Preparation and characterization of a co-electrospun nanofibrous gelatin/polyurethane composite scaffold for cardiac tissue engineering

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1. MOTIVATION

- **Cardiovascular diseases remain a leading cause of death[1].**
- **Cardiac tissue engineering is an alternative approach to the repair and regeneration of damaged myocardium[2].**
- **Degradable polar hydrophobic ionic polyurethane (D-PHI) is an innovative biomaterial that has immunomodulatory function which minimizes macrophage pro-inflammatory activation[3].**
- **An aligned nanofibrous scaffold (Figure 1) produced by electrospinning D-PHI and a degradable linear polycarbonate polyurethane (PCNU) enabled effective attachment and healthy growth of human pluripotent stem cell derived cardiomyocytes (Figure 2).**

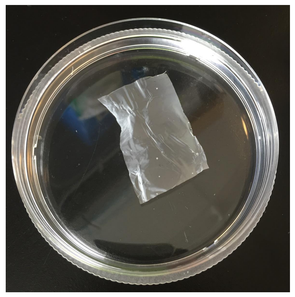

Figure 1: Nonfibrous D-PHI/PCNU scaffold.

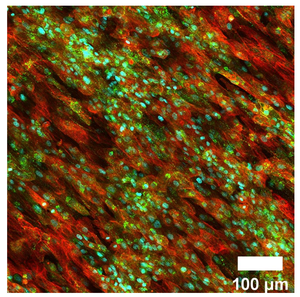

Figure 2: Immunostaining of hiPSC-CMs (sarcomere actinin, connexin, nucleus) on the D-PHI/PCNU scaffold.

- **However, such scaffolds take too long to degrade (>3 months) and are too stiff (~55 MPa) for cardiac tissue engineering[4].**

2. OBJECTIVES

Central objective: To generate a co-electrospun gelatin/polyurethane composite nanofibrous scaffold with a reduced stiffness (ideal: 20-500 KPa[2]) and degradation time (ideal: 3-6 months[5]), when compared to the D-PHI/PCNU scaffold

Research Questions:

- **How do different electrospinning parameters (voltage, flowrate, solution viscosity, and PCNU molecular weight) affect gelatin fibre morphologies?**
- **To what extent does the incorporation of gelatin impacts the mechanical properties and the scaffold degradation rate?**

3. METHODS

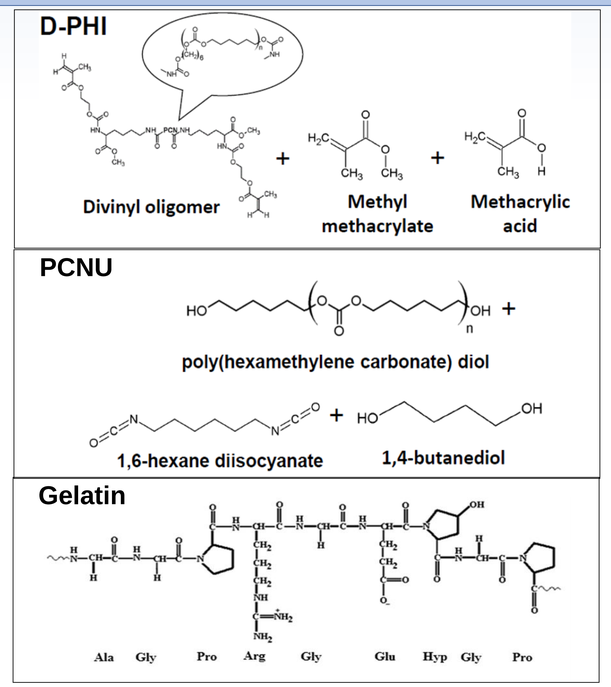

Figure 3: Structure of D-PHI.

Figure 4: Structure of PCNU.

Figure 5: Structure of gelatin[6].

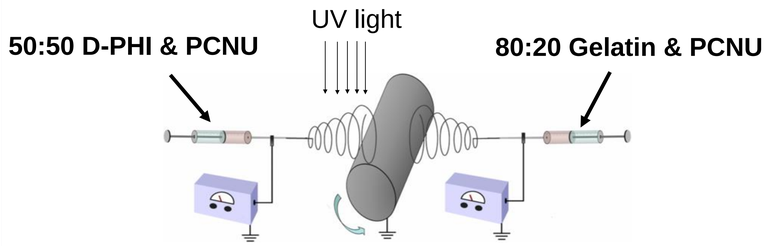

Figure 6: Schematic of the co-electrospinning setup.

