

Decellularized retinal matrix based biomaterials as novel cell delivery platforms

Joydip Kundu¹, Andrew Michaelson¹, Petr Baranov², Michael J. Young² and Rebecca L. Carrier¹

¹Chemical Engineering, Northeastern University, 360 Huntington Avenue, Boston, MA 02115

²Schepens Eye Research Institute, an affiliate of Harvard Medical School, Boston, MA 02114

Introduction: Retinal degenerative diseases impact the lives of millions of people worldwide. Photoreceptor cell transplantation holds great promise for management of vision loss¹. Cell based retinal regeneration is limited due to high cell death and low integration². Subretinal implantation of hRPC (human retinal progenitor cell) on scaffolds improved cell survival rate, but integration still is very low (~1%)³. This motivates the development of the material that is permissive to human retinal progenitor cell (hRPC) survival, differentiation, and integration within the host retina to restore vision. Extracellular matrix (ECM) can be used to develop naturally occurring scaffold materials, which provide cues for *in vivo* cell migration, differentiation and integration within host tissue. The decellularized retina (decell-retina) mimics the environment of the retina, and thus we hypothesize it will promote (hRPC) migration, differentiation and integration into host retina. In this study, we explored the prospects of decell-retina based substrates for delivery of hRPCs to the subretinal space.

Methods: Retinas were isolated from bovine eyes, decellularized with 1% sodium dodecyl sulfate (SDS), and purified using dialysis. The decell-retina was pepsin digested and assayed for its biochemical composition (collagen, host DNA, hyaluronic acid, sulfated GAGs) and growth factors. Decell-retina solutions were cast dried to develop thin films and copolymerized with collagen at 37°C to develop thermoresponsive hybrid gels. The films were characterized for their surface topology and hybrid gels were studied for their gelation kinetics. hRPCs were seeded onto decell-retina films to determine cell viability, morphology and proliferation using live-dead assay, F-actin staining and CyQUANT cell proliferation assay, respectively.

Results: Biochemical analysis of decell-retina compared to native retina indicated the bulk of cellular material was removed, while the majority of collagen, hyaluronic acid, and sulfated GAGs were retained after the decellularization process (Fig. 1). Growth factor (basic fibroblastic growth factor (b-FGF), epidermal growth factor (EGF) and nerve growth factor (NGF)) analysis of decell-retina relative to native retina indicated high retention even after the decellularization process (Fig. 2). The decell-retina films are

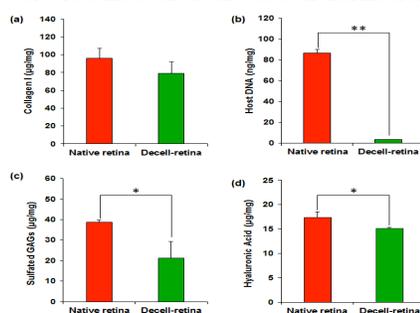


Fig 1: The biochemical composition of the native and decell-retina isolated from bovine eyes.

pealable, clear, easy to handle and thus amenable to subretinal surgical implantation. Thermosensitive hybrid gels with varying ratios of decell-retina and collagen were developed, and their gelation kinetics depend on the varying amount of retina

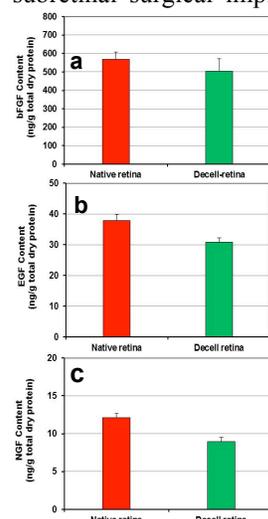


Fig 2: Growth factor assay of native and decell-retina.

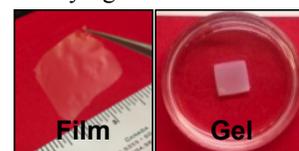


Fig 3: Decell-retina matrix based substrates.

ECM, with the gelation time ranging from 10-15 mins.

Cultured hRPCs demonstrated good survival rate and well-defined morphology (Fig. 4) on decell-retinal matrices. The hRPCs appeared to align with structural features in decell-

retina. The hRPCs were found to proliferate (50% increase) on the decell-retina matrices from day 1 to day 7 of culture.

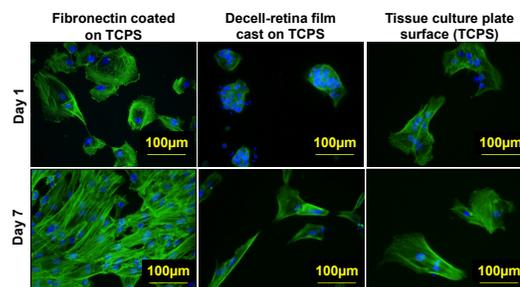


Fig 4: Cell morphology on decell-retina film after 1 and 7 days of culture.

Conclusions: Decell-retina provides a novel biomimetic material supporting attachment, viability and proliferation of hRPCs. These results corroborate the investigation of decell-retina as a prospective novel cell delivery platform to treat retinal degenerative diseases.

References:

- Maclaren et al., Nature 2006, 444, 203-207.
- Klassen et al., Invest Ophthalmol Vis Sci, 2004, 45, 4167-73.
- Yao et al., Polymers 2011, 3, 899-914.

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