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Interaction of Benzoquinones with Q_A^- and Q_B^- in Oxygen-Evolving Photosystem II Particles from the Thermophilic Cyanobacterium *Synechococcus elongatus*

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The interactions of benzoquinones with the reduced forms of the bound plastoquinone acceptors, Q_A and Q_B, were studied with oxygen-evolving photosystem II (PS II) particles from the thermophilic cyanobacterium *Synechococcus elongatus*, which largely lack pool plastoquinone molecules [Takahashi and Katoh (1986) *Biochim. Biophys. Acta* 845: 183]. Oxygen evolution in the presence of various electron acceptors was determined and flash-induced changes in absorbance in the blue region were analyzed in terms of difference spectra, dependence on the concentration of benzoquinone and on temperature, and sensitivity to 3-(3,4-dichlorophenyl)-1,1-dimethylurea (DCMU). The more hydrophobic the quinone molecule, the higher was the rate of oxygen evolution, and the maximum rate of 3,000 μmoles $O_2 \cdot (\text{mg chlorophyll})^{-1} \cdot h^{-1}$ was recorded in the presence of phenyl- and dichloro-*p*-benzoquinones. DCMU inhibited oxygen evolution by more than 95%. However, spectrophotometric studies revealed that, even though electrons were transferred to benzoquinones predominantly via Q_B^{2-}, the direct oxidation of Q_A^- by added benzoquinones occurred in such a way as to indicate that about 40% of PS II reaction centers were not associated with functional Q_B sites. Q_B^- was very stable in the presence of ferricyanide. However, benzoquinones induced the slow oxidation of Q_B^-. The characteristics of the benzoquinone reductioin in the PS II preparation is discussed.

Key words: Benzoquinone — Electron acceptor — Oxygen evolution — Q_A — Q_B binding site — *Synechococcus elongatus*.

A preparation of PS II that was active in the evolution of oxygen was first isolated from the thermophilic cyanobacterium *Phormidium laminosum* (Stewart and Bendall 1979). Since then, a number of oxygen-evolving PS II preparation have been isolated from various cyanobacteria and higher plants (England and Evans 1981, Berthold et al. 1981, Yamamoto et al. 1982, Kuwabara and Murata 1982, Schatz and Witt 1984, Satoh and Katoh 1985) and they have provided a wealth of information about electron transport on the water side of the PS II reaction center (see reviews by Govindjee et al. 1985, Dismukes 1986, Homann 1988). Less is known, however, about electron transport on the reducing side of the reaction center in such preparations. Various oxidants, including several benzoquinones, can serve as electron acceptors (Satoh and Katoh 1985, Ikeuchi and Inoue 1986) but details of the interactions between exogenously added oxidants and an endogenous reductant(s) produced in the light remain to be elucidated.

The endogenous electron acceptors of PS II include two bound plastoquinone molecules and a pool of free plastoquinone molecules that are present in the fluid lipid phase of the thylakoid membranes (Bouges-Bocquet 1973, Vermas and Govindjee 1981, Crofts and Wraight 1983). On illumination, the first bound plastoquinone molecule, Q_A, accepts an electron from the primary donor chlorophyll P680 via pheophytin and in turn is oxidized by the second bound plastoquinone molecule, Q_B. Q_A is a one-electron carrier, whereas O_B accumulates two negative charges: Q_B^- produced by the transfer of one electron from Q_A^- binds strongly to its binding site but Q_B^{2-} generated by

Abbreviations: Q_A, primary quinone electron acceptor; Q_B, secondary quinone electron acceptor; DCIP, 2,6-dichlorophenolindophenol.

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the second turnover of Q_A is replaced by a pool plastoquinone molecule.

The number and function of plastoquinone molecules associated with an oxygen-evolving PS II preparation from *Synechococcus elongatus* have been studied in detail (Takahashi and Katoh 1986). Functional Q_A and Q_B moieties are present in the preparation which contains about three plastoquinone molecules per Q_A. Thus the preparation lack any significant pool of plastoquinone. Reduction of exogenously added benzoquinones in the cyanobacterial PS II preparation, therefore, serves as an interesting model system for studies of the electron transfer to the plastoquinone pool in situ.

In the present study, the interactions between various added benzoquinones and reduced forms of Q_A and Q_B in PS II preparation from *Synechococcus* was examined. First, rates of oxygen evolution with various electron acceptors were determined. Then, flash-induced absorption changes of Q_A and Q_B were analyzed in detail in order to characterize electron transfer from the bound plastoquinones to the added benzoquinones.

Materials and Methods

Thylakoid membranes were prepared from two-day grown cells of *Synechococcus elongatus*. (Yamaoka et al. 1978, Hirano et al. 1980) and oxygen-evolving PS II preparations were isolated with β-octylglucoside by the method described previously (Satoh and Katoh 1985).

Oxygen evolution was monitored with a Clark-type oxygen electrode (Yamaoka et al. 1978). Photoreduction of DCIP was determined by measuring light-induced decrease in absorbance at 580 nm with a Hitachi 356 spectrophotometer (Yamagishi and Katoh 1984). The actinic light used was saturating. The basal reaction medium contained 1 M sucrose, 50 mM MES/NaOH (pH 6.5), 10 mM NaCl, 5 mM $MgCl_2$ and PS II preparations that contained 2–3 g chlorophyll *a*, except in the case of ferricyanide-supported oxygen evolution, which was determined in the presence of 50 mM MES/NaOH (pH 5.0). Final concentrations exogenously added electron acceptors were 0.4 mM in the case of benzoquinones and ferricyanide and 0.2 mM in the case of DCIP.

