

## **GOLD NANOPARTICLES (GNP) OR ADENO-ASSOCIATED VIRUS (AAV) MEDIATED GENE DELIVERY IN HUMAN CORNEAL ENDOTHELIAL CELLS**

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**Purpose:** Healthy Corneal endothelium is essential for corneal clarity. Treatment of defective corneal endothelial function often requires corneal transplant. Gene therapy is an attractive approach to treat/cure corneal endothelial diseases. We evaluated gene transfer efficacy of AAV and GNP vectors for delivering therapeutic genes into human corneal endothelial cells.

**Methods:** Cultures of HPV16-E6/E7 transformed human corneal endothelial cells were incubated with AAV2/6, AAV2/8, AAV2/9 or GNP vector expressing reporter gene for 6 hours. Transfection efficiency was quantified with cytochemical or immuno staining and real-time polymerase chain reactions techniques. Commercial kits were used to prepare RNA and cDNA. Cellular viability was tested using Trypan blue assay.

**Results:** Immunocytochemical quantification of GFP-positive cells in cultures revealed 30% transgene delivery into endothelial cells with gold nanoparticles. The three tested AAV serotypes showed up to 2% endothelial transduction. The AAV6 was found most efficacious among 3 AAVs. These findings were complimented by the Real-time PCR data. AAV vectors did not alter cellular viability whereas notable decrease in cellular viability was noted with GNP vector.

**Conclusions:** AAV6 and GNP vectors are efficient at introducing therapeutic genes into human corneal endothelial cells. More studies are required to define dose and safety of tested vectors for corneal endothelial gene therapy.

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