New Delhi



Prof. K. Dharmalingam at NHPS 2025 seminar, Kariavattom





AMRF team at ARVO-India 2025, Chennai

31st Annual meeting of ARVO-India 2025

Chennai, 26-27 July 2025

Faculty members and Research scholars of AMRF participated at the 31st annual meeting of the ARVO-India 2025 was held at Sankara Nethralaya, Chennai.

Dr. Gowri Priya Chidambaranathan

- Towards a stem cell-based therapy for patients with primary open angle glaucoma (Key Note lecture)

Dr. Bharanidharan Devarajan

- Enhancing Genomic Discovery with a Novel Computational Framework for Eye Disease-Specific Pathogenic Variants (Invited Talk)

Dr. P. Sundaresan

- Session: Genetics, Molecular and Cell Biology Session (Moderator)
- Dr. S. Senthilkumari served as a judge for poster session and free paper session.

Gopika S. Kumar

- Pigmentation loss reflects EMT and Functional decline in iPSC-derived RPE cells (Oral presentation)

Dharsini Nandakumar

- Prospective evaluation of RID-MYC assay for POC diagnosis and clinical management in smear and culture-negative keratitis with IVCM correlation (Oral presentation)

Dhanu Prakash Muthupandi

- Distinct Tear fluid macrophage Migration Inhibitory Factor (MIF) and inflammatory mediator profiles in uveitis patients (Poster presentation)

Anusha Erulandi

- Altered tear fluid immuno-inflammatory profiles in patients with scleritis (Poster presentation)

Mathan L

- Proteomic Analysis of Plasma Small Extracellular Vesicles (SEVs) In Patients with Diabetic Retinopathy (DR) In the Indian Population (Poster presentation)

Dr. Daipayan Banerjee

- Plasma and vitreous extracellular vesicle-derived retinol binding protein 3 as a molecular signature of proliferative diabetic retinopathy (Poster presentation)

Shiva Sankari Sundar

- Leber Congenital Amaurosis: Genetic spectrum and genotype-phenotype correlations for precise molecular diagnosis (Poster presentation)

Hari Vignesh S

- Exploring mitochondrial genetic variants in primary open angle glaucoma (Poster presentation)

Karthik Alagarsamy

- Localisation of multiple isoforms of human tear lysozyme in the extracellular vesicle (Poster presentation)

Kanthimathi R

- Spatial transcriptomic analysis of adult human retinal pigment epithelium (Poster presentation)

Lakshmi Devi Priya J.

- Demonstrating the anti-fibrotic property of Relaxin-2 (RLN2) In TGF- β 2 induced fibrosis model of glaucoma using human organ cultured anterior segment (Poster presentation)

Thushmitha P

- Alteration in the YAP signalling pathway in cataractous human lens epithelium (Poster presentation)

Sneha Nair

- Unravelling MicroRNA signatures in human trabecular meshwork stem cells: Implications with ageing and in glaucoma (Poster presentation)

Dr. Senthilkumari Srinivasan

- Regulatory role of Micrnas in Wnt signalling pathways associated with steroid-induced ocular hypertension: An insight from Ex Vivo studies (Poster presentation)

Kanmani M

- Development of an integrated RPA-EXO probe assay for low complexity and rapid detection of fungal DNA in ocular infections (Poster presentation)

Hariharan Gnanam

- Isolation and characterisation of extracellular vesicles from *aspergillus flavus* cultures exposed to stress (Poster presentation)

Praveenkumar R

- MiRNA profiling reveals novel mechanistic insights on the role of neurotrophin signalling in *pseudomonas aeruginosa* keratitis (Poster presentation)

Shreya Dinesh

- Comparative host transcriptome analysis to identify core and pathogen-specific immune response in fungal keratitis (Poster presentation)

Dr. Swagata Ghosh

- Distinct corneal gene expression profiles associate with differential disease trajectories in *fusarium* and *aspergillus keratitis* (Poster presentation)

G. Balaji

- CRISPR-Based RID-Myc assay versus conventional culture in a postoperative fungal endophthalmitis cluster: Diagnostic and therapeutic implication (Poster presentation)

PRIME Summit 2025

Narayana Nethralaya, Bengaluru, 7-9 November 2025

Faculty members and Research scholars of AMRF participated at the PRIME Summit 2025 (Progress in Regenerative & Investigational Medicine in Eye) - Workshop & Conference organised by Narayana Nethralaya Foundation, Bengaluru. All the faculty members served on the judging panel for the poster session.