Flash-induced absorption changes were determined at 25°C with a single-beam spectrophotometer (Koike et al. 1978, Hirano et al. 1980). A dye laser (Phase R DL-1100) with Red 6 (oxazine 720) in ethanol was used as the source of saturating 690 nm flashes with a half peak-height duration of 300 ns. Flashes were passed through Toshiba VR 65 and VR 66 filters and the photomultiplier was protected with two Corning 4-96 filters. The repetition rate of flashes was 1 Hz, unless otherwise stated, and signals were averaged with a microcomputer attached to the spectrophotometer. Averaged signals were transfered to and analyzed by another microcomputer (NEC PC9801 VM2). The reaction medium was the same as that for the assays of oxygen evolution with the exception that 50 mM MES/NaOH (pH 5.5) and PS II preparations containing 8 μM chlorophyll/ml were used. The temperature of the reaction medium was controlled by the circulation of thermostated water through the water jacket of the cuvette and monitored with a calibrated copper-constantan thermocouple.

Results

Oxygen evolution in the presence of various benzoquinones—Rates of oxygen evolution in *Synechococcus* PS II preparations were determined in the presence of various electron accpetors and the results obtained are summarized in Table 1. The activity was determined at 30°C because oxygen evolution with benzoquinone acceptors were unstable at higher temperatures. The rate of oxygen evolution in the presence of ferricyanide was maximum at pH 5.0, whereas the optimum pH for oxygen evolution in the presence of benzoquinones and DCIP was between 6.0 and 7.5 in each case (Fig. 1). The rates of oxygen evolution were measured at or near respective optimal pH values. Digitonin, which has been shown to stimulate oxygen evolution in the presence of ferricyanide at 45°C (Satoh and Katoh 1985) was less effective under the present experimental conditions and often slightly inhibited the evolution of oxygen. The activity was, therefore, determined in the absence of this detergent.

The rate of oxygen evolution varied markedly with the electron acceptor added, indicating that the overall electron transport is limited at the step of acceptor reduction. The highest rates of oxygen evolution as much as 3,000 μmoles $O_2 \cdot (\text{mg Chl})^{-1} \cdot h^{-1}$ were obtained with phenyl-*p*-benzoquinone and dichloro-*p*-benzoquinone. Note that the rate of oxygen evolution increased in following order: *p*-benzoquinone, methyl-*p*-benzoquinone, dimethyl-*p*-ben-

Table 1 Rates of oxygen evolution in the presence of various electron acceptors

Acceptors	(μmoles $O_2 \cdot (\text{mg Chl})^{-1} \cdot h^{-1}$) No addition	10 μM DCMU
Phenyl-*p*-benzoquinone	2,940	110
2,6-Dichloro-*p*-benzoquinone	2,660	110
Tetramethyl-*p*-benzoquinone	1,410	~0
Ferricyanide	1,210	70
2,6-Dimethyl-*p*-benzoquinone	520	~0
2,6-Dichlorophenolindophenol	500	190
2,5-Dimethyl-*p*-benzoquinone	490	—
p-Benzoquinone	450	~0

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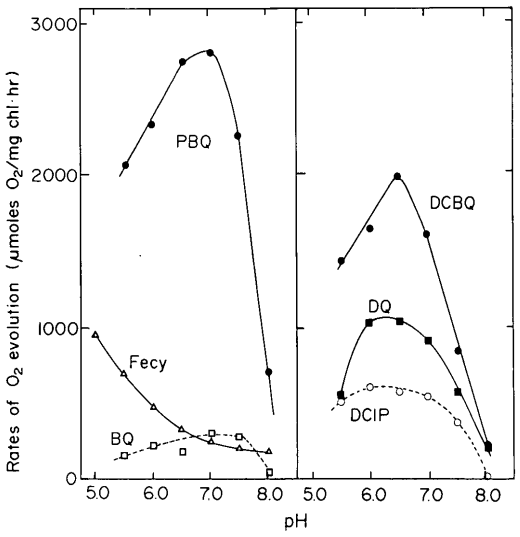

Fig. 1 Dependence on pH of oxygen evolution in the presence of various electron acceptors. Buffers used were 50 mM MES/NaOH between pH 5.0 and 6.5 and 50 mM HEPES/NaOH between pH 7.0 and 8.0. PBQ, phenyl-*p*-benzoquinone; Fecy, ferricyanide; BQ, benzoquinone; DCBQ, 2,6-dichloro-*p*-benzoquinone; DQ, duroquinone (tetramethyl-*p*-benzoquinone).

