Chemical and Structural Properties of Polyethyleneimine Film Coated on a SiO₂ Substrate in Different Concentrations

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Angle-resolved X-ray photoelectron spectroscopy (AR-XPS) and atomic force microscopy (AFM) were employed to investigate the detailed surface bonding structure and morphology of polyethyleneimine (PEI) layers coated on a SiO₂ substrate in concentrations of 0.5–10.0 mass%. With an increase in the PEI concentration, the C–N bonds corresponding to the imine group (–NH) became dominant in the PEI layers, implying a characteristic feature to verify the immobilization of PEI on the SiO₂ surface. The thickness of PEI layer calculated using AR-XPS gradually increased with increasing PEI concentration up to a maximum 5 mass%, above which it became saturated. AFM results showed that an increase in PEI concentration led to a decrease in the root mean square (RMS) roughness of PEI layers. Such a saturation tendency of PEI thickness combined with the strongly dependence of the surface morphology behavior of PEI layers on the PEI concentration indicated that SiO₂ surface was fully covered by monolayer PEI with a smooth surface morphology at PEI concentration in excess of 5 mass%.


(Received January 9, 2014; Accepted February 18, 2014; Published March 28, 2014)

Keywords: SiO₂, polyethyleneimine, angle-resolved X-ray photoelectron spectroscopy, atomic force microscopy, imine group

1. Introduction

In order to realize highly sensitive, specific, selective, and reliable biosensors, surface linker materials should be employed to immobilize biomolecules such as protein or DNA on a solid surface.¹,² Until now, (3-Aminopropyl)triethoxysilane (APTES),³ polyethyleneimine (PEI),¹–³ polyethylene glycol (PEG)⁴ and polyethylene (PE)¹,⁴ have been proposed as a surface linker material for the sensing or recognition of biological elements in biosensors. Among these materials, PEI has been considered as a viable surface linker owing to its simple controllability and biocompatibility.²,⁶ However, detailed knowledge of the chemical and structural properties of PEI is essential for further enhancement in biosensor performance. Moreover, since the charge transfer through direct linkages between PEI and substrate is a key factor for the operation of biosensors, the optimization of PEI film controlled by its concentration is required to realize the maximum surface coverage of PEI directly adsorbed on substrate. Previously, considerable efforts have been made to understand the nature of PEI film on various substrates. Tang et al. investigated the absorption of citrate-stabilized CdSe or CdSe/CdS core–shell nanoparticles (NPs) on Si wafers, showed that the adsorption layer of the weak poly electrolyte PEI could alter its chain distribution by partial wrapping around the NPs.⁵ Kim et al. showed that a biotinylated gradient PE surface, prepared by the reaction of PEI and biotin, provided a continuous gradient of streptavidin–RITC caused by the avidin-biotin binding.⁶ Reybier et al. demonstrated the detection of a potentiometric pesticide using different electrodes, modified with butyrylcholinesterase immobilized by co-reticulation in the PEI matrix, which was electrochemically deposited on the electrode surface.⁷ In the present study, we investigated the chemical bonding properties of polyethyleneimine coated on a SiO₂ substrate in different concentrations, by using angle-resolved X-ray photoelectron spectroscopy (AR-XPS) and atomic force microscopy (AFM). It will be shown that a > 5 mass% increase in the PEI concentration leads to the formation of a PEI monolayer with a smooth surface morphology, indicating full surface coverage of the SiO₂ substrate by the PEI layer at PEI concentration in excess of 5 mass%.

2. Experimental Procedure

A 4 nm-thick SiO₂ film thermally grown on the p-type Si (001) wafer was used as a starting material. Initially, organic and metallic contaminants were removed by the standard wet cleaning process. The cleaned SiO₂ surfaces were confirmed by XPS measurements. The surfaces were chemically modified with a silanol group (–OH), obtained using a mixture of MeOH and HCl, for 30 min at room temperature.⁸ The branched PEI with a molecular weight of 25,000 g/mol was coated on the SiO₂ surface in different concentrations (0.5, 1.0, 5.0, and 10.0 mass% in 50 mM Na₂CO₃) by the dipping method at pH 8.2 for 3 h.⁹ Finally, PEI was removed using deionized (DI) water, and this was followed by drying using nitrogen gas. The chemical bonding states and quantification of the PEI films coated on SiO₂ surface were analyzed using AR-XPS, which was conducted using a monochromatic Al-Kα radiation source (1486.6 eV) with a hemispherical analyzer. The resolution of the analyzer was 0.47 eV as measured from the full-width-half-maximum (FWHM) of the Ag 3d₅/₂ peak. In order to avoid charging effects, the measured binding energy was calibrated by the C 1s line (hydrocarbon contamination). To increase surface sensitivity, AR-XPS was performed by varying the take-off angle (TOA) in the range of 30–90° with respect to the
electron analyzer. The surface morphologies of PEI layers formed on the SiO2 surfaces were characterized using AFM in contact mode.

