Electrode Potential Diagrams and their Use in the Hill-Bendall or Z-Scheme for Photosynthesis

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Electrode potential diagrams provide a common way of representing biochemical electron-transfer processes in photosynthesis (*1–3*) and respiration (*4*). However, we have found that when students approach these diagrams from the viewpoint of physical chemistry or photochemistry they become confused by their apparent similarity to molecular energy-level diagrams such as the Jablonski diagram.

Also, the diagrams are difficult to understand in themselves, and confusions arise for several reasons: the use of a reverse scale; the use of the term "oxidation/reduction potential" to describe the ordinate; the depiction of single substances on the diagram rather than reactions; misunderstanding of the meaning of various arrows; and misunderstanding of the nature of the "energy" which can be calculated from the height.

The aim of this article is to help clear away some of the confusions by providing a description of electrode potential diagrams in terms of a simple and probably familiar example, that for zinc and copper sulfate. Various cautions are offered and then a brief description of the Hill-Bendall or Z-scheme for photosynthesis is given to emphasize the main points.

While we have used an up-to-date diagram for photosynthesis, it is not the aim here to provide a current view of the topic; our object is simply to present a correct physical-chemical description of the diagram.

An Electrode Potential Diagram for the Zn/Cu^{2+} System

The example chosen to illustrate the principles is the Zn/CuSO$_4$ system. It will be recalled that zinc reacts with aqueous copper(II) sulfate to give metallic copper and zinc sulfate:

$$Zn(s) + Cu^{2+}(aq) \rightarrow Cu(s) + Zn^{2+}(aq) \tag{1}$$
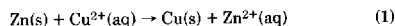
In the laboratory the reaction goes to completion for all ordinary concentrations of Cu^{2+}. The standard change in the Gibbs function[1] for the reaction, $\Delta G^{\ominus}$, is -212.8 kJ mol^{-1}.

If the system is set up as an electrochemical cell (the Daniell Cell)

$$Zn(s) \mid Zn^{2+}(aq) \mid Cu^{2+}(aq) \mid Cu(s)$$
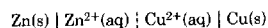
then the *EMF* for the standard activities is $+1.103$ V, the copper electrode being positive. In this case, with unit activities of reactants and products, the *EMF* would be $E^{\ominus}$, the standard *EMF* of the reaction, which in turn is related to the standard change in free energy by

$$E^{\ominus} = -\Delta G^{\ominus}/nF$$

where F is the Faraday constant and n is the number of electrons exchanged in the reaction. Using such a cell, work could be obtained from the reaction.

Now the work, w, in a process is related to the change in Gibbs function, at constant temperature and pressure, by the inequality

$$w > \Delta G$$

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[1] The Gibbs Function is now the recommended name (*5, 6*) for the more commonly used "free energy."
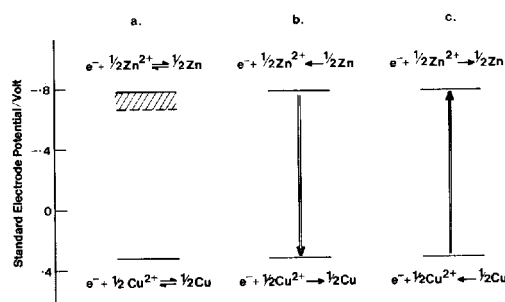

Figure 1. (a) An electrode potential diagram for the Zn/Cu system, showing the levels, half reactions and the effect of a hundredfold change in concentration. (b) The diagram showing the spontaneous reaction with electron transfer from Zn to Cu (note the directions of the arrows). The height gives the maximum work available from the reaction. (c) The nonspontaneous reaction: work is required to drive the reaction and the height gives the minimum quality needed. Again note the direction of the arrows.

so that extra work must always be done on the system to achieve a given increase in the Gibbs function.

For a spontaneous reaction ΔG is negative so that if any work is obtained from the process, it will be *less* than that given by the value of ΔG; i.e., for reaction (1) with standard activities, the work obtained ($= -w$) will be less than 212.8 kJ mol^{-1}.

Reaction (1) can be driven backwards in an electrochemical cell

$$Cu(s) + Zn^{2+}(aq) \rightarrow Cu^{2+}(aq) + Zn(s) \tag{2}$$
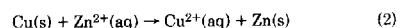
by applying a higher opposing voltage, in other words by doing work on the cell. Now the calculated work required,

$$w_{min} = \text{minimum work required} = \Delta G$$

is a minimum quantity and in practice, more work would be needed to drive the reaction back.