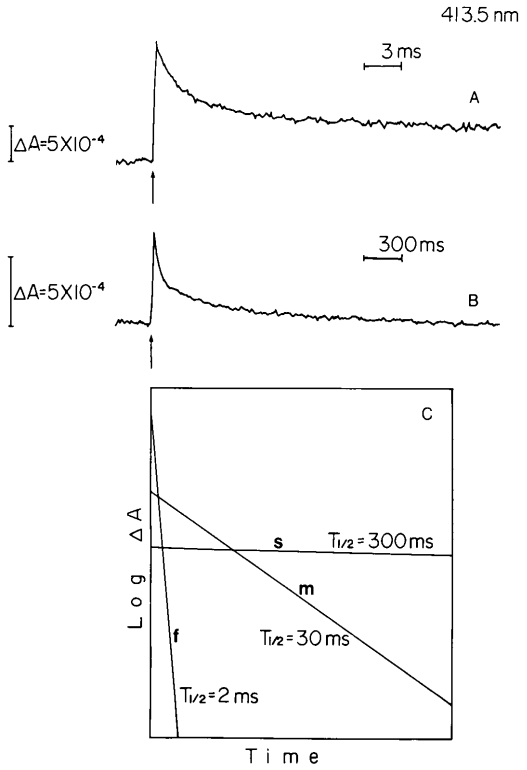

Fig. 2 Kinetics of flash-induced change in absorption at 413.5 nm. Absorption changes were determined in the presence of 0.1 mM dichloro-*p*-benzoquinone. A, 30 flashes were fired at 1 Hz; B, 10 flashes were fired with an interval of 6 s between each flash. C, Deconvolution of decay kinetics.

zoquinones and tetramethyl-*p*-benzoquinone. Clearly, the more hydrophobic the acceptor molecule, the higher is the rate of oxygen evolution. However, the efficiency of an electron acceptor is not determined solely by the hydrophobicity of the acceptor molecule. Ferricyanide, which carries strongly negative charges, supported oxygen evolution at a rate similar to that observed in the presence of tetramethyl-*p*-benzoquinone.

DCMU was strongly inhibitory on oxygen evolution with all the acceptors tested (Table 1). High rates of oxygen evolution supported by phenyl-*p*-benzoquinone and dichloro-*p*-benzoquinone were depressed by more than 95% in the presence of 10 μM DCMU. This observation, together with high rates of oxygen evolution observed with several benzoquinones, suggests that the cyanobacterial preparation of PS II has a relatively intact electron-transport system on the reducing side of the PS II reaction center and that benzoquinones are reduced via Q_B (but see below).

Spectrophotometric investigations—Interactions of benzoquinones with Q_A and Q_B were studied by measuring flash-induced changes in absorption in the blue region. Figure 2A shows the kinetics of absorption changes at 413.5 nm determined for two different time scales in the presence of dichloro-*p*-benzoquinone. Flash excitation induced a very rapid absorbance increase, which was followed by a multiphasic decay to the dark level. The decay kinetics could be resolved into three exponential components with half times of 1.3 to 2.0 ms, 13 to 30 ms and about 300 ms in the presence of 0.1 mM dichloro-*p*-benzoquinone and at 25°C. The three exponential components, which are called the f, m and s components in order of their appearance, accounted for about 50%, 40% and 10% of the total relaxation, respectively (see Fig. 4 B).

The magnitudes of each kinetic component are plotted against wavelength in Figure 3. The difference spectrum for the f component shows maxima at 395, 415 and 440 nm and a trough at 430 nm. These features are similar to those of the difference spectrum for the reduction of Q_A that can be ascribed, for the most part, to electrochromic shifts of pigments responding to the reduction of Q_A (Dekker et al. 1984, Schatz and van Gorkom 1985). A large peak at 430 nm may be explained if we assume electron transfer from Q_A to Q_B because, whereas reduction of Q_A is accompanied by no change or only a small change in absorption, reduction of Q_B results in a considerable increase in absorption at this wavelength (Schatz and var Gorkom 1985). The f component cannot be related to a back reaction of Q_A^- with a donor-side component because this component was eliminated in the presence of DCMU (data not shown). The half time of the f component was longer than that of the $Q_A^-Q_B$ to $Q_AQ_B^-$ transition in other photosynthet-

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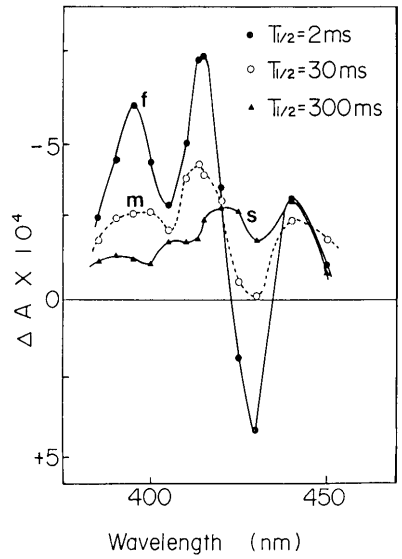

Fig. 3 Difference spectra of the three kinetic components. Experiments were carried out as described in the legend to Fig. 2. To permit convenient comaprisons, decreases in absorption are plotted upwards.

ic organisms, which ranges from 0.1 to 0.6 ms (Stiehl and Witt 1969, Robinson and Crofts 1983), but this difference can be ascribed to the assay temperature which was approximately 30°C below the growth temperature of the cyanobacterium (see below). However, a contribution of redox components on the donor side of PS II to this rapid-decay phase cannot be totally excluded (Schatz and van Gorkom 1985).

The m component is also related to Q_A because its difference spectrum, without a negative peak at 430 nm, resembles that of the reduction of Q_A. The spectrum of the s component is different from those of the faster components but similar to that of the reduction of Q_B (Schatz and van Gorkom 1985). Thus, the m and s components can be ascribed to oxidation of Q_A^- and Q_B^-, respectively, either with dichloro-*p*-benzoquinone added or by back-transfer of electrons to an oxidant produced by the flash on the oxidizing side of the PS II reaction center.

