The Study of the Nanocontact Imprint with Aminosilane for Bio Application

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Abstract

In this paper, we used soft HSQ (hydrogen silsesquioxane) mold to transfer aminosilane ink on HSQ and glass substrates by nano-contact imprint technology. And the aminosilane pattern line bound with Glutraic dialdehyde (GA) and Affinipure Goat Anti-Mouse igG (H+L) protein sequentially for bio application. The width/height of nano-line before and after bound GA and protein are 133/9nm and 164nm/14nm, respectively. On glass substrate, the width/height of nano-line before and after bound GA and protein are 390/50nm and 429nm/55nm. The layer thickness of GA with protein is about 5 nm. This indicates an approximate monolayer of absorbing igG (H+L) protein on the GA bound to patterned aminosilane nano-line.

Keywords: nano-contact imprint, HSQ, aminosilane, Anti-Mouse igG(H+L) protein

Introduction

The ability to pattern biomolecules on solid substrates has become increasingly important for the development of molecular and cellular biosensors. From nanoscale devices, gene chips and biosensors improve their surface density and decrease the probe volumes, and fabrication of nano-electrode arrays increases the information obtained in electrophysiological studies. And a major advantage of the nanoimprint techniqueis that the feature size can be reduced to the nanoscale to create high-density arrays, or potentially control placement of individual proteins, while still retaining high throughput and reproducibility. A. Ruiz et al[1]. fabricated PDMS mold of nano-domes structure. The stamp was inked with poly-L-lysine and then put it contact with PEG layer on glass substrate. The patterns created by pressing the stamps with 50g weight and have a diameter 300nm. Mateu Pla-Roca et al.[2] used PMMA stamps with line/space ratio of 1:2 immersed into the protein solution then to print line of Streptavidin protein ~150nm. The contact printing pressure is 7.5×10^5 pa. J. Damon Hoff et al.[3]used present a flexible technique for selectively patterning bioactive proteins with nanoscale resolution using nanoimprint lithography. They used Si mold and an aminosilane monolayer is covalently attached to the patterned regions. Biotin and streptavidin were sequentially linked to the aminosilane layer. Finally, the biotinylated target protein is bound to the streptavidin layer. Their feature sizes could be down to 75 nm.

For our study, we developed nano-contact imprint technology by using soft HSQ (hydrogen silsesquioxane) mold transfer aminosilane ink on HSQ and glass substrates. Soft HSQ molds were fabricated by low-dose e-beam lithography[4]. A flexible stamp is in favor of quality contact printing. And the HSQ mold is also durability and repeatability. The nano-features of aminosilane on the HSQ stamps were transferred onto the substrates with low printing pressure. The surface of the substrate was modified by O_2 plasma with various power. Then aminosilane nano-line can be bound with Glutraic dialdehyde (GA) and Affinipure Goat Anti-Mouse igG (H+L) protein sequentially for bio application. Then we used AFM to measure the nano-line before and after soaking with GA and Anti-Mouse igG(H+L) protein. The mold fabrication, printing process and experimental results are presented in the following sections.

Experimental

Fig. 1 is the process flow. In fig. 1(a), we defined the pattern on HSQ (Fox-15, Dow Corning) by the e-beam lithography of low dose with the ratio of line width and space is 1:1 and 1:10. The e-beam dose is 360μC/cm^2, and 280μC/cm^2 for the 1:1 and 1:10 space/width ratio, respectively. The HSQ film spin-coated on six inch wafer and prebake temperature is 120^{0}C, 3min. Then different TMAH developer concentration was used for wet etching for 1:1 and 1:10 mold individually. Table 1 is the conditions of template fabrication. HSQ molds were fabricated with 100nm~200nm wires width.

In fig. 1(b), aminosilane was studied as ink for nano-contact imprint. We use the mixture of aminosilane (Sigma-Aldrich) and ethanol. The rate of mixture is 0.02c.c : 10c.c for 30min stirring. A thin layer of ink was spin-coated onto a SiO_2 inker pad. The HSQ mold was press against the inker pad by NX-2000 system (Nanonex corp.), then ready for printing.

In order to enhance the adhesion, surface modification on HSQ and glass substrate is needed. There are two conditions of O_2 plasma to treat HSQ and glass substrate. The HSQ and glass substrate are treated with O_2 plasma power 350W and 450W for 30s in fig. 1(c).

In fig. 1(d), we use NX-2000 system (Nanonex corp.) to transfer the aminosilane ink from mold to the HSQ and glass substrate. The conditions are 35psi and 10psi for 30s on HSQ and glass substrate individually.