Figure 1a shows an electrode potential diagram for the Zn/Cu^{2+} system. The reactions are shown as half-cell reactions; values of $E^{\ominus}$, the standard electrode potentials, are taken from tables of standard values (*7*).

Each potential refers to the *EMF* of a cell in which the left-hand electrode is a hydrogen electrode, e.g.,

$$H_2(g; 1\ atm)Pt \mid H^+(aq) \mid Zn^{2+}(aq) \mid Zn(s)$$
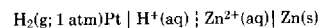
for which the overall reaction is

$$H_2(g) + Zn^{2+}(g) \rightarrow 2H^+(aq) + Zn(s)$$
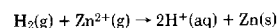
The half-cell reaction is an abbreviated form of this equation.

The actual value of $E^{\ominus}$ given refers only to this reaction and so, when using the diagram, only differences in the values of the *EMF* are important. Absolute values have no special significance other than for the reaction with hydrogen.

Figure 1b shows a representation of the spontaneous reaction (1). The vertical arrow shows the direction of electron transfer, from zinc to copper. With reversible reactions at the top and bottom of the vertical arrow there is an ambiguity in the direction of the overall reaction. This is eliminated if it is noticed that the reaction at the arrow head is proceeding in the *normal* direction (left to right) depicted here by a single arrow, while that at the tail is in the reverse direction.

So, when electron transfer occurs, the electrons are produced in the (tail) reaction $(Zn \rightarrow Zn^{2+} + 2e^-)$ and consumed in the (head) reaction $(Cu^{2+} + 2e^- \rightarrow Cu)$. If reaction takes place in an electrochemical cell, electrons flow from the negative electrode to the positive electrode.

The downward direction of the vertical arrow shows that the reaction is spontaneous. The convention in physical chemistry is to use a downward arrow in an energy diagram to represent energy evolved and thus, usually, a spontaneous reaction.

The *EMF* of the cell, for reactants and products at standard activity, is $E^\ominus$(head) $- E^\ominus$(tail); i.e.,

$$EMF = E^\ominus(\text{head}) - E^\ominus(\text{tail}) = 0.3402 - (-0.7628) = +1.103 \text{ V}$$

which is the standard electrode potential for the overall reaction.

For a spontaneous reaction (downward arrow), the work is then

$$w = \Delta G^\ominus = -nFE^\ominus$$

or for two-electron transfer in reaction (1),

$$w = -212.8 \text{ kJ mol}^{-1}$$

The negative sign indicates that work is available and the equality between w and $\Delta G^\ominus$ shows that the maximum work available has been calculated.

The reverse process is shown in Figure 1c by an upward arrow. Again electron transfer is shown by the arrow direction with the reaction at the head proceeding normally, i.e., left to right, consuming electrons, and that at the tail proceeding right to left, producing electrons. The *EMF* however is now negative ($E^\ominus$(head) $- E^\ominus$(tail) $= -1.103$ V) and so w is a positive quantity (+212.8 kJ mol^{-1}). Thus work must be done on the system to drive the reaction in the direction indicated by the arrow and the value of w, calculated from the change in Gibbs function, is the minimum work required.

The Electrode Potential Diagram in Teaching Electrochemistry

Diagrams such as those shown in Figure 1 could be of much help in teaching electrochemistry where sign conventions often seem to be a stumbling block to understanding. The conventions are, of course, implied in the diagram and in the reaction directions assumed for head and tail of the electron-transfer arrow, but the correct sign is yielded easily and the treatment seems to operate in a more natural way.

The principal disadvantage is the scale, which is reversed, with the positive end at the bottom. The diagram is constructed in the same sense as we use for energy diagrams, with the notion of "energy in," represented by an upward arrow. The electrode potential diagram is thus essentially a free energy diagram with the upward arrow representing minimum work required as well as electron transfer. Since $\Delta G^\ominus$ and $E^\ominus$ have opposite signs the standard electrode potential scale must be a reverse scale.

Free energy diagrams are met with elsewhere in physical chemistry, for example in the Ellingham diagram representing the reduction of metal oxides to metals (8).

