Developing Biomaterial Systems to Enhance Cell Engraftment

Krishna Patel, Adam Dundas, I Ning Lee, Ricky Wildman, Derek Irvine, and Lisa White
School of Pharmacy, Biodiscovery Institute and Centre for Additive Manufacturing, University of Nottingham

1. Introduction

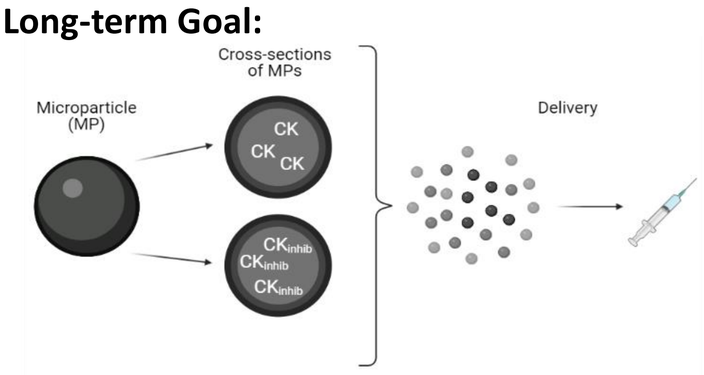
Currently, islet transplantation is used as the treatment of type 1 diabetes mellitus when patients struggle to maintain glycaemic control despite insulin therapy.

The Problem:

Approximately 60-70% of islets fail to engraft. This is thought to be due to:

1. Poor revascularisation
2. The instant blood-mediated inflammatory reaction

Long-term Goal:



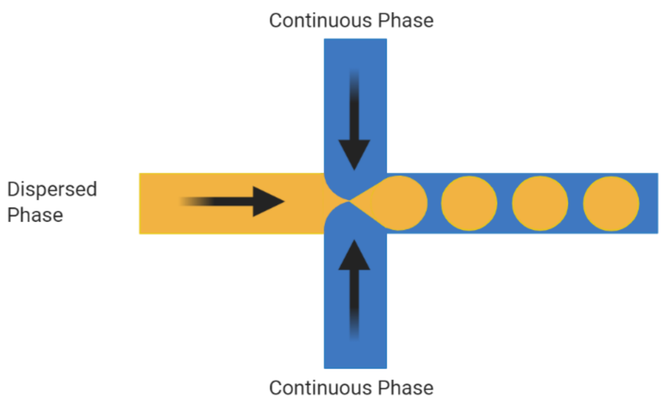
Deliver a localised and controlled release dosage of cytokines including growth factors, or cytokine inhibitors within polymeric microparticles to aid cell engraftment at the hepatic site.

Microparticles can be fabricated using a variety of methods. In particular, microfluidics uses active forces such as flow-focusing and T-junction systems which offers greater control in droplet manipulation.

2. Methods

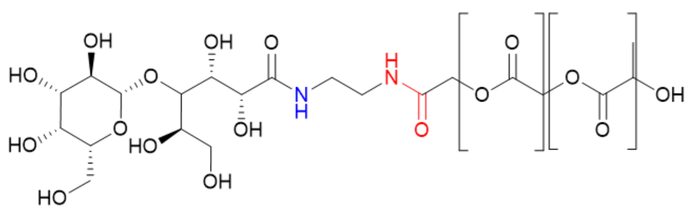
All experiments:

- Used a 100µm 3D flow-focusing hydrophilic chip
- Aimed to form an oil-in-water emulsion
- Used flow rates of 4ml/hr for the continuous phase and 0.15ml/hr for the dispersed phase



Materials

- **Solvent: Dichloromethane (DCM)** Mw 85Da, used for all experiments
- **Surfactant: Poly(vinyl alcohol-co-vinyl acetate) (PVA)** 88% hydrolysed, Mw 25kDa, used for all experiment at varying concentrations
- **Poly(D,L-Lactide-co-Glycolide) (PLGA)** 50:50, Mw 61kDa
- **Gal-PLGA:** Synthesised using D-(+)-Galactose Mw 180Da, and PLGA