AMRF group at PRIME summit 2025, Bengaluru

Dr. P. Sundaresan

- Paving the Future of Regenerative Medicine: Preparing for gene and cell therapy in India (Moderator at a session and panel discussion)

Dr. Gowri Priya C

- Small extracellular vesicles for human trabecular meshwork regeneration in primary open-angle glaucoma (Invited talk)

Dr. Daipayan Banerjee

- EVs on the road to biomarker discovery in diabetic retinopathy: Retinal RBP3 in plasma small extracellular vesicles (Invited talk)

Dr. Swagata Ghosh

- Host-response transcript profiling in the patient cornea: an early lead for predicting treatment outcome in fungal keratitis (Invited talk)

Shiva Sankari

- Clinical and Genetic Analysis of CRB1: Associated Leber Congenital Amaurosis (Poster presentation)

Hari Vignesh

- Mitochondrial dysfunction drives primary open-angle glaucoma: Insights from whole mitochondrial genome sequencing (Poster presentation)

UNICORA 2025 – National Conference and Workshop on Advances in Microscopy for Diagnosis and Research

Trichy, 7-8 November 2025

Dr. Lalitha Prajna and Dr. Gowri Priya

Chidambaranathan participated and delivered invited talks at the two-day national conference and workshop on Advances in Microscopy for Diagnosis and Research was conducted by the Department of Microbiology, SRM Medical College, and the



Dr.Lalitha and Dr.C.Gowri Priya received the memento at UNICORA 2025, Trichy

Department of Biotechnology, SRM Institute of Science and Technology, Trichy.

Dr. Lalitha Prajna

- Applications of light microscopy

Dr. Gowri Priya Chidambaranathan

- Confocal microscopy and its applications

Integrated Single Cell and Spatial Workshop

Adyar Cancer Institute (WIA), Chennai, 20–21 November 2025

R. Kanthimathi, Project Research Scientist I and P. Thushmitha, Senior Research Fellow, Immunology and Stem Cell Biology, AMRF, attended the Integrated Single Cell and Spatial Workshop, organised by 10x Genomics and GeneX India Bioscience Pvt. Ltd. The two-day programme, held at Adyar Cancer Institute (WIA), Chennai, combined basic theory, demonstrations, hands-on training and analysis related to single-cell sequencing and spatial transcriptomics.

Discussion in integrated single cell and Spatial workshop, Chennai



Special Lecture at the 25th Popular Science Lecture Madurai, 5th December 2025

Prof. K. Dharmalingam delivered a special lecture at the Kalaingar Centenary Library, Madurai as part of the 25th Popular Science Lecture on the topic, "Our unseen friends: Microbiome and us". The event was jointly organised by The Kalaingar Centenary Library, Madurai and the Tamil Nadu Science Forum, Madurai.

The importance of the microbes as beneficial organisms was the main theme, explained in simple terms during the talk. How microbes made the earth liveable, how microbes help humans in all aspects of the human activities were the points emphasised. The omnipresence of microbes, their communication with human cells and how the harmless microbes become pathogens were the other aspects of the talk. The take home message was that microbes are our co-inhabitants and we must take good care of our microbiome to live healthy life. There was extensive interaction and large number of questions from a young girl to very old people.

Special lecture by Prof.K.Dharmalingam at Kalaingar Centenary Library





Dr.C.Gowri Priya delivered inaugural lecture at Lady Doak College, Madurai

Workshop on Animal Cell Culture Techniques

Department of Biotechnology, Lady Doak College, Madurai, 29th January 2026

Dr. Gowri Priya Chidambaranathan, Scientist, Department of Immunology and Stem Cell Biology, Aravind Medical Research Foundation delivered the inaugural lecture on "Role of Cell Culture in Regenerative Medicine" in the workshop on Animal Cell Culture Techniques at the Department of Biotechnology, Lady Doak College, Madurai.