Cautions in Using the Diagram

One difficulty that has been mentioned already is that when the height is used to calculate the work for a given reaction then minimum (upward) or maximum (downward) quantities

are given. The processes by which the work is obtained, be they direct such as drawing electric current, or indirect such as the coupling between this reaction and another via an enzymic mechanism, are subject to the limitations of the second law of thermodynamics. All real processes involve loss of work as heat and so to drive a reaction more work than that indicated by the height of the arrow will be needed, whereas for a spontaneous reaction less work would be obtained.

Another problem arises in how the work which is available from a spontaneous reaction is used to drive another reaction. An example of this will appear with the synthesis of ATP in the photosynthetic scheme. Imagine we have one reaction

$$A \rightarrow B$$

which is spontaneous, with a large decrease in the Gibbs function, and another

$$C \rightarrow D$$

which has a small increase in the Gibbs function.

If both are independent of one another so that energy can only be transferred from one to another in the form of heat, then, even if the reactions are occurring together in the same pot, the second cannot proceed since no work has been provided.

If, however, there is a mechanism, such as

$$A + C \rightarrow [AC] \rightarrow B + D$$

in which both steps are spontaneous, then both B and D will be formed.

In particular, if the reactions are coupled enzymically through a membrane so there is some sort of common intermediate, then both the reactions will proceed. It is in this sense that biochemical reactions are coupled so that reactions which are not spontaneous can be driven by those that are.

Also there is the effect of concentration. The values in Figure 1 obtained from the tables are those for the hypothetical standard state with standard molality and fugacity. In using such values in qualitative discussion, the effect of non-ideality may be small so that activity can be replaced by concentration, but the effect of concentration can be appreciable. Thus for the reaction

$$\tfrac{1}{2}H_2 + \text{oxidized form} \rightarrow \text{reduced form} + H^+$$
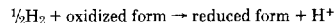
then if H^+ and H_2 are in their standard states,

$$E = E^\ominus - \frac{RT}{nF} \ln \frac{[\text{reduced}]}{[\text{oxidized}]}$$

The shaded portion in Figure 1 a shows the effect of decreasing the Zn^{2+} concentration by a factor of 100.

Provided the half-cell reactions have large differences in standard electrode potentials, valid qualitative conclusions as to the feasibility of a particular reaction can be drawn from the diagram. If, however, the two half-cell reactions have similar potentials, then changes in concentration can render a spontaneous reaction impossible.

Photosynthesis

The overall process in photosynthesis in green plants is the absorption of CO_2 and H_2O in the presence of light to give carbohydrates and oxygen

$$CO_2(g) + H_2O(l) + \text{Light} \rightarrow (CH_2O) + O_2(g) \qquad (4)$$
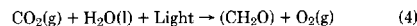
(CH_2O) represents a "proto-carbohydrate."
Photosynthesis can be divided in three steps (1).

1) Light collection via chlorophyll and other pigments and conveying the energy to a reaction center.
2) The oxidation of H_2O to O_2 and the reduction of NADP$^+$ to NADPH (nicotinamide adenine dinucleotide phosphate). The overall equation is

$$2NADP^+ + 2H_2O + \text{light} \rightarrow 2NADPH + 2H^+ + O_2 \qquad (5)$$
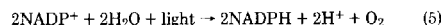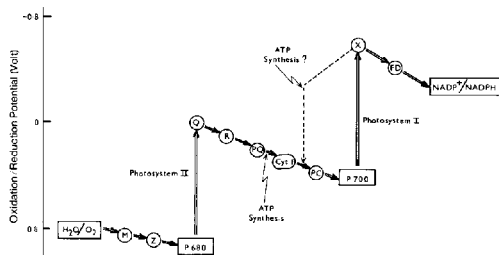
Figure 2. A Hill-Bendall or Z-diagram for electron transport in a chloroplast. The details are discussed in the text together with some suggestions for improving the clarity. See also the addendum concerning the pheophytin intermediate.

ATP (adenosine triphosphate) is also formed from the diphosphate, ADP, in this step.
3) The absorption of CO_2, oxidation of NADPH and formation of (CH_2O):

$$2NADPH + 2H^+ + CO_2 \rightarrow (CH_2O) + 2NADP^+ + H_2O \qquad (6)$$

The process requires three molecules of ATP.