Further characterization of the three kinetic components was carried out via measurements of absorption changes in the presence of various concentrations of dichloro-*p*-benzoquinone. The half time of the f component was totally independent of the concentration of quinone (Fig. 4 A-a). This result is consistent with the above conclusion that the f component represents electron transfer from Q_A to Q_B. In contrast, the half time of the m component decreased significantly with increasing concentrations of the electron acceptor (Fig. 4 A-b). Thus, the m component represents the chemical oxidation of Q_A^- by dichloro-*p*-benzoquinone and not by a back reaction. In fact, this kinetic component was totally insensitive to DCMU (Fig. 4 A-b), which strongly suppressed the other decay components (data not shown). This result also strongly suggests that Q_A^- is oxidized by benzoquinone molecules that are not bound to the Q_B site. The magnitude of the total absorption change was only slightly reduced with increasing concentrations of quinone (not shown) and the results in Figure 4 (right) indicate that the relative magnitude of the m component remained constant at all the concentrations of quinone examined. Thus, unexpectedly, about 40% of PS II reaction centers are not associated with the functional Q_B site and donate electrons directly from Q_A to benzoquinone in the cyanobacterial preparations used here.

Figure 4-c shows that the s component was also accelerated in the presence of benzoquinone, suggesting the direct oxidation of Q_B^- by exogenously added dichloro-*p*-benzoquinone. This was also unexpected because Q_B^- is considered to be a very stable plastosemiquinone that is unable to give electrons to free plastoquinone molecules (Crofts and Wraight 1983). However, the results of the following experiments confirmed a quinone-dependent oxidation of Q_B^-. Figure 5A shows absorption changes at 413.5 nm that were induced by four successive flashes fired at 1 Hz in the presence of 0.4 mM ferricyanide. After each flash, the signal decayed to a constant level. Note that the levels attained after the first and third flashes above the dark level recorded prior to the flash excitation, whereas the levels fell below the dark level after the second and fourth flashes. The results are consistent with the two-electron gate model of Q_B (Bouges-Bocquet 1973, Crofts and Wraight 1983) which predicts that even numbers of flashes produce Q_B^{2-}, which is rapidly replaced by a free plastoquinone molecule, whereas odd numbers of flashes result in formation of Q_B^- which remains stably bound to the binding site. The difference spectra indicate that the dominant species that contribute to absorbance changes at 2 and 70 ms after the first flash are in fact Q_A^- and Q_B^-, respectively (Fig. 6). A diminished amplitude of the oscillation was also observed in the presence of 0.2 mM DCIP (data not shown). In contract, no binary oscillation in levels of Q_B^- was observed in the presence of 0.4 mM dichloro-*p*-benzoquinone (Fig. 5b) or of phenyl-*p*-benzoquinone (not shown). These results suggest that Q_B^- is oxidized by benzoquinones, but not by ferricyanide, during the 1-s interval between flashes. Figure 7 shows that this is indeed the cases. In this experiment, only a single flash was given and absorption changes were monitored for an extended period of 6 s. Although there was no appreciable decay of Q_B^- in the presence of ferricyanide, Q_B^- was oxidized with a half time of 300–400 ms in the presence of quinone acceptors. DCIP also induced the slow oxidation of Q_B^-.

All the above experiments were carried out at 25°C, which is about 30°C below the growth temperature of the thermophilic cyanobacterium (Yamaoka et al. 1978).

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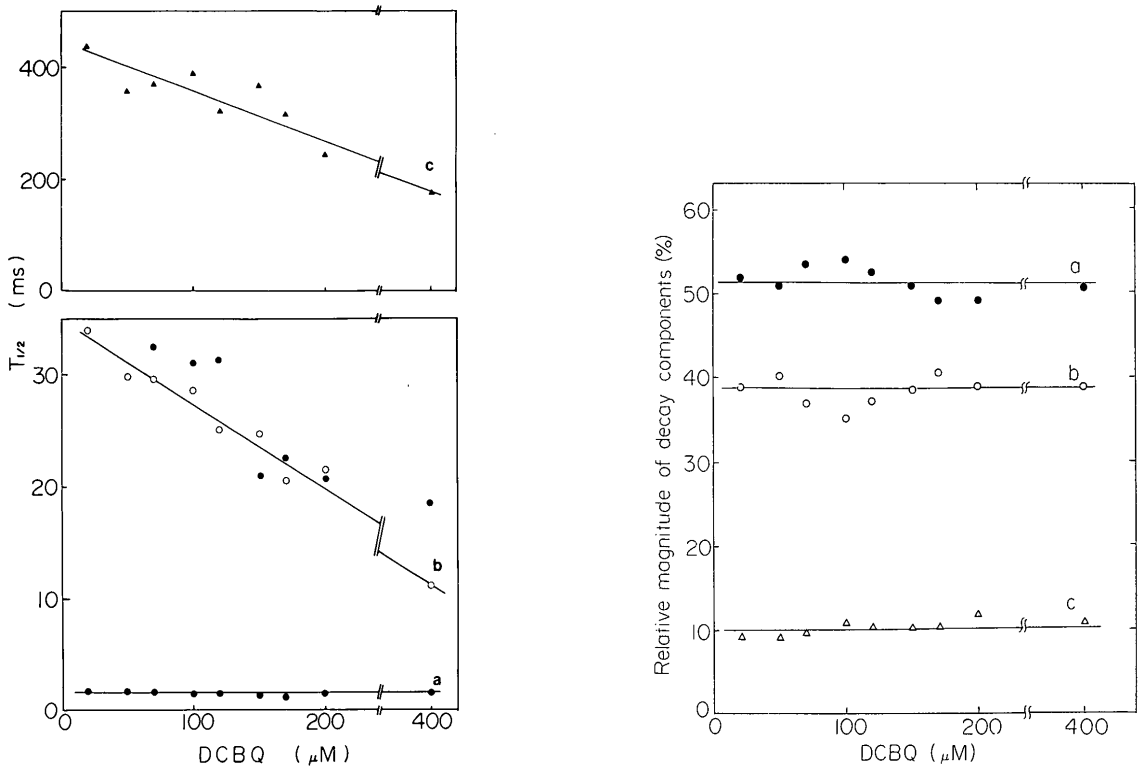

