

Endothelial Cell Adhesion on Highly Controllable Compared to Random Nanostructured Titanium Surface Features

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Statement of Purpose: Vascular stenting is the procedure of implanting a thin tube into the site of a narrow or blocked artery due to atherosclerosis. Currently, the application of vascular stents using conventional metals is limited because the implantation process will cause significant injury to the vascular wall and endothelium. Numerous studies have focus on how to accelerate the endothelialization of the stents. In this study, a method was used to modify the surface features of the stents to achieve this objective. Since highly controllable nanostructured surface features mimic the surface of the natural endothelium, it is thought to be a good choice. The objective of this study is thus to fabricate highly controllable nanostructured surface features and to investigate subsequent endothelial cell function on them.

Methods:

Materials and treatment: Two methods were developed to produce Ti highly controllable nanostructured surface features: lithographic technique and E-beam evaporation. Multiplexed chips were created by the first method and were composed of commercially pure titanium (99.60%Ti). Briefly, Each sample is made up of an array of nine 5×5 mm patterned sub-regions. The dimensions of the lines and spaces of each pattern range from 100 μm to 0.75 μm. By E-beam evaporation, another kind of Ti nanopatterns with the help of Au-coated grids with grooves (width of 30 μm, SPI Supplies) was created placed on Ti-coated silicon wafers. Compacts with random nanostructured surface features and etched glass cover slips were used as controls. All the surface features were characterized using scanning electronic microscopy.

Cell experiment: Rat aortic endothelial cells (RAEC) cultured in MCDB-131 Complete Medium (VEC Technologies) were used in cell adhesion and 1, 3 and 5 day proliferation experiments (seeding area: 4500 cells/cm² for cell adhesion test and 2000 cells/cm² for cell proliferation test). Cell viability and morphology were analyzed by a Live/Dead Assay with fluorescence microscope.

Results/Discussion:

Ti highly controllable nanostructured surface features and random nanostructured surface features used in this study demonstrated the ability to enhance cell adhesion compares with conventional Ti surface features. Cell adhesion and proliferation results indicated improved cell viability on highly controllable nanostructured surface features. RAEC were also well spread and aligned along

the nanolines.

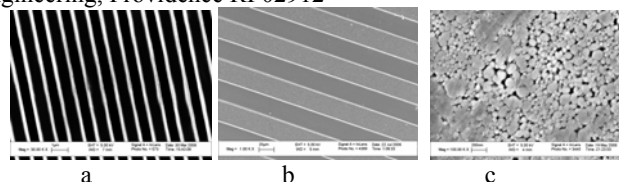


Figure 1: Different kinds of surface features created in the study. a: highly controllable nanostructured surface features by lithography (bar=1μm); b: highly controllable nanostructured surface features by E-beam evaporation (bar=20μm); c: random nanostructured surface features (bar=200 nm)

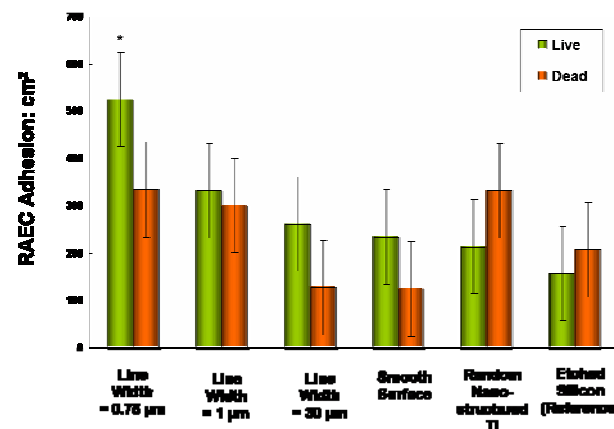


Figure 2: Increased rat aortic endothelial cell (RAEC) adhesion on highly controllable nanostructured surface features compared with random nanostructured features. *p<0.1 compared with random nanostructured surfaces and micropatterned surface

Conclusions: The results of this study could help in promoting endothelial cells to adhere and, thus, form a monolayer with an elongated morphology that mimics that observed in the natural vascular for improved titanium stent applications. Future studies will focus on how to improve highly controllable nanostructured surface features fabrication process and the relation between endothelial cell spreading degree (the extent to which endothelial cells can spread) and nanopatterned dimensions.

References:

1. Choudhary S, Berhe, M Haberstroh KM, and Webster TJ. International Journal of Nanomedicine 2006; 1(1): 41-49
2. Khang D, Sato M, et al. International Journal of Nanomedicine 2006; 1(1): 65-72