INTEGRATED pH AND OXYGEN SENSOR ARRAY PREPARED BY MICROCONTACT DOUBLE PRINTING
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ABSTRACT
The integrated optical pH and oxygen sensor array on which pH and oxygen sensor spots whose diameter is 10 μm were arranged alternately on one chip, was developed by using microcontact “double” printing. FITC based pH sensor and ruthenium complex based oxygen sensor was stamped onto a DLC sputtered slide glasses. After the printing of oxygen sensor spots, pH sensor spots were stamped between oxygen sensor spots by using the same PDMS stamp. The response was measured by using a micro array scanner. pH sensor spots responded to only pH changes and oxygen sensor spots responded to only oxygen concentration changes.

KEYWORDS: Microcontact printing, pH sensor, Oxygen sensor, Sensor array

INTRODUCTION
We have developed microarrayed chemical sensors for parallel monitoring of single cell activity [1]. These sensors consist of optical sensor film for pH or oxygen, and micro well array prepared with PDMS. By arranging these chemical sensor film as a million of 10 μm i.d. spots on one chip, novel chemical imaging device might be realized. Integration of several kinds of such sensor spots on one chip might open new bio imaging. Microcontact printing technology [2] using PDMS stamps is easy and simple technique for micro patterning of functional chemicals. In this study, 10 μm i.d. pH and oxygen sensor spots were arranged alternately on one chip by using this technique.

EXPERIMENTAL
PDMS stamp was replicated from a mold. Molds were prepared with SU-8 25 (MicroChem Corp.) by photolithography. Prepared mold is 10 μm i.d. micro well array and well pitch is 40 μm. The prepared stamp was set to the micro contact printer (PA 400, Nanotech Corp.). 0.2 mg/ml FITC solution (pH9.5, 0.05M carbonate buffer ) was used as a pH sensor reagent. 0.5 mg/ml dichlorotris(1,10-phenanthro-line) ruthenium (II) solution (2.5% Nafion solution) was used as an oxygen sensor reagent. These sensor reagents were stamped onto a DLC (diamond like carbon) sputtered slide glass (Gene slide, Toyo Kohan Co.Ltd.). Fluorescence intensity of each spot was measured by using a laser confocal high resolution microarray scanner (CRBIOIle-FITC, Hitachi Soft Corp.)(Ex. 473nm, Em. 535nm for FITC and 585nm for ruthenium complex).
RESULTS AND DISCUSSION

For microcontact printing of pH sensor spots, sensor reagent was infiltrated into a lens cleaning tissue (Whatman) and it was used as an ink pad. Averaged spot size was 10.0±0.5 μm (n=48). Response to 0.05M phosphate buffer of pH 6~8 were measured. Figure 1 shows averaged fluorescence intensity of 100 spots normalized with the value at pH 7. Good correlation could be observed between pH and fluorescence intensity. For microcontact printing of oxygen sensor spots, sensor reagent was dropped onto a PDMS stamp, and was developed by using an air gun. Averaged spot size was 9.9±0.6 μm (n=25). Sensor response was evaluated by using distilled water aerated with oxygen or nitrogen, and 5% sodium sulfite solution. Figure 2 shows averaged fluorescence intensity of 99 spots normalized with the value at normal distilled water. Good correlation could be observed between oxygen concentration and fluorescence intensity. Fluorescence micrograph of oxygen sensor array was shown in Figure 3.

![Figure 1. pH profile of microcontact printed pH sensor array (n=100)](image1)

![Figure 2. Responses of O2 sensor array prepared by microcontact printing (n=99)](image2)

Finally, after the printing of oxygen sensor spots, pH sensor spots were stamped between oxygen sensor spots by using the same PDMS stamp. Figure 4 shows the pseudo-colored fluorescence image of a prepared integrated sensor array. The response of the prepared integrated pH and oxygen sensor array was measured by using a micro array scanner. pH sensor spots responded to only pH changes (Fig.5(a)) and oxygen sensor spots responded to only oxygen concentration changes(Fig.5(b)).
Figure 4. Fluorescence image of integrated pH and O₂ sensor array (ex.473nm, em.535nm)
Darker spots: pH sensors, Brighter spots: O₂ sensors

(a) pH sensor spots

(b) Oxygen sensor spots

Figure 5. Responses of integrated pH and oxygen sensor array
pH7(1), pH7(2) & pH7(3) represent O₂ aerated-, no treated-, N₂ aerated phosphate buffer (pH7), respectively.

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REFERENCES