Title: In vivo evaluation of injected and bioprinted hyaluronic acid-based bioink in corneal stromal pocket

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Abstract
The corneal stroma contained specialized stromal keratocytes (CSKs) that preserved corneal transparency and homogeneity. Stromal scarring and opacities are major causes of vision loss worldwide, and while corneal transplantation remains the gold standard, donor shortages limit its availability. Cell-based therapies using stromal cells are promising but remain donor-dependent. Human adipose tissue–derived stem cells (hASCs) provide an abundant alternative, with the capacity to differentiate into CSK-like cells (hASC-CSKs). A three-dimensional (3D) tissue matrix was essential for mimicking native tissue and supporting stromal regeneration. Hyaluronic acid (HA)-based matrices emerged as promising stromal substitutes. In this study, we aimed to investigate the biocompatibility of HA-based bioink *in vivo*, both as injectable formulations and bioprinted constructs containing hASC-CSKs. *In vitro*, bioprinted HA constructs exhibited high cell viability, organized structure, and maintained transparency. *In vivo*, the bioink integrated progressively into the corneal stroma, considerably reducing stromal thickness in two weeks. Transplanted cells retained CSK phenotype, evident by lumican expression. Although inflammatory responses were observed, the bioink shielded transplanted cells from immediate immune response, promoting graft survival and integration. Moreover, the transplanted eyes showed improved corneal clarity and reduced inflammatory responses in two weeks. These findings demonstrated that HA-based bioink serves as a biocompatible scaffold for cell delivery, supporting stromal regeneration and highlighting its potential for future corneal therapies.