International Conference on AI for Genomics and Drug Discovery

Coimbatore, 28-30 January 2026

Research scholars from the Department of Immunology and Stem Cell Biology, Aravind Medical Research Foundation, participated in the International Conference on AI for Genomics and Drug Discovery (ICAGDD-2026), organised by the Department of Bioinformatics, Bharathiar University, Coimbatore. The scholars delivered oral presentations and were recipients of travel grant awards.

Thushmitha P., ICMR-Senior Research Fellow

- Data mining of single-nucleus RNA sequencing identifies stem-like populations in the adult human lens

Sneha Nair, LTMT-Junior Research Fellow

- Digital spatial transcriptomic profiling reveals molecular dysregulation of the human trabecular meshwork in glaucoma (Best Oral Presentation Award under Category II: Integrative OMICS Data Analysis)

Kanthimathi R., CSIR-Senior Research Fellow

- Spatial transcriptomic analysis confirms the presence of stem cells in the peripheral retinal pigment epithelium



Sneha Nair received best oral presentation award

National Pharmacy Education Day Celebration

KM College of Pharmacy, Uthangudi, Madurai, 6th March 2026

Dr. S. Senthilkumari has been invited as a Chief Guest and to deliver a talk in the National Pharmacy Education Day Celebration at KM College of Pharmacy, Uthangudi, Madurai on 6th March 2026. She has delivered a talk on the topic entitled, "My Journey (Research) from Undergrad Student to Independent Scientist" which was well received by the pharmacy students who were participated in the event.

84th Annual Conference of the All-India Ophthalmological Society (AIOC 2026)

Jaipur, 12-15 March 2026

Dr. P. Sundaresan, ICMR Emeritus Scientist participated and delivered two talks at the 84th Annual Conference of the All India Ophthalmological Society (AIOC 2026), held at the Jaipur Exhibition & Convention Centre, Jaipur.

Dr. S. Senthilkumari at KM College of Pharmacy





Dr. P. Sundaresan at AIOC 2026 conference, Jaipur

- Genetics of glaucoma
- Exploring mito-nuclear genetic factors in Leber's hereditary optic neuropathy

Scientific and Motivational Talk at the University of Gujarat

Ahmedabad, 16th March 2026

Dr. P. Sundaresan delivered a scientific and motivational talk to postgraduate students from the Departments of Human Genetics, Zoology, and Biomedical Technology at the University of Gujarat, Ahmedabad.

Popular Science Lecture

Kalaighar Centenary Library, Madurai, 20th March 2026

Dr. Gowri Priya Chidambaranathan, Scientist 4, Aravind Medical Research Foundation delivered the 32nd Popular Science Lecture on "Stem Cells in Regenerative Medicine", organized by The Tamil Nadu Science Forum, Madurai and the Kalaighar Centenary Library, Madurai on 20.03.2026.

Dr. C. Gowri Priya delivered a lecture at Kalaighar Centenary Library



Dr. P. Sundaresan delivered Engaging talk to children at Kalaighar Centenary Library

Engaging Talk to children

Kalaighar Centenary Library, Madurai, 22nd March 2026

Dr. P. Sundaresan, ICMR Emeritus Scientist delivered an engaging talk titled "Children! You will wake up to protect your eyes" under the program "Nool Arumbugal." The event was held at the Kalaighar Centenary Library on 22 March 2026.

CONFERENCES ORGANISED

Three-Day Workshop on "Extracellular Vesicles: Biology and Applications"

AMRF, 6-8 October 2025

As part of the October Summit 2025, a three-day workshop on "Extracellular Vesicles: Biology and Applications" was organised at AMRF. The event brought together 16 participants, including faculty members, Ph.D. scholars, and M.Sc. students representing 11 institutes from Tamil Nadu, Karnataka, and Telangana. The workshop provided extensive hands-on training on the isolation of EVs from plasma and commercial milk using ultracentrifugation. Participants also learned EV characterisation using Nanoparticle Tracking Analysis (NTA), protein isolation and profiling by SDS-PAGE, and marker evaluation through Western blotting. In addition, a demonstration session was held on mass spectrometry for EV proteomic analysis.

Guest lectures were delivered by Dr. Jitendra Shandilya, Application Specialist from Beckman Coulter, on flow cytometry, and Dr. Sebastian Raja, Application Specialist from Leica, on confocal microscopy. Participants also witnessed live demonstrations of how these technologies can be applied to EV research.