3. Results and Discussions

Figure 1 shows high-resolution C 1s AR-XPS spectra at a TOA of 60° for PEI films formed at different concentrations. The dotted and solid lines represent the experimental data and the curve fitted results, respectively. The spectra were fitted by the Shirley method. For all samples, the deconvolution of C 1s AR-XPS spectra clearly revealed the presence of four distinct peaks indicated by C-1, C-2, C-3, and C-4. The assignments of the individual peaks were obtained from previous studies.10-16) The C-1 peak at 285.0 eV could be attributed in part to the C–C bond of PEI and in part to hydrocarbon contamination occurring during atmospheric exposure.12,13,15) The C-2 peak at 286.4 eV could be associated with the C–N bond of PEI corresponding to the amine group (–NH) mainly constituting the structure of PEI.9,12,14,15) In other words, the presence of peak at 286.4 eV is a characteristic feature to verify the immobilization of PEI on the SiO2 surface. The C-3 peak at 287.9 eV corresponds to a chemical bonding state originating from the aldehyde (C=O) group.10-12) The highest binding energy at 288.9 eV (C-4) is mainly due to the carboxylic acid (O–C=O) functional group on the surface.10,11,14,15) The amide group (C=O) and the carboxyl group (O=COH) could have originated from the interaction between reactive PEI radicals and H2O and/or O2 molecules in air. It should be noted here that as shown in Fig. 2, the C–N bond intensity gradually increased with increasing PEI concentration, while the variation in the intensities of other peaks were insignificant. This indicates that an increase in PEI concentration resulted in the improvement of immobilization behavior of PEI on the SiO2 surface.

The effective sampling depth of PEI films formed on the SiO2 surfaces under different PEI concentrations was investigated using XPS with changing TOA. Theoretically, the effective sampling depth d of XPS measurements can be determined using the following equation.

\[ d = 3\lambda \sin \theta \]  

where \( \lambda \) and \( \theta \) are the effective attenuation length and TOA (angle between the photoelectron emission direction and the plane of the sample), respectively.17) In the case of \( \lambda = 2.5 \text{ nm} \) for the C 1s photoelectron excited by Al Ka X-rays in organic materials, the effective sampling depths were calculated to be 3.8 and 7.5 nm for \( \theta = 30^\circ \) and 90°, respectively.17,18) This provides a means to determine the compositional gradients in the outer surface layers. On the basis of this technique, the distribution of the elemental composition and ratio of PEI films formed at PEI concen-
Table 1 Elemental composition and ratio of PEI film formed at PEI concentrations in the range of 0.5–10.0 mass% calculated using AR-XPS with changing TOA.

<table>
<thead>
<tr>
<th>PEI Concentration</th>
<th>TOA (°)</th>
<th>Elemental composition (%)</th>
<th>Elemental ratio</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>O</td>
<td>N</td>
</tr>
<tr>
<td>0.5 mass%</td>
<td>90°</td>
<td>51.93</td>
<td>3.01</td>
</tr>
<tr>
<td></td>
<td>60°</td>
<td>53.78</td>
<td>3.06</td>
</tr>
<tr>
<td></td>
<td>30°</td>
<td>51.8</td>
<td>3.8</td>
</tr>
<tr>
<td></td>
<td>90°</td>
<td>51.32</td>
<td>4.04</td>
</tr>
<tr>
<td>1.0 mass%</td>
<td>60°</td>
<td>50.68</td>
<td>4.17</td>
</tr>
<tr>
<td></td>
<td>30°</td>
<td>47.47</td>
<td>5.07</td>
</tr>
<tr>
<td></td>
<td>90°</td>
<td>50.51</td>
<td>5.76</td>
</tr>
<tr>
<td>5.0 mass%</td>
<td>60°</td>
<td>48.95</td>
<td>5.99</td>
</tr>
<tr>
<td></td>
<td>30°</td>
<td>42.83</td>
<td>7.56</td>
</tr>
<tr>
<td></td>
<td>90°</td>
<td>40.8</td>
<td>8.79</td>
</tr>
<tr>
<td>10.0 mass%</td>
<td>60°</td>
<td>39.53</td>
<td>9.79</td>
</tr>
<tr>
<td></td>
<td>30°</td>
<td>32.74</td>
<td>11.34</td>
</tr>
</tbody>
</table>

Concentrations in the range of 0.5–10.0 mass% were calculated and summarized in Table 1. It is clear that a decrease in TOA led to an increase in the amount of C and N atoms, which are the main constituents of the imine group in PEI, while the amount of O and Si atoms that mainly originated from the SiO₂ film gradually increased with increasing TOA. This indicated that a decrease in TOA is effective in revealing the chemical bonding structure and surface morphology of PEI. In particular, the N/C ratio increased with increasing PEI concentration up to 5.0 mass%, above which any change in the N/C ratio was insignificant. This suggested saturation in the growth of PEI films formed at PEI concentrations in excess of 5.0 mass%, which will be discussed later.

