Immobilization of DNA on arrayed SiO_2 dots prepared by AFM anodic oxidation

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Introduction

Patterning of biomolecules on Si has been reported in relation to fabrication of biosensors [1]. In recent years, there is also an emerging interest in DNA / protein chips and microfluidic devices. For miniaturization and integration of such devices, patterning of biomolecules at a high spatial resolution is required.

In this study, protocols for patterning of biomolecules based on the AFM (atomic force microscope) anodic oxidation process [2-4] were developed.

AFM Anodic Oxidation

By applying a dc voltage between the Si substrate and the conductive AFM tip, the surface of the Si substrate is locally oxidized by anodic oxidation mediated by the adsorbed water (Figs.1-3).

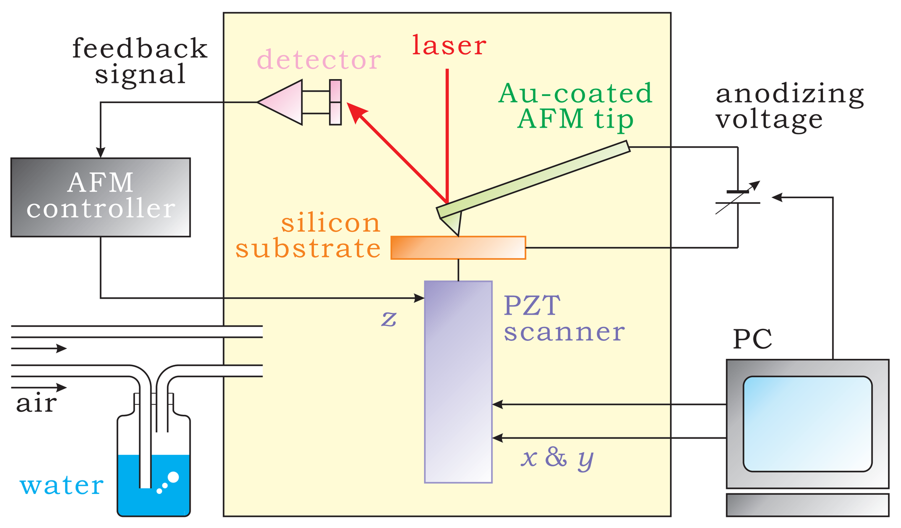

Fig.1 The block diagram of the AFM anodic oxidation system.

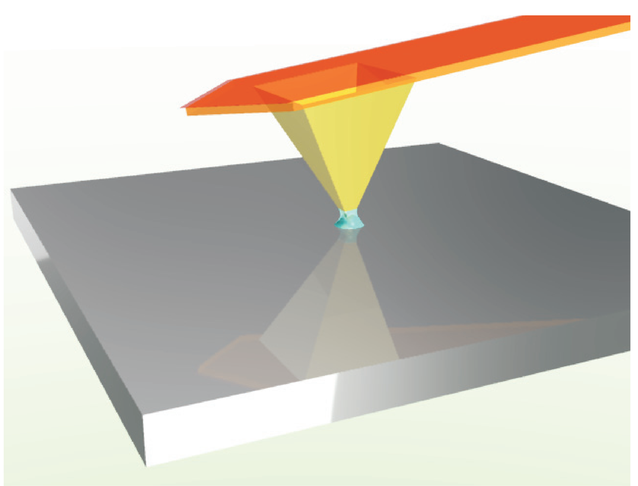

Fig.2 Schematic drawing of adsorbed water between the Si surface and the AFM tip.

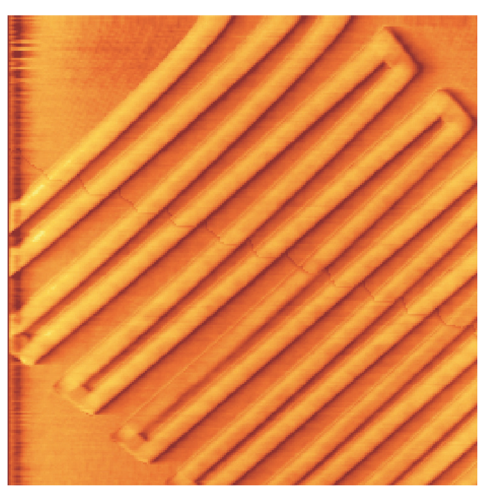

Fig.3 Example of oxide pattern drawn on Si by AFM anodic oxidation.

