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AAV Delivery of Genes for Secreted Cell Penetrating Proteins to Treat Retinal Inflammation

Cristhian J. Ildefonso[1], Virginia White[2], Qiuhong Li[1], and **Alfred S. Lewin**[2]

Departments of Ophthalmology[1] and Molecular Genetics and Microbiology[2], University of Florida, Gainesville, FL, USA

The eye has been shown to be an excellent place to develop viral mediated gene therapy. The retina is a multilamelar structure, and access its deeper layers typically involves a subretinal injection that may permanently injure a diseased retina. Our objective has been to use adeno associated virus (AAV) delivered to the vitreous chamber of the eye to produce secreted, cell penetrating peptides that gain access to the entire retina. Chronic inflammation is a contributing factor or a cause of retinal diseases such as age related macular degeneration and uveitis. Cell types contributing to the production of inflammatory cytokines include the retinal pigment epithelium (RPE), microglia and Müller glia, which are not directly exposed to the vitreous humor. Using an AAV2 capsid with three tyrosine to phenylalanine and one threonine to valine modifications, we have produced a vector in which anti-inflammatory proteins are coupled with a cell penetrating peptide and expressed as a fusion with a secreted protein. Among these, we have tested a cell penetrating version of M013 protein of myxoma virus. M013 inhibits inflammation by (1) interfering with the nuclear transport of Nf-κB and (2) by binding to and inhibiting the ASC protein of the NLRP3 inflammasome. Thus, both the synthesis of pro-IL-1β and its processing to mature IL-1β are blocked. We tested this vector in a model of acute inflammation (endotoxin induced uveitis) and in a model of chronic RPE oxidative stress (RPE-specific ablation of Mn superoxide dismutase). In the uveitis model, AAV-TatM013 reduced production of IL-1β and the accumulation of inflammatory infiltrates. In the chronic model, reducing inflammation protected photoreceptors and preserved retinal function over several months. We believe that this approach has the potential provide treatment for recurrent uveitis and for age related macular degeneration.