Feedback mode-based electrochemical imaging of conductivity and topography for large substrate surfaces using an LSI-based amperometric chip device with 400 sensors

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1 **Abstract**

2 Feedback mode-based electrochemical imaging of conductivity and topography
3 for large substrate surfaces is presented using a large-scale integration (LSI)-based
4 amperometric chip device with 400 sensors at a pitch of 250 μm . The LSI-based chip
5 device has enabled rapid electrochemical imaging of large substrate surfaces, compared
6 to scanning electrochemical microscope (SECM). Substrates modified with conductive
7 and insulating materials were placed onto the device to acquire electrochemical signals
8 from the substrate surface using positive and negative feedback signals. The
9 conductivity and topography of the substrate were successfully imaged, indicating that
10 the feedback mode-based electrochemical imaging with such a device is useful to
11 characterize large-area substrate surfaces.

12
13 **Keywords:**

14 LSI-based amperometric sensor

15 Electrode array

16 Electrochemical imaging

17 Feedback mode-based electrochemical detection

18 Chip device

19

1 Introduction

2 The conductivity and topography of a sample can be electrochemically imaged
3 using scanning electrochemical microscopy (SECM) with a feedback mode for local
4 surface characterization by electrode probe scanning.^[1-2] In the feedback mode of SECM,
5 a redox active species is electrogenerated from a precursor species in solution at the
6 microelectrode tip positioned close to an interface, which then diffuses away from the
7 tip towards the interface. If the electrogenerated species is converted to the original
8 form at the interface (e.g., conductive material and enzyme), then the original form
9 diffuses back to the tip and enhances the current at the electrode by establishing a
10 feedback cycle, which is termed positive feedback. If the interface is inert with respect
11 to the electrogenerated species (e.g., insulating material), then diffusion of the species
12 from the bulk to the microelectrode tip is blocked by the interface, thereby decreasing
13 the current at the electrode compared to that in the bulk, which is termed negative
14 feedback. The feedback mode can be used to provide information on the topography of a
15 sample. Thus, feedback mode-based electrochemical imaging of SECM is useful to
16 evaluate the conductivity and topography of a substrate surface. However, the SECM
17 technique may take a long time to obtain an electrochemical image due to time required
18 for scanning with the probe. Therefore, SECM is unsuitable for rapid electrochemical
19 imaging of the conductivity and topography of a large substrate surface.

20 To solve these problems, several devices with electrode arrays have been
21 developed.^[3-7] Barker et al. reported a probe-based electrode array for SECM imaging
22 with 16 individually addressable electrodes, and large-area imaging for conductive
23 materials was achieved by scanning with a probe.^[3] Chip-based electrode arrays have
24 been developed; however, it is difficult to incorporate many electrochemical sensors into
25 a small area, due to large electrode areas that include lead connectors and connector
26 pads when electrodes are simply arranged onto devices.

27 Large-scale integration (LSI)-based chip devices have been developed to
28 incorporate many amperometric sensors into a small area.^[8-12] We have previously
29 developed an LSI-based amperometric device that contains 400 sensors with a pitch of
30 250 μm .^[8,9] The device is based on complementary metal-oxide-semiconductor (CMOS)
31 technology and each sensor contains an operational amplifier with a switched-capacitor
32 type I - V converter for in-pixel signal amplification.^[8] The device has been applied to
33 real-time imaging of enzyme activity^[8] and array-based cell analysis.^[9] This device has
34 enabled rapid electrochemical imaging of large substrates (5×5 mm²).

35 In the present study, the LSI-based chip device was applied to imaging the
36 conductivity and topography of large substrate surfaces. Substrates modified with

1 conductive materials and insulating materials were placed onto the device, and their
2 conductivity and topography were imaged using positive and negative feedback signals.
3
4

Materials and Methods

Device fabrication

Detailed information on the device has been described previously.^[8,9] Briefly, the device consists of 400 Pt electrodes and 400 SU-8 (SU-8 3005, MicroChem, USA) microwells (Fig. 1). A Pt electrode is located at the bottom of each microwell (18 μm diameter and 5 μm deep). A polycarbonate reservoir (Japan Aviation Electronics Industry) was bonded onto the device using poly(dimethylsiloxane) (PDMS) to insulate the connector electrodes. The reservoir was used to keep sample solutions.

Electrochemical detection

We prepared an X-shaped Pt electrode substrate, an Au electrode substrate modified with a nonconductive material (ink: Zebra Co., Ltd., Japan) and a glass substrate modified with windmill-shaped SU-8 layers (SU-8 3025, MicroChem).

