Therapeutic Potential of Extracellular Vesicles Derived from Human Placenta for the Treatment of Corneal Transplant Graft Rejection and Corneal Injuries
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BACKGROUND

In response to trauma, infection or chemical exposure, the corneal immune tolerance breaks down and the cornea undergoes neovascularization and scarring.

There is a lack of therapies to treat corneal neovascularization and scarring. Corneal transplant can restore vision in these eyes, but comes with the risk of transplant rejection.

Human leukocyte antigen G (HLA-G) is a molecule expressed in the cornea and involved in maintaining the cornea’s immune tolerance. HLA-G is also highly expressed in the placenta, and secreted in placental extracellular vesicles (EVs).

HLA-G gene therapy to the cornea has been shown to reduce fibrosis and neovascularization following chemical injury to rabbit corneas.¹

OBJECTIVES

- Isolate HLA-G+ EVs from placental tissues and cells
- Study the therapeutic potential of placenta tissue and cell-derived HLA-G+ EVs to
  - 1) reduce corneal transplant rejection
  - 2) reduce corneal scarring and neovascularization after chemical injury

METHODS

Sources of HLA-G:

- Conditioned media from placental explants
- Conditioned media from HTR-8 (immortalized trophoblast) cells

Enriching for HLA-G+ EVs:

Aqueous two-phase separation (ATPS):

1. A mixture of dextran (DEX) and polyethylene glycol (PEG) is added to the conditioned media
2. The mixture is centrifuged at 200 g for 15 minutes, which allows for separation into two phases (DEX phase and PEG phase) by density

RESULTS

HLA-G+ EVs are detected in the conditioned media from placental explants and HTR-8 cells using nanoscale flow cytometry (nFC)

Figure 1: HLA-G+ EVs from placentas at different gestational ages

Figure 2: HLA-G+ EVs from HTR-8 cells under various oxygen conditions

Figure 3: HLA-G+ EVs from placental explants are enriched 5.9X following ATPS

Figure 4: nFC shows HLA-G+ EV populations before (50 µL loaded) and after (5 µL loaded) ATPS.

CONCLUSION

- HLA-G is involved in the immune tolerance of the cornea and placenta
- We have detected HLA-G+ EVs in the conditioned media from placental villous explants and a trophoblast cell line, and have enriched for HLA-G+ EVs
- Our next steps involve evaluating corneal epithelial cell viability, fibrosis and neovascularization with topically applied HLA-G+ EVs

REFERENCES