Development of a label-free needle type ion image sensor for real-time monitoring of potassium ion distributions
Yusuke Nakamura1, Tatsuya Iwata1, Kazuhiro Takahashi1,2, Kazuaki Sawada1,2
1Toyohashi University of Technology, Japan
2Electronics-Inspired Interdisciplinary Research Institute, Japan
E-mail: nakamura-y@int.ee.tut.ac.jp

INTRODUCTION
A charge transfer type hydrogen ion image sensor developed using CMOS technology that visualize two dimensional imaging of extracellular proton concentration with label free have developed [1-3]. Furthermore, a potassium ion image sensor (PIS) was fabricated by applying a potassium ion selective membrane. This sensor enabled real-time imaging of the K+ concentration gradient in solution [4, 5]. By using the K+ image sensor, potassium ion released from hippocampal slices was observed [6]. However, these image sensor were only use in in-vitro condition. We are developing a 32 × 128 pixel needle type pH image sensor that can insertion into the body that can real-time image the Hydrogen ion concentration gradient in the living body [7]. We have newly developed a needle type potassium ion image sensor for in-vivo conditions that can observe potassium ion concentration gradient distributions in real time.

MATERIALS AND METHODS
Details of the needle type pH image sensor used in this study are described in other literatures [7]. PIS is fabricated by coating a potassium ion sensitive membrane on a sensing area of needle type pH image sensor [4]. Potassium ion sensitive membranes are made from plasticized PVC. Procedures for the preparation of potassium ion sensitive membranes are described in other literatures [2, 3]. The membrane components were composed of 42 mg plasticized PVC, 1.0 mg valinomycin, which is a potassium ion-selective ionophore, 0.4 mg TFPB, 23 mg DOS and 30 mg PSS-methacryl substituted. The above reagents were dissolved in 0.4 mL tetrahydrofuran (THF). Then the THF solution was cast onto a sensing area surface. The PVC-based membrane was dried for more than 12 h.

Using this sensor, the potassium ion concentration gradient was measured in solution and agarose gel. Using KCl and HEPES 5 mM (adjusted to pH 7.2 with NaOH) as a solvent, potassium ion sensitivity curves in solution and agarose gel were obtained by varying the concentration of KCl. The formula of HEPES used in this study is C6H13N2O8S. Also, using the agarose gel for the lower layer and the solution for the upper layer, this sensor was inserted from the solution into the agarose gel at a speed of 0.5 μm / sec and then pulled out. The change in K+ concentration at that time was measured. At this time, the agarose gel of the lower layer was HEPES 5 mM, KCl 100 mM was used as a solvent, and the upper layer solution was HEPES 5 mM as a solvent.

RESULTS AND DISCUSSION
Fig. 1 shows the K+ sensitivity curve in the solution measured using the sensor fabricated. Fig. 2 shows the potassium sensitivity curve in agarose gel. Each plot is the calculated value of the average of five pixels in the center area of the sensor. Potassium sensitivity was 50 mV / decade at physiological potassium concentration of 10 -3 to 10 -1 M. Fig. 3 shows a photograph of the insertion experiment. Fig. 4 shows the snapshot image of the potassium ion during insertion into an agarose gel from solution. Fig. 5 shows the change in the output signal voltage of one pixel when it inserted into the agarose gel from the solution. It was confirmed that the output signal voltage of the pixel inserted into the agarose gel was increased and monitored K+ concentration in a deep part of the agarose gel. The potassium ion sensitive membrane did not peel off even after the inserting experiment. After the insertion experiment, it was confirmed that it was sensitive to potassium ions. These results suggest that this sensor can image the potassium concentration gradient in-vivo.

ACKNOWLEDGEMENTS
This work was supported by JPT Grant Number JPMJCR14G2.

REFERENCES
Fig. 1 Standard curve of K$^+$ with the output signal voltage in solution.

Fig. 2 Standard curve of K$^+$ with the output signal voltage in Agarose gel.

Fig. 3 Photograph of insertion experiment.

Fig. 4 K$^+$ image during insertion into Agarose gel from solution.

Fig. 5 K$^+$ changes with the output signal voltage.