The impact of RAGE/Diaph1 deletion on reducing retina and optic nerve damage in diabetes

In this project, I plan to determine the role of RAGE (receptor for advanced glycation end-products) and Diaph1 (protein diaphanous homolog 1) signaling in the development of eyesight impairment caused by retina and optic nerve deterioration in long term diabetes. Over the past few decades, diabetes gradually has become one the top non-communicable disorders that affected 476.0 million in 2017 and predicted to reach 570.9 million people in 2025. Of those one in three diabetic patients is predicted to develop eyesight impairments of varying degrees of severity over the course of the disease. A range of pathological factors such as microvascular and neuronal changes, enhanced local inflammation, cytoskeleton protein glycation, oxidative stress, and defective ciliary transport contribute to diabetes triggered vision impairment. Numerous evidence shows that both RAGE and Diaph1 are involved in diabetes driven neurovascular, structural and metabolic changes in cells and tissues most susceptible to hyperglycemia and glucose toxicity. Based on current knowledge, I propose an innovative, translation approach to studies of molecular mechanisms underlying the development of diabetic retina and optic nerve degeneration leading to vision impairment, combing most recent research evidence on RAGE-Diaph1 signaling with my research experience, preliminary data and my own earlier work on the role of RAGE in neurodegeneration in animal models and patients with neurodegenerative diseases.

RAGE is a signal transduction receptor, whose activation triggers an increase in proinflammatory molecules, oxidative stressors and cytokines. RAGE was first identified as a cell surface receptor for Advanced Glycation End-products (AGEs) formed during nonenzymatic glycation and oxidation of proteins/ lipids that accumulate during physiological aging, but also in diabetes, inflammatory and neurodegenerative disease. Diaph1 belongs to the Rho-GTPase formins, known for its regulatory role in structural modification of actin, microtubulin and related cytoskeleton proteins, thus affecting cellular morphology, motility and secretion. Diaph1 is mainly involved in regulation of cytoskeleton machinery, binding to major proteins of cellular skeleton. In recent years it was discovered that RAGE and Diaph1 work together, with RAGE binding to Diaph1 via its cytosolic tail. RAGE belongs to a group of pattern recognition receptors, and, similar to Diaph1, it is also a part of RhoA signaling cascade. Together with Diaph1 it has been found to play a primary role in the pathogenesis of diabetes and its related complications.

I hypothesize that a double, RAGE-Diaph1, gene deletion, will slow down the progression of eyesight deterioration by effectively blocking the cascade of RAGE-Diaph1 signaling driven metabolic changes in diabetes. In order to test this hypothesis, I will employ a translational approach and examine samples from both diabetic patients and wildtype and RAGE-Diaph1 knockout animals with pharmacologically induced diabetes. In the first phase of the project, I will study in vivo the effects of diabetes on retina and optic nerve degeneration, vascular proliferation and inflammation by ophthalmological evaluation followed by ex vivo comparative analysis of protein and gene expression of RAGE, Diaph1 and their signaling partners in blood and tear samples both in patients and experimental animals at specified time points over the course of the disease. In the second phase, I will test my hypothesis by pharmaceutical blockade of RAGE-Diaph1 signaling in commercially available immortalized genotypically and phenotypically normal human retinal cell cultures grown in normo- and hyperglycemic environment. Finally, as a third step I plan to validate obtained results by treating diabetic wildtype mice with RAGE-Diaph1 inhibitor and assess the efficiency of such treatment as compared to RAGE-Diaph1 genetic knockout. If my hypothesis proves correct, obtained results will provide a background for further translation, pre-clinical and clinical studies of RAGE-Diaph1 signaling in prevention and treatment of diabetes triggered retina and optic nerve damage.

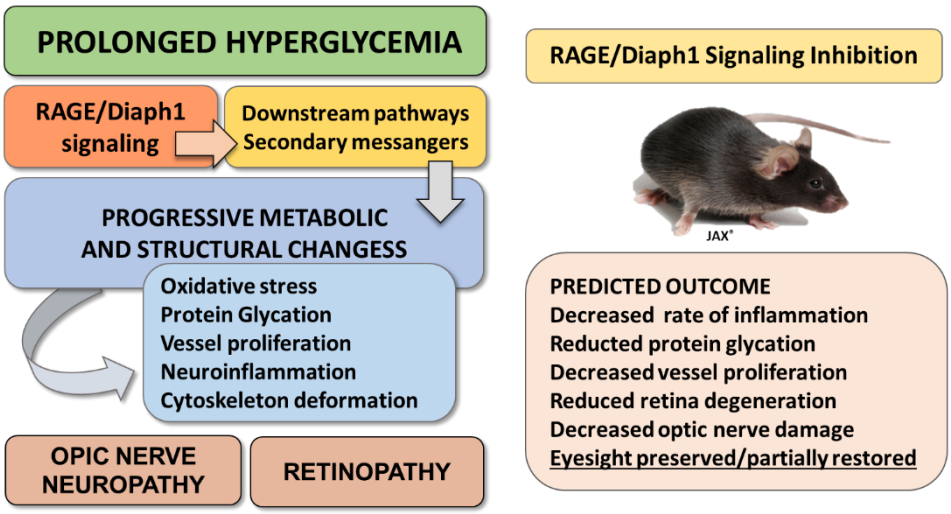

Fig 1. Illustration of hypothesis. RAGE-Diaph1 interactions play a prominent role in multiple downstream metabolic pathways involved in hyperglycemia triggered molecular and structural changes leading to retina and optic nerve damage, that if not treated, result in vision impairment and ultimately vision loss; we predict that blocking RAGE-Diaph1 signaling will halt or at least slow down the progression of eye pathological changes preserving the eyesight and preventing from vision loss.