During the inaugural session, Prof. K. Dharmalingam highlighted the emerging importance



Prof. K. Dharmalingam delivered inaugural talk in workshop on "Extracellular Vesicles: Biology and Applications"

of EV research in diagnostics and therapeutics. Dr. Gowripriya spoke on the role of EVs in regenerative medicine; Dr. Bharanidharan, Scientist, discussed EV-derived microRNAs in host-pathogen interactions; and Dr. Daipayan Banerjee, Scientist, presented on the use of EVs as biomarkers in diabetic retinopathy.

The workshop concluded with a certificate distribution ceremony, where Prof. Dharmalingam commended the participants and encouraged them to pursue research in this rapidly evolving field.

Mini Symposium on Eye Health and Diabetes

AMRF, 30th March 2026

As part of the 50th Jubilee Year Celebration of the Aravind Eye Care System (AECS), the Aravind Medical Research Foundation (AMRF) organised a mini symposium on "Eye Health and Diabetes." This symposium was designed for Master's students in Life Sciences, Biotechnology, and Pharmaceutical Sciences, offering them an in-depth understanding of

the critical links between diabetes and eye disorders, along with current and emerging clinical perspectives, while emphasising the importance of preventing eye complications associated with diabetes.

The session began with an introductory note by Dr. S. Senthilkumari, Convenor, followed by the inaugural address by Prof. K. Dharmalingam, Director - Research, AMRF. The symposium featured invited talks that covered broad perspectives on diabetes and its associated ocular complications. Dr. Anand Annamalai, Endocrinologist, Ashwin Hospital, Madurai, presented on the fundamental understanding of diabetes. An overview of diabetes-associated complications such as glaucoma and retinal diseases was provided by clinicians Dr. R. Krishnadas, Senior Medical Consultant, Glaucoma Services, Aravind-Madurai, Dr. T.P. Vignesh, Medical Consultant, Retina-Vitreous Services, Aravind-Madurai, and Dr. S. Bhavani, Medical Consultant, Retina-Vitreous Services, Aravind-Madurai.

Participants of Mini symposium on Eye Health and Diabetes



The role of Artificial Intelligence in the diagnosis of diabetic retinopathy was highlighted by Dr. R. Kim, CMO, Aravind-Madurai. Dr. A. Vanniarajan, Scientist, Molecular Genetics, AMRF, Dr. D. Bharanidharan, Scientist, Microbiology, AMRF, and Dr. Daipayan Banerjee, Scientist, Proteomics, AMRF, spoke on the basic research contributions of AMRF, including the genetic and mechanistic understanding of glaucoma pathogenesis, the role of microRNA as a biomarker, and the potential of extracellular vesicles for early diagnosis of diabetic retinopathy.

The symposium was attended by postgraduate students from various institutions in and around Madurai, including Thiagarajar College of Arts and Science, Lady Doak College, and KM College of Pharmacy. There was also representation from Alagappa University, Karaikudi, and Kalasalingam University, Krishnankoil. Research scholars and faculty members from AMRF also participated in the event.

AWARDS & RECOGNITIONS

Appreciation certificate at the 83rd Annual Conference of AIOS - 2025

New Delhi, 3-6 April 2025

Dr. P. Sundaresan, ICMR Emeritus Scientist, received an appreciation certificate for Best Publication in the Indian Journal of Ophthalmology (IJO) for the year 2024. Title: Genotypic and Phenotypic diversity in X linked retinoschisis: Findings from a South Indian patient cohort- Indian J Ophthalmol 72(6): p 902-911, June 2024". Co-authors: Susmita Chowdhury,

Dr. P. Sundaresan received an appreciation certificate for best publication in the Indian Journal of Ophthalmology for the year 2024



Gopika S. Kumar received Young Investigator award

Prakash Chermakani, Girish Baliga, Rupa Anjanamurthy, Sundaresan P

Young Investigator Award for paper presentation 31st Annual meeting of ARVO-India 2025

