Numerical Modeling and Cell Adhesion of a Tissue Engineered Loop of Henle Device

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Statement of Purpose: Current renal tissue engineering designs promise to restore renal function(1). However, low concentrations of solutes in the filtrate stream require devices to use large volumes of filtrate, thereby limiting device miniaturization and consequent implantation or portability. Here, modeling of a tissue engineered loop of Heinle (LOH) device shows the potential to significantly concentrate waste solutes in the filtrate stream. The microfabricated device design enables incorporation of a membrane in conjunction with a cellularized biomaterial layer in a well-controlled microfabricated device in order to mimic LOH functionality. Device material selection and surface treatments indicate potential cellularization with HEK293 kidney cells.

Methods: The numerical model represented the basic geometries of a LOH device incorporated into a synthetic nephron with a rectilinear mesh. Figure 1 shows the cross-section and the rectilinear mesh approximation of the LOH device section of the nephron. The LOH device consisted of an extracellular fluid channel coupled to a blood and a filtrate channel via a membrane and cellularized layer. Device fabrication will employ micro-molding, enabling the use of polymeric biomaterials. The model calculated the fluid and solute motion in two separate steps. Volumetric flux was calculated using hydrostatic and osmotic pressure-driven flow in a rectangular channel. Darcy membrane permeability governed mesh point interaction for membrane-connected mesh points. After volumetric flux calculation, solute motion was calculated incorporating diffusivity and mixing effects due to blood cells. Four solutes were considered: albumin, urea, Na^+, and Cl^-. Inlet blood flow rate to the entry of the device at distance 0 m was set at 0.025 ml/hr, the flow rate to a single nephron based on a kidney comprised of 1.2 million nephrons receiving a total flow rate of 0.5 L/min (2). The concentration inlet conditions were assumed to have concentrations typical of normal blood, and conditions at the inlet to the LOH section were based on simulations of the upstream components.

The cell adhesion assay characterized cell adhesion to the device biomaterials, thereby indicating potential cellularization of the device. Human kidney cells from the HEK293 cell line were cultured in proliferative conditions on 4 substrate types: uncoated and coated polydimethylsiloxane (PDMS) substrates and polystyrene sheets. Cells were imaged via brightfield microscopy and adherence, expansion, and morphology were assessed at 24 and 72 hours after seeding.

Results: The device design model indicated significant concentration of urea in the filtrate stream for the given levels of flow rate and solute concentration. Figure 1 shows the results of the numerical modeling of pressure,

flow rate, and urea concentration along the distance of the microfluidic channel within the device. Geometries were iteratively adjusted such that filtrate urea concentration was 200-400 mM. Urea concentration increased sharply as the filtrate flowed into the collecting duct, reabsorbed fluid and exchanged NaCl for urea.

The treatment of both PDMS and polystyrene with an adsorbed collagen coating resulted in an increase in adherent cell density. Cell surface coverage appeared to reach a confluent level at seeding densities of 1.8 – 2.4 x 10^5 cells/mL for collagen coated substrates. Cellular morphology was rounded on uncoated PDMS substrates, and well spread on all other substrates.
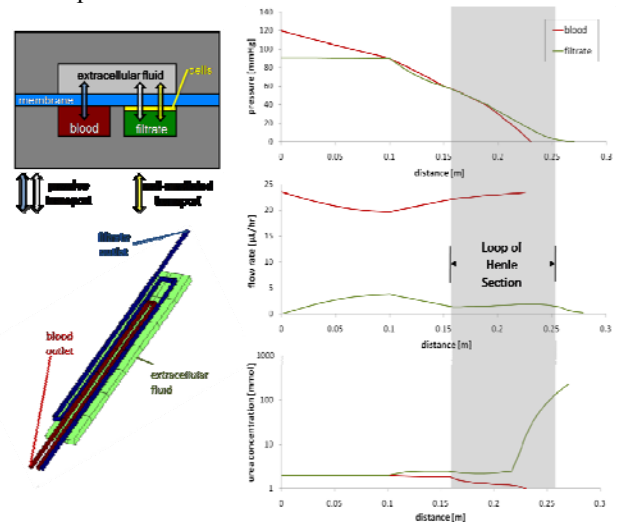

Figure 1. The LOH device cross section (upper left) shows passive transport through a membrane and active transport via a cell layer. The rectilinear mesh model (lower left) provided numerical simulations indicating appropriate pressure, flow rate, and urea concentration within the device (right).

Conclusions: Numerical modeling of a MEMS-based synthetic LOH device indicates a biologically appropriate concentration of solutes in the filtrate stream due to fluid and solute motion. Both membrane properties and cellular effects contribute to the design parameters and resulting simulated performance. Polystyrene and PDMS treated with an adsorbed collagen layer permit cell adhesion, justifying their use as appropriate biomaterials for manufacture of the LOH device. The characterized design and biomaterial selection presented here enable a viable tissue engineered LOH device and consequent renal tissue engineering technologies.

References:
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