Modulation of Cell Behaviour using Self–Assembled Binary Colloidal Crystals

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Statement of Purpose: The control of cell behaviour on surfaces is the key to a broad range of biomedical applications. Biomaterial surfaces with tuneable surface topographies and chemistries can profoundly influence the development of advanced biomaterials used in applications including tissue engineering and regenerative medicine. Recently, we developed an elaborate and feasible method to display an ordered surface topography with tuneable surface chemistry using binary colloidal crystal particles.[1] Using this binary colloidal system, various combinations of particle size and surface chemistry can be readily employed. In this study, two combinations of binary colloidal crystals, i.e. PS-COOH (2 μm)/PMMA (0.4 μm) and SiO_2 (2 μm)/PMMA (0.4 μm) were assembled on ozone-treated silicon wafers. The morphology of L929 fibroblasts was studied after a 24h cell culture period.

Methods: Silicon wafer cut into 1 cm^2 pieces were cleaned by 15 min of sonication in ethanol, followed by drying with N_2 gas. Substrates were then UV-ozone treated to remove organic contamination. Highly purified deionized water was used as the solvent during colloidal assembly. The volume of colloidal particle solution, number of the particles in the solution, and subsequently, their volume fractions were calculated assuming the area of substrate encircled by the rubber ring, diameter, and % wt solid content of particles employed. We have also determined the number ratio of large to small particles for different binary colloidal assemblies used in the work. In this method, we used very low amounts of colloidal particles. Rubber rings were cleaned thoroughly by sonication in ethanol for 15 min. After the rubber ring was fixed to the substrate, the colloidal particles suspension was dropped carefully inside the ring. The substrate was then kept at room temperature until complete evaporation of the solvent.

Results: Cells had a small projection area rather than fully spread morphology on the binary crystal surfaces compared with the flat control. However, an abundance of cell protrusions called filopodia were observed using scanning electron microscopy (SEM). Furthermore, cells had long and thin extended filopodia on the PS/PMMA binary crystal surfaces, while they had short and thick filopodia on the SiO_2/PMMA crystal surface. Regarding the surface chemistry, both SiO_2 and PMMA particles were not as favourable as the PS-COOH particles for cell attachment, and resulted in the average cell projection area on the PS/PMMA being larger compared to the SiO_2/PMMA binary colloid assembly.

Conclusions: These results show for the first time that cell-substrate interactions can be easily controlled by precise positioning of different particles with various sizes and chemistries. The present results will help gain a more thorough understanding of cell-material interactions benefiting the development of advanced biomaterials and materials for tissue engineering.

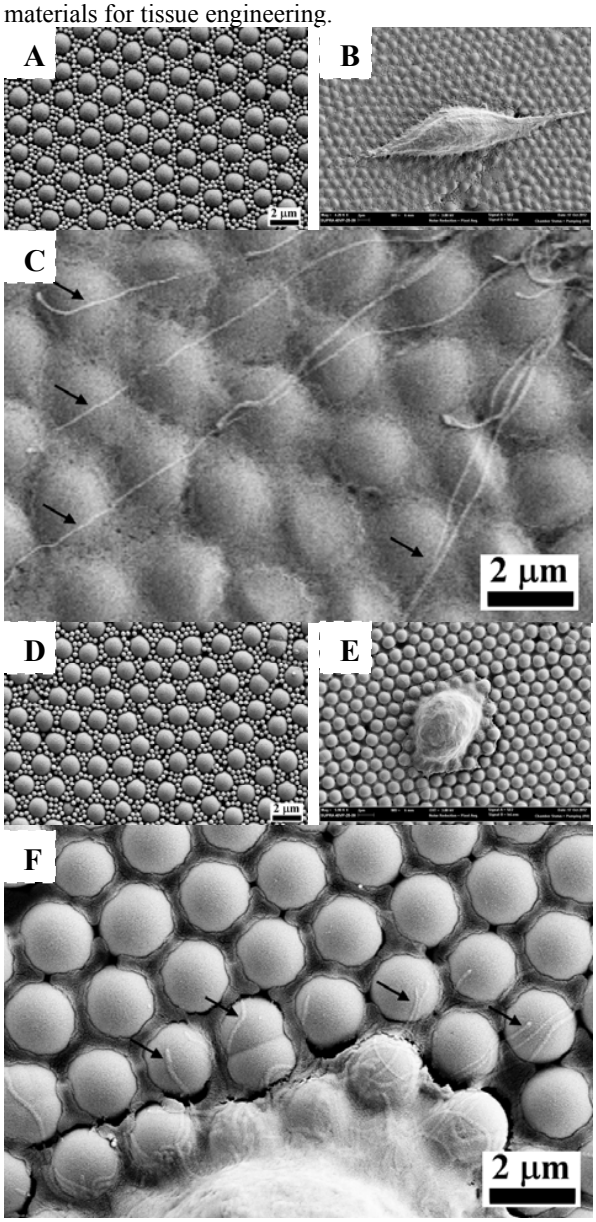

Figure 1. Binary colloidal crystals self-assembled on the surface acting as a substrate for L929 fibroblasts attachment (after 24 h). Cells attached on (A-C) PS (2 μm)/PMMA (0.4 μm) and (D-F) SiO_2(2 μm)/PMMA (0.4 μm) colloidal crystal surfaces. Cells have longer filopodia (arrows) and show more spreading on the PS/PMMA surface compared to the SiO_2/PMMA surfaces (inserted images). Cell spreading on the colloidal crystals was reduced compared to a flat Si control surface.

References: Singh G, Pillai S, Arpanaei A, Kingshott P. Soft Matter, 2011:7:3290