Electronic Supplementary Material (ESI) for Journal of Materials Chemistry B.
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Supporting information

Signal amplified two-dimensional photonic crystal biosensor immobilized with glyco-nanoparticles

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1. Evaluation of the holes in the structural-color films using AFM

The holes in the surfaces of the structural-color films were evaluated using AFM2 (Fig. S1 and S2). Regularly arranged hole patterns were observed in the unmodified structural-color film (Fig. S1 (a)). The diameter and depth of the holes were 230 nm and 200 nm, respectively. Regularly arranged hole patterns were observed in the ManHP- and ManNP-immobilized structural-color films, similar to the unmodified structural-color film (Fig. S1 (b) and (c)). Similar behaviors were observed after the glycopolymer-immobilized films were immersed in a 1 g L^{-1} ConA solution (Fig. S2).

The initial diameter and depth of the holes were 230 nm and 200 nm, respectively. Even after the immobilization of the glycopolymers, no drastic change was observed in the diameter and depth of the holes. The immobilization of the glycopolymers on the structural-color films was demonstrated in the XPS and AFM results (Figs. 2, 3, and 4). It is likely that the glycopolymers were immobilized on the top surface, and on the bottom of the holes. This retention of the holes is important in biosensors with structural color, because the expression of the structural color is due to the regularly arranged microstructure. The retention of the holes in the glycopolymer-immobilized structural-color films suggested that the sensing surface had a sufficient ability to express structural color. The holes still remained even after the glycopolymer-immobilized films were immersed in a 1 g L^{-1} ConA solution. This suggested that high concentrations of ConA would still be detectable.

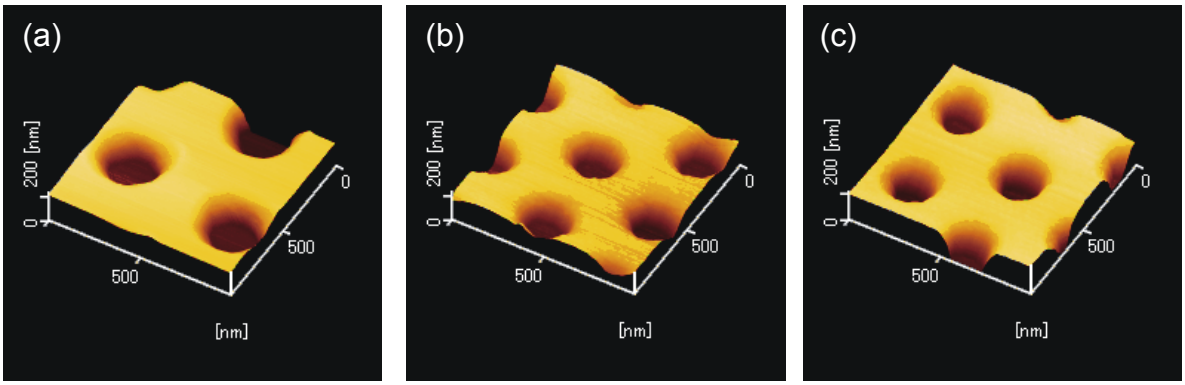

Fig. S1 AFM images of (a) unmodified, (b) ManHP-immobilized, and (c) ManNP-immobilized structural-color films.

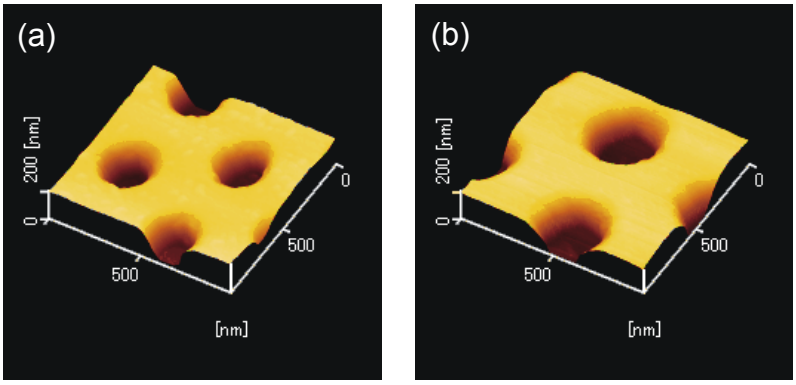

Fig. S2 AFM images of (a) ManHP- and (b) ManNP-immobilized structural-color film surfaces after the adsorption of ConA.

2. Analysis of the interaction between the mannose-incorporating polymers and proteins using QCM

QCM measurement results with unit of x-axis transformed into g L^{-1} is shown in Fig. S3.