Fig. 4 Dependence upon the concentration of dichloro-*p*-benzoquinone of half times (left) and relative magnitudes (right) of the three kinetic components. Experiments were carried out as in Fig. 2. a, f component; b, m component (in right panel, open and solid circles indicate the m component determined in the absence and presence of 10 μM DCMU, respectively); c, s component.

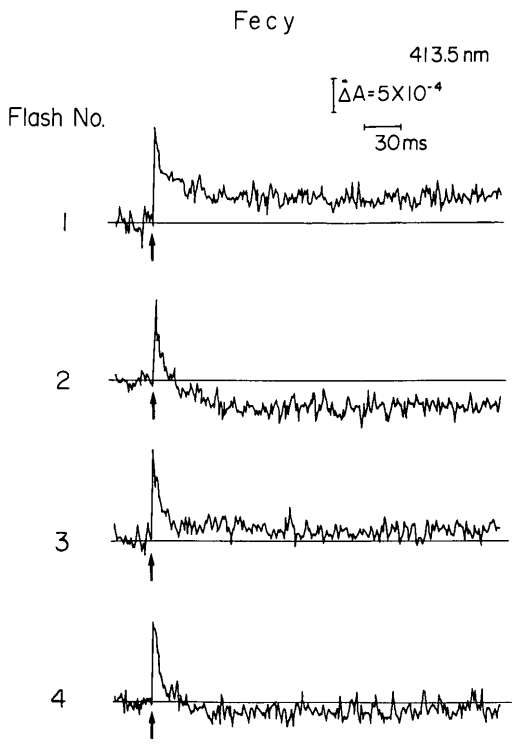

Fig. 5A

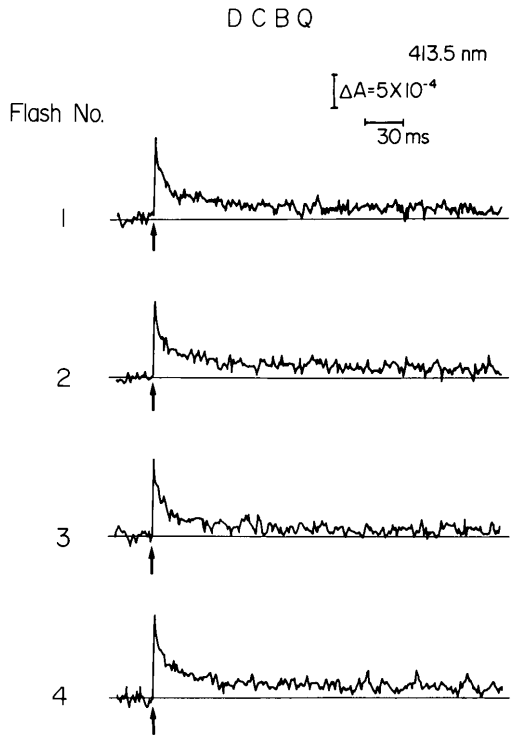

Fig. 5B

Fig. 5 Dependence of absorption changes on the number (odd or even) of flash in the presence of 0.4 mM ferricyanide (A) and dichloro-*p*-benzoquinone (DCBQ) (B). Experiments were carried out as in Fig. 2. For other details, see text.

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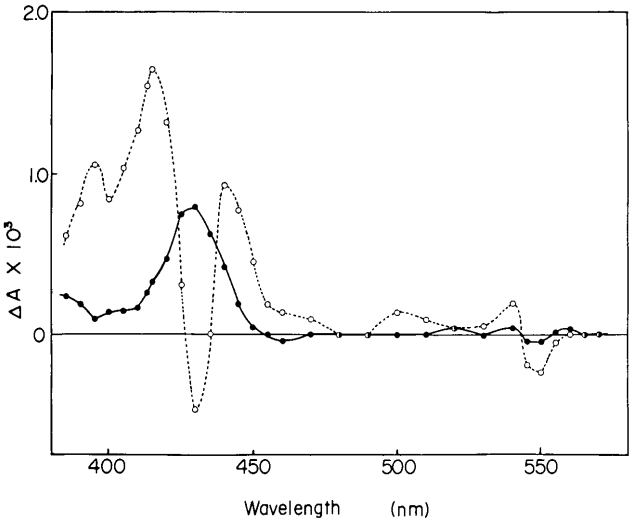

Fig. 6 Difference spectra for absorption changes determined 2 ms (open circles) and 70 ms (solid circles) after excitation by a single flash in the presence of 0.4 mM ferricyanide. Experimental conditions were as in Fig. 5A.