Materials

substrate: n-type Si(111)
8-12 Ωcm, 250 μm

protein: ferritin
4.6×10^5 Da, ϕ=12nm

DNA: λ-phage DNA
48.5kbp, L=16μm

Protocols

The oxide patterns drawn by AFM anodic oxidation are used as templates for immobilization of biomolecules with the help of linker molecules.

Amino and aldehyde groups are introduced with γ-aminopropyltriethoxysilane (γ-APTES) and glutaraldehyde, respectively. To prevent adhesion of biomolecules on the Si surface, octadecyltrichlorosilane (OTS) is used.

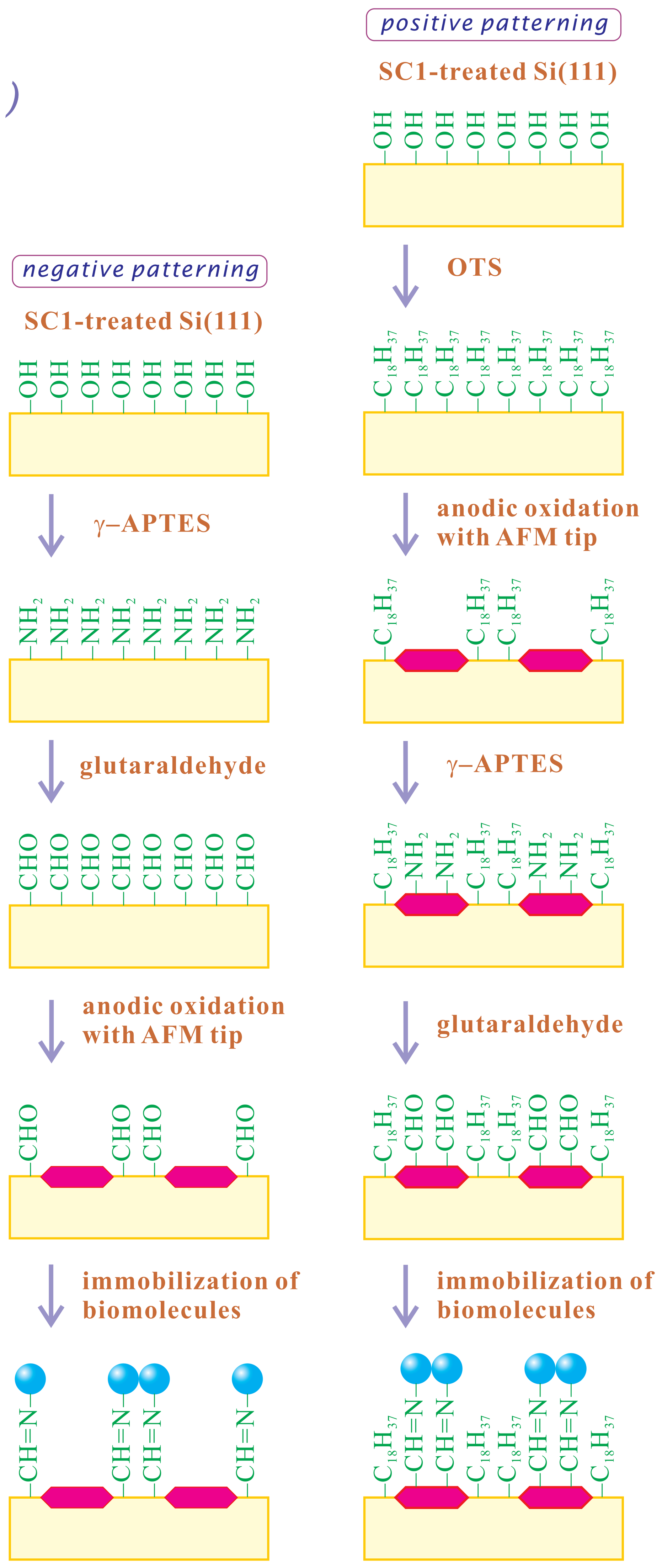

Fig.4 Protocols for negative (left) and positive (right) patterning of biomolecules.

