Summary

Pseudoexfoliation (PEX) is a prevalent, age-related systemic disorder and a leading identifiable cause of secondary glaucoma, yet its underlying mechanisms remain poorly defined. Clinically, PEX progresses from pseudoexfoliation syndrome (PEXS) to pseudoexfoliation glaucoma (PEXG), a vision-threatening condition associated with elevated intraocular pressure and irreversible optic nerve damage. The marked variability in disease onset and progression, despite known genetic associations, underscores a critical gap in understanding how systemic and cellular factors converge to drive pathology. Addressing this gap is essential for improving early diagnosis and developing targeted therapeutic strategies.

This thesis advances the field by establishing a systems-level framework that integrates multi-level biomarker profiling with mechanistic insights into disease biology. Through the combined analysis of proteins, microRNAs, and metabolites, the study demonstrates that PEX is not driven by isolated molecular defects but by coordinated dysregulation across interconnected pathways. Protein profiling identifies activation of the ROCK2–TGF-β1–VIM axis, reflecting enhanced fibrotic signalling and cytoskeletal remodelling, while concurrent downregulation of miR-320a-3p and miR-223-3p indicates loss of post-transcriptional regulatory control over inflammatory and fibrotic responses.

Metabolomic profiling further reveals stage-specific alterations, with early lipid and mitochondrial dysfunction in PEXS and a shift toward oxidative stress and amino acid imbalance in PEXG. Central to these changes is disruption of one-carbon metabolism, characterized by elevated homocysteine (Hcy), reduced arginine (Arg), and decreased phosphatidylcholine. A core biomarker panel comprising Hcy, Arg, TGF-β1, and VIM robustly distinguishes disease states, with multi-marker models significantly outperforming individual biomarkers. These findings highlight the clinical potential of integrated biomarker strategies for early detection and disease stratification.

At a mechanistic level, the study identifies late-stage autophagy impairment and lysosomal dysfunction as pivotal events linking systemic metabolic imbalance to tissue pathology. Despite preserved autophagy initiation, defects in lysosomal biogenesis, endosomal trafficking, and proteolytic capacity disrupt autophagic flux, leading to intracellular aggregate accumulation and extracellular matrix deposition. Cytoskeletal disorganization further exacerbates these defects, reinforcing impaired cellular clearance.

A key contribution of this work is the identification of Hcy as a central upstream driver that functionally links metabolic stress to downstream signalling and proteostatic failure. Elevated Hcy disrupts redox balance and nitric oxide (NO) signalling while activating TGF-β/ROCK2 and mTORC1 pathways, resulting in TFEB inhibition and compromised lysosomal function. These alterations, compounded by miRNA dysregulation, establish a self-perpetuating pathogenic loop that sustains disease progression.

Importantly, this mechanistic framework is supported by in vitro evidence demonstrating that Hcy recapitulates core features of PEX pathology. Moreover, a dual therapeutic approach targeting both metabolic imbalance (vitamin B12) and signalling pathways (ROCK inhibition) shows synergistic potential in restoring cellular homeostasis.

This study provides a unifying and translationally relevant model of PEX pathogenesis, bridging the gap between biomarker discovery and disease mechanism. By identifying actionable molecular targets and robust diagnostic signatures, it lays the groundwork for biomarker-guided, mechanism-based interventions, with significant implications for early diagnosis, risk stratification, and therapeutic development in PEX.