Process (3) is the Calvin-Bassham cycle, which involves a large number of carbohydrates and their various mono- and diphosphates. Process (1), the light-collection step, is the subject of much current work since the nature of the energy transfer steps and the mechanism by which the photoreceptors act is still not well-understood.

It is process (2) to form NADPH which we are concerned with here. The mechanistic scheme was first suggested by Hill and Bendall (9), and it is often known as the Hill-Bendall scheme or as the Z-scheme because of its general shape.

The Hill-Bendall or Z-scheme

A simplified but recent version of the diagram is shown in Figure 2. It can be seen that Figure 2, like Figure 1, is an electrode potential diagram.

Immediately, several points can be made.
1) The majority of the substances are depicted as single entities. In the electrode potential diagram, Figure 1, each level represents not a single species but a half reaction between at least two species.
In Figure 2, most of the materials are not completely characterized chemically, and so presumably, from the viewpoint of those familiar with field, for the electron acceptor in photosystem II, the term Q represents both the reduced and oxidized forms. However, from a novice's viewpoint, single substances with an arrow between them represent a chemical transformation, but this is not the case at all here. The vertical arrows represent electron transfer in the sense of Figure 1.

Ideally each level should show the reaction with hydrogen but, practically it would seem preferable to depict Q as

$$Q_{ox} + e^- \rightleftarrows Q_{red}$$
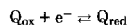
or perhaps as Q_{ox}/Q_{red} when the position of the charges is still not clear and as Q^+/Q or Q/Q^- when the position of the charges is known. It should not be shown as simply Q.
Further, in some representations, the reductions are written in the wrong sense, i.e.,

$$H_2O \rightarrow \tfrac{1}{2}O_2 + 2H^+ + 2e^-$$
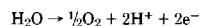
This is to convey the particular direction of the reaction during electron transfer but it can lead to difficulties in calculation if the $E^\ominus$ value is taken, mistakenly, to refer to this direction.
2) The axis is labelled "oxidation/reduction potential,"

presumably because the reactions are reversible. In fact, a scale of standard electrode potentials is used.
3) For those reactions which are well-characterized (e.g., O_2/H_2O) the values given in the diagram are $E^{\ominus\prime}$ values rather than $E^\ominus$ values. Because it is usual in biochemical systems (4, 10) to determine quantities at the physiologically convenient pH of 7, $E^{\ominus\prime}$ and $\Delta G^{\ominus\prime}$ values refer to standard quantities at this pH.
4) For the remaining reactions (e.g., Q_{ox}/Q_{red}, etc.) the values are known approximately from practical measurements and are therefore simply electrode potentials, E values, for the unknown concentrations in the cell.
In a complicated system, under active study, the mixing of precise and imprecise information is unavoidable in order to gain the overall understanding which the diagram gives.
5) The vertical arrows represent two things: (a) the direction of electron transport between the reduced form of one substance and that of another; (b) the quantity of work required to effect the transfer in the upward direction or the work available from a transfer in the downward, spontaneous, direction as indicated in Figure 1. In particular, the upward arrows do not represent the difference in energy between molecular energy levels in the chlorophyll molecule such as would be found in a Jablonski diagram (11). The energy absorbed by the chlorophyll certainly provides the work to effect the transformations in photosystems I and II but the energy used must be greater than the work required.

An Outline Mechanism

The Z-scheme is a suggested mechanism for the overall reaction. It is simplest to follow an electron through the sequence but it must be understood that in the dark all the various reactants and products are at concentrations determined by the equilibrium constants for the reactions (in the absence of side processes). Illumination shifts the equilibria to steady state values which will depend on the light intensity. The process can be looked at sequentially as follows.
1) Light is entrapped by photosystem II (PSII) in which it is thought a complex of the photo-receptor, P680, with a quinone electron acceptor, Q, is excited and undergoes charge transfer
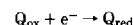
$$(P680_{red} \cdot Q_{ox}) + h\nu \rightarrow P680_{ox} + Q_{red}$$
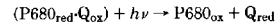
The oxidized form, $P680_{ox}$ undergoes, via intermediate reactions, a reaction with water

$$P680_{ox} + \tfrac{1}{2}H_2O \rightarrow P680_{red} + H^+ + \tfrac{1}{4}O_2$$
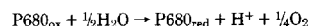
So the net effect is the transfer of an electron as water is oxidized

$$\tfrac{1}{2}H_2O \rightarrow H^+ + \tfrac{1}{4}O_2 + e^-$$
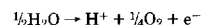
and Q_{ox} reduced

$$Q_{ox} + e^- \rightarrow Q_{red}$$

As the diagram shows, this requires work to be done on the system, the minimum work being

$$w_{min} = \Delta G = -nF[E(Q_{ox}/Q_{red}) - E^{\ominus\prime}(O_2/H_2O)]$$
$$\simeq 77 \text{ kJ mol}^{-1}$$