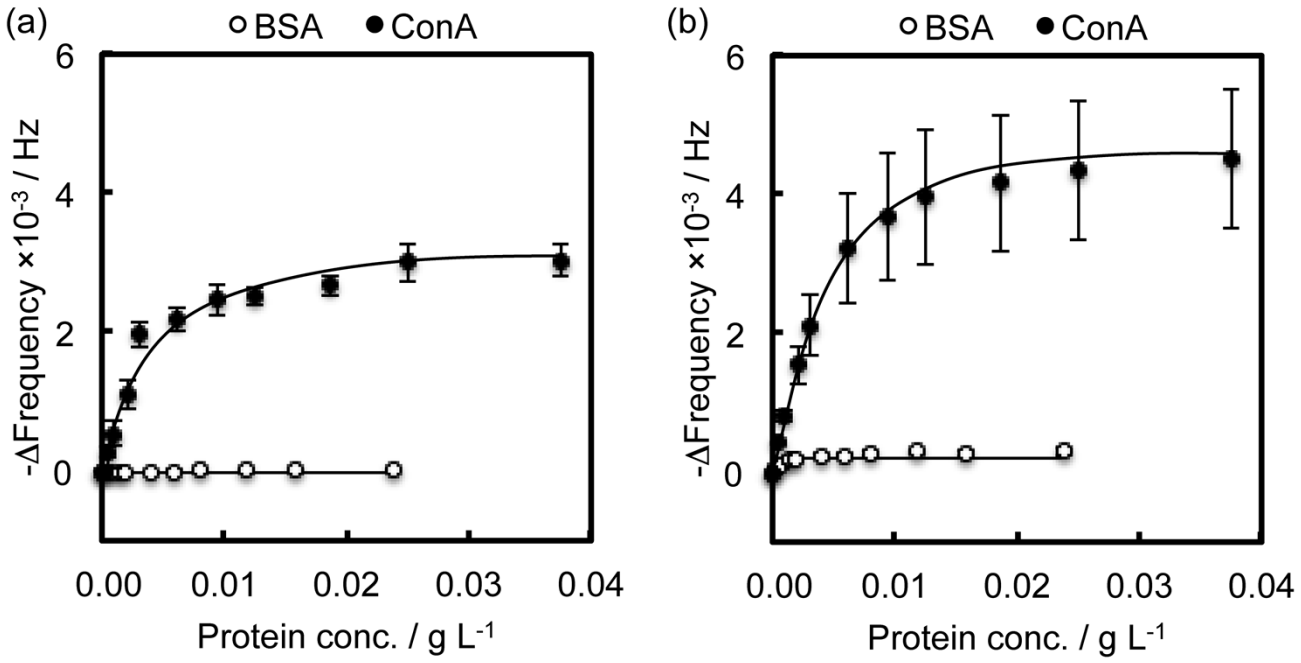

Fig. S3 QCM frequency changes produced by the adsorption of proteins on (a) ManHP- and (b) ManNP-immobilized Au electrodes.

3. Detection of protein adsorption on structural-color films via the monitoring of changes in the reflection intensity of the structural color

To achieve the detection of proteins using structural-color films without glycopolymers, the changes in the reflection intensity of the structural color were measured (Fig. S4). In the unmodified structural-color films, the reflection intensity increased from 10^{-4} g L^{-1} to 1 g L^{-1} with the addition of both ConA and BSA (Fig. S4 (a)). In the UV/O_3-treated structural-color films, no change in the reflection intensity of the structural color was observed with the addition of proteins (Fig. S4 (b)). In the ConA-immobilized structural-color films, no changes in the reflection intensity of the structural color were observed with the addition of proteins (Fig. S4 (c)).

Both ConA and BSA adsorbed onto the unmodified structural-color films. The structural-color films were composed of cyclo-olefin polymers, and the surfaces of the structural-color films were therefore hydrophobic. Proteins were non-specifically adsorbed on the structural-color films via hydrophobic interactions. In the UV/O_3-treated structural-color films, no change in the reflection intensity was observed with the addition of either ConA or BSA. After UV/O_3 treatment, the structural-color films were oxidized, and hydroxyl groups were formed on the surfaces. The surfaces of the structural-color films thus became hydrophilic, and the proteins could not adsorb onto the

structural-color films via hydrophobic interactions. In the ConA-immobilized structural-color films, no change in the reflection intensity was observed with the addition of either ConA or BSA. This suggested that no succinimide groups remained on the ConA-immobilized structural-color film surface. Succinimide groups were used to bind the ConA, and were also hydrolyzed. These results suggested that specific adsorption of proteins could not be observed without the immobilization of glycopolymers on the structural-color films. Glycopolymers are necessary for the specific detection of proteins.

