Surface Patterning of Protein Matrix Basement

H. Mekaru
National Institute of Advanced Industrial Science and Technology (AIST),
1-2-1 Namiki, Tsukuba, Ibaraki, Japan 305-8564
h-mekaru@aist.go.jp

We looked at the patterning of fine structures directly on the surface of a basement membrane matrix as a grid mesh for the recording of adhesion positions of a growing cell. Thermal nanoimprint lithography (NIL) is now being applied on thermoplastics with glass transition temperatures, where the press operation is executed at high temperatures of 100-200 °C. It is well known that a high dimension protein structure can be easily damaged at higher temperatures. In this presentation, a possibility of thermal-NIL technology being applied to a protein is explored.

Two 35-mm-square ceramic heaters were mounted on the upper and bottom ends of a press device driven by a servomotor in a desktop thermal NIL system NI-273 (Nano Craft Technologies). To fit to the size of the ceramic heater, BioCoat Matrigel[1] (Corning Life Sciences), coated on a 35-mm-diameter dish at 100 μg/cm^2, was chosen as a gelatinous protein mixture basement membrane (Fig. 1). The protein thin layer was then gelled according to the manufacturer's specs, followed by placing the dish on the bottom ceramic heater. An actual inside growth surface diameter of the dish was 33.9 mm. Therefore, a 100-μm-wide grid mesh and area numbers were processed using photolithography and reactive-ion-etching on a 1-inch Si wafer. The height of convex structures on the Si mold was 25 μm. To avoid any intrusion by a 10-mm-high circular sidewall of the dish, a 1-inch-diameter copper mold mount 22 mm high was prepared. The Si mold was fixed by a double-faced tape on the mold mount. The difference between the temperatures of the Si mold surface and of the upper ceramic heater was measured, and adjustments were made to attain a preset temperature before the imprinting.

Figure 2 shows the results of observation on the surface of the Matrigel thin film imprinted at 75, 100, and 125 ^{0}C for 30, 60, and 90 s using an optical microscope. An area number "B2" was located at the center of the grid mesh. When an imprint temperature was 75 ^{0}C, an imprinted pattern was barely observable. On the other hand, if the mold was heated up to 100 ^{0}C or more, the imprinted grid mesh and the printed number on the Matrigel was clearly observed. However, a heating at 125 ^{0}C seemed to have damaged the protein. Figure 3 shows linewidths and depths of the grid, as measured by a five-line confocal-microscope Optelics S130 (Lasertec), was directly proportional to the imprinting time. The linewidth of the imprinted patterns was spread in the case with the imprint temperature of 125 ^{0}C, and the depth became shallow. The protein around the imprinted patterns was also deformed by a heat transfer; and that resulted in the boundary of the patterns becoming indistinguishable.

[1] S. Vukicevic, *et al.*, Exp. Cell. Res., 202, 1 (1992).

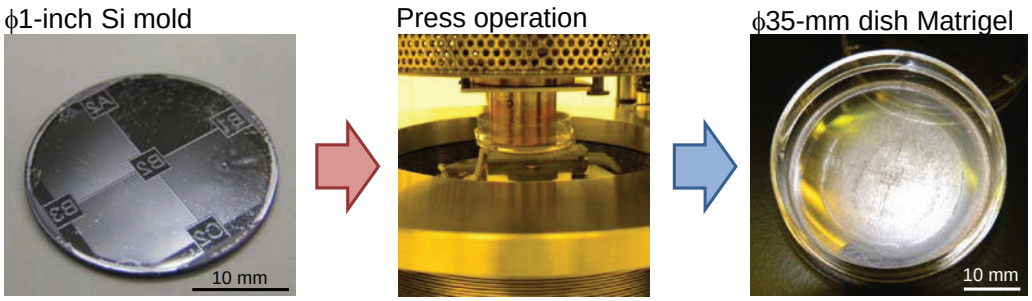

Figure 1: Experimental procedure in thermal imprinting on Matrigel.

Figure 2: Optical micrographs of the imprinted surface of Matrigel.

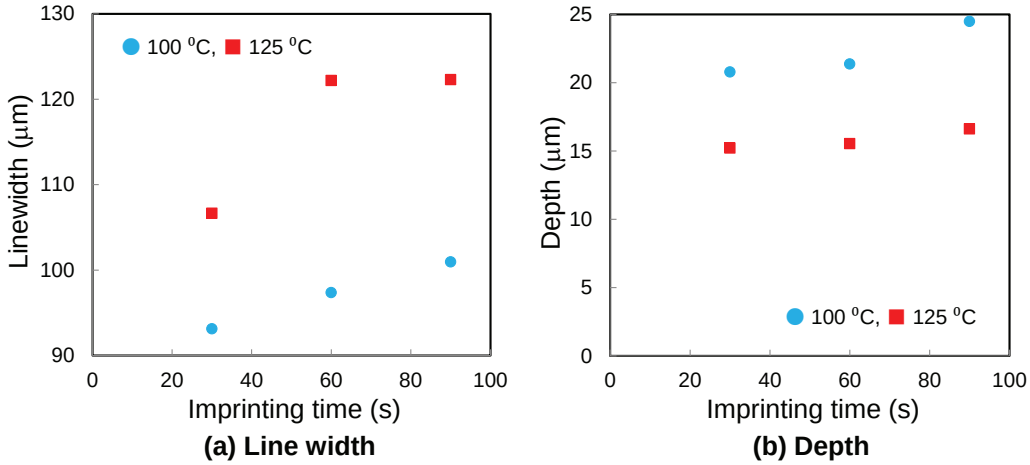

Figure 3: Measured (a) linewidth and (b) depth of imprinted pattern on Matrigel.