It is the photon which provides the energy for the work to be done.
2) The reduced quinone (Q_{red}^-) reacts with a plastocyanin via a series of reactions involving a quinone, a plastoquinone and a cytochrome; again an electron is transferred

$$Q_{red} \rightarrow Q_{ox} + e^-$$
$$PC_{ox} + e^- \rightarrow PC_{red}$$
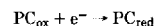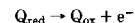
The transfer is a downhill process so it is a spontaneous step and work is available

$$w_{max} = \Delta G$$
$$= -38.6 \text{ kJ mol}^{-1}.$$

Some of the available work is thought to be used for the conversion of adenosine diphosphate (ADP) to the triphosphate

$$ADP + P \rightarrow ATP$$
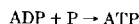
for which the standard change in Gibbs function is

$$\Delta G^{\ominus'} = 30.5 \text{ kJ mol}^{-1}$$

It is known that half a mole of ATP is formed per mole of electrons transferred and so it appears that less than half of the available work is used in this conversion.

The synthesis of ATP presents a further problem in using the electrode potential diagram: it is not a redox reaction and so cannot be represented by electron transfer or by a standard electrode potential and so cannot be shown by a level on the diagram.

3) In photosystem I, the photoreceptor P700 reacts in an electron-transfer reaction with the substance, X,

$$(P700_{red} \cdot X_{ox}) + h\nu \rightarrow P700_{ox} + X_{red}$$
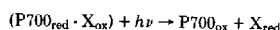
and the oxidized, $P700_{ox}$ reacts with the reduced plastocyanin

$$P700_{ox} + PC_{red} \rightarrow P700_{red} + PC_{ox}$$
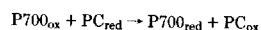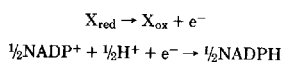
so that the overall process is electron transfer from PQ_{red} to X_{red}.

The minimum work required is

$$w = \Delta G = -nF[E(X_{ox}/X_{red}) - E(PQ_{ox}/PQ_{red})]$$
$$\simeq +96.5 \text{ kJ mol}^{-1}$$

which is provided by the photon.

4) The final steps involve the reduction of $NADP^+$ and oxidation of X_{red} again through a sequence involving a ferridoxin

$$X_{red} \rightarrow X_{ox} + e^-$$

$$\tfrac{1}{2}NADP^+ + \tfrac{1}{2}H^+ + e^- \rightarrow \tfrac{1}{2}NADPH$$

The work here is

$$w = \Delta G = -nF[E^{\ominus'}(NADP^+/NADPH) - E(X_{ox}/X_{red})]$$
$$= -26.6 \text{ kJ mol}^{-1}.$$

The process is spontaneous but the work available is not apparently used.

The mechanism has been described sequentially in a chemist's way, but, in fact, all the reactants and products together with the enzymes which catalyse the reactions are part of a membrane in the chloroplast which absorbs light thus providing the energy for proton transfer through the membrane. The protons are used to synthesize ATP. The NADPH produced is used in CO_2 fixation.

Quantum Requirement and Energy Yield

The diagram can be used to calculate the minimum quantum requirement and the maximum energy yield.

To produce 1 mole of O_2 and 2 moles of NADPH at an intermediate stage and absorb 1 mole of CO_2, 4 moles of electrons are required, and these must pass through the sequence of reactions outlined. At the wavelength corresponding to the threshold for photosynthetic activity (~700 nm) the energy absorbed (171 kJ mol^{-1}) for each photon is more than sufficient to raise 1 electron in either photosystem. Much of the energy is lost in the initial steps by vibrational relaxation and by second law effects, but, if we assume that 1 photon per electron is required, then there is a minimum quantum requirement of 8 for the passage of 4 electrons through the two photosystems.

