Enhancement the Growth of Human Endothelial Cells by Surface Roughness at Nanometer Scale

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Abstract

This study investigated whether a nanometer scale of surface roughness could improve the adhesion and growth of human endothelial cells on a biomaterial surface. Different molecular weights or chain lengths of polyethylene glycol (PEG) were mixed and then grafted to a polyurethane (PU) surface, a model smooth surface, to form a nanometer (nm) scale of roughness for PU-PEG surfaces (PU-PEG(mix)) while PEG with a molecular weight of 2000 was also grafted to PU to form PU-PEG(2000) for comparison. In addition, the concept was tested on cell-adhesive peptide Gly-Arg-Gly-Asp (GRGD) that was photochemically grafted to PU-PEG(mix) and PU-PEG(2000) surfaces (e.g., PU-PEG(mix)-GRGD and PU-PEG(2000)-GRGD surfaces, respectively). To prepare GRGD-grafted PU-PEG(mix) and PU-PEG(2000) surface, 0.025M of GRGD-SANPAH (N-Succinimidyl-6-[4'-azido-2'-nitrophenylamino]-hexanoate) solutions was grafted to PU-PEG(mix) and PU-PEG(2000) by surface adsorption of the peptide and subsequent ultraviolet (UV) irradiation for photoreaction. The grafting efficiencies for GRGD to PU-PEG(mix) and PU-PEG(2000) surfaces were about 67% for both surfaces, semi-quantitatively analyzed by an HPLC. The surface roughness, presented with a roughness parameter, R(a), and the topography of the tested surfaces were both measured and imaged by an atomic force microscope (AFM). Among the R(a) values of the films, PU was the smoothest (e.g., R(a)=1.53+/-0.20 nm, n=3) while PU-PEG(mix) was the roughest (e.g., R(a)=39.79+/-10.48 nm, n=4). Moreover, R(a) values for PU-PEG(mix) and PU-PEG(mix)-GRGD surfaces were about 20 nm larger than those for PU-PEG(2000) and PU-PEG(2000)-GRGD, respectively, which were consistent with the topographies of the films. Human umbilical vein endothelial cells (HUVECs) were adhered and grown on the tested surfaces after 36 h of incubation. Among the films, HUVEC's adhesion on the surface of PU-PEG(mix)-GRGD was the densest while that on the surface of PU-PEG(2000) was the sparsest. Also, the adhesion and growth of HUVECs for the roughness surfaces were statistically significantly better than that of smooth surface for both GRGD grafted and un-grafted surfaces, respectively. The viability for the growth of HUVECs on the tested surfaces analyzed by MTT assay also confirmed the efficacy of

the increased surface roughness.In conclusion, increased surface roughness of biomaterial surfaces even at 10-10(2) nm scale could enhance the adhesion and growth of HUVECs on roughness surfaces that could be useful for applications of tissue engineering.