Finally, the aminosilane nano-line can be bound with Glutraic dialdehyde (GA) and Affinipure Goat Anti-Mouse igG (H+L) protein sequentially. During the protein chemical absorption process, dropped the 5%GA on the aminosilane line waiting for two hours and then rinsed with DI water. Then we dropped the 0.0125 mg/c.c protein to bind on the GA waiting for four hours and rinsed with DI water again. After N_2 drying, the AFM are also performed in order to observe to protein absorption test. The width and thickness of transferred feature can be measured by using AFM.

Table 1 Conditions of HSQ mold fabrication

Mold	Dose	Develop condition
1:1	360μC/cm^2	25% TMAH, 47^{0}C,25s
1:10	280μC/cm^2	5% TMAH, RT ,20s

Fig1. (a)HSQ mold fabrication

Fig1. (b) Ink preparation:aminosilane with ethanol (0.02C.C+10 C.C.)

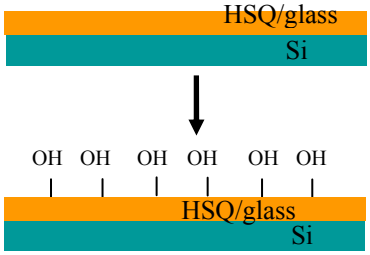


Fig1. (c) O_2 plasma treatment

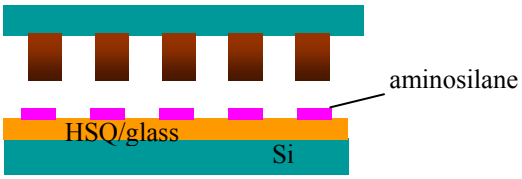


Fig1. (d) Nanocontact imprinting

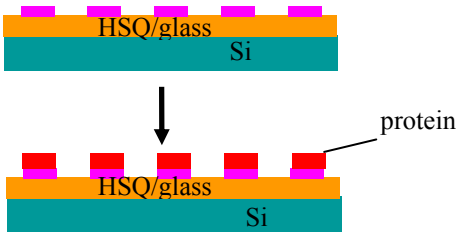


Fig1. (e) Aminosilane bound with GA and protein

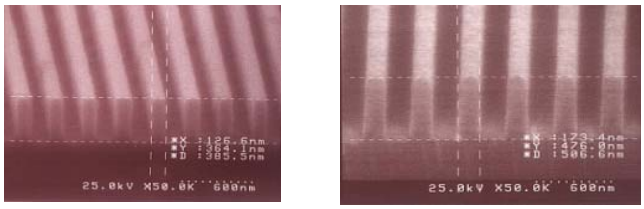
Fig. 1 Process flow

Results and Discussions

HSQ template

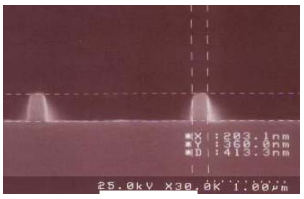
Figure 2 (a) and (b) show HSQ template with line/space ratio of 1:1 with line width 126nm and 173nm. And figure 2 (c) shows template with line/space ratio of 1:10 with line width 203nm.

From fig.2 and 3, because the modulation transfer function (MTF) will decrease as line width getting smaller for dense pattern. So that higher developer concentration and temperature must be used to increase etching rate for 1:1 mold. And in order to obtain the vertical sidewall, rapid development time is also needed.



(a) (b)

Figure. 2 SEM images of dense HSQ template with line/ space ratio of 1:1 (a) 126nm (b) 173nm

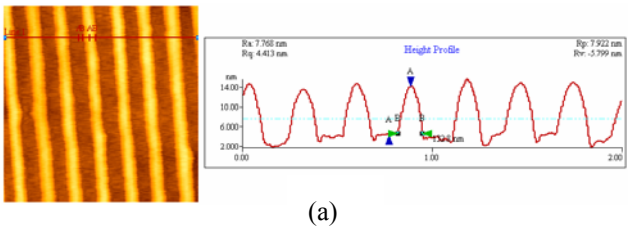


(c)

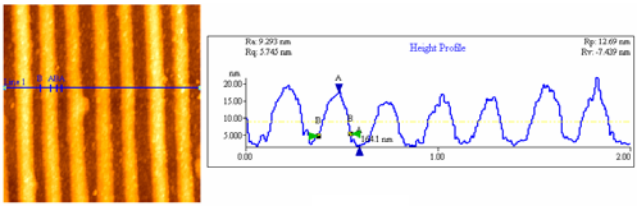
Nano-contact imprint

To form the aminosilane nano-line, the ink molecules must bind covalently to the HSQ and glass substrate surface. The linkage between the ink molecules to the substrate surface is through the reaction between the methoxy groups of the ink and the hydroxyl groups on the substrate. The O_2 plasma treatment can break the Si-H bond and creates the Si-OH and H-OH bonds on the HSQ film[5] to help the ink adhesion. Besides O_2 plasma intensity, imprinting force and time are also the critical factors at the ink-susbtrate interface.

