ELSEVIER

Biochimica et Biophysica Acta 1553 (2002) 302–308

BIOCHIMICA ET BIOPHYSICA ACTA
BBA
www.bba-direct.com

The rate of charge recombination in Photosystem II

Rik de Wijn, Hans J. van Gorkom *

Department of Biophysics, Huygens Laboratory, Leiden University, P.O. Box 9504, 2300 RA Leiden, The Netherlands

Received 25 October 2001; received in revised form 7 January 2002; accepted 9 January 2002

Abstract

Loss by recombination of the charge separated state $P_{680}^{+}Q_{A}^{-}$ limits the performance of Photosystem II (PS II) as a photochemical energy converter. Time constants reported in literature for this process are mostly either near 0.17 ms or near 1.4 ms. The shorter time is found in plant PS II when reduction of P_{680}^{+} by the secondary electron donor Tyrosine Z cannot occur because Y_Z is already oxidized. The 1.4 ms recombination is seen in Y_Z-less mutants of the cyanobacterium *Synechocystis*. However, the rate of $P_{680}^{+}Q_{A}^{-}$ recombination that actually competes with the stabilization of the charge separation has not been previously reported. We have measured the kinetics of the flash-induced fluorescence yield changes in the microsecond time domain in Tris-washed spinach chloroplasts. In this way the kinetics and yield of P_{680}^{+} reduction by Y_Z were obtained, and the rate of the competing $P_{680}^{+}Q_{A}^{-}$ recombination could be evaluated. The recombination time was less than 0.5 ms; the best-fitting time constant was 0.1 ms. The presence of Y_Z^{ox} slightly decreased the efficiency of excitation trapping but did not seem to accelerate $P_{680}^{+}Q_{A}^{-}$ recombination. The two $P_{680}^{+}Q_{A}^{-}$ lifetimes in the literature probably reflect a significant difference between plant and cyanobacterial PS II.

Keywords: Photosystem II; Charge recombination; P_{680}; Tyrosine Z; Fluorescence yield

1. Introduction

Any photosynthetic system is engineered to efficiently convert the energy of optical excitations into a chemical form that is stable enough to allow its use in biochemical processes. Photosystem II (PS II), reviewed in [1], oxidizes water to oxygen and reduces plastoquinone to plastoquinol, simultaneously serving as the ultimate electron source for biosynthesis of organic matter and contributing to the pH gradient over the photosynthetic membrane for ATP synthesis. The electron transfer reactions take place in the reaction center (RC), where excitations in the pigment antenna are trapped by electron transfer from the singlet excited state of the primary donor chlorophyll, P_{680}*, via a pheophytin molecule, Pheo, to a permanently bound plastoquinone molecule Q_A. The lifetime of the resulting charge-separated state $P_{680}^{+}Q_{A}^{-}$, which ultimately limits the lifetime of any

---

Abbreviations: DCMU, 3-(3′,4′-dichlorophenyl)-1,1-dimethylurea; HEPES, 4-(2-hydroxyethyl)-1-piperazineethanesulfonic acid; MES, 2-(*N*-morpholino)ethanesulfonic acid; P_{680}, primary electron donor; Pheo, pheophytin, primary electron acceptor; PS II, Photosystem II; Q_A, first quinone acceptor; Q_B, second quinone acceptor; RC, reaction center; TPB, tetraphenylboron; Tricine, *N*-tris(hydroxymethyl)methylglycine; Tris, tris(hydroxymethyl)aminomethane; Y_Z, tyrosine-Z (Tyr-D1–161), secondary electron donor

* Corresponding author. Fax: +31-71-527-5819.

E-mail address: vangorkom@biophys.leidenuniv.nl (H.J. van Gorkom).

0005-2728/02/\$ – see front matter
PII: S0005-2728(02)00183-4

product generated by PS II [2,3], is the subject of this study.

Normally the loss of the charge separation by recombination of $P_{680}^{+}Q_{A}^{-}$ is prevented because P_{680}^{+} oxidizes Y_Z, tyrosine D1–161, with polyphasic kinetics in the 10^{-8} to 10^{-4} s time range [4]. As a result, the charge recombination time is well established only in conditions where this reaction is blocked, e.g., by destruction of the oxygen evolving complex by Tris-treatment and pre-illumination to bring Y_Z in the oxidized state. A time constant for $P_{680}^{+}Q_{A}^{-}$ recombination of 0.17 ms is then observed [5–7]. In Y_Z-less mutants of a cyanobacterium the recombination time appears to be slower by almost an order of magnitude [8–10], suggesting that either the destructive sample treatment or the presence of the oxidized form of Y_Z in the earlier measurements may have accelerated the recombination of $P_{680}^{+}Q_{A}^{-}$. We found no evidence for that when comparing the decreased stabilization yield and the slower P_{680}^{+} reduction by Y_Z in D_2O, but this concerned only a 10% effect due to a minor slow phase in P_{680}^{+} reduction in only one of the oxidation states of the oxygen-evolving complex [11]. The alternative possibility, that the recombination is not much dependent on Y_Z^{ox} but intrinsically slower in cyanobacteria than in plants, is also suggested by the 1.2 ms time constant reported for Tris-washed PS II preparations from the cyanobacteria *Phormidium laminosum* [7] and *Synechococcus elongatus* [12].

