



<b>Title</b>	<b>Electrospun fibrous tissue engineering scaffolds: topographic cues and their influence on cell behavior</b>
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## ELECTROSPUN FIBROUS TISSUE ENGINEERING SCAFFOLDS: TOPOGRAPHIC CUES AND THEIR INFLUENCE ON CELL BEHAVIOR

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### INTRODUCTION

Electrospinning can fabricate nanofibers that mimic the natural extracellular matrix of most human tissues. Electrospun fibrous scaffolds have thus been widely investigated for regenerating different human tissues such as bone, skin, ligament, etc [1]. Topographic cues of fibrous scaffolds can play an important role in influencing cell behavior and hence tissue formation. The present study aimed to investigate cell behavior on electrospun fibers having different diameters, surface topography and alignments. Being a natural, biocompatible and biodegradable polymer suitable for tissue engineering, poly(hydroxybutyrate-co-hydroxyvalerate) (PHBV) was used as a model biopolymer in this investigation.

### METHODS

PHBV was dissolved in chloroform and the PHBV solution was electrospun into microfibrillar scaffolds using the setup described in our previous study [2]. With the aid of benzyl triethylammonium chloride (BTEAC), submicron fibrous scaffolds were electrospun. Surface-porous fibrous scaffolds were electrospun when a mixture of chloroform and dichloromethane (DCM) was used as a solvent for dissolving PHBV. Scaffolds consisting of aligned fibers were fabricated using our previously developed technique [3]. A human osteoblast-like cell-line (SaOS-2) was used and the cells were seeded onto the aforementioned four types of fibrous scaffolds. At different cell culture time points (days 2, 7, and 14), the cell morphology and alignment, proliferation and expression of alkaline phosphatase (ALP) activities were evaluated.

### RESULTS AND DISCUSSION

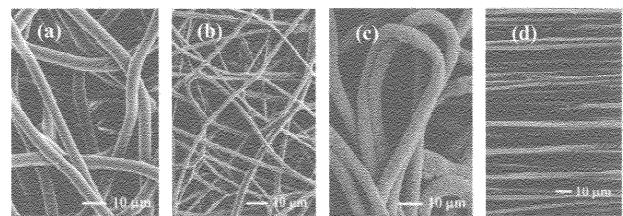
Electrospun microfibrils (Fig.1a) and submicron fibers (Fig.1b) exhibited fiber diameters of 3000-5000 nm and 200-800 nm, respectively. Surface-porous fibers (Fig.1c) exhibited surface nano-pores having diameters of about 200 nm. The aligned fibers (Fig.1d) exhibited micron-sized diameters and smooth fiber surfaces.

The proliferation rates of SaOS-2 seeded on non-woven scaffolds consisting of microfibrils, submicron fibers and surface-porous fibers were comparable to that of cells seeded on tissue culture plate (*i.e.* control), implying that these non-woven fibrous scaffolds support cell proliferation. The proliferation rate of SaOS-2 seeded on the aligned fibrous scaffolds was higher than that on the non-woven fibrous scaffolds. Cao *et al.* reported similar observations, though they did not deal with the cell proliferation issue [4].

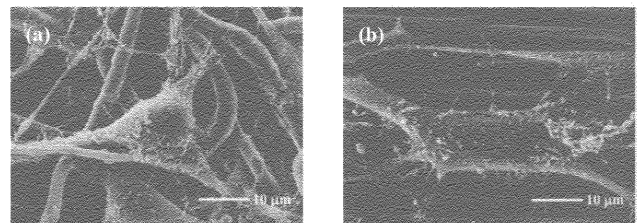
ALP activities of SaOS-2 cells seeded on all types of fibrous scaffolds investigated increased dramatically from day 7 to day 14. As ALP production is one of the steps within the whole differentiation sequence of bone tissue, significant ALP expressed by proliferated cells seeded on scaffolds for

bone tissue engineering is important. The enhanced ALP activity expression on these scaffolds suggested their high potential to direct cell behavior towards bone regeneration.

The cells seeded on the non-woven fibrous scaffolds (including microfibrils, submicron fibers and surface-porous fibers) spread randomly in all directions (Fig.2a) but the cells spread along the fiber direction on the aligned fibrous scaffolds (Fig.2b), indicating that the aligned fibers may affect the cell behavior and hence tissue formation by contact guidance.



**Figure 1:** Electrospun PHBV scaffolds consisting of (a) microfibrils, (b) submicron fibers, (c) surface-porous fibers, (d) aligned fibers.



**Figure 2:** SaOS-2 cells on electrospun PHBV scaffolds consisting of (a) non-woven fibers, (b) aligned fibers.

### CONCLUSIONS

Fibers having different diameters, surface topography and alignments were successfully fabricated by electrospinning. The electrospun PHBV fibrous scaffolds consisting of microfibrils, submicron fibers, surface-porous fibers and aligned fibers could all support cell proliferation and elicit expression of ALP activity. Fiber alignment was an important topographic cue for influencing SaOS-2 cell spreading, proliferation, and probably bone tissue formation.

### REFERENCES

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