PROPERTIES OF, AND SUB-MICROMETER CONTACT PRINTING WITH HYBRID AMINOSILANE-AGAROSE HYDROGELS.

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Hydrogels doped with an amino-silane have been prepared and used for the production of submicrometer stamps, which allowed transferring biofunctional submicrometer motives to flat Si (100) surfaces.

Soft lithographic techniques compatible with the preparation of biomedical surfaces are emerging mainly due to the increasing demand of bioanalytical and biotechnological techniques. The interest in these materials is expanding due to the possibility to use them as stamps for the transfer onto surfaces of different functional molecules [1], proteins [2] or even cells [3]. Many hydrogel types have been already explored for the realization of micropatterns on surfaces: agarose, acrylamide, hydroxyethyl-methacrylate, polyethylene-glycol-diacrylate, carboximethylated dextran or polyvinyl methyl ether have been used in a very wide margin of applications and micropatterning configurations. 2D and 3D structures were prepared by different techniques implying PDMS masters, inkjet printing or selective polymerization induced by electron beams, UV photons or X-ray radiation. In this work we have applied colloidal lithography for the production of submicrometer biofunctional motives on surfaces. The organometallic 3-aminopropyl-triethoxysilane (APTS), a common biofunctionalizing molecule generally deposited by self assembly, is used as a dopant agent to form a hybrid hydrogel.

The preparation of the hydrogels involved the conventional melting of agarose in water and the addition of 3-aminopropyl-triethoxysilane (APTS) as biofunctional dopant. The hybrid gels were characterized by Fourirer transform infrared spectroscopy. Silicon surfaces were activated previous to contact printing with the hybrid hydrogels in a piranha solution. Homogeneous functionalization of surfaces was performed to measure the wettability by water contact angle. Submicrometer microcontact printing was carried out by preparing a polystyrene colloidal monolayer, which shaped the hydrogel surface. After microcontact printing, biofunctionalized structures were observed in a Fluorescence microscope by the immobilization of an FITC labelled antibody. A topographic view of the generated structures during stamp fabrication was obtained by using atomic force microscopy (AFM).

The physicochemical characterization of the mixed APTS-agarose hydrogels denotes the formation of an hybrid gel as deduced to the presence of new bands in the FTIR spectra not present in the independent APTS or agarose spectra.
The biofuntionalization induced by microcontact printing of piranha solution activated silicon substrates was patent after water contact angle measurements, which showed that surfaces had become much more hydrophilic.
During stamp fabrication with submicrometer colloidal monolayers as moulds, a series of structures were obtained. AFM images confirmed that the colloidal moulds were effective in transferring the hexagonal structure to the hydrogels. The masters were used as pealed to define biofunctionalized motives in the silicon substrates. The transferred motives were evidenced by fluorescence microscopy after FITC labelled Igg immobilization. Typical structures show a hexagonal arrangement of activated sumbmicrometer dots (figure 1).

The formation of hybrid gels and their transformation into masters by using well structured colloidal monolayers as moulds is a promising method for the selective topographic biofunctionalization of surfaces in the submicrometer scale, which could be of interest in both biomolecular and cellular assays.

References:

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Figures:

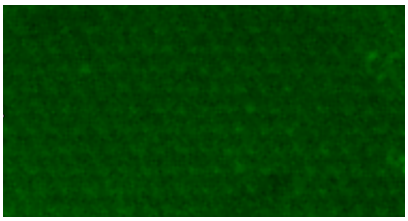

Figure 1: Fluorescence microscopy image of the bio functionalized dots with hexagonal structure.