Spectrophotometric experiments were repeated over a wide range of temperatures and the results obtained are illustrated in Fig. 8. The rate of the f component was markedly accelerated as the temperature was raised, showing an apparent activation energy of about 42 kJ/mol. Comparable values have been reported for the apparent activation energy of electron transfer from Q_A to Q_B in photosynthetic bacteria (Chamorovsky et al. 1976, Mancino et al. 1984). When extrapolated to 55°C, the half time decreases to 300 μs, which is in the range of the half times reported for the $Q_A^- Q_B$ to $Q_A Q_B^-$ transition in other photosynthetic organisms (Stiehl and Witt 1969, Robinson and Crofts 1983). Both the m and the s components were less sensitive than the f component to temperature. Although the data points were scattered, the activation energies of the two components were estimated to be in the range of 12 to 16 kJ/mol.

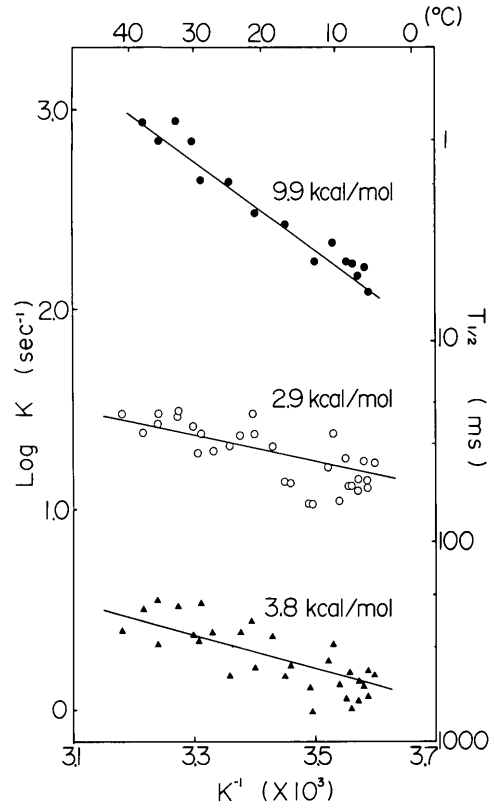

Fig. 8 Dependence on temperature of the three kinetic components. Solid circles, f component; open circles, m component; triangles, s component.

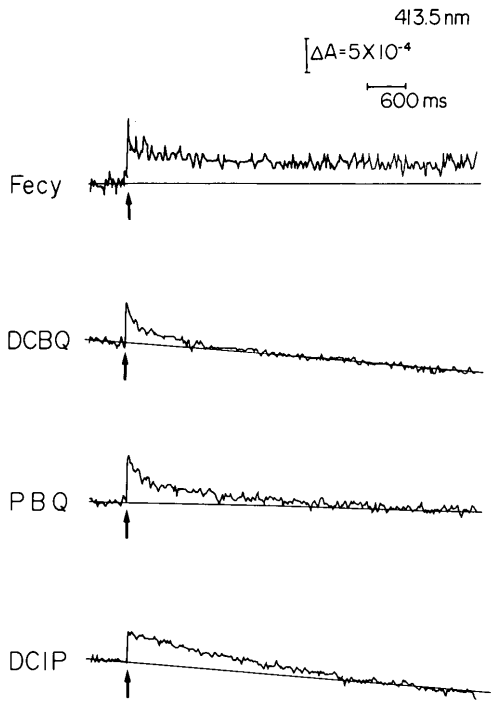

Fig. 7 Decay kinetics of absorption changes induced by excitation with a single flash in the presence of 0.4 mM ferricyanide (Fecy), dichloro-*p*-benzoquinone (DCBQ), phenyl-*p*-benzoquinone (PBQ) or 0.2 mM DCIP.

Discussion

The PS II particles from *Synechococcus* used in the present studies were highly competent in terms of the evolution of oxygen with benzoquinone as electron acceptor. DCMU strongly inhibited the benzoquinone-supported oxygen evolution. Because DCMU specifically inhibits the oxidation of Q_A^- by Q_B by blocking binding of plastoquinone to the Q_B site (Velthuys 1981, Croft and Wraight 1983), the strong inhibition of oxygen evolution by the inhi-

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bition is considered to be indicative of the functioning of Q_B in electron transport on the reducing side of PS II. The present results indicate, however, that this assumption does not always hold true. Exogenously added benzoquinone accepted electrons not only from Q_B but also from Q_A in the PS II preparations and the kinetic data indicated that approximately 40% of the PS II reaction centers have no functional Q_B site and, hence, donate electrons directly from Q_A to benzoquinone.

The rate of electron transfer from Q_B^{2-} to benzoquinones could not be determined spectrophotometrically. However, because the overall electron transport from water to benzoquinone is limited by the rate of reduction of benzoquinone, the rate of electron transfer from Q_B^{2-} to phenyl- or dichloro-*p*-benzoquinone can be estimated from the steady-state rate of oxygen evolution. If we assume a chlorophyll to Q_A ratio of 50 in the PS II preparation (Satoh and Katoh 1985, Satoh et al. 1985), the maximum rate of 3,000 μmoles $O_2 \cdot (\text{mg Chl})^{-1} \cdot h^{-1}$ corresponds to an electron transfer rate of 152 s^{-1} at 30°C. Thus, the half time required for an electron to be transferred to benzoquinones is 4.6 ms. However, about 40% of the PS II reaction centers have no Q_B site and donate electrons from Q_A with half times of about 30 ms at temperatures in the region of 25–30°C. When this percentage is taken into account, the half time required for an electron to be transferred is 2.7 ms. The steady-state rate of electron transfer from Q_B^{2-} to benzoquinone is, therefore, at least one order of the magnitude greater than that from Q_A^-. Thus, benzoquinones accept electrons predominantly via Q_B^{2-}. This conclusion explains why, in spite of the absence of Q_B from about 40% of the PS II reaction centers, DCMU inhibits the steady-state rate of oxygen evolution by more than 95%. Thus, the strong inhibition of oxygen evolution by DCMU cannot be regarded as solid evidence for the integrity of the electron-transport system on the reducing side of PS II.