Chennai, 26-27 July 2025

- Gopika S. Kumar, Project Associate, AMRF received a certificate for winning the Young Investigator Award for paper presentation. Title: *Pigmentation loss reflects EMT and functional decline in iPSC-derived RPE cells.*
- Dharsini Nandhakumar, Project Assistant II, AMRF received a certificate for winning the Young Investigator Award for paper presentation. Title: *Prospective Evaluation of RID-MyC Assay for Diagnosis and Clinical Management in Smear and Culture-negative Keratitis with IVCM Correlation*

Dharsini Nandhakumar received Young Investigator award



**Dr. Vidya Ravi Memorial Award for Women Scientist
at the National Conference on Advances in Burns
Treatment – Thermal and Radiation**

Trichy, 28th September 2025.

Dr. Gowri Priya Chidambaranathan was awarded the Dr. Vidya Ravi Memorial Award for Women Scientist for proving existence of stem cells in trabecular meshwork and its deficiency leading to glaucoma at the National Conference, held at Trichy.



Dr.C.Gowri Priya received Dr. Vidya Ravi Memorial Award for Women Scientist

GUEST LECTURE



14th November 2025

Dr. Mala Shanmugam, Ph.D, Associate Professor, & Co-leader, Translational Cancer Metabolism Initiative, Department of Hematology and Medical Oncology, Winship Cancer Institute of Emory University, Atlanta, GA 30322

Topic : “Metabolism in Cancer: From Mechanisms to Precision Medicine”



19th July 2025

Dr. Manmohan Bajaj, Product Manager, BIOGENE-INDIA, New Delhi

Topic: “BioLegend's LEGENDplex multiplex ELISA: Principle and applications”.



2nd February 2026

Dr. Arijit Mukhopadhyay, Professor of Precision Health, University of Salford, UK

Topic : “Glaucoma genetics and management”

Ph.D awarded by Alagappa University



P. Saranya

Department of Immunology &
Stem Cell Biology

Thesis: Characterisation of adult human lens epithelial stem cells in the maintenance of tissue homeostasis and their functional status in cataractous lens.

Guide: Dr. Gowri Priya
Chidambaranathan



A. Waseema

Department of Immunology &
Stem Cell Biology

Thesis: Identification, Characterization and Maintenance of Stem Cells in Adult Human Retinal Pigment Epithelium.

Guide: Dr. Gowri Priya
Chidambaranathan

Ph.D awarded by Madurai Kamaraj University



Susmita Chowdhury

Department of Genetics

Thesis: Molecular genetics of Juvenile X-linked Retinoschisis in South Indian population.

Guide: Dr. P. Sundaresan



Swathi Chadawalada

Department of Bioinformatics

Thesis: Identification of dysregulated microRNAs in ocular fluids as diagnostic markers for intraocular tuberculosis

Guide: Dr. D. Bharanidharan

Prof. VR. Muthukkaruppan Endowment Award 2025

Saraswathi K, Department of Molecular Genetics, won the Prof. VR. Muthukkaruppan Award for her outstanding research work on, "Molecular characterisation of ocular lymphoma for improved disease prognosis".

This Endowment Award, which includes a certificate and a cash prize, was established in 2014 by the students and colleagues of Prof. VR. Muthukkaruppan under an endowment created in his name. From this endowment fund, an annual award is presented to the institute's best researcher, selected on the basis of the scientific merit of their research abstracts and presentations.



RADHAKRISHNAN ISWARYA, SUBBIAH KRISHNADAS, KUPPAMUTHU DHARMALINGAM, CHIDAMBARANATHAN GOWRI PRIYA

- *"Human trabecular meshwork stem cell-derived small extracellular vesicles enhance trabecular meshwork cell survival and proliferation"*

Experimental Eye Research 253 (2025) 110281

MATHAN L, TEJASWI PRASAD, MOHAMMED HAMEED ASLAM, AADHITHIYA T. GR, BHARANIDHARAN DEVARAJAN, N. VENKATESH PRAJNA, K. DHARMALINGAM, DAIPAYAN BANERJEE

- *"Genome-wide methylation analysis unveils genes and pathways with altered methylation profiles in pterygium"*

Experimental Eye Research 2025

RATHINAVEL SETHU NAGARAJAN, USHA KIM, RADHAKRISHNAN SHANTHI, AYYASAMY VANNIARAJAN

- *"Investigation of GD2 synthase as a potential marker for retinoblastoma poor prognosis"*

