

**In order of Importance, list of 10 best papers of the applicant highlighting the important discoveries/contributions described in them briefly (Max. 1 MB) \***

***Understanding the key mechanisms involved in ROP pathogenesis and their role in early disease risk prediction and monitoring the progression:***

Retinopathy of prematurity is major ocular complication of preterm birth especially in the Indian subcontinent. It is considered an oxygen induced retinopathy however it's not clear why it affects only a subset of preterm infants (20-50% preterm infants worldwide). We undertook a comprehensive genomics analysis of blood, vitreous humor and tear samples of preterm infant with ROP and No-ROP to identify the genetic pathways involved in disease pathogenesis. Our study identified aberrant activation of the alternate complement pathways and extracellular matrix remodelling under hypoxic stress (1). Further, we demonstrated that aberrant complement activated proteins and MMPs expression is significantly increased in the vitreous of ROP patients and this could also be reliably seen in tear samples and thus can be used as reliable tool for community-based screening and risk prediction among preterm born infants. (1,2). A central role of microglial cell in complement activation and secreting proinflammatory cytokines was also noted (1). Our next efforts demonstrated how activated microglia under hypoxia secretes MMP9 that degrades Opticin an anti-inflammatory protein in the retina and vitreous and this can be rescued by treatment with dexamethasone a corticosteroid (2). We further assess the role of lipid (polyunsaturated fatty acids) metabolism in ROP pathogenesis. Most of the enzymes involved in the PUFA metabolism showed a higher expression and activity in ROP vitreous except EPHX2 which was downregulated leading eventually to a higher activation of Notch signalling and abnormal angiogenesis in the retina (3).

1. Rath S, Jalai S, Patnaik SB, Shahulhameed S, Musada GR, Balakrishnan D, Rani PK, Kekunnaya R, Chhablani PP, Swain S, Giri L, Chakrabarti S, **Kaur I\***. Aberrant microglia-mediated complement activation and inflammation in the pathogenesis of retinopathy of prematurity. *Front Immunol* 2017; 8: 1868.
2. Patnaik, S., Rai, M., Jalali, S., Agarwal, K., Badakere, A., Puppala, L., Vishwakarma, S., Balakrishnan, D., Rani, P.K., Kekunnaya, R...& **Kaur I\***. 2021. An interplay of microglia and matrix metalloproteinase MMP9 under hypoxic stress regulates the opticin expression in retina. *Scientific reports*, 11(1), p.7444.
3. Kumar, S., Patnaik, S., Joshi, M. B., Sharma, N., Kaur, T., Jalali, S., Kekunnaya, R., Mahajan, A., Chakrabarti, S., & **Kaur I\***. (2024). Arachidonic acid metabolism regulates the development of retinopathy of prematurity among preterm infants. *Journal of Neurochemistry*, 00, 1–17. <https://doi.org/10.1111/jnc.16190>

***New methods developed:***

In these two publications, we have established new protocols for undertaking the metabolomics and lipidomics of vitreous humor (4, 5).

4. Kumar S, Joshi MB, **Kaur I\***. Protocol and Methods Applicable to Retinal Vascular Diseases. *Lipidomics: Methods and Protocols* 2023 Jan 19 (pp. 71-78). New York, NY: Springer US.

5. Patnaik S, Jalali S, Joshi MB, Satyamoorthy K, **Kaur I\***. Metabolomics applicable to retinal vascular diseases. *Metabolomics: Methods and Protocols*. 2019:325-31.

### ***From Bench to Bedside:***

Taking the finding from our studies (1,2 &3) for clinical translation, we next developed a nanosensor for a simultaneous estimation of complement C3, MMP 3 and 9 in the tear samples of preterm infants for an early disease risk prediction (6). Towards the development of more efficacious and cost effective treatment for retinal vascular diseases, we tested the potential of microglia under hyperglycemic(7) and hypoxic (8) stress by using electrospun fibres based meshes coated with Dexamethasone. Our study demonstrated the microglial cells grown on the elctrospun meshes adopt an anti-inflammatory phenotype and thus could be utilised for an efficient, simultaneous and sustained delivery of drugs (anti-VEGF and Corticosteroids) to combat inflammation and neovascularization as seen in ROP and diabetic retinopathy (7, 8).

6. Ghosh, T.N., Rotake, D., Kumar, S., **Kaur, I\***. and Singh, S.G., 2023. Tear-based MMP-9 detection: A rapid antigen test for ocular inflammatory disorders using vanadium disulfide nanowires assisted chemi-resistive biosensor. *Analytica Chimica Acta*, 1263, p.341281.
7. Venugopal, D., Vishwakarma, S., Sharma, N., **Kaur, I\***. and Samavedi, S., 2024. Evaluating the protective effects of dexamethasone and electrospun mesh combination on primary human mixed retinal cells under hyperglycemic stress. *International Journal of Pharmaceutics*, 651, p.123768.
8. Venugopal D, Vishwakarma S, **Kaur I\*** Samavedi S. Electrospun meshes intrinsically promote M2 polarization of microglia under hypoxia and offer protection from hypoxia-driven cell death. *Biomedical Materials*. 2021 Jun 29;16(4):045049.

### ***Exploring the role of Alternate complement pathway in retinal vascular conditions:***

Alternate complement pathways genes were found to be associated with ocular neovascularization in age-related macular degeneration in Caucasian cohort. We identified a significant association of these genes in Indian AMD patients (9). Since these genes are known involved in inflammation and neovascularization, we further demonstrated their functional involvement in the pathogenesis of diabetic retinopathy (10).

9. **Kaur I**, Hussain A, Hussain N, Das TP, Pathangay A, Mathai A, Hussain A, Nutheti R, Nirmalan PK, Chakrabarti S. Analysis of *CFH*, *TLR4* and *APOE* polymorphisms in India suggests the Tyr402His variant of *CFH* to be a global marker for age-related macular degeneration. *Invest Ophthalmol Vis Sci* 2006; 47: 3729-3735.
10. Shahulhameed S, Vishwakarma S, Chhablani J, Tyagi M, Pappuru RR, Jakati S, Chakrabarti S, **Kaur I\***. A Systematic Investigation on Complement Pathway Activation in Diabetic Retinopathy. *Front Immunol*. 2020;11:154.

*\*Corresponding author*