1.0 mM ferrocenemethanol (FcCH_2OH) solution containing 0.1 M KCl was introduced onto the device, and Ag/AgCl reference and Pt counter electrodes were inserted into the solution. Glass substrates modified with conductive and/or insulating materials were then placed onto the device under compression to achieve contact between the substrate and the device. For all experiments, the potential of the 400 electrodes was stepped from 0.00 V to 0.50 V to oxidize FcCH_2OH to FcCH_2OH^+ . Electrochemical images consisting of 400 oxidation currents of FcCH_2OH were acquired for periods from 0 to 120 s at 200 ms intervals.

Current simulation

To investigate the effect of the substrate-electrode distance on the device, redox currents at the 400 electrodes were calculated by simulation. Detailed information on the simulation conditions is given in the Supporting Information.

Results and discussion

Electrochemical imaging for conductive materials

To evaluate electrochemical imaging for conductive materials using the device, an X-shaped Pt electrode was fabricated onto a glass substrate. The substrate was electrochemically imaged based on positive and negative feedback signals (Fig. 2A). The electrochemical image shown in Fig. 2B clearly displays the Pt pattern on the substrate. The image lasted for at least several minutes after the potential step, although the currents from the sensors then gradually decreased. A part of the metal pattern on the substrate covered non-sensor areas (Fig. S1), which might be why the positive feedback sustained for such a long time in this geometrical configuration. Also, the reason might be that the diameter of the sensors (18 μm) was smaller than the pitch between the sensors (250 μm). To describe the reason clearly, further experiments and simulations on positive feedback are required. Figure 2C shows chronoamperograms from the uncovered sensor, the sensor under the Pt electrode, and the sensor under the glass. At 120 s after the potential step, the sensor current under the Pt electrode (1.01 nA) was higher than that under the glass (0.32 nA) because positive feedback occurred under the metal pattern. In contrast, these sensor currents were considerably lower than the current from the uncovered sensors (1.52 nA at 120 s after the potential step) because the supply of FcCH_2OH from the bulk was blocked by the substrate (negative feedback). It should be noted that the sensor currents under the glass were not sufficiently small, because FcCH_2OH diffused into the sensor areas through the gap between the device and substrate. The effect of distance between the device and the substrate is discussed in the following simulation section. For the present device, the currents from the sensors under the metal pattern were lower than those from the uncovered sensors (Figs. 2B and 2C). This phenomenon contrasts with that observed in typical SECM measurements, where the current increases when a SECM probe approaches a conductive region due to the positive feedback. The driving force for positive feedback is derived from the difference between the concentrations of the redox species on the unbiased conductive surface near and far from the working electrode. Recycling of the redox mediator on the unbiased conductive substrate proceeds effectively when the area of the conductive surface is larger than the size of the sensor electrodes. However, in the present study, there was insufficient positive feedback because the area of the X-patterned electrode was insufficient compared with the area of the high-density sensor array.

To evaluate the conductivity of a substrate surface, an Au electrode substrate modified with a nonconductive material was electrochemically imaged using positive

1 feedback signals (Fig. 3A). The letter “C” was written on the substrate using an ink as a
2 nonconductive material. Figure 3B shows that the positive feedback is prevented by the
3 ink, so that the letter “C” appears in the electrochemical image. Figure 3C shows
4 chronoamperograms from the sensor under Au electrodes with and without the ink; the
5 currents from these sensors after 120 s of potential step were 0.71 and 1.69 nA,
6 respectively. In this case, there was sufficient positive feedback compared to that for the
7 imaging shown in Fig. 2 because the electrode area was larger than the previous
8 substrate. Thus, the conductivity of the substrate surface could be successfully imaged
9 using feedback signals.

10 Although these images were obtained every 200 ms in the present study, one
11 image consisting of 400 points can be obtained within 18 ms when using a suitable
12 current measuring range.^[8]

14 **Electrochemical imaging for substrate topography**

15 To demonstrate the utility of this device for the investigation of substrate
16 topography, a glass substrate modified with windmill-shaped SU-8 layers was
17 electrochemically imaged using negative feedback signals (Fig. 4A). The height of the
18 SU-8 layers was approximately 25 μm . The topography of the substrate is observed in
19 the electrochemical image (Fig. 4B). Figure 4C shows chronoamperograms from the
20 uncovered sensor, the sensor under the glass, and the sensor under the SU-8 layer. The
21 currents from these sensors after 120 s of potential step were 1.56, 0.89, and 0.37 nA,
22 respectively. At 120 s after the potential step, the sensor current under the SU-8 layer
23 was lower (0.37 nA) than that under the glass (0.89 nA), due to the strong negative
24 feedback under the SU-8 layer. The difference in responsivity is caused by the difference
25 in the negative feedback effect, which is dependent on the sensor-substrate distance.
26 These results show that negative feedback mode-based electrochemical imaging can be
27 applied to image the topography of a large substrate surface.

