Polyhydroxyalkanoates: A Family of Natural Polymers, for Medical Implant Development and Disease Modelling

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Statement of Purpose: Polyhydroxyalkanoates or PHAs are a family of naturally occurring intracellular polymers synthesized by a variety of microorganisms. PHAs can be broadly classified into two types such as short chain length PHAs (scl-PHAs), containing 3-5 carbon atoms and medium chain length PHAs (mcl-PHAs) containing 6-14 carbon atoms within their monomer units. Their physical properties differ based on their type. Scl-PHAs, except for Poly(4-hydroxybutyrate) are brittle, stiff and have a high melting point and glass transition temperature, whereas mcl-PHAs are soft, elastomeric and have low crystallinity, melting point and glass transition temperature. PHAs are known to be biocompatible and biodegradable in nature. They degrade into non-toxic byproducts via surface erosion thereby maintaining its bulk properties. They are currently being explored for several biomedical applications such as medical devices, tissue engineering and drug delivery systems[1,2,3]. In this study, a range of novel PHAs were produced, characterized and used for 3 different medical applications such as – (1) Development of coronary artery stents, (2) Nerve guidance conduits and (3) Cancer disease modelling.

Methods: Novel PHAs were produced via bacterial fermentation using *Bacillus* and *Pseudomonas* strains. They were extracted and purified using the soxhlet extraction. They were characterized using Gas Chromatography-Mass Spectrometry (GC-MS) and Nuclear Magnetic Resonance (NMR). Biocompatibility studies were carried out to ensure that the PHA produced were non-toxic in nature. Coronary artery stent prototypes were developed using the dip moulding process. Dip moulded PHA tubes were mechanically characterized and laser cut by means of a picosecond pulsed laseto achieve a specific design and strut dimensions. Nerve guidance conduits were developed using the micro extrusion process. Adhesion assay was carried out to investigate the biocompatibility of the PHA based nerve conduits. *In vivo* studies were carried out where PHA based nerve conduits were implanted in the rat model of sciatic nerve injury. Finally, PHA based 3D scaffolds were developed using the particulate leaching method. These porous scaffolds were used to understand the cellular behavior and mechanisms of breast cancer using the Breast cancer cell line MDA-MB 231. Cell viability and their morphology was assessed using Alamar blue assay, confocal microscopy and scanning electron microscopy.

Results: A range of novel PHAs were produced as a result of extensive screening experiments. Based on their physical properties, purified PHAs were chosen for specific applications. Cell viability studies demonstrated that the PHAs produced were highly cytocompatible. For the development of coronary artery stents, PHA tubes were successfully produced using the optimised dip moulding and extrusion process. Their mechanical properties demonstrated significant post yield plastic deformation and elongation at break, while maintaining high stiffness and tensile strength. The tubes were successfully laser cut to achieve a specified design and strut dimensions. For the development of PHA based nerve guidance conduits, PHA tubes were successfully developed using the melt extrusion method. PHA based nerve conduits were cytocompatible and promoted neurite formation. *In vivo* studies demonstrated that, PHA based nerve guidance conduits promoted nerve regeneration in a rat model of sciatic nerve injury with a 10 mm gap. Finally, 3D scaffolds were successfully developed using a P(3HB)/mclPHA blend. The breast cancer cell line MDA MB 231 was used which demonstrated *in vivo* like clustered-rounded morphology when cultured on the 3D porous scaffolds.

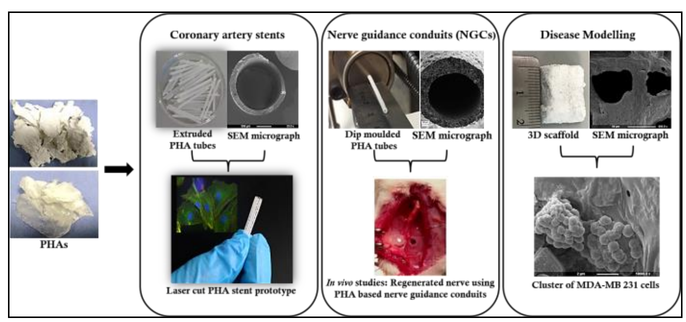

Figure1: Development of PHA based coronary artery stent prototype, Nerve guidance conduits and cancer disease model.

Conclusions: A range of novel PHAs were produced and characterized. These biocompatible and biodegradable PHAs were successfully used in the development of coronary artery stent prototypes, nerve guidance conduits and a cancer disease model.

References: **1.** Rai, R *et al.*, 2011. Biomacromolecules, 12, 2126-2136. **2**. Basnett, P., *et al.*, 2017. *Microbial Biotechnology*, *10*(6), pp.1384-1399. **3**.Misra, S.K., *et al.*, 2006. *Biomacromolecules*, *7*(8), pp.2249-2258.