Immobilization of oligonucleotide molecule on oxide surface through organic layers

Ayumi Narita^{1,*}, Kentaro Fujii¹, Akinari Yokoya¹, Yuji Baba² and Iwao Shimoyama² ¹Advanced Science Research Center, Japan Atomic Energy Agency, Tokai 319-1195, Japan ²Quantum Beam Science Directorate, Japan Atomic Energy Agency, Tokai 319-1195, Japan

1 Introduction

DNA damage induced by radiation is thought to cause serious biological effects such as cell lethality or mutation induction. Monochromatic soft X-rays have been used as a probe to control the initial process of DNA damage induction. Fujii et al. reported that yields of radiation damage in DNA including strand breaks and oxidative lesions of nucleobases depend strongly on the soft X-ray energy tuned around K-edges of DNA constituent atoms, namely carbon (280 eV), nitrogen (400 eV) or oxygen (530 eV)[1]. They used the thin layer samples of DNA dropped and dried on an inorganic substrate. The chemical state of the thin layer DNA is substantially different from that in vivo DNA which twines round histone proteins. In order to simulate in vivo DNA, we suggest a new model of the DNA sample using selfassembled monolayer (SAM), which are films voluntarily formed by strong chemical bond and molecular interaction between the molecule and the substrate. Namely, the SAM on the inorganic substrate is likened to histone proteins. For the first step, we present immobilization of oligonucleotide molecules on the inorganic surface through the organic SAM.

2 Experiment

We used mercaptopropyltrimetoxisilane(MPTS) molecules (HS(CH₂)₃Si(OCH₃)₃) as organic layer between the substrate and the oligonucleotide. MPTS molecules have thiol group (SH) and silicon alkoxide (Si(OCH₃)₃) at the respective terminal sites of the alkyl backbone. MPTS molecules were adsorbed on sapphire C-plane by immersing the substrate in 10% ethanol solution. Then, 1 μ M oligonucleotide dissolved in 10 mM Tris-HCl was dropped on the MPTS film. The samples were measured by XPS (X-ray photoelectron spectroscopy) and NEXAFS (near edge X-ray absorption fine structure).

3 Results and Discussion

For S K-edge NEXAFS spectra of the MPTS film on the sapphire surface (Fig.1(a)), a sharp peak appeared at 2472.1 eV. This peak is the resonance from S 1s to σ^* orbital localized in the S-C bond. On the other hand, as to the sample for the oligonucleotide on MPTS films (Fig.1(b)), the intensity of the peak for the S-C bond decreased. This means that the location of the S-C bonds became relatively close to the substrate surface by depositing the oligonucleotide. Concerning XPS spectra of the MPTS film, the clear peaks of S 1s and Si 1s were confirmed (Fig. 2(a)(b)).After depositing the oligonucleotide solution, the intensity of these two peaks decreased, while that of the P 1s peak increased (Fig.

2(c)). These results show that the oligonucleotide molecules are strongly immobilized on the sapphire surface through the MPTS film.

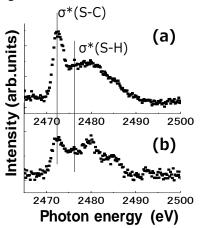


Fig. 1: NEXAFS spectra of S *K*-edge for MPTS film on sapphire (a) and oligonucleotide immobilized on MPTS film (b).

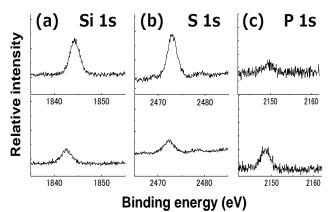


Fig. 2: XPS spectra of Si 1s, S 1s and P 1s for MPTS film on sapphire (upper side) and oligonucleotide immobilized on MPTS film (lower side).

Acknowledgement

This work was supported by JSPS KAKENHI Grant Number 25740020.

References

 K. Fujii, N. Shikazono, A. Yokoya, J. Phys. Chem. B 113, 16007 (2009).

* narita.ayumi@jaea.go.jp