The Photoelectrolytic Reaction of Chlorophyll–β-Carotene–Quinone Electrodes

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The NAD^+ (nicotinamide adenine dinucleotide) reduction was carried out under illumination by using a platinum electrode coated with chlorophyll and quinone. Various quinones and β-carotene were used as components of the cathode layer to obtain a higher current density. The highest current density, 13 $\mu A/cm^2$, was obtained in the case in which the cathode layer contained 0.01 $\mu mol/cm^2$ of chlorophyll and the molar ratio of chlorophyll: chloranil : β-carotene was 1 : 1 : 2. The role of quinone and β-carotene in the cathode layer was discussed.

Since the photochemical reaction in chloroplasts can be compared with an electrochemical reaction, Photosystem I and Photosystem II can be regarded as a cathode and an anode, respectively. It was reported that the photoelectrolysis was carried out by the system connecting the chlorophyll-1,4-naphthoquinone electrode and the chlorophyll-9,10-anthracenediol electrode under illumination.[1] The photoexcited chlorophyll-1,4-naphthoquinone electrode and chlorophyll-9,10-anthracenediol electrode behaved as a cathode to reduce NAD^+ (nicotinamide adenine dinucleotide) and as an anode to oxidize hexacyanoferrate(II) (The expression "chlorophyll-redox compound electrode" is used for an electrode lined with a chlorophyll/redox compound mixture. Similar expressions are used where a different compound is specified.)

The redox potentials of plastocyanin and cytochrome f in Photosystem I (+0.37 V)[2] are more positive than that of plastoquinone in Photosystem II (*ca.* 0 V).[2] Therefore, plastocyanin and cytochrome f in Photosystem I are able to oxidize the reduced plastoquinone in Photosystem II. In turn, the reduced plastocyanin and cytochrome f transfer the electron to the chlorophyll cation in Photosystem I, and plastoquinone gains the electron from the photoexcited chlorophyll in Photosystem II. It was assumed that the electron transfer from Photosystem II to Photosystem I might occur with the driving force caused by the difference of redox potentials of the redox compounds in the photosystems. Therefore it was expected that a higher potential difference between two chlorophyll electrodes could be obtained under illumination if a redox compound which exhibited a more positive redox potential than 1,4-naphthoquinone was used as a component in the cathode layer.

β-Carotene, one of carotenoids in the photosystems, is known to exhibit a photoconductive property[3] and to be utilized in the mixture of chlorophyll, albumin, and 2-methyl-1,4-naphthoquinone to make an electron transfer easier under illumination.[4] An energy transfer from β-carotene to chlorophyll is known from a study of photoexcitation with a black lipid membrane containing chlorophyll and β-carotene.[5] From these facts it was thought that the addition of β-carotene as one of components in the electrode layer might make the current density higher.

This paper will present some recent results on the photoelectrolysis by using chlorophyll electrodes and the speculation on the role of quinone and β-carotene in the electrode layer under illumination.

Experimental

Chlorophyll, which was a mixture of type a and b, was prepared from spinach leaves by the method of Jacobs *et al.*[6] Various quinones were purified by sublimation. NAD^+ used was made by the Oriental Yeast Co. The other chemicals, including β-carotene, were of reagent grade or the best commercially available grade. The conducting adhesive used was Fujikura-kasei Dotite D-362.

Catholyte (anolyte): 5 mM NAD^+ (5 mM potassium hexacyanoferrate(II) in M/30 phosphate buffer (pH 6.9).* For the photoelectrolysis 60 ml of catholyte and anolyte were used.