Immobilization of Protein Molecules

Figure 5 shows various examples of protein patterns obtained by the present method.

In the case of negative patterning, protein molecules could be confined inside narrow lines as thin as 50 nm.

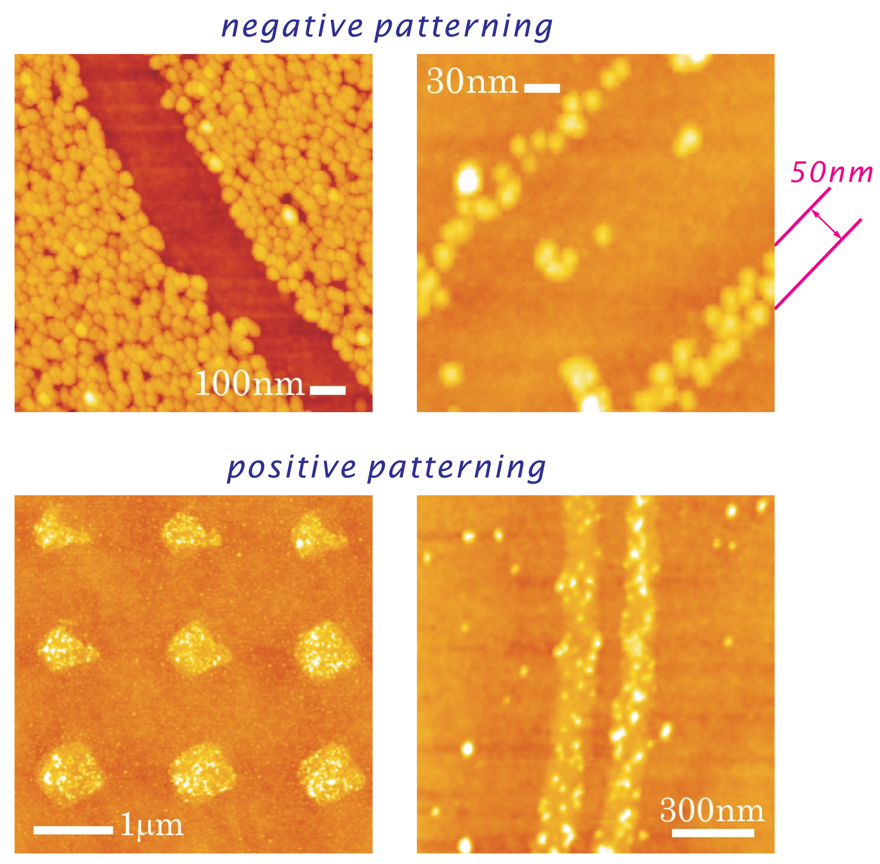

Fig.5 Negative and positive patterning of protein molecules.

Immobilization of DNA Molecules

The present method was applied to immobilization of DNA molecules on arrayed SiO_2 dots. When the dots were treated with γ-APTES and GA, the "DNA wire" structure was formed.