In higher plants, it has been suggested that PS II reaction centers are heterogeneous in terms of the secondary electron acceptor, one part being connected with Q_B and another part being nonfunctional in Q_B reduction (Lavergne 1982, Melis 1985, Chylla and Whitmarsh 1989). The present results suggest that a similar situation occurs in the cyanobacterium. However, there is no evidence for any heterogeneity in situ of PS II reaction centers in cyanobacteria. The possibility still exists, therefore, that about 40% of the binding sites for Q_B are damaged during preparation of the PS II particles with detergents. It should also be noted that the f component is ascribed to the transition of $Q_A^- Q_B$ to $Q_A Q_B^-$ on the basis of its difference spectrum, sensitivity to DCMU and insensitivity to the concentration of quinone, but the f component may additionally incorporate a contribution from donor-side component(s). If such were the case, the fraction of PS II reaction centers without the Q_B site would be slightly larger than estimated above.

An unexpected finding was the slow oxidation of Q_B^- in the presence of benzoquinones. Q_B^- is generally considered not to be oxidized by free plastoquinone molecules, so that a characteristic oscillation in level of Q_B^- occurs under repetitive flash excitations. In fact, a clear binary oscillation was observed in the presence of ferricyanide. It is, therefore, remarkable that Q_B^- undergoes slow oxidation in the presence of benzoquinones. The rate of quinone-dependent oxidation of Q_B^- is much lower than that of Q_A^- but it is high enough to abolish the binary oscillation of Q_B with intervals of 1 s between flashes.

A question arises as to why Q_B^-, which cannot donate its electron to endogenous plastoquinone, is oxidized by exogenously added benzoquinones. We suggest that added benzoquinones bind to the Q_B sites that are associated with about 60% of PS II reaction centers and accpet electron directly from Q_A^-. (This type of benzoquinone reduction must be distinguihed from the m component which represents the reduction of benzoquinone that is not bound to the Q_B site.) Semiquinones formed in this way have lower affinities for the Q_B site than does plastosemiquinone so that an apparent oxidation of Q_B^- occurs as the semiquinones are slowly released from the binding sites and dismutate. The slow oxidation of Q_B^- in the presence of DCIP also suggests reduction of the dye at the Q_B site. The binding of several benzoquinones to the Q_B site has been suggested previously to explain quinone-dependent photooxidation of iron (Fe II) located near Q_A and Q_B (Zimmerman and Rutherford 1986, Diner and Petrouleas 1987). Benzoquinones have also been shown to reactivate a Q_B function in spinach thylakoids from which Q_B had been extracted (Wydrzynski and Inoue 1987).

Replacement of plastoquinone by a benzoquinone would not be expected to cause a large change in the difference spectrum of the oxidation or reduction of $Q_{B'}$. However, the rate of oxidation of Q_A^- may depend upon the species of quinone bound to the Q_B site. We observed consistently that the half time of the f component was slightly shorter in the presence of dichloro-*p*-benzoquinone than in the presence of ferricyanide (data not shown). More detaild studies of the interactions of various quinones with the Q_B site in cyanobacterial PS II preparations are in progress.

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References

Berthold, D. A., Babcock, G. T. and Yocum, C. F. (1981) A highly resolved, oxygen evolving Photosystem II preparation

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from spinach thylakoid membranes. EPR and electron-transport properties. *FEBS Lett.* 134: 231–234.

Bouges-Bocquet, B. (1973) Kinetic models for the electron donors of photosystem II of photosynthesis. *Biochim. Biophys. Acta* 594: 58–103.

Chamorovsky, S. K., Remennikov, S. M., Konoenko, A. A., Venediktov, P. S. and Rubin, A. B. (1976) New experimental approach to the estimation of rate of electron transfer from the primary to secondary acceptors in the photosynthetic electron transport chain of purple bacteria. *Biochim. Biophys. Acta* 430: 62–70.

Chylla, R. A. and Whitmarsh, J. (1989) Inactive photosystem II complexes in leaves. Turnover rate and quantitation. *Plant Physiol.* 90: 765–772.

Crofts, A. R. and Wraight, C. A. (1983) The electrochemical domain of photosynthesis. *Biochim. Biophys. Acta* 762: 149–185.

Dekker, J. P., van Gorkom, H. J., Wensink, J. and Ouwehand, L. (1984) Absorbance difference spectra of the successive redox states of the oxygen-evolving apparatus of photosynthesis. *Biochim. Biophys. Acta* 767: 1–9.

Diner, B. A. and Petrouleas, V. (1987) Light-induced oxidation of the acceptor-side Fe (II) of Photosystem II by exogenous quinones acting through the Q_B binding site. II. Blockage by inhibitors and their effects on the Fe (III) EPR spectra. *Biochim. Biophys. Acta* 893: 138–148.

Dismukes, G. C. (1986) The metal centers of the photosynthetic oxygen-evolving complex. *Photochem. Photobiol.* 43: 99–115.

England, R. R. and Evans, E. H. (1981) A rapid method for extraction of oxygen-evolving Photosystem 2 preparation from cyanobacteria. *FEBS Lett.* 138: 175–177.

