Surfactant-assisted Imprinting for Fabricating Bioactive Gel Micropatterns

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Statement of Purpose: Hydrophilic polymer patterns are uneasy to handle in the traditional photolithography techniques, because the developing reagents are aqueous solutions. Attempts to etch the hydrophilic patterns usually lead to severe contamination of etchant and probable harm to the biomolecules. Also, the polymer carrying some bioactive molecules should not endure photoirradiation and etching. In the study, a flexible process was adopted to fabricate hydrophilic gel pattern without residual layer. By adhering an amphiphilic surfactant, phosphatidylcholine (PC), on the protrusion areas of the stamp pattern, the stamp was then able to selectively adsorb poly(vinyl pyrrolidone) (PVP), alginic acid (AA), and chitosan (CS) which were transferred on a silicon plate via imprinting process.

Methods:
1. Enrichment of PC in phospholipids: Extraction of PC-enriched phospholipids has been developed well under liquid chromatography analysis. The commercial Lecithin (for food) was precipitated in acetone, extracted by ethanol, and put into a mixed acetonitrile/methanol solvent (4/1 (V/V)) for 30 min of vigorous stir. Those insoluable lipids and most PE were filtered out, giving a PC-enriched, clear-yellow solution. After evaporating the solvent at 40 ^{o}C, the extract was dissolved in 30 ml of ethanol. Zinc chloride (60 wt%) solution was then dropped into the extract solution under stirring to form ivory PC-$ZnCl_2$ complex precipitation.
2. Polymer transfer process: The PDMS stamp fabrication can be referred from the other published literature.[1] By lifting to separate the polymer, the stamp with the complementary pattern was ready for printing polymers. Besides, a clean silicon wafer was treated with 1 mM of octadecyltrichlorosilane (OTS) in toluene solution on the surface for easy releasing before being immersed into a 0.1 wt% of crude phosphatidylcholine (PC) in n-hexane solution for 30 s and then was dried by nitrogen flow. The stamp was briefly exposed to O_2 plasma (AR-100 PC) using simple plasma equipment. By impressing the stamp on the PC-coated silicon plate for 3 min, the PC-adhered stamp was then coated with a water-soluble polymer by spinning. Upon the conditions of a given hydrostatic pressure and a temperature on the hot plate, the hydrophilic polymer was transferred on a silicon wafer and gave a no-residual-layered pattern.
3. Analysis: Contact anglometer (Magic Drop AT) was used to check the contact angle changes of a no-pattern plate during the process. Optical microscope (OM) and scanning electron microscope (SEM) was directly evaluated for the polymeric thin layers and patterns.

Results/Discussion: The OTS-pretreated silicon plate assisted in PC transfer from the plate to the convex of the PDMS pattern, causing the water contact angle change from 120.7^{o} to 50.5^{o}. Another contrast test with a freshly cleaned silicon plate (no OTS-treatment) was strongly adhered by PC layer which was not transferred any more. Optical microscopic observation was employed to ensure the PC transferred pattern. Therefore, the convex patterns revealed more hydrophilic than the concave ones. Hydrophilic polymers: PVP, AA, or CS, in a diluted solution were able to adsorb on the convex patterns. That is why no residual layer found. Imprinting process was then carried out under a slightly normal pressure to force the polymers to adhere the silicon wafer. In Guo's publication, a high temperature was applied to soften the given polymers, leading to the dimension shrinkage after cooling. [2] We chose the gentle condition under O2 plasma treatment or not to imprint the polymers at ambient temperature, giving no apparent shrinkage of patterns. The thickness of transferred chitosan was about 180 nm, dependent on the concentrations of chitosan solution and immersion time.

Conclusions: The hydrophilic polymers—PVP, AA and CS were successfully transferred onto the wafer as a cell-adhesion layer using imprinting techniques. The PDMS stamp pre-adhered with PC on the protruded patterns and followed by adsorbing gels to imprint the chitosan layer on the wafer. Plasma activation on the stamp, adsorbed chitosan, and/or the substrate were able to provide an accessible method to obtain the nanothick polymer pattern without residual layers.

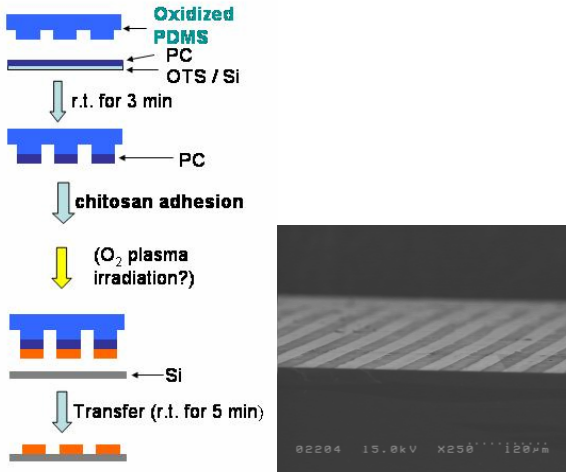

Figure 1. The schematic process for the surfactant-assisted imprinting technique (left), and the imprinted chitosan micropattern (right).

References:
1. Xia Y, Whitesides GM. Angew Chem Int Ed. 1998; 37: 550.
2. Guo, LJ. J Phys D : Appl Phys. 2004; 37: R123.