

INVESTIGATION OF CELL ATTACHMENT ON THE SCAFFOLDS MANUFACTURED BY ELECTROSPUN PCL-HYALURONAN BLENDS

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INTRODUCTION. Recently, nanofibre-based scaffolds were developed to mimic natural extracellular matrix (ECM). Various synthetic and natural polymers have been used (1-3). Polycaprolactone (PCL) is one of the commonly studied synthetic polymers (4). However, PCL is associated with high hydrophobicity. Among other factors such as topography or the biological receptors on the scaffold surface, hydrophilicity is an important factor for cell attachment. It is known that hyaluronan is an important natural material found in ECM. But it suffers from low mechanical properties. Thus, to combine mechanical strong structure provided by PCL and the hydrophilicity provided by hyaluronan, a blend of PCL and hyaluronan was hypothesized to be a better option for cell attachment and function.

MATERIALS AND METHODS Either acetic acid or a mixture of acetic acid with 26% chloroform was used to dissolve PCL. To half of specimens 5% of hyaluronan was added to PCL solution. Both pure PCL and PCL-hyaluronan blend solutions were then electrospun using a voltage source (20 kV) to produce non-woven structures that were collected on glass coverslides. UV light was used for sterilisation of the scaffolds. Osteofibroblast cell line (MG63) was seeded on the scaffolds and incubated. Constructs were examined at 12h, 48h and 1 week. Actin staining was made after one week of culture and examined by fluorescent microscopy.

RESULTS AND DISCUSSION Scaffolds made from PCL-hyaluronan blends have formed more homogenous nanofibers as compared to neat PCL. Fibre diameter of neat and blend PCL fibres ranged from 150 to 250 nm. Cell attachment on the blend fibres was much faster and earlier and appeared healthier shape compared to that on neat PCL nanofibres. After 12 and 48 hour cell seeding, round shape cells were still seen on neat PCL nanofiber scaffold but not on blend fibres (Fig. 1). No relation between the type of used solvent and cell morphology could be demonstrated. Actin staining demonstrated the presence of dense filaments and elongated cell bodies on PCL blend scaffolds mesh indicating better cell attachment and focal adhesion (Fig. 2). It seems that hyaluronan which is naturally occurring polysaccharide present in connective tissues with high hydrophilicity can be beneficial for enhancing cell growth and cell attachment on PCL scaffolds. Although, in gel form, pure hyaluronan was found not to favour cell attachment [5]. However, when PCL was blended with small portion of hyaluronan, cell attachment was improved as compared to hydrophobic neat PCL. UV sterilization of the scaffolds in the current study may have affected hyaluronan structure, and contributed to improved cell attachment. However, such explanations need to be studied further, which is the subject of undergoing studies. From the current results, one can conclude that hyaluronan blended with PCL in 5% can improve osteofibroblast cell attachment and spread on electrospun nanofibre based scaffolds. This can further applied in tissue engineering.

References:

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Figures:

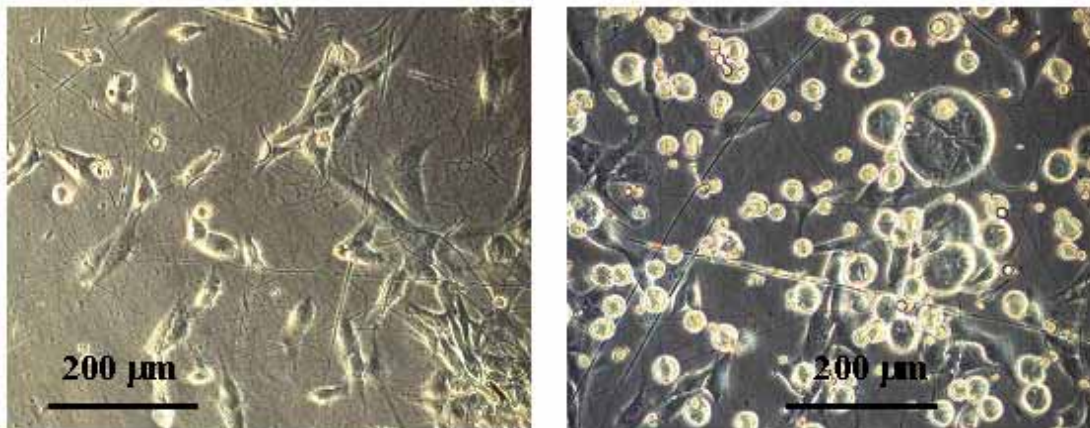


Figure 1 Light microscopic images of MG63 cells grown on nanofiber based scaffolds after 12 hour of culture on either neat PCL (Left) or PCL hyaluronan blend (Right).

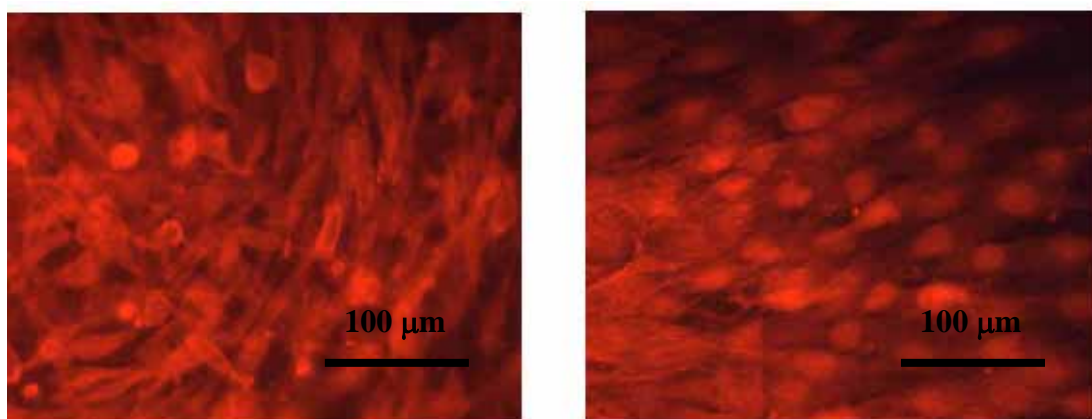


Figure 2 Actin stain images of MG63 cells grown on nanofiber based scaffolds after 1 week of culture on either neat PCL (Left) or PCL hyaluronan blend (Right).