Chlorophyll-redox compound electrodes were prepared by the method described in the previous paper:[1] The cathode layer on a platinum plate (2 cm × 4 cm) consisted of 2.5 μmol of chlorophyll, 2.5 μmol of one of the quinones, and 0.1 g of conducting adhesive. The anode layer was prepared in the same manner as the cathode, except that 2.5 μmol of 9,10-anthracenediol was added in place of the quinone. Two-layer electrodes:** A platinum wire (1 cm long with a diameter of 0.1 cm) connected with a sealed wire was used as an electrode. Two drops (0.1 ml) of 5 mM chlorophyll solution in hexane spread on the water surface. A thin layer of chlorophyll remained after evaporation of the hexane on the water surface. The thickness of the chlorophyll layer was estimated to be *ca.* 0.05 μm because the diameter of the chlorophyll layer which remained on the water surface was about 5 cm. Two grams of conducting adhesive and 10 mg of chloranil were added into 50 ml of hexane to make a suspensoid. Two drops of the suspensoid were spread on the water surface in another beaker to make a layer containing conducting adhesive and chloranil. A two-layer electrode was prepared by the following method: The platinum wire was dipped into the chlorophyll layer spread on the water surface very quietly and then pulled up. The platinum wire was thus covered by a chlorophyll layer. In turn the platinum wire coated by the chlorophyll layer was dipped into the chloroanil layer spread on the water surface. Thus the chloranil layer was coated on the chlorophyll layer. The electrode coated with chloranil first and then with a chlorophyll layer was prepared in the same manner.

The same electric cell as reported in the previous paper was used for all the experiments. The photoelectrolytic reaction was carried out under illumination (1×10^4 lx) through a heat absorber with a photoreflector lamp (100 V, 500 W). The entire electric cell including the calomel electrode was placed in a thermostat and was controlled at 20 °C. Nitrogen gas was bubbling into the electrolyte during the

* $M = 1$ mol dm^{-3}.

** We defined "two-layer" as putting one layer upon another.

experiments.

The measurements of potential and current, and the determination of NAD^+ and hexacyanoferrate(II) were carried out in the same manner as described in the previous paper.[1)]

Results and Discussion

Various Chlorophyll–Quinone Electrodes. To make the current density of the photoelectric reaction increase, various quinones were used as a component of one layer of the chlorophyll–quinone electrode.

The photoelectrolytic reaction was carried out with the systems of chlorophyll–quinone electrode in NAD^+ solution and chlorophyll–9,10-anthracenediol electrode in hexacyanoferrate (II) solution. The photovoltage and photocurrent were determined when both electrodes were illuminated at 1×10^4 lx. The photovoltage was found to increase with quinones exhibiting a more positive redox potential, as shown in Fig. 1 (above). The cathodic potential and current were observed at the chlorophyll–quinone electrode under illumination. When the light was turned off, the chlorophyll–quinone electrode became a nanode and the chlorophyll–9,10-anthracenediol electrode became a cathode. The potential difference of the chlorophyll–quinone *vs.* the chlorophyll–9,10-anthracenediol electrode in the dark increased with a positive shift of the redox potential of quinone as shown in Fig. 1 (below). The electrode potentials of the chlorophyll–1,4-naphthoquinone electrode and the chlorophyll–chloranil electrode *vs.* calomel electrode were obtained to be +80 and +90 mV in the dark, and −46 and −93 mV under illumination.

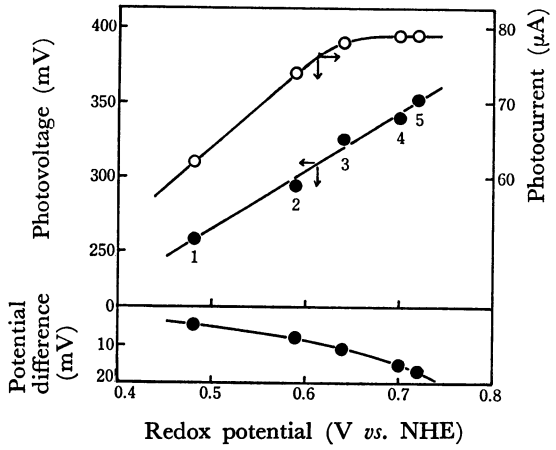


Fig. 1. Various chlorophyll–quinone electrodes.
1: 1,4-Naphthoquinone, 2: dimethyl-*p*-benzoquinone, 3: *p*-toluquinone, 4: chloranil, 5: 2,5-dichloro-*p*-benzoquinone. The values of redox potential of various quinones were cited from Ref. 7.