By assuming that X-ray intensity does not diminish within the XPS sampling depth, the film thickness can be evaluated by measuring the peak intensities in XPS spectra for the film and substrate at different TOAs, as proposed by That et al. The relation between the film thickness t and XPS peak intensities for the film and substrate can be described as

\[
\ln \left( \frac{I_t}{I_s} \cdot \frac{1}{K} + 1 \right) = \frac{t}{\lambda} \cdot \sec \theta 
\]

where \(I_t\) and \(I_s\) are the XPS peak intensities for the film and substrate, respectively. Neglecting the hydrogen molecules that are not detectable by XPS, both \(I_{N_1s}\) and \(I_{Si2p}\) are found to be 1/3 upon considering the stoichiometric molar fractions of N and Si species in PEI and SiO₂ structures. The values of the relative sensitivity factor (RSF) are 0.477 for N 1s and 0.328 for Si 2p. A plot of \(\ln \left( \frac{I_t}{I_s} \cdot \frac{1}{K} + 1 \right)\) as a function of sec \(\theta\) should be a straight line with a slope corresponding to the film thickness. From the slopes of the fitted lines in the \(\ln \left( \frac{I_t}{I_s} \cdot \frac{1}{K} + 1 \right)\) vs. sec \(\theta\) plots (Fig. 3(a)), the thicknesses of the PEI films were calculated to be 0.28, 0.44, 0.59 and 0.60 nm at the PEI concentrations of 0.5, 1.0, 5.0 and 10.0 mass%, respectively. It should be noted here that an increase in the PEI concentration led to an increase in the PEI film thickness with a strong tendency toward saturation above the PEI concentration of 5.0 mass%, as shown in Fig. 3(b). In particular, the saturated PEI film thickness was comparable to the thickness of one monolayer of copolymer (VDF-TrFE) formed according to Langmuir–Blodgett method and calculated using XPS. In other words, the PEI film grew linearly at lower PEI concentrations, eventually forming a PEI monolayer at higher PEI concentrations.

The formation of PEI monolayer at PEI concentrations above 5.0 mass% was also confirmed by the AFM measurements. Figure 4 shows the AFM images of PEI films formed at PEI concentrations in the range of 0.5–10.0 mass%. Clearly, the PEI films formed at higher PEI concentrations showed better surface morphologies than those formed at lower PEI concentrations. For instance, as shown in Fig. 5, the corresponding root mean square (RMS) roughness of a film surface monotonously decreased with increasing PEI concentrations up to 5.0 mass%, above which its variation was negligible. In other words, at PEI concentrations in excess of 5.0 mass%, the amount of absorbed PEI molecules was enough to fully cover the SiO₂ surface, implying the formation of a monolayer PEI film with a smooth surface morphology.

4. Conclusion

We investigated the effects of PEI concentration on the chemical bonding structure and surface morphology of PEI...
films formed on a SiO₂ surface, by using AR-XPS and AFM analyses. An increase in the PEI concentration led to an increase in the intensity of the C–N bonding originating from the imine group. This implied an improvement in the immobilization behavior of the PEI film formed on the SiO₂ surface. The thickness of the PEI film increased with increasing PEI concentration up to 5 mass%; however, above this value, the PEI film thickness became comparable to that of one monolayer of the copolymer. AFM measurements showed that the RMS roughness of the PEI film formed on the SiO₂ surface gradually decreased until the PEI concentration reached 5 mass%, above which its variation was insignificant. By taking into account the saturation tendency of the PEI thickness coupled with the surface morphological evolution caused by variations in the PEI concentration, it can be stated that a PEI monolayer with a smooth surface morphology was fully absorbed on the SiO₂ surface at PEI concentration in excess of 5 mass%.

Acknowledgments

This research was supported by Human Resource Development Program (20124030200080) of the Korea Institute of Energy Technology Evaluation and Planning (KETEP) grant and Industrial Core Technology Program (10045216) funded by the Ministry of Trade, Industry and Energy, Republic of Korea. It was also supported by Technology Innovation Program (Grant No. 10041066) funded by the Ministry of Science, ICT & Future Planning, Republic of Korea.

REFERENCES