Figure 4 (a) shows the AFM image of a 1:1 mold with 126nm to transfer 133nm nm width and 9nm height aminosilane line on HSQ substrate, and used with conditions of 350W O_2 plasma treatment for 30 s and 35 psi imprinting force for 30s. Figure 4 (b) shows the AFM image of a 133nm line transferred pattern of aminosilane after the Glutraic dialdehyde (GA) and Affinipure Goat Anti-Mouse igG(H+L) protein dropping onto the line pattern. The width and height protein line are 164nm and 14nm. Because aminosilane is composed of amino function group and hydrolysable alkoxy group. The former can interact with the GA and protein. The GA is the cross–linker between aminosilane and Anti-Mouse igG (H+L) protein[6]. The layer thickness of GA with protein is about 5 nm. It can be clear seen that the protein preferentially to the aminosilane molecules.



(a)



(b)

Figure 4. AFM image of (a) transferred pattern with aminosilane line on HSQ. (b) the line patterns after the chemical absorption of GA and protein on HSQ.

And a 1:1 mold with 173nm to transfer 179 nm width and 9nm height aminosilane line on HSQ substrate also be done. The O_2 plasma treatment and imprinting conditions are the same with figure 4. The 179nm aminosilane line after the GA and protein dropping, the width and height protein line are 188nm and 14nm.

NSTI-Nanotech 2008, www.nsti.org, ISBN 978-1-4200-8504-4 Vol. 2

Besides HSQ substrate, the 1:10 mold with 203nm also transfer 390 nm width and 50nm height aminosilane line on glass substrate. The condition of O_2 plasma treatment is 450W for 30 s and 10 psi imprinting force for 30s. The 390nm aminosilane line after bound with the GA and protein, the width and height protein line are 429nm and 55nm. Table 2 collects the AFM results of nano line before and after bound Glutraic dialdehyde (GA) and Anti-Mouse igG(H+L) protein on HSQ and glass substrate.

Table 2 Dimension of nano line before and after bound GA and protein

Substrate	Nano line formed by contact imprint	Line width	Line height
HSQ	Aminosliane	133nm	9nm
	Aminosliane+GA+protein	164nm	14nm
	Aminosliane	179nm	9nm
	Aminosliane+GA+protein	188nm	14nm
Glass	Aminosliane	390nm	50nm
	Aminosliane+GA+protein	429nm	55nm

Because aminosilane is composed of amino function group and hydrolysable alkoxy group. The later can interact with the hydroxyl groups on a hydroxylated surface. From the AFM results, the hydroxyl groups of this aminosilane ink ensured the well-formed nano-line by contact printing technology. Then we defined the patterning of proteins on a planar substrate by amino function group of aminosilane pattern line to immobilize biomolecules. The thickness of GA and protein is about 5nm. As we know, the size of the Anti-Mouse protein is 5nm×5nm×15nm. And the thickness of GA is also less than 1nm. This indicates an approximate monolayer of absorbing igG (H+L) protein on the GA bound to patterned aminosilane nano-line.

Compare the results of nano-line dimension on the HSQ and glass substrate. The feature sizes of nano-lines on glass substrate were larger than others. Because we used a larger O_2 plasma power treatment on glass substrate. That will induce more hydroxyl group to enhance the adhesion between the ink and substrate to increase the line width in our obvious study[7]. In order to scale down the aminosilane line width on glass substrate, we will reduce the O_2 plasma power on substrate. And after binding protein, we must clean the substrate to remove excess protein without bound to aminosilane. If it was not removed completely, that will influence the line feature size, too. To fit the nano feature requirement, the conditions of O_2 plasma power, stamping force and protein absorption must be optimized.

Conclusions

In this paper, nanocontact printing using HSQ soft mold and ink aminosilane was studied. The molds were fabricated by low-dose e-beam lithography with 100nm~200nm width, the ratio of line width and space is 1:1 and 1:10. The HSQ stamp is in favor of quality contact printing. For aminosilane ink, a strong O_2 plasma treatment on the HSQ and glass substrate provides a possible nanoscale contact printing. The transfer line dimension increased with increasing plasma intensity. The O_2 plasma power and imprinting force will be optimized for our experiment. We also made the protein absorption on aminosilane ink successfully. In the future, nano-feature size for bio application on sensors and facilitate studying will be developed further by using nano-contact imprint technology.

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