The value of the photocurrent reached a plateau in the case that chloranil and 2,5-dichloro-*p*-benzoquinone were used as components of the cathode layer.

The Effect of β-Carotene. To make the electric resistance of the chlorophyll electrode lower under illumination, β-carotene was added as a component of both anode and cathode layers. The chlorophyll-chloranil electrode was used as a cathode. The quantity of chlorophyll and chloranil (9,10-anthracenediol) of the electrode layer was the same as in the previous section, except for the addition of 0—12.5 μmol of β-carotene.

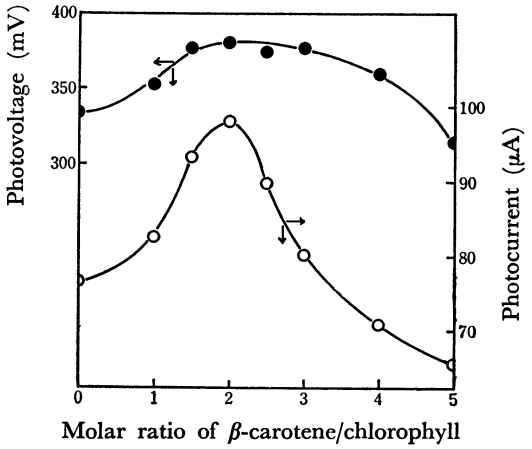


Fig. 2. The effect of β-carotene.

By using the chlorophyll–chloranil–β-carotene electrode as a cathode and the chlorophyll–9,10-anthracenediol-β-carotene electrode as an anode, the photocurrent was measured as shown in Fig. 2. The maximum photocurrent was obtained at the molar ratio of chlorophyll : chloranil (9,10-anthracenediol) : β-carotene = 1 : 1 : 2. The maximum photovoltage was about 50 mV higher than in the case where β-carotene was not included as a component of the electrode layers.

It is plausible that in the electrode layer chlorophyll, chloranil, and β-carotene may form a ternary complex which might exhibit a photoconductive property.

The Thickness of the Electrode Layer. The thickness of the electrode layer had to be considered as one of the factors which influences the electric resistance of the electrode. It was expected to obtain larger potential differences and current densities by reducing the thickness of the electrode layer.

The chlorophyll electrodes which were coated by different quantities of electrode layers were prepared with a mixture of chlorophyll, chloranil (9,10-anthracenediol), β-carotene (molar ratio of 1 : 1 : 2),

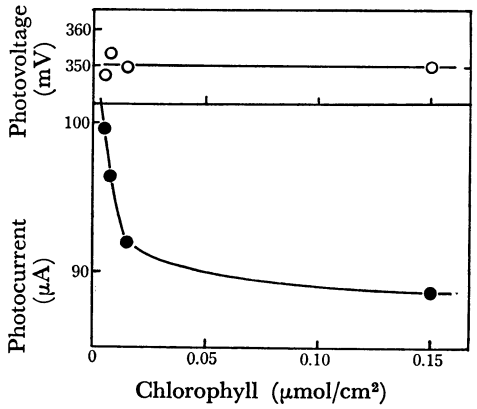


Fig. 3. The thickness of electrode layer.

and conducting adhesive (the mixing ratio: 0.1 g of conducting adhesive/2.5 μmol of chlorophyll).

The photocurrent density was found to increase by decreasing the quantity of the electrode layer from 0.16 to 0.005 μmol of chlorophyll/cm² (Fig. 3). A constant photocurrent could not be produced below 0.005 μmol/cm². The optimum thickness of the electrode layer was found to be about 0.01 μmol of chlorophyll/cm² (Fig. 3).

