Capture and Release of Endothelial Progenitor Cells in a Microfluidic Channel

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Over 400,000 coronary bypass surgeries are performed every year in the United States annually. Endothelial progenitor cells (EPCs), which are naturally present in circulating blood, are an especially interesting cell type because they have the ability to repair damaged blood vessels. EPCs have been utilized as precursors in the in vitro cultivation of vascular grafts [1]. The conventional technique of isolating EPCs involves centrifugation followed by pre-plating. As tissue engineering and cell-based therapeutics begin the transition from the laboratory to clinical applications, the availability of robust and simple cell isolation techniques becomes significant. Microfluidic devices have recently been recognized as effective tools for separation. These devices can be fabricated using soft lithography techniques. This enables devices to be produced en masse in a cost effective and straightforward nature. The use of antibody coated channels for cell capture in microfluidic devices has recently been applied to several applications.

The principal goal of the present investigation is to create microfluidic cell separations systems to rapidly and efficiently isolate or enrich key cell types and use them for tissue engineering applications. In tissue engineering functional cell types must be enriched prior to seeding onto scaffolds. In cell-based approaches to tissue repair and regeneration stem and progenitor cells present in certain types must be isolated and characterized prior to use. The adhesion of cells to a functionalized surface is the basis for this type of separation. By virtue of their micro scale geometries microfluidic devices have large surface area to volume ratios; this characteristic makes them particularly suitable for adhesion based separation processes. Highly specific cell-ligand interactions have been identified and can be utilized to enact cell separations for specific subpopulations.

The ability to release captured cells has been a challenge [2]. Recently alginate has been shown to be an effective means of release [3]. Alginate is useful due to the ability to physically cross link the gel with divalent cations such as calcium, the gel can then be dissolved by use of a chelating agent such as ethylenediaminetetraacetic acid (EDTA). Selectivity and efficiency can be improved by incorporation of 4-arm amine terminated poly(ethylene glycol) (PEG) molecules into the alginate hydrogel. The gel can be further improved by optimizing the mixing parameters. Results show that alginate hydrogels conjugated with 4-arm PEG molecules is effective in achieving highly selective capture of rare cell populations (see Figure 1a-b). The dissolution of the gel by introduction of a chelating agent results in a facile release mechanism for purified cell populations. Applying these gels as surface coatings within a microfluidic channel provides the ability to isolate EPCs from untreated whole blood in a single pass. These cell populations retain viability as well as ability to proliferate in culture as shown in Figure 2. This technique for rapid isolation of viable cells can enable the use of such cells for vascular grafts and cell based treatments.

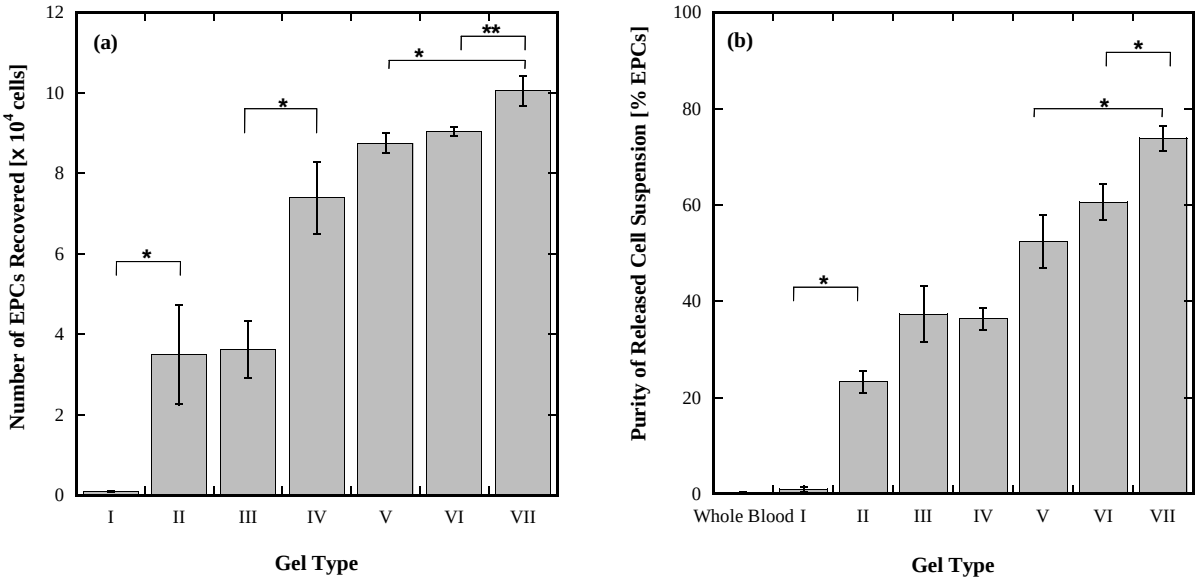

Figure 1. Yield (a) and purity (b) of EPCs captured from whole blood within microfluidic devices coated with PEG- and antibody-functionalized hydrogels.

Figure 2. (a) Released cells at day 2 and (b) day 10. The cells are adhering to the substrate and dividing to demonstrate the ability of released cells to proliferate.

References:

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