Three moles of ATP are also required to fix 1 mole CO_2 in the Calvin cycle. The passage of 4 electrons will produce 2 molecules of ATP in step (2) above, so a further molecule is required. Ideas on where this is formed have varied; one suggestion is that it is formed in an alternative pathway to the production of NADPH in a cycle which includes photosystem I. The second possibility would require extra photons so the requirement would then be more than 8. The actual value is difficult to measure but is thought (12) to lie between 8 and 12.

The value of 8 can be used to calculate the maximum energy yield for the Z-scheme. The 8 photons at 700 nm provide an equivalent energy of 1368 kJ mol$^-$. The minimum work required to reduce H_2O and form NADP is given by

$$2H_2O + 2NADP^+ \rightarrow 2NADPH + 2H^+ + O_2$$

$$w = \Delta G^{\ominus'} = -nF[E^{\ominus'}(NADP^+/NADPH) - E^{\ominus'}(O_2/H_2O)]$$
$$= +440 \text{ kJ mol}^{-1}$$

for 4 electrons and so the maximum yield is 32%.

Some of the energy is used in the formation of ATP. The remainder appears as heat, with losses occurring at every stage: in the rapid vibrational relaxation in the excited singlet state of chlorophyll, in the energy transfer to the reaction center, and in all the various reactions which, because they occur in an actual system, are subject to second law losses. There will also be substantial further losses in the Calvin Cycle.

Conclusion

This article has outlined the use of electrode potential diagrams in a simple system, and in the Hill-Bendall scheme for photosynthesis, for which the diagram provides a basis for understanding the mechanism.

Emphasis has been placed (1) on the use of arrows to depict electron transfer, with a rule to give the correct direction of the coupled reactions; (2) on the fact that the diagram is a free-energy diagram and not an energy diagram, and (3) on the fact that the energy quantities corresponding to heights represent the maximum work available or the minimum work required for particular processes.

It has been suggested that to help people less familiar with these diagrams, the scales should be labelled as electrode potentials and that each level should be shown as a redox reaction rather than as a single substance.

While the mechanism has been described in outline, the purpose has not been to give the experimental evidence or the fine details which are well described elsewhere (1–3).

The various conclusions could also be applied to the description of the cytochrome electron-transport scheme in respiration (4).

Addendum

The referee has kindly pointed out a recent suggestion that a pheophytin (Ph) acts as a further intermediate between P680 and Q in photosystem II. Two measurements have indicated that electrode potential for the reduction of the pheophytin ($e^- + Ph^- \rightarrow Ph$) is ~610 mV. The implication of these findings is that the presently accepted mechanism, outlined in Figure 2, may need substantial revision. However, the conclusions of the present article are still valid since it is concerned with the interpretation of electrode potential diagrams rather than with the detailed mechanism.

Literature Cited

(1) Govindjee and Jursinic, P. A., Photochem. Photobiol. Rev. 4, 125 (1979).
(2) Arnon, D. I., Proc. Natl. Acad. Sci. (U.S.A.), 68, 2883 (1971).
(3) Levine, R. P., Sci. Amer., 58 (Dec. 1969).
(4) Lehninger, A. L., "Short Course in Biochemistry," Worth, New York, 1975.
(5) McGlashan, M. L., "Chemical Thermodynamics," Academic Press, London 1979.
(6) Atkins, P. W., "Physical Chemistry," Oxford Press 1979.
(7) Weast, R. C., "Handbook of Chemistry and Physics," Chem. Rubber Publ. Co., Cleveland, 1975.
(8) Dannat, C. W., and Ellingham, H. J. T., Discuss. Faraday Soc., 4, 126 (1948).
(9) Hill, R., and Bendall, F., Nature, 186, 136 (1960).
(10) Sober, H., "Handbook of Biochemistry," Chem. Rubber Publ. Co., Cleveland, 1978.
(11) Calvert, J. G., and Pitts, J. N., "Photochemistry," John Wiley, New York, 1966.
(12) Fong, F. K., J. Amer. Chem. Soc., 98, 7840 (1976).
(13) Rutherford, A. W., Mullet, J. E., and Crofts, A. R., F.E.B.S. Letters, 123, 235 (1981).
(14) Klimov, V. V., Allakverdiev, S. I., Demeter, S., and Krasnovskii, A. A., Dokl. Akad. Nauk. SSSR (Biophys.), 249, 227 (1979).