Photoelectrolysis. The photoelectrolysis was carried out by using the electrodes which were prepared with the optimum conditions of quantity (0.01 μmol/cm² of chlorophyll) and mixing ratio as in the previous section. The potential-time curves and the photocurrent-time curve are shown in Fig. 4. The potential difference and the current density were found to be 0.35 V and 13 μA/cm² under illumination. These values are higher than those in the case of the chlorophyll–1,4-naphthoquinone electrode (0.25 V, 8 μA/cm²) in the previous paper.[1] The quantity of electricity amounted to 5.29 C (corresponding to 54.8×10^{-6} F where F is the Faraday constant) after 12 h. This is much greater than the quantity of electricity passed for the reduction (oxidation) of chloranil (9,10-anthracenediol) in the electrode layer which contained 0.13 μmol of chloranil (9,10-anthracenediol). Meanwhile 7.4% NAD^+ was reduced and 14.2% hexacyanoferrate(II) was oxidized. The current efficiencies were 72% and 71%, respectively. When the light was turned off while the external circuit was kept closed, the reverse potential and the reverse current were observed to be 0.11 V and 10 μA/cm². The current decreased with time.

From the potential-current curves of the chlorophyll–chloranil electrode and the chlorophyll–chloranil-β-carotene electrode (Fig. 5), two reduction waves were observed, at −0.2 and −0.8 V *vs.* SCE; these were assigned to chloranil and NAD^+. The cathode potential in Fig. 4 (−0.1 V *vs.* SCE) was more positive than that obtained in Fig. 5. These results suggest that chloranil in the cathode layer could not be reduced during photoelectrolysis.

Two-layer Electrodes. To clarify the process of electron transfer on the cathode, the photoelectrolytic reaction was carried out with two-layer electrodes. The two-layer electrodes used were prepared as described in the experimental section. As the counter electrode a platinum electrode (2 cm×4 cm) was used.

In the case of the two-layer electrode coated with chloranil on a chlorophyll layer, very little current was produced, as shown in Fig. 6 (curve D). A higher current production by the two-layer electrode coated with chlorophyll on a chloranil layer in curve B was observed, as compared with curve D and with curve C, which was obtained by the single layer electrode coated with chloranil only.

It is deduced that the photoexcited chlorophyll can not, but chloranil can gain an electron from the counter electrode. It seems that the electron may

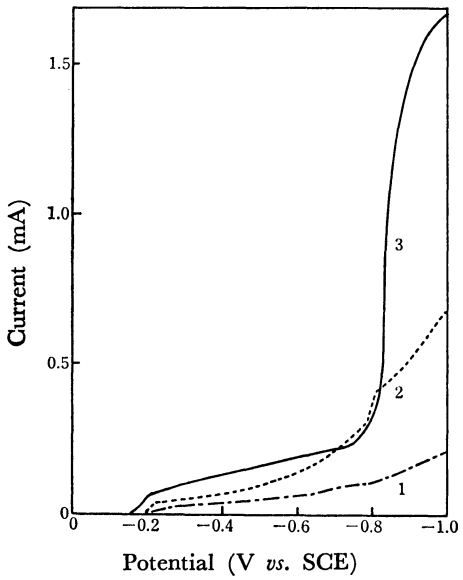


Fig. 5. The potential-current curves.
1: Chlorophyll-chloranil electrode (molar ratio of 1 : 1) in buffer solution. 2: the same electrode as 1 in NAD^+ solution, 3: chlorophyll-chloranil-β-carotene electrode (molar ratio of 1 : 1 : 2) in NAD^+ solution. A platinum plate (2 cm×4 cm) was used for the counter electrode. The potential-current curves were measured in the dark.