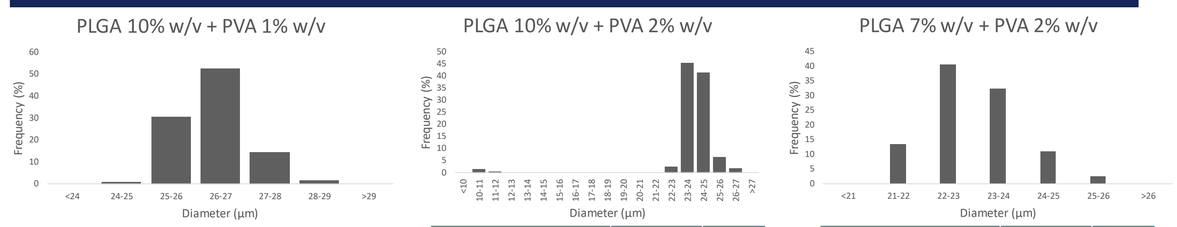
Chemical structure of Gal-PLGA

Galactose binds to the ASGP receptor on hepatocytes[1], therefore, can serve as an hepatocyte-targeting material for microparticle fabrication.

3. Results

Microparticles can be fabricated using 10% PLGA as the material and 2% PVA as the surfactant. SEM analysis along with an image processing software showed that the average particle size was 24µm.

Monodispersity



Average Diameter (µm)	PDI	CoV %
26.11	0.0009	2.93

Average Diameter (µm)	PDI	CoV %
23.52	0.0071	8.42

Average Diameter (µm)	PDI	CoV %
22.62	0.0019	4.4

Microparticles are considered monodisperse if:

1. Polydispersity Index (PDI) is <0.1[2]
2. Coefficient of Variance (CoV) is ≤3.04%[3]

$$PDI = \left(\frac{\sigma}{d}\right)^2$$

$$CoV = \left(\frac{\sigma}{d}\right) x\ 100$$

Hence, whilst these batches of microparticles cannot all be classed as monodisperse, they demonstrate low polydispersity.

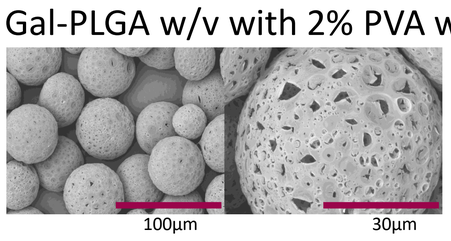
Surfactant Concentration with 10% PLGA

PVA Concentration	Average Diameter (µm)	PDI	CoV %
2 % w/v	23.52	0.0071	8.42
1 % w/v	26.11	0.00086	2.93
0.5 % w/v	30.74	0.031	17.73

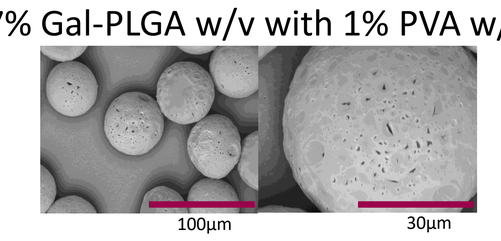
The reduction in PVA concentration caused an increase in the average particle size. Low system stability was also observed with 0.5% PVA.

Hepatocyte-targeting Material: Galactosylated PLGA

7% Gal-PLGA w/v with 2% PVA w/v



7% Gal-PLGA w/v with 1% PVA w/v



Gal-PLGA microparticles were successfully fabricated using microfluidics. Upon analysis notable pores were identified therefore the concentration of PVA was reduced. Porosity was lessened by this but still remained present.

4. Conclusions

1. Microfluidics can produce microparticles with low polydispersity
2. Optimisations with surfactant concentrations are required to maintain a stable microfluidic system
3. Galactosylated PLGA can be used with the microfluidic system, however, further optimisation and material characterisation is needed to eliminate porosity

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Krishna.patel@nottingham.ac.uk
www.linkedin.com/in/krishna-patel2
@_Krishna_P