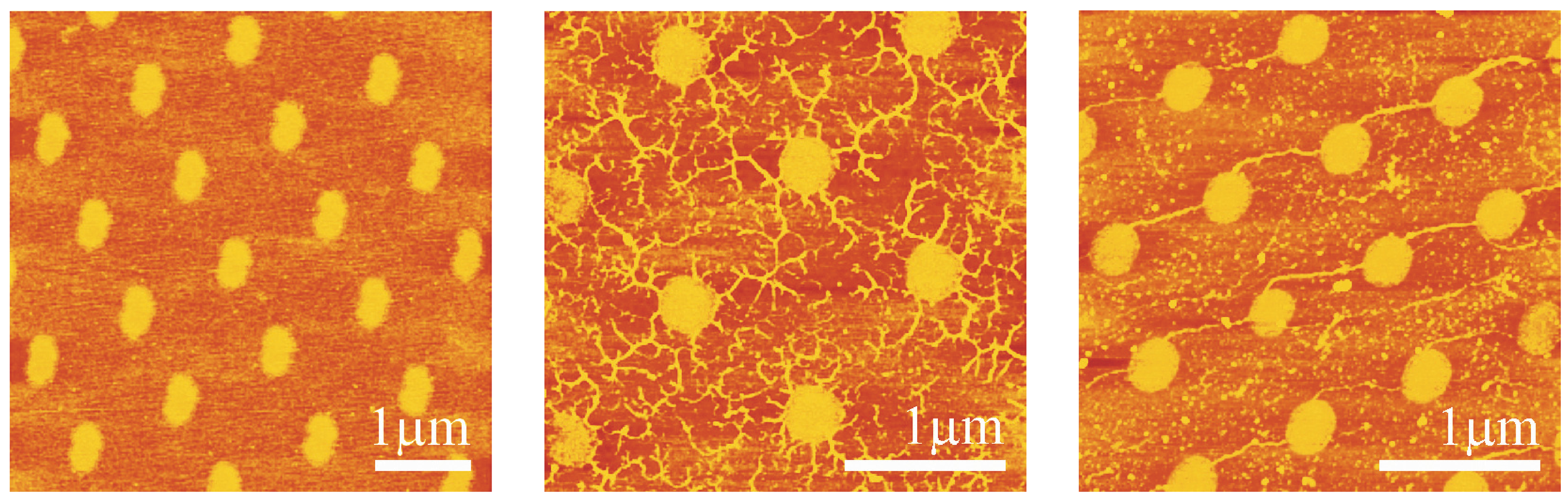

Fig.6 Immobilization of DNA molecules on arrayed SiO_2 dots. (left) without linker (middle) treated with γ-APTES (right) treated with γ-APTES and GA.

Figure 7 compares DNA molecules immobilized on arrayed SiO_2 dots with different densities. The DNA molecules form straight or zigzag lines depending on the density.

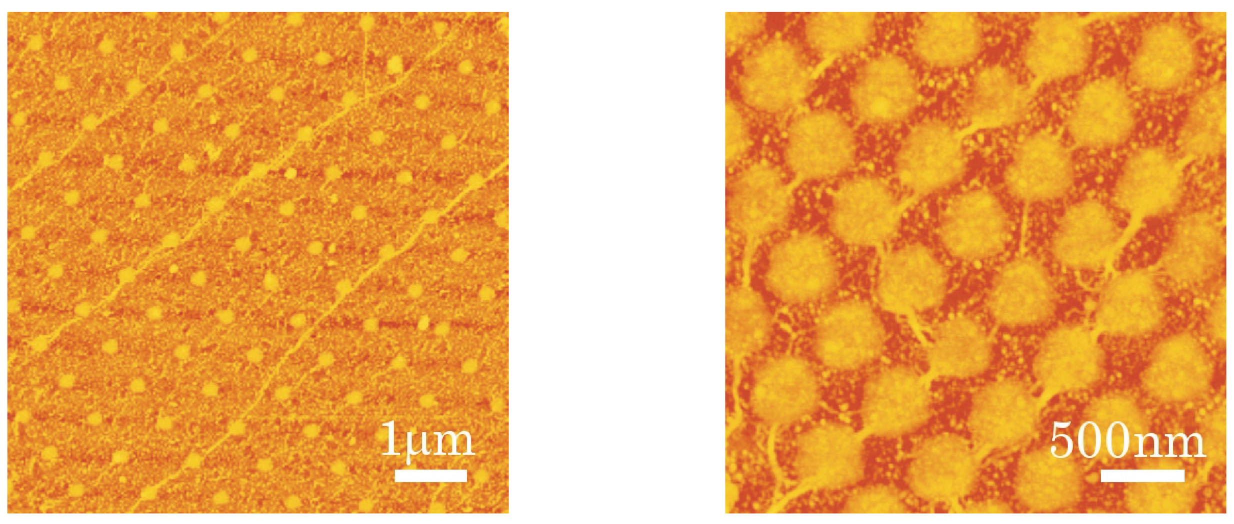

Fig.7 AFM images of λ-phage DNA molecules immobilized on arrays of oxide dots with different densities.

By using the SiO_2 dots as "stepping stones", wiring of two points with DNA along an arbitrary path on the surface may be possible.

Summary

The oxide patterns drawn by anodic oxidation with an AFM tip can be used as templates for immobilization of biomolecules. "DNA wire" structures were obtained on arrayed SiO_2 dots.

References

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