Molecular Biology Reports (2025) 52:372

AADHITHIYA T. GR, S. BHAVANI, SAHANA KUPPURAJ, SAGNIK SEN, PRITHVIRAJ UDAYA, NARESH BABU KANNAN, KIM RAMASAMY, KUPPAMUTHU DHARMALINGAM, DAIPAYAN BANERJEE

- *"Retinol-binding protein 3 in vitreous and plasma-derived small extracellular vesicles is reduced in proliferative diabetic retinopathy"*

Experimental Eye Research 255 (2025) 110385

PREMKUMAR S, SHIVA SANKARI S, SANDHRA G, MANJU R. PILLAI, KRISHNADAS SR, SUNDARESAN P

- *"A novel 5bp deletion in HPS4 gene associates with Hermansky-Pudlak syndrome"*

Ophthalmic Genetics 2025 Oct;46(5):474-478

MOHAMED HAMEED ASLAM AR, VANNIARAJAN A, USHA KIM, BHARANIDHARAN D

- *"Analysing Differential Alternative Splicing events and their impact on Retinoblastoma progression using RNA-Seq Metadata"*

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- *"Mesenchymal stem cell-derived miRNAs: key immunomodulators to prevent ocular tissue degeneration"*

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- *"Folate receptor targeting amino modified Niobium Mxene for the delivery of Doxorubicin to cancer cells"*

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ONGOING RESEARCH PROJECTS

No	Projects	Funded by	Investigators	Research Scholar
ICMR - CENTRE FOR ADVANCED RESEARCH (CAR)				
1.	Early Detection of Open Angle Glaucoma in Diabetes Patients with Increasing Age and Family History of Glaucoma	ICMR 2025-2030	Dr. S.R. Krishnadas Dr. K. Dharmalingam Dr. A. Vanniarajan Dr. D. Bharanidharan	Dr. K. Lavanya Dr. R. Santhosh R. Kaviya T. Saravanakumar V. Ramya K. Petchimuthu T. Maheshwari
PROTEOMICS				
2.	Characterisation of the secretome and secretome - derived Extracellular Vesicles from <i>Aspergillus flavus</i> saprophyte and clinical isolates.	AMRF 2025-2026	Prof. K.Dharmalingam Dr. N. Venkatesh Prajna Dr. Lalitha Prajna	G. Hariharan K.S. Vedhavarshini
3.	Functional characterisation and identification of the cargo of <i>A.flavus</i> EVs	AMRF 2025-2026	Prof.K. Dharmalingam Dr. N.V. Prajna Dr. Lalitha Prajna	Dr. K. Lavanya
4.	Characterisation of Cell-Derived Nanovesicles from Human Corneal Epithelial Cells incubated with Fungal Extracellular Vesicles and Spores	AMRF 2025-2026	Prof. K.Dharmalingam Dr. N.V. Prajna Dr. Lalitha rajna	Karthik Alagarsamy Dr. K. Lavanya
5.	Evaluating the clinical potential of small extracellular vesicle-derived RBP3 (SEV-RBP3) in Diabetic Macular Edema: Inflammation modulation and Anti-VEGF response prediction	ICMR November 2025-October 2028	Dr. Daipayan Banerjee Dr. K. Naresh Babu Dr. R. Kim Prof.K. Dharmalingam	L. Mathan T. Anu Kiruthiga
6.	Identification of regulatory tear microRNAs during keratoconus progression and evaluation of affected microRNAs in keratoconus corneas by the application of a therapeutic cross-linker	ICMR 2026-2029	Dr. O.G.Ramprasad Dr. D. Bharanidharan Dr. Venkatesh Prajna	N.S. Makesh Raja Priya Lavanya S
7.	Comparative analysis of the outcome of large macular holes operated with Platelet Rich Plasma derived exosomes versus Platelet Rich Plasma	2024-2025	Dr. Naresh Babu K Prof.K. Dharmalingam Dr. Gowri Priya Chidambaranathan Dr. V. Muthukrishnan Dr. Athul S. Puthalath	
MICROBIOLOGY				
8.	Development and validation of a non-invasive point-of-care diagnostic tool for fungal keratitis	VELUX STIFTUNG 01.08.2021 – 31.07.2025	Dr. N. Venkatesh Prajna Dr. Lalitha Prajna Dr. K.Dharmalingam Dr. Thulasiraj Ravilla Dr. N. Siddharth	C.Niraja J.Deepa N.Dharsini C.Dharani G.Balaji M.Kanmani