Here we compare the yield and kinetics of P_{680}^{+} reduction by electron transfer from Y_Z in Tris-treated chloroplasts. In this case all P_{680}^{+} is reduced in the microsecond time range and the stabilization yield is decreased substantially, allowing quantification of the competing charge recombination process.

2. Materials and methods

Thylakoids ('broken chloroplasts') were isolated from laboratory-grown spinach leaves essentially according to [13] in a buffer containing 0.4 M sucrose, 15 mM NaCl, 5 mM $MgCl_2$ and 25 mM Tricine (pH 7.8). Manganese depletion by Tris-washing was performed by mixing the thylakoid suspension 1:1 with a solution containing 1.6 M Tris (pH 8.3) and 0.4 M sucrose and incubating for 30 min at 4°C under dim light. The Tris-treated thylakoids were collected by centrifugation, resuspended in the isolation buffer and stored until use in small portions at 77 K. Shortly before measurement, the thylakoids were diluted to a chlorophyll *a+b* concentration of 15 μg/ml in a buffer similar to the isolation buffer but containing MES/NaOH (pH 6.5, 5.5) or HEPES/HCl (pH 7.8) instead of Tricine. For measurements in the presence of TPB and DCMU, the thylakoids were dark-adapted for 15 min to minimize the presence of Q_A^{-} before addition of 25 μM TPB (Sigma) and 5 μM DCMU (Lancaster). Before measurement the thylakoids were dark-adapted for 15 min.

Chlorophyll fluorescence yield changes induced by saturating 10 ns, 532-nm flashes were measured using 2 MHz measuring light modulation and lock-in detection, on the instrument described in [14]. The instrument response time was 2 μs or 10 μs for fast or slow measurement conditions, respectively. For each measured flash series a fresh dark-adapted sample was taken. Measurements where averaged 3–4 times. Least squares fitting of the result of model calculations to the data was performed using the routine e04jaf of the NAG toolbox for Matlab (MathWorks, Natick, MA).

3. Results

Fig. 1 shows chlorophyll fluorescence yield changes induced by a series of saturating flashes, fired at 10 Hz, in Tris-treated thylakoids at pH 6.5. In these conditions P_{680}^{+} reduction by Y_Z takes place almost entirely in the microsecond time domain [5,9,15]. Since quenching by P_{680}^{+} prevents the fluorescence yield increase associated with Q_A reduction, its reduction is reflected in a rise of the fluorescence yield after the first flash (shown enlarged in Fig. 1A). In Tris-washed PS II, Q_A^{-} can still reduce a plastoquinone bound at the Q_B site, decreasing the fluorescence yield again, but Y_Z^{ox} reduction is blocked, so the final product of the first flash is the state $Y_Z^{ox}Q_B^{-}$. The lifetime of this state is longer than the 100 ms flash spacing used in these experiments. The second and subsequent charge separations can only generate the charge pair $P_{680}^{+}Q_{A}^{-}$, which in these conditions is well known to recombine with a time constant of 150–200 μs [5,12], so the fluorescence yield remains

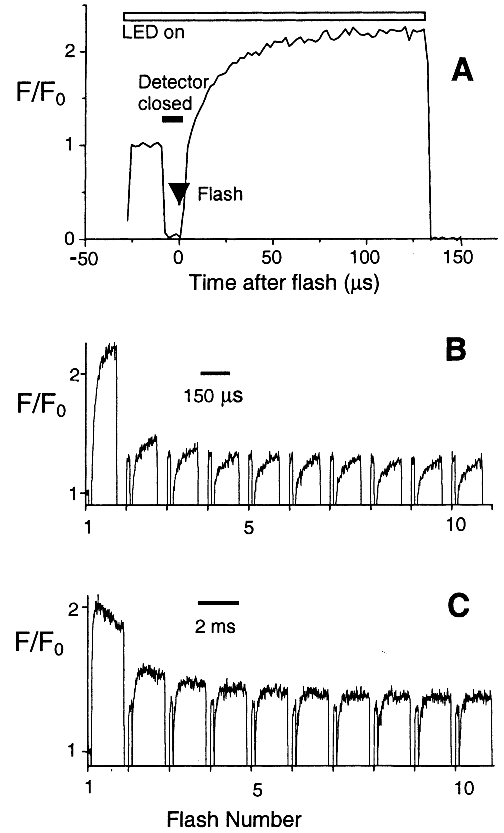

Fig. 1. Chlorophyll fluorescence yield changes induced in Tris-washed chloroplasts (pH 6.5) by a train of 10 saturating flashes fired at 10 Hz (B). The timing of measuring light, detector gate, and actinic flash is indicated in A. In C, a 12-times longer measurement time per flash was used, as indicated.