29 **Current simulation**

30 To investigate the effect of the substrate-electrode distance on the device, redox
31 currents at the 400 electrodes were calculated by simulation. Figure 5A shows a
32 simulation model. Briefly, three-dimensional models with 400 microwells were
33 constructed, and an insulation layer was placed 5-105 μm above the electrodes.
34 Chronoamperograms from the uncovered sensor show that the currents were estimated
35 to approximately 1.44 nA, 120 s after the potential step when the distance between the
36 electrodes and the electrode-substrate distance in the device were 25 and 105 μm ,

1 respectively (Fig. 5B). In contrast, the current decreased significantly from 1.13 to 0.32
2 nA when the distance was changed from 105 to 25 μm (Fig. 5B). This is because the
3 diffusion of FcCH_2OH was prevented by the insulating substrate (negative feedback).
4 The relationship between the substrate-electrode distance and the current is shown in
5 Fig. 5C. The actual distance between the substrate and the electrode cannot be
6 determined; however, if the experimental results are compared with the simulation
7 results, then the substrate-electrode distance of the present device (Figs. 2 and 4) is
8 estimated to be approximately 25 μm .

9 Thus, the conductivity and topography of a large substrate surfaces could be
10 successfully evaluated using the feedback mode-based electrochemical imaging with the
11 LSI-based amperometric chip device.

12 **Conclusion**

13 Feedback mode-based electrochemical imaging using an amperometric
14 LSI-based device was presented for evaluation of the conductivity and topography of
15 large substrate surfaces. Electrochemical imaging with 400 pixels for a large substrate
16 surface was rapidly achieved using the device with 400 sensors at 250 μm pitch. The
17 device is suitable for rapid electrochemical imaging of large substrate surfaces,
18 compared to SECM. The electrochemical imaging was based on positive and negative
19 feedback signals, and conductivity and topography of the substrate were successfully
20 imaged. These results demonstrate that feedback mode-based electrochemical imaging
21 with such a device can be successfully applied to characterize the conductivity and
22 topography of large-area substrate surfaces.

23 **Acknowledgement**

24 This work was supported in part by a Grant-in-Aid for Scientific Research (A)
25 (No. 25248032), a Grant-in-Aid for challenging Exploratory Research (No. 26600057)
26 and JSPS Invitation Fellowship Program for Research in Japan (Long Term) from the
27 Japan Society for the Promotion of Science (JSPS). This research was partly supported
28 by Special Coordination Funds for Promoting Science and Technology, Creation of
29 Innovation Centers for Advanced Interdisciplinary Research Areas Program from the
30 Japan Science and Technology Agency.

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Figure 1

Images of the LSI-based amperometric chip device.

Figure 2

Electrochemical imaging for conductive and insulating substrates. (A) Detection scheme. (B) Optical and electrochemical images of the substrate with an X-patterned Pt electrode above the device. The electrochemical image was acquired after 40 and 120 s of the potential step. (C) Chronoamperograms from the sensor under the glass, the sensor under the Pt electrode, and the uncovered sensor. These sensors are indicated by the $\times$ symbols in Fig. 2B (red: uncovered area, green: Pt electrode, blue: glass).

Figure 3

Electrochemical imaging for the conductivity of a substrate surface modified with oil-based ink. (A) Detection scheme. (B) Optical and electrochemical images of the Au electrode modified with the ink. The electrochemical image was acquired after 120 s of the potential step. (C) Chronoamperograms from the sensors indicated by the × symbols (blue: no ink, red: ink) in Fig. 3B.

Figure 4

Electrochemical imaging for the topography of an insulating substrate. (A) Detection scheme and illustration of the insulating substrate modified with SU-8. (B) Optical and electrochemical images of the insulating substrate above the chip device. The electrochemical image was acquired after 120 s of the potential step. (C) Chronoamperograms from the sensor under the glass, the sensor under the SU-8 layers and the uncovered sensor. These sensors are indicated by the $\times$ symbols (red: uncovered area, green: glass, blue: SU-8) in Fig. 4B.

Figure 5

Current simulation based on the diffusion of a redox compound. (A) Configuration of 400 electrodes, 400 microwells and an insulating substrate (not to scale). (B) Chronoamperograms of 1.0 mM FcCH_2OH at the sensor under the insulating substrate and the uncovered sensor for electrode-substrate distances of 25 and 105 μm . These sensors are indicated by the $\times$ symbols in Fig. 5A (red: uncovered area, blue: glass). (C) Relationship between the electrode-substrate distance and the current after 120 s of the potential step.