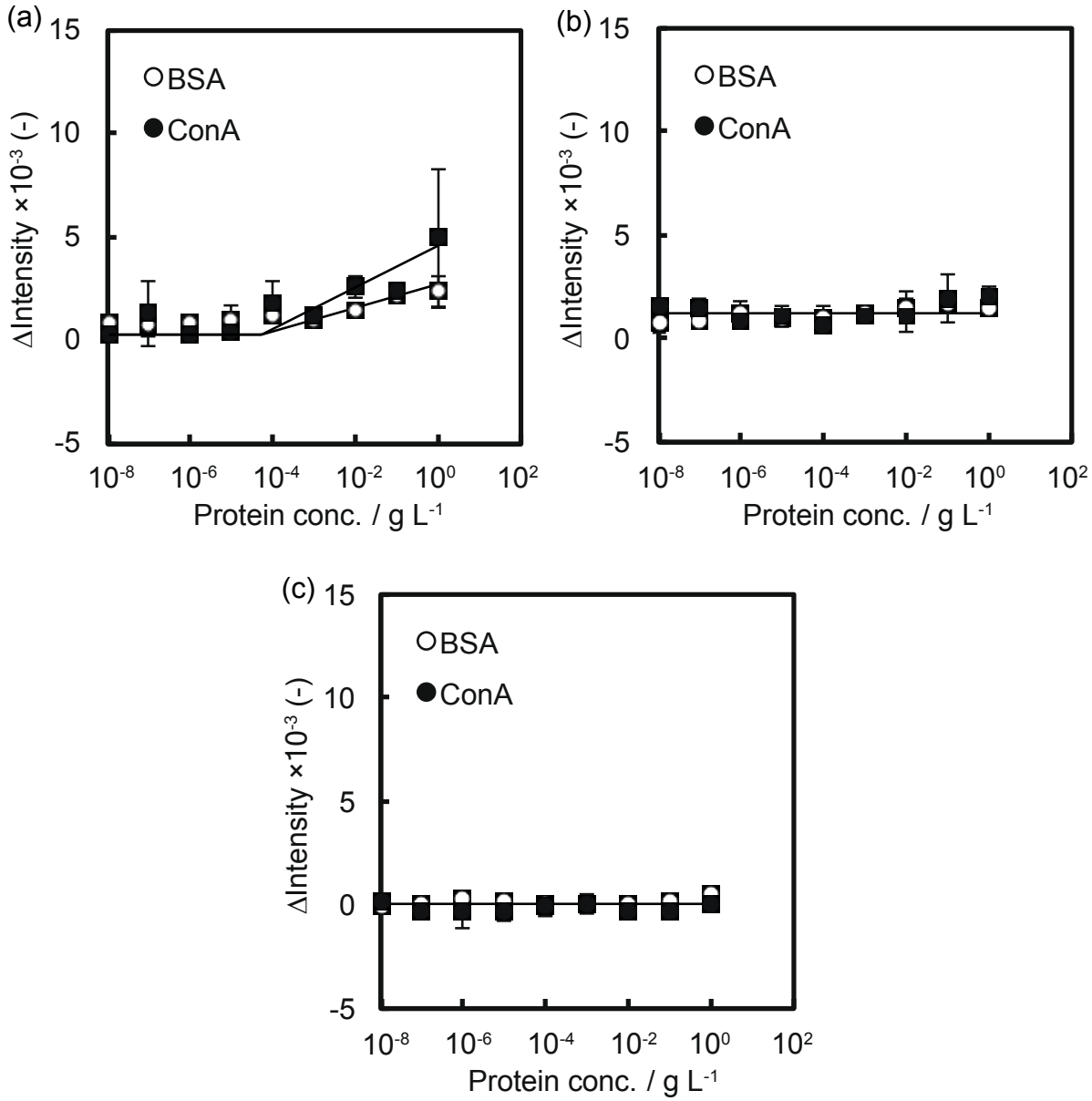

Fig. S4 Changes in the reflection intensity of the structural color produced by the adsorption of proteins on (a) unmodified, (b) UV/O_3-treated, and (c) ConA-immobilized structural-color films.

4. Reflection spectrum of the PhC film of experimental setup for optical charactarization

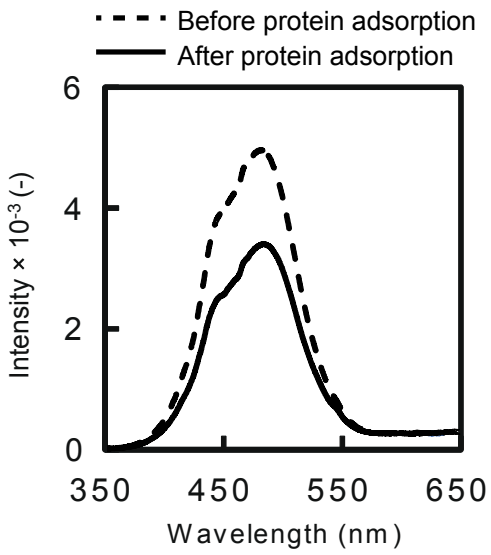

Fig. S5 Reflection spectrum of the PhC film of experimental setup for optical characterization. The broken line shows the reflection spectrum of **ManNP**-immobilized PhC film, and the normal line shows the reflection spectrum of **ManNP**-immobilized PhC film after immersing into 1 g L^{-1} of ConA solution. The peak wavelength could be observed in the visible region.