9.	Ocular Macrophage Migration Inhibitory Factor-based stratification and prognostication of patients with vision threatening ocular inflammatory conditions	ICMR 05.02.2025 04.02.2028	Dr. D. Bharanidharan Dr. K. Dharmalingam Dr. N. Venkatesh Prajna Dr. Lalitha Prajna	Dhanu Prakash M Anusha Erulandi
10.	Attenuation of drug-resistance in Ocular Multidrug-Resistant Pseudomonas aeruginosa using human corneal miRNA cargo of Extracellular Vesicles	ICMR 2025-2028	Dr. D. Bharanidharan Dr. K. Dharmalingam Dr. N. Venkatesh Prajna Dr. Lalitha Prajna Dr. Lalan kumar Arya Dr. Rakhi Kusumesh Dr. Bibhuti P. Sinha	Hariharan Gnanam Sri Sanjana. S.V.
11.	Dysregulated human Corneal miRNAs and their role in disease progression	ICMR-SRF 2022-2025	Dr. D. Bharanidharan	Shreya Dinesh
12.	Epidemiological and Molecular Insights Into The Estrogen - Molecular Chaperone Axis In Cataract Prevalence and Progression In Menopausal Women	ICMR- Extramural Small grant 2025-2028	Dr. Siddharth Narendran Dr. Daipayan Banerjee Dr. Jaishree Pandian	Akshaya
13.	Retinal Disease Phenotyping in South India: The Aravind Inherited Eye Disease Registry	GKD – Charity trust 2023-2026	Dr. Siddharth Narendran	Jeyashree
14.	Multicentric Diagnostic and Therapeutic Evaluation of a Rapid Molecular Platform for Fungal Endophthalmitis and Metagenomic Sequencing to Characterise the Microbial Spectrum in Endophthalmitis	ICMR – Intermediate grant 2025-2028	Dr. Siddharth Narendran Dr. Kim Ramasamy Dr. Lalitha Prajna Dr. V.R. Saravanan Dr. Karthik Srinivasan Dr. Bharanidharan Devarajan Dr. Jaishree Pandian	Balaji Seethalakshmi Brindha Niveditha
MOLECULAR GENETICS				
15.	Investigation of nuclear genes involvement in a Mitochondrial Disorder: Leber's Hereditary Optic Neuropathy	SERB 18.08.2023 – 17.08.2026	Dr. P. Sundaresan Dr. S. Mahesh Kumar	-
16.	Decoding the Unknown Genetic Etiology to Ameliorate the Molecular Diagnosis of Leber's Congenital Amaurosis	DHR-GIA, ICMR 06.11.2023 – 05.11.2026	Dr. P. Sundaresan Dr. S. Senthilkumari Dr. Rupa Anjanamurthy	S. Shiva Sankari
17.	Mitochondrial Association with Transcriptomic Signatures for Trabecular Meshwork Damage in Glaucoma: A tissue-specific omics approach.	ICMR 2025-2027	Dr. P. Sundaresan Dr. S.R. Krishnadas Dr. Ann Mary Mathews	S. Hari Vignesh
18.	Molecular Genetics in Ocular diseases: Laneway for Personalised medicine especially Leber's Congenital Amaurosis (LCA) and Leber's Hereditary Optic Neuropathy (LHON)	ICMR 04-12-2024 – 13-05-2028	Dr. P. Sundaresan Emeritus Scientist	-