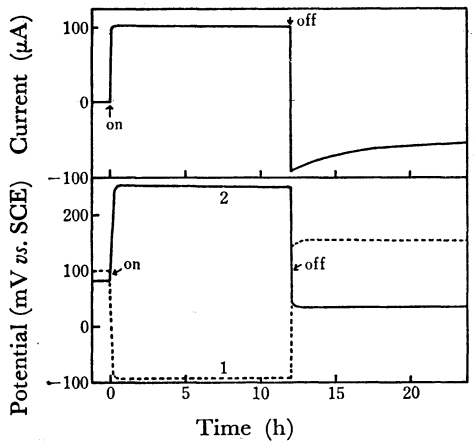


Fig. 4. The photoelectrolysis.
1: Chlorophyll-chloranil-β-carotene electrode, 2: chlorophyll-9,10-anthracenediol-β-carotene electrode.

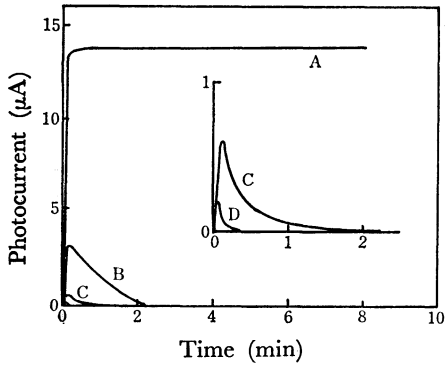
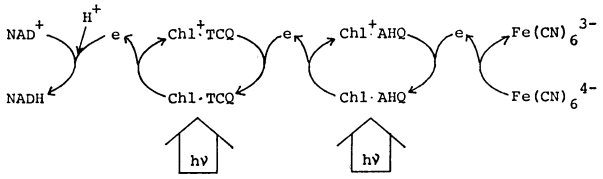


Fig. 6. Two layer electrodes.
A: Mixture of chlorophyll and chloranil on Pt, B: chlorophyll on chloranil layer on Pt, C: chloranil on Pt, D: chloranil on chlorophyll layer on Pt.

transfer from the reduced chloranil to the photoexcited chlorophyll. However, the reduced chloranil can not release an electron to NAD^+. On the other hand, the single layer electrode prepared from the mixture of chlorophyll and chloranil was able to keep producing much higher and constant current during the time tested (curve A).

The Process of Electron Transfer. It is known that the reduced plastocyanin and the reduced cytochrome f can reduce P700 under illumination.[8,9] It has been reported that a semiconductor electrode prepared by depositing chlorophyll on a single crystal of zinc oxide can exhibit an anodic behavior under illumination to transfer an electron from hydroquinone in electrolyte to zinc oxide through chlorophyll.[10] On the contrary it seems that an electron may not transfer in the order platinum, chloranil, chlorophyll, and NAD^+ in the case of a two-layer electrode. As a probable speculation, chlorophyll was thought to form a complex with chloranil in the cathode layer under illumination, and a transient state of the complex might drive the electron transfer from the layer of chlorophyll–9,10-anthracenediol in the anode to that of chlorophyll–chroanil in the cathode through platinum. A contact potential barrier, which was known to exist at the chlorophyll–metal contact,[11] might not influence the direction of current flow in the case of our chlorophyll electrodes. Because both cathode and anode were prepared in the same condition on a platinum plate, the contact potential barrier between chlorophyll and platinum at the cathode would offset that at the anode. The process of electron transfer can be expressed as the following: where Chl·TCQ and Chl·AHQ respectively represent the chlorophyll complexes of chloranil and 9,10-anthracenediol, and $Chl^+ \cdot TCQ$ and $Chl^+ \cdot AHQ$ mean the cations of these chlorophyll complexes The driving force of electron transfer from anode to cathode through the external circuit was found to increase as the redox potential of quinone in the cathode was made more positive, using the results in Fig. 1. The mechanism of electron transfer from the cathode to the catholyte will be discussed in another paper.



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