

## Z-AXIS (DEPTH) CONTROL OF CELL SPREADING ON MICROPATTERNED SURFACES

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The control of the proliferation and differentiation by the cell reaction to a surface was reported recently [1]. However, the mechanisms involved in the cell contact to micro- and nano-topographies have not been fully understood. Also the study of cell morphology when seeded in surface micropatterns with different depths was not yet quantitatively analysed. In this study, the response in terms of cell morphology and alignment to micro-scale topographical structures was evaluated. A range of parallel grooved surfaces with equidistant spaces (3.38-11.36  $\mu\text{m}$ ) and various depths (1.35-4.95  $\mu\text{m}$ ) were prepared in poly(caprolactone) (PCL) membranes using the hot embossing technique. The response of osteoblastic cells to those patterns after fluorescence labeling of the cytoskeletons was evaluated. The results were quantitatively analyzed by image analysis.

As shown in Figure 1 (a) and (b), it was found that the cell area and maximum length were marginally affected by the pattern dimension varying from 3.35 to 11.36  $\mu\text{m}$ . In contrast, the cell area and length changed significantly when the pattern depth was varied between 1.35 to 4.95  $\mu\text{m}$ , being the effect more pronounced in terms of cell length.

Figure 2 (a) shows the compiled data from the previous parameters, where a very distinct transition in the cell morphology parameter is shown with depth. The cell area/cell length ratio was significantly more affected by the smaller in-plane dimensions and by the deepest patterns. In addition, at the lowest depth, the ratio decreased directly with the pattern dimension from 3.35 to 11.36  $\mu\text{m}$ . As the pattern depth increase, the ratio dropped drastically and the differences caused by the in-plane dimension variation were not so clear. The decrease in the ratio cell area/cell length indicates that the cell behavior changed from round spread shape into highly aligned morphology, which could be confirmed by the microscopical analysis (Fig 2 (b)). The effect of the pattern depth over the cell morphology was 2-4 times larger than that caused by the variation of pattern size in the plane, thus being strongly effective in controlling the cell alignment.

### References:

[1] D. M. Pirone, W. F. Liu, S. A. Ruiz, L. Gao, S. Ragdavan, C. A. Lemmon, L. H. Romer, C. S. Chen. *J Cell Biol*, (2006) 277-287.

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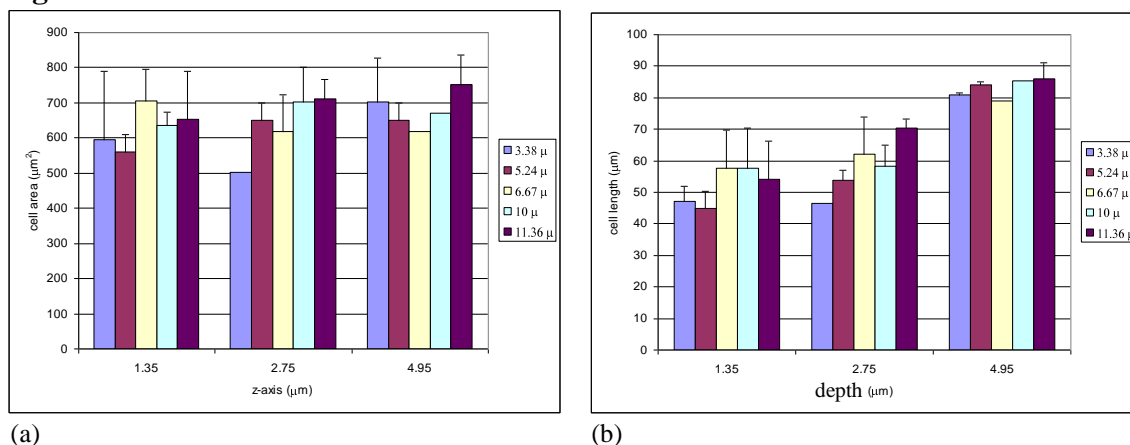
**Figures:**

Figure 1. In-plane pattern dimensions and pattern depth effect over the average cell area (a) and cell length (b).

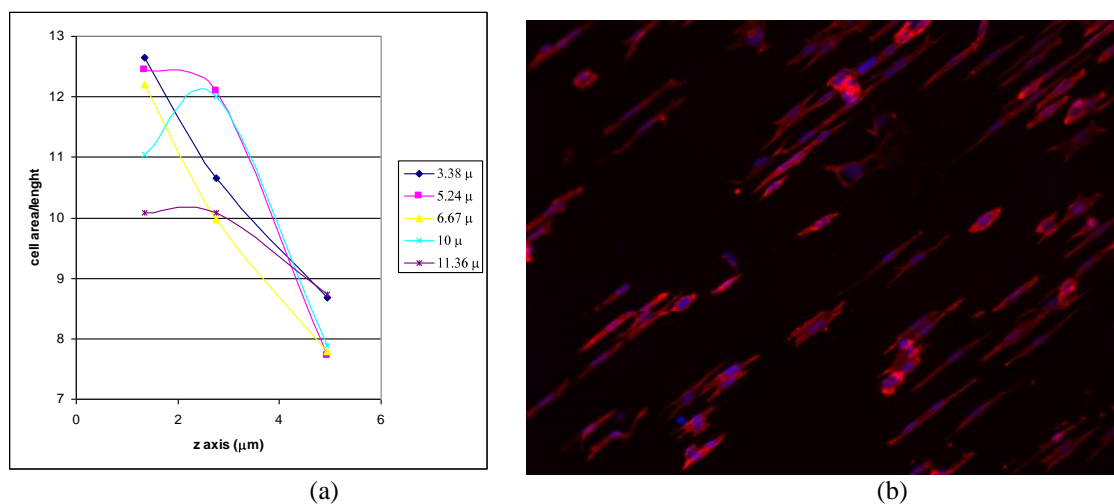


Figure 2. Variation of morphology parameters (cell area/cell length ratio) with depth for each pattern dimension (a). Fluorescence light microscope image of highly elongated and aligned SeOs-2 cells on PCL surface with 11.36  $\mu\text{m}$  ridge and 4.95  $\mu\text{m}$  deepness (b).