19.	Molecular characterisation of ocular lymphoma for improved disease prognosis	Lady Tata Memorial Trust 01.08.2020 31.07.2025	Dr. A. Vanniarajan	K. Saraswathi
20.	Elucidating the role of cancer stem cells in chemoresistant retinoblastoma and their therapeutic implications	ICMR-SRF 2022-2025	Dr. A. Vanniarajan	R. Sethu Nagarajan
21.	Functional repair of mutant RB1 by CRISPR-mediated gene editing in retinal tumour cells of bilateral RB patients	ICMR 2025-2028	Dr. A. Vanniarajan Dr. C. Gowri Priya Dr. V. Thirunavukkarasu & Dr. S. Selvakumar, Bharathiar University, Coimbatore	-
IMMUNOLOGY AND STEM CELL BIOLOGY				
22.	Identification and Characterisation of adult human retinal pigment epithelial stem cells	Part time PhD from 17.03.2022	Dr. Gowri Priya Chidambaranathan	A. Waseema
23.	Adult stem cell derived extracellular vesicular miRNAs for trabecular meshwork regeneration in glaucoma	UGC-SRF 03.06.2022– 02.06.2025	Dr. Gowri Priya Chidambaranathan Dr. S.R. Krishnadas Prof. VR. Muthukkaruppan	R. Iswarya
24.	Molecular regulators associated with the maintenance of human trabecular meshwork stem cells in relation to their reduction in ageing and glaucoma	LTMT –JRF Dec 2024 – Dec 2026	Dr. Gowri Priya Chidambaranathan	Sneha Nair
25.	Molecular characterisation of human retinal pigment epithelial stem cells and their role in age related macular degeneration	ICMR Dec 2022-Dec 2025 CSIR-SRF Dec 2025 Onwards	Dr. Gowri Priya Chidambaranathan Dr. D. Bharanidharan Dr. R. Kim Dr. Siddharth Narendran	R. Kanthimathi
26.	Molecular regulation of adult human lens epithelial stem cells: changes with aging and cataract	ICMR-JRF Oct 2023- Oct 2025 SRF Nov 2025 Onwards	Dr. Gowri Priya Chidambaranathan	P. Thushmitha
27.	Adult stem cell derived exosomes: a cell-free therapy for glaucoma.	ICMR-IG March 2025 – Feb 2028	Dr. Gowri Priya Chidambaranathan Prof.K.Dharmalingam Dr. S.R. Krishnadas Dr. Nirmal Jayabalan, Birla Institute of Technology and Sciences, Pilani, Hyderabad Campus	Dr.K. Balachander
28.	A scalable system for the production of human induced pluripotent stem cell derived functional retinal pigmented epithelial cells	SERB 04.10.2023 – 03.10.2026	Dr. P. Anwar Azad Dr. C. Gowri Priya	S. Gopika J. Srijan

OCULAR PHARMACOLOGY				
29.	Evaluation of IOP lowering property and Anti-fibrotic property of Relaxin on TGFβ2-induced Elevated IOP Ex vivo model of glaucoma using Human Organ Cultured Anterior Segment (HOCAS)	SERB-CRG grant (2023-2026)	Dr. S. Senthilkumari Dr. R. Sharmila	Lakshmi Devi Priya Mohanaejeyapriya
30.	'Relaxin' the pressure in glaucoma	Fight for Sight Ph.D. Studentship 2022 programme (UK) (Oct 2023-Oct 2026)	Prof. Colin Willoughby (UK) Dr. S. Senthilkumari Dr Kazuhiro Yamamoto (UK)	Ms. Eva Mihalovova
31.	Role of dopamine and serotonin levels in myopia development and progression	AEH, Madurai	Dr. Sahithya Dr. Elackiya Dr. P. Vijayalakshmi Dr. S. Senthilkumari Prof. K.Dharmalingam	
32.	Prospective Study Analysing the Role of Cytokine- Mediated IOP reduction in POAG /OHTN Patients Receiving Therapeutic Ultrasound for Glaucoma (TUG) Treatment	EyeSonix, USA	Dr. Sharmila Rajendrababu Dr. S.Senthilkumari Prof.K.Dharmalingam Dr. Donald Schwartz Mr. Eric Schultz	M. Fazil
BIOINFORMATICS				
33.	Expression profiling of human corneal miRNAs and their role in Pseudomonas aeruginosa induced keratitis	DBT-BET 2022-2027	Dr. D. Bharanidharan	R.Praveen kumar
34.	Identification and Analysis of Alternative Transcripts in Retinoblastoma Progression	ICMR-SRF 2022-2025	Dr. D. Bharanidharan	Mohd Hameed Aslam

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