Jpn. J. Appl. Phys. Vol. 39 (2000) pp. L 269–L 270
Part 2, No. 4A, 1 April 2000

Express Letter

A New Self-Fabrication of Large-Scale Deoxyribonucleic Acid Network on Mica Surfaces

Takashi KANNO, Hiroyuki TANAKA, Norio MIYOSHI[1] and Tomoji KAWAI
The Institute of Scientific and Industrial Research, Osaka University, Mihogaoka, Ibaraki, Osaka 567-0047, Japan
[1]*Department of Pathology, Fukui Medical University, Yoshida-gun, Fukui 910-1193, Japan*

(Received March 1, 2000; accepted for publication March 8, 2000)

We have successfully fabricated large-scale deoxyribonucleic acid (DNA) networks on mica surfaces using a simple and easy fabrication method for the first time. Sample drops of poly(dA-dT)·poly(dA-dT) which is a synthetic linear DNA were spotted on freshly cleaved mica and blown off with air. At low DNA concentrations, clusters of DNA molecules were separated from each other. However at high concentrations, substrates were covered with a two-dimensional DNA network measuring more than 12 mm laterally. The DNA network discovered in our study seems highly practical due to its simple and easy fabrication method and its length. We believe this DNA network has the potential to serve as a biomaterial for medical, engineering and environmental applications.

KEYWORDS: deoxyribonucleic acid (DNA), poly(dA-dT)·poly(dA-dT), mica, surface atomic force microscopy (AFM), DNA network, DNA film

1. Introduction

Studies of deoxyribonucleic acid (DNA) on the genetic level are making dramatic progress along with genetic engineering and molecular biology. DNA is the most important material in life science. However, DNA can also be regarded as a naturally occurring and highly specific functional biopolymer. In Nishi's studies, DNA immobilized on substrates and fibers can be utilized as a captor for endocrine disruptors,[1)] as an antigen for detecting anti-DNA antibodies[2–4)] and as an antibacterial film.[5)] When it has high conductivity, DNA can be utilized as an electric circuit in itself. Even with low conductivity, DNA can be utilized as an ultra-minute molecular circuit after modification using other molecules.[6)] DNA film and networks therefore have the potential to serve as biomaterials for medical, engineering and environmental applications. Although many researchers have tried to fabricate DNA film and/or networks, a simple and easy method of fabricating DNA film and/or network uniformly over the entire substrate surface has not been found. For example, although Seeman reported the fabrication of DNA networks,[7,8)] in his studies the edges left for applying paste (special single-strand DNA, well designed in base sequence) are necessary to link the DNA molecule with another molecule to form a network. Moreover, the DNA network fabricated by Seeman measured $2000 \times 2000\,\mathrm{nm}^2$, which is too small for practical use.

Our method of fabricating DNA networks involves simply spotting a high-concentration poly(dA-dT)·poly(dA-dT) solution on freshly cleaved mica and blowing it off with air. In addition, our DNA network is sufficiently large-scale to cover entire substrates ($12 \times 12\,\mathrm{mm}^2$). Most significantly, the DNA network discovered in our study is highly practical due to both its simple and easy fabrication method and its large scale. This makes it highly suitable for industrial applications as well as scientific research.

2. Experimental

The DNA sample used in our study was poly(dA-dT)·poly(dA-dT) (Amersham Pharmacia Biotech Co., Tokyo, Japan). The mean length of poly(dA-dT)·poly(dA-dT) was 1534 bp. We diluted it with 17.4 MΩ deionized water to concentrations of 25 and 250 μg/ml. Freshly cleaved mica (muscovite green mica, Nilaco Co., Japan) was used as substrates for atomic force microscopy (AFM) observation. AFM images were obtained using an SPI3700-SPA300 (Seiko Instruments, Chiba, Japan) in "dynamic force microscope" mode. The scanning tip used was an Si microcantilever 450-μm long with a force constant of 1.8 N/m at a scan rate of 1–2 Hz.[9)] A 10-μl sample drop was spotted on freshly cleaved mica and spread over a diameter of approximately 12 mm. The sample solution was allowed to remain on the substrates for about one minute, after which it was blown off with air and studied using AFM. All the experiments, sample preparation and AFM imaging were conducted in air.

3. Results and Discussions

Figure 1 shows AFM images of poly(dA-dT)·poly(dA-dT) at concentration of 25 μg/ml. Clusters of DNA molecules were separated from each other (Figs. 1(a) and 1(b)). The mean height of DNA was 1.0 nm (Figs. 1(a) and 1(b)), indicating that the clusters mainly comprised a single layer of DNA molecules.[10–12)] As the molecule density is high at the edge of the sample drop,[13)] the DNA molecules are linked with another molecules (Figs. 1(c) and 1(d)). It should be mentioned that for plasmid DNA, its two-dimensional network is not linked with another molecules even at the edge of the sample drop.[13)] The major difference between poly(dA-dT)·poly(dA-dT) and plasmid DNA is the topological conformation: the former has a linear structure with two ends whereas the latter has a circular structure with no end. This structural difference may be responsible for the network formation.

