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In vitro retinal neuron differentiation by a two-step approach from peripheral blood mononuclear cell derived monocytes

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The retina is one of the most complex tissues with high metabolic activity. Gradual loss of photoreceptors or other retina related cells is the leading cause of retinal degeneration leading to severe visual impairment. The adult retina lacks stem cells and the cells lost are never regenerated. Thus, it is desirable to have an *in vitro* system of reprogramming pluripotent cells into retinal cells that may be further utilized for repopulating the retina with the required cells to subside the level of degeneration. Peripheral blood is an easy and accessible source of adult stem cells. Subsequent studies show that PBMCs have been successfully used to generate multitude of cell lineages. The ethical concerns, mitogenic risks and the difficult isolation procedure involved in ESC, iPSC and BMC respectively make them unsuitable for their use in regenerative medicine. In the present study, PBMC derived monocytic cells are induced with plasticity and reprogrammed into retinal neuron like cells. The cells are primed by treating with a cocktail of growth factors in a low serum environment. These primed cells acquire properties of stem cells like proliferation. Further incubation with a re-differentiation cocktail that includes an array of growth factors like EGF, b-FGF, B27 supplement, SCF, IGF, taurine and retinoic acid re-differentiates them into retinal neuron like cells exhibiting morphological, phenotypic and functional resemblance to retinal cells suggesting a lineage shift of these cells.

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Assessment of adhesion response to 3D printed materials for ophthalmic device development

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Introduction: Glaucoma is the leading cause of irreversible visual impairment worldwide. Glaucoma surgical devices fail due to a scarring response results in fibrous encapsulation surrounding the device to prevent aqueous humour drainage. 3D printing technology has the potential to develop personalized ophthalmic devices or organs with improved cost effectiveness and productivity. Limited experimental data exists as to the biocompatibility response of 3D printed photopolymers.

Aim: Aim of this study is to perform cell adhesion and protein adsorption studies of 3D printed photopolymers compared to materials used in current ophthalmic devices [Silicone, Polytetrafluoroethylene (PTFE) and Poly (methyl methacrylate) (PMMA)] to assess 3D printed materials as a potential route for ophthalmic device development.

Methods: 3D printed materials (n=6) were developed using a high-resolution, desktop stereolithography (SLA), 3D printer and compared to materials used in current ophthalmic devices. Protein adsorption was quantified using a micro bicinchoninic acid (Micro BCA) assay and fluorescein-conjugated bovine serum albumin (FITC-BSA) adsorption. Cell adhesion (monocytes, fibroblasts) was assessed using Alamar Blue, CyQUANT and Live/Dead assays. Data were compared using a two-tailed unpaired t-Test.

Results: 3D printed materials demonstrated low cell adhesion and protein adsorption. Results were similar to those found with materials used in current ophthalmic devices ($P>0.05$). However, it was noted that 3D printed materials demonstrated increased cytotoxicity ($P<0.05$).

Conclusion: 3D printed photopolymer materials demonstrated a similar biocompatibility response to currently used materials and may allow for the development of customisable ophthalmic devices or organs. Subsequent testing will determine the adhesion response to 3D printed materials containing anti-scarring agents.

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