5. RESULTS & DISCUSSION

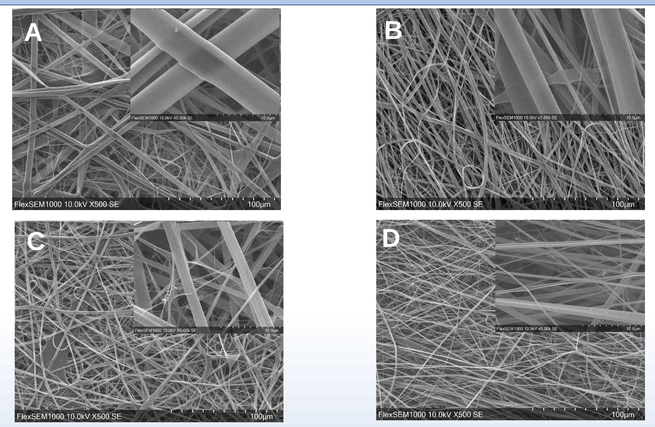

Figure 7A-7D: SEM images of the single-spun 80:20 gelatin/PCNU scaffold with parameters listed in Table 1 (see the third panel).

Figure	Voltage (kV)	Viscosity (s)	PCNU molecular weight (Dalton)	Fibre Diameter (µm) (n=60) (mean ± SD)
A	18	60	90,000	3.10 ± 2.15
B	18	60	140,000	2.20 ± 0.80
C	36	45	140,000	0.89 ± 0.52
D	36	25	140,000	0.68 ± 0.56

Table 1: Electrospinning parameters and fibre diameter of fibres in Figure 7.

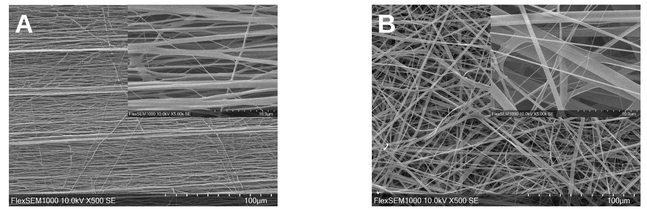

Figure 8A & 8B: SEM images of single-spun D-PHI/PCNU (left) and co-spun gelatin/D-PHI/PCNU scaffold (right). Fibres are defined below in Table 2.

Figure	Fibre Diameter (µm) (n=60) (mean ± SD)	Porosity (%) (n=3) (mean ± SD)
A: 50:50 D-PHI/PCNU SINGLE-spun	0.54 ± 0.45	13.54 ± 1.30 *
B: D-PHI/PCNU Gelatin/PCNU CO-spun (20% D-PHI + 25% PCNU + 55% gelatin)	0.78 ± 0.70	21.49 ± 5.51 *

Table 2: Fibre diameter and porosity of fibres in Figure 8.

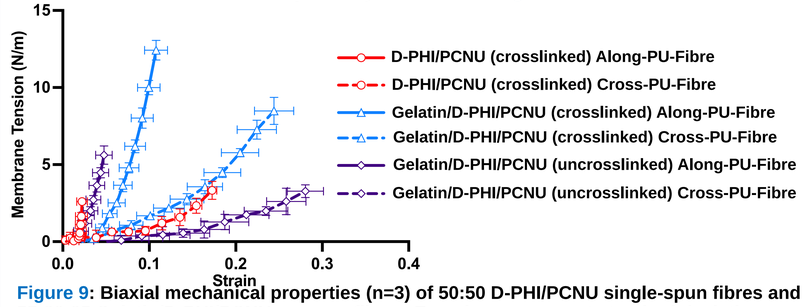

Figure 9: Biaxial mechanical properties (n=3) of 50:50 D-PHI/PCNU single-spun fibres and 55:20:25 Gelatin/D-PHI/PCNU co-spun fibres. Error bars represent standard deviations.

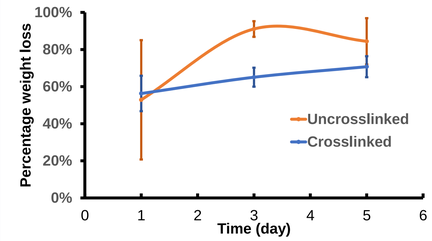

Figure 10: Collagenase (0.01 unit/mL) accelerated degradation of 55:20:25 Gelatin/D-PHI/PCNU co-spun scaffolds(n=6). Error bars represent standard deviations.

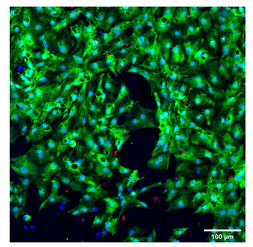

Figure 11: hiPSC-CMs viability (n=1) (>96%) on Matrigel coated 55:20:25 gelatin/D-PHI/PCNU scaffolds.

6. CONCLUSIONS AND FUTURE WORK

- **Gelatin/D-PHI/PCNU scaffolds have a higher porosity which would enable easy nutrient diffusion and cell infiltration.**
- **Incorporation of gelatin reduces the stiffness at least by half in the PU fiber direction, and significantly accelerated degradation.**
- **hiPSC-CM compatibility is currently being investigated.**

Reference:

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(3) 3) Sharifpoor et al., *Biomacromolecules* 2009, *10* (10), 2729–2739.

(4) Chan et al., *Acta Biomater.* 2019, *96*, 161–174.

(5) Anderson et al., *Semin Immunol.* 2008 Apr; 20(2): 86–100.

(6) Devi et al., *Advances in Colloid and Interface Science 2016, 239*

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