Figure 2 shows AFM images of poly(dA-dT)·poly(dA-dT) at concentration of 250 μg/ml. The substrates are covered with a two-dimensional DNA network which spreads over about 12 mm uniformly. The histogram reveals that the height of DNA was about 1.8 nm, indicating that the network comprises a few DNA molecules oriented perpendicularly to the substrate surface.

4. Conclusions

We have successfully fabricated large-scale ($12 \times 12\,\mathrm{mm}^2$) DNA networks on mica surfaces using a simple and easy

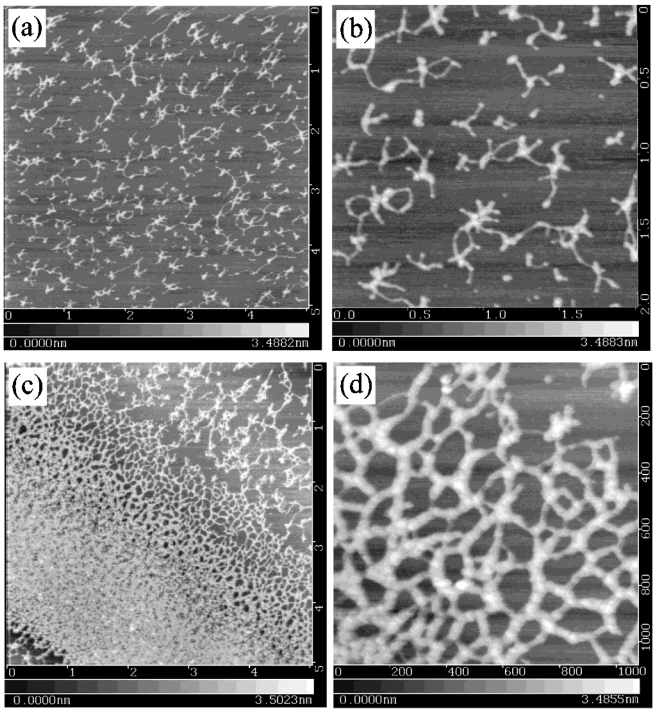

Fig. 1. AFM images of poly(dA-dT)·poly(dA-dT) spread on mica. The DNA concentration was 25 μg/ml. (a) Clusters of DNA molecules were separated from each other ($5000 \times 5000 \times 3.5\,\mathrm{nm}^3$). (b) A magnified image ($2000 \times 2000 \times 3.5\,\mathrm{nm}^3$) of (a). The mean height of DNA was 1.0 nm. (c) At the edge of the sample drop with a very high molecule density ($5000 \times 5000 \times 3.5\,\mathrm{nm}^3$). (d) A magnified image ($1000 \times 1000 \times 3.5\,\mathrm{nm}^3$) of (c). DNA molecules were linked each other.

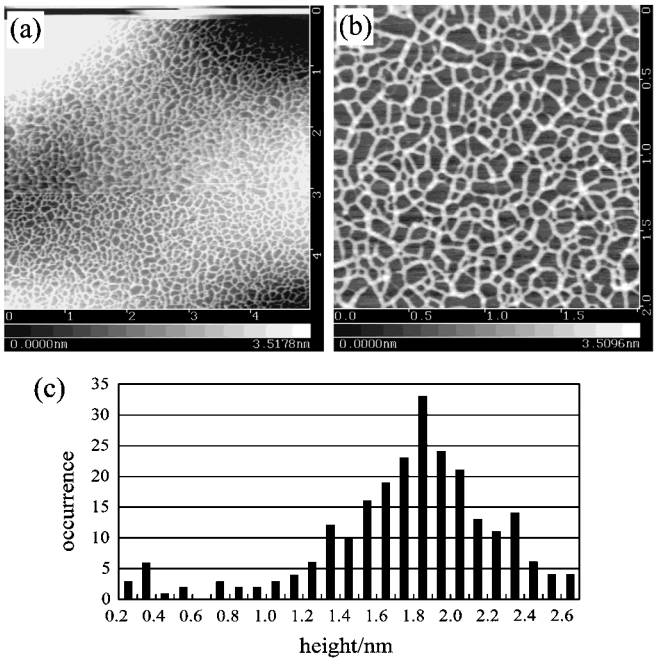

Fig. 2. AFM images of poly(dA-dT)·poly(dA-dT) spread on mica. The DNA concentration was 250 μg/ml. (a) Two-dimensional DNA network covered substrates ($5000 \times 5000 \times 3.5\,\mathrm{nm}^3$). (b) A magnified image ($2000 \times 2000 \times 3.5\,\mathrm{nm}^3$) of (a). The ditch diameter was approximately 100 nm. (c) A histogram of the DNA network. The mean height of DNA was 1.8 nm.

fabrication method for the first time. The sample drops of poly(dA-dT)·poly(dA-dT) were spotted on freshly cleaved mica and blown off with air. We believe that this simple and easy fabrication method and its large scale make it suitable for practical applications such as biomaterials for medical engineering and environmental purposes as well as scientific research.

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