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Hyaluronic acid based hydrogel droplets: A potential injectable cell culture scaffold

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Introduction

Cell culture scaffolds such as hydrogels give support and structure for cultured cells in 3D environments that better mimic *in vivo* conditions [1]. Hyaluronic acid (HA) derived hydrogels are particularly attractive scaffold materials, due to their high water content, and its high presence in the extracellular matrix of a multitude of tissues in the human body [2]. Adequate diffusion of oxygen and nutrients however, is generally limited to a depth of 200 μm in bulk hydrogels [3], heavily limiting their applicability to relatively large size constructs. We propose the use of droplet-based microfluidics to produce monodisperse HA-derived injectable microgel droplets which could enable the diffusion of nutrients and metabolites, while maintaining a size in which encapsulating sufficient cells to allow cell-cell interactions and proliferation would be possible.

Experimental results

Hyaluronic acid acrylamide (HA-am) was synthesized by partially modifying high molecular weight sodium hyaluronan with a N-(2-aminoethyl)acrylamide linker. Degree of modification was confirmed by NMR to be of 20%. HA-am bulk hydrogels were formed by exposing a solution of HA-am and photoinitiator Irgacure 2959 (0.4 % w/v) to a UV light source of 365 nm wavelength. Gel droplets were produced in a PDMS microfluidic device designed in a flow focusing geometry. In order to simulate cell encapsulation in the microgel, hydrogel precursor mixtures were prepared as for bulk hydrogels with the addition of polystyrene beads (10μm in diameter) at a concentration of 10 million beads ml^{-1}. For the oil phase, a fluorinated oil (Novec 7500, 3M) with 0.5% surfactant (PicoSurf 1) was used. The flow rates for the oil phase and aqueous phase were adjusted to 15 and 5 μl min^{-1}, respectively to produce highly monodisperse droplets of 151 μm in average diameter. Collected droplets were polymerized by exposing to UV light, washed and transferred to an aqueous solution.

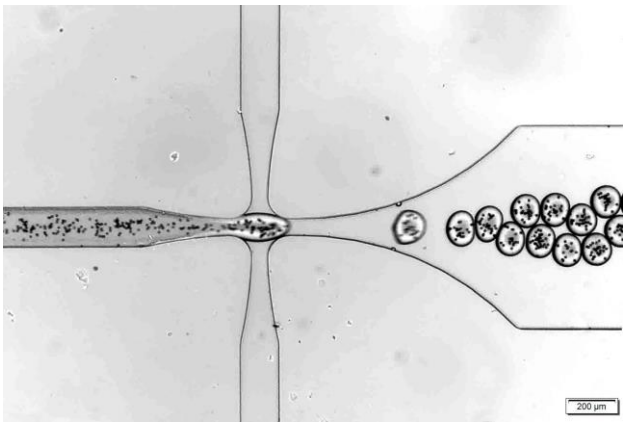

Figure 1: Microfluidic production of HA-am droplets with 10 μm sized polystyrene beads with flows of 5 μl min^{-1} on the aqueous phase and 15 μl min^{-1} of the fluorinated oil phase per each of the 2 inlets. Scale bars correspond to 200 μm.

Conclusion

Highly monodisperse microgels containing microbeads were obtained. We demonstrate that photocrosslinkable hydrogel droplets can be produced from HA-am in a microfluidic flow-focusing chip which could enable the encapsulation of cells and the use of the droplets as injectable cell culture scaffolds.

References

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