Govindjee, Kambara, T. and Coleman, W. (1985) The electron donor side of photosystem II: The oxygen evolving complex. *Photochem. Photobiol.* 42: 187–210.

Hirano, M., Satoh, K. and Katoh, S. (1980) Plastoquinone as a common link between photosynthesis and respiration in a blue-green alga. *Photosyn. Res.* 1: 149–162.

Homann, P. H. (1988) Explorations in the "inner sanctum of the photosynthetic process" the water-oxidizing system. *Plant Physiol.* 88: 1–5.

Ikeuchi, M. and Inoue, Y. (1986) Characterization of O_2 evolution by a wheat photosystem II reaction center complex isolated by a simplified method: Disjunction of secondary acceptor quinone and enhanced Ca^{2+} demand. *Arch. Biochem. Biophys.* 247: 97–107.

Koike, H., Satoh, K. and Katoh, S. (1978) Effects of dibromothymoquinone and bathophenanthroline on flash-induced cytochrome *f* oxidation in spinach chloroplasts. *Plant Cell Physiol.* 19: 1371–1380.

Kuwabara, T. and Murata, N. (1982) Inactivation of photosynthetic oxygen evolution and concomitant release of three polypeptides in the photosystem II particles of spinach chloroplasts. *Plant Cell Physiol.*, 23: 533–539.

Lavergne, J. (1982) Two types of primary acceptor in chloroplasts photosystems II: I. Different recombination properties. *Photobiochem. Photobiophys.* 3: 257–271.

Mancino, L. J., Dean, D. P. and Blankenship, R. E. (1984) Kinetics and thermodynamics of the $P870^+Q_A^-$–$P870^+Q_B^-$ reaction in isolated reaction centers from the photosynthetic bacterium *Rhodopseudomonas spheroides. Biochim. Biophys. Acta* 764: 46–54.

Melis, A. (1985) Functional properties of PS IIb in spinach chloroplasts. *Biochim. Biophys. Acta* 808: 335–342.

Robinson, H. H. and Croft, A. R. (1983) Kinetics of the oxidation-reduction reactions of the photosystem II quinone acceptor complex, and the pathway of deactivation. *FEBS Lett.* 53: 221–226.

Satoh, K. and Katoh, S. (1985) Inhibition by ethylenediamine tetraacetate and restoration by Mn^{2+} and Ca^{2+} of oxygen-evolving activity in Photosystem II preparation from the thermophilic cyanobacterium, *Synechococcus* sp. *Biochim. Biophys. Acta* 806: 211–229.

Satoh, K., Ohno, T. and Katoh, S. (1985) An oxygen-evolving complex with a simple subunit structure—"a water-plastoquinone oxidoreductase"—from the thermophilic cyanobacterium *Synechococcus* sp. *FEBS Lett.* 180: 326–330.

Schatz, G. H. and van Gorkom, H. J. (1985) Absorbance difference spectra upon charge transfer to secondary donors and acceptors in photosystem II. *Biochim. Biophys. Acta* 810: 283–294.

Schatz, G. H. and Witt, H. T. (1984) Extraction and characterization of oxygen-evolving photosystem II complexes from a thermophilic cyanobacterium *Synechococcus* sp. *Photobiochem. Photobiophys.* 7: 1–14.

Stewart, A. C. and Bendall, D. S. (1979) Preparation of an active oxygen-evolving photosystem 2 particle from a blue-green alga. *FEBS Lett.* 107: 308–312.

Stiehl, H. H. and Witt, H. T. (1969) Quantitative treatment of the function of plastoquinone in photosynthesis. *Z. Naturforshung.* 24b: 1588–1598.

Takahashi, Y. and Katoh, S. (1986) Numbers and functions of plastoquinone molecules associated with photosystem II preparation from *Synechococcus* sp. *Biochim. Biophys. Acta* 845: 183–192.

Velthuys, B. R. (1981) Electron-dependent competition between plastoquinone and inhibitors for binding to photosystem II. *FEBS Lett.* 126: 277–281.

Vermas, W. F. J. and Govindjee (1981) The acceptor side of photosystem II in photosynthesis. *Photochem. Photobiol.* 34: 775–793.

Wydrzynski, T. and Inoue, Y. (1987) Modified photosystem II acceptor side properties upon replacement of the quinone at the Q_B site with 2,5-dimethyl-*p*-benzoquinone and phenyl-*p*-benzoquinone. *Biochim. Biophys. Acta* 893: 33–42.

Yamagishi, A. and Katoh, S. (1984) A photoactive protosystem II reaction center complex lacking a chlorophyll-binding 40-kilodalton subunit from the thermophilic cyanobacterium *Synechococcus* sp. *Biopchim. Biophys. Acta* 765: 118–124.

Yamamoto, Y., Ueda, T., Shinkai, H. and Nishimura, M. (1982) Preparation of Q_2-evolving photosystem-II subchloroplasts

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from spinach. *Biochim. Biophys. Acta* 79: 347–350.

Yamaoka, T., Satoh, K. and Katoh, S. (1978) Photosynthetic activities of a thermophilic blue-green alga. *Plant Cell Physiol.* 19: 943–945.

Zimmerman, J.-L. and Rutherford, A. W. (1986) Photoreductant-induced oxidation of Fe^{2+} in the electron acceptor complex of Photosystem II. *Biochim. Biophys. Acta* 851: 138–148.

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