

## TWO-DIMENSIONAL CONTROL OVER PROTEIN AND CELL ATTACHMENT ON MODIFIED POLY(ETHYLENE) SURFACES BY LASER ABLATION

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**INTRODUCTION:** In the field of biomaterials and related research, there is currently an increasing amount of interest in obtaining two-dimensional control over the location of cells and bioactive compounds on a surface by manipulation of the surface chemistry and topography. Having control over location of proteins or cells on a surface with high resolution is of great importance for applications such as diagnostic tests, cell based sensors and tissue-engineered medical devices<sup>1,2</sup>.

**METHODS:** Spatially controlled surface chemistries were created as follows; a poly(ethylene) co-polymer [PE-co-GMA] (Atofina Chemicals) with a high degree of glycidyl methacrylate functionalities was used to provide a surface suitable for cell attachment. Coupling of thiol functionalised poly(ethylene oxide) [PEO] (Sigma-Aldrich) at a pH of 9.0 under cloud point conditions was used to create a surface resistant to protein adsorption.

Patterning of the surface chemistry of injection moulded samples was achieved by laser ablation using a 248 nm excimer laser. X-ray photoelectron spectroscopy (XPS) and atomic force microscopy were used to characterize the surface topography and surface chemistries. Protein adsorption tests were performed on the surfaces using collagen [Coll] (Vitrogen<sup>®</sup>) and the biological response to two-dimensional patterned substrates was analysed in cell culture experiments using bovine corneal epithelial cells [BCEp]. After fixation of the cells with formal saline and staining cell attachment patterns were imaged using a confocal scanning laser microscope.

**RESULTS:** XPS experiments were performed to monitor protein adsorption on the PE-co-GMA substrate material and PEO functionalised surface. The presence of adsorbed proteins is clearly evident from analysis of the C1s high resolution spectra of the PE-co-GMA-Coll sample as seen in Figure 1. A broad shoulder representing amine/hydroxyl and amide components (286.5 eV and 288.5 eV respectively) of the protein are clearly visible. However, no protein is detected after collagen adsorption to PE-co-GMA-PEO surfaces indicating low fouling characteristics.

Figures 2A and 2B show light microscopy images of a tissue culture poly(styrene) surface after collagen adsorption and BCEp cell culture and an ablated PE-co-GMA-PEO sample respectively. The ablated rectangular area is clearly visible and well resolved. Cell attachment clearly follows the pattern provided by excimer laser ablation, while the non-ablated area only shows very few cells with little or no spreading.

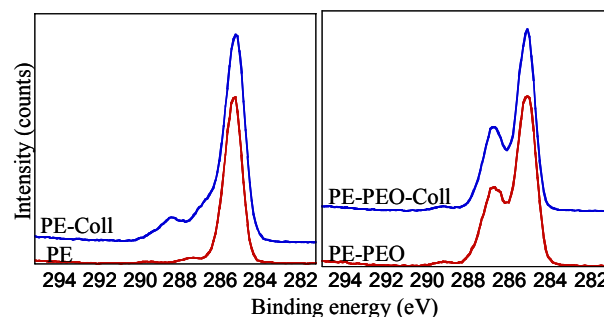


Fig. 1: (A) PE-co-GMA before and after adsorption of collagen; (B) PE-co-GMA – PEO before and after adsorption of collagen

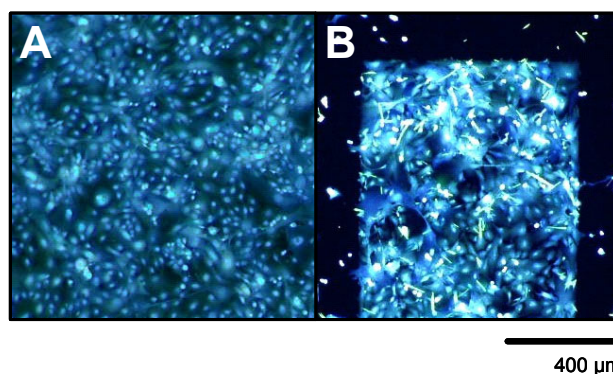


Fig. 2: Microscope images of (A) TCPS control surface and (B) PE-co-GMA-PEO after laser ablation and BCEp cell attachment.

**DISCUSSION & CONCLUSIONS:** Cell culture experiments using bovine corneal epithelial cells confirmed that cell attachment is controlled by the surface chemistry pattern. The method is an effective tool for use in a number of *in vitro* and *in vivo* applications.

**REFERENCES:** <sup>1</sup>Rudolph A S and Reasor J (2001) *Biosensors & Bioelectronics* **16** 429. <sup>2</sup>Desai T A 2000, *Medical Engineering and Physics* **22** 595.