low. In fact these flashes appear to cause a small additional quenching that disappears in such a time, much slower than that by P^+_{680} on the first flash (Fig. 1B). The fluorescence yield decreases from 1.3 times F_0 (the dark-adapted level) just before the flash to 1.0 times F_0 just after. This decrease upon formation of $P^+_{680}Q^-_A$ indicates that the increased fluorescence yield remaining at 100 ms after a flash was not due to the presence of some $P_{680}Q^-_A$, e.g., in equilibrium with $P_{680}\ Q_AQ^-_B$, but to a less efficient excitation trapping by $Y^{ox}_ZP_{680}Q_A$ than by $Y^{ox}_ZP^+_{680}Q^-_A$ or $Y_ZP_{680}Q_A$. If Y^{ox}_Z can decrease the quantum yield of charge separation, it might well accelerate the charge recombination.

Fig. 2 shows the same measurements after addition of DCMU, which prevents plastoquinone binding at the Q_B site, and TPB, an efficient electron donor to Y^{ox}_Z. In this case a charge separation will produce the non-quenching state $Y_ZP_{680}Q^-_A$ almost irreversibly. This is clearly illustrated by the fluorescence yield increase induced by the measuring light alone (Fig. 3). These curves are reminiscent of classical fluorescence induction by continuous light, but note that the illumination here is by 0.15-ms pulses spaced at 100 ms. It took several saturating flashes to close all PS II centers in the non-quenching state and the fluorescence yield reached after the first flash remained far below the maximum. The actual 'miss probability' is smaller than the fluorescence yield suggests, due to the hyperbolic relationship between fluorescence yield and concentration of quenching centers (see below). Nevertheless, a considerable fraction of $P^+_{680}Q^-_A$ must have been lost by recombination before

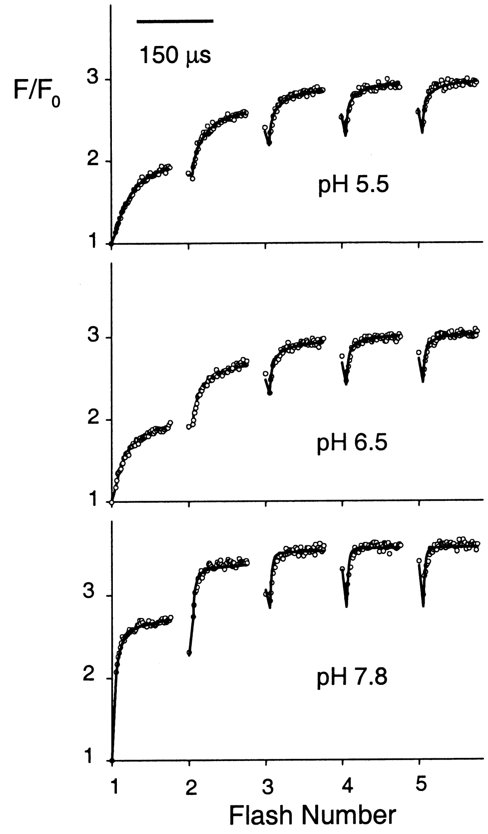

Fig. 2. Kinetics of the flash-induced fluorescence yield change in the presence of 10 μM DCMU and 25 μM TPB after the first 5 flashes (fired at 10 Hz) of a series at pH 5.5, 6.5 and 7.8 (as indicated). The first point of each trace indicates the level just before the flash, the next point is taken 6 μs after the flash (after that a regular spacing of 2 μs). The line represents a typical fit to the data (see text).

stabilization of the charge separation by Y_Z oxidation had occurred.

The same measurements also reflect the kinetics of this process. The reduction of P_{680}^{+} by Y_Z is known to be faster at high than at low pH in Tris-washed PS II, whereas the rate of $P_{680}^{+}Q_A^{-}$ recombination is pH independent in this range [5,8,9]. Fig. 2 confirms that faster P_{680}^{+} reduction kinetics led to a lower miss probability, so the rate $P_{680}^{+}Q_A^{-}$ recombination competing with P_{680}^{+} reduction by Y_Z may be assessed by analysis of these data. A complicating factor, however, is that the rise of the fluorescence is not monophasic. At pH 5.5 and 6.5, (at least) two phases occur in the microseconds time range. At pH 7.8, an unresolved fast phase is seen in addition, consistent with the appearance of a 1-μs phase in P_{680}^{+} reduction at pH > 7 reported by Ahlbrink et al. [15]. The data in Fig. 2 also show that conversion to the non-quenching state $Y_ZP_{680}Q_A^{-}$ was not always irreversible. At pH 5.5 and 6.5, there is some decay of the fluorescence yield between flashes, indicating a slow leak to an electron acceptor, in spite of the DCMU block. Also at pH 7.8 a fluorescence decrease between flashes occurs, but in this case mainly after the first flash and to a lesser extent after later flashes. Apparently Y_Z^{ox} reduction by TPB is much slower at pH 7.8 than at low pH and could not fully prevent $Y_Z^{ox}P_{680}Q_A^{-}$ recombination. These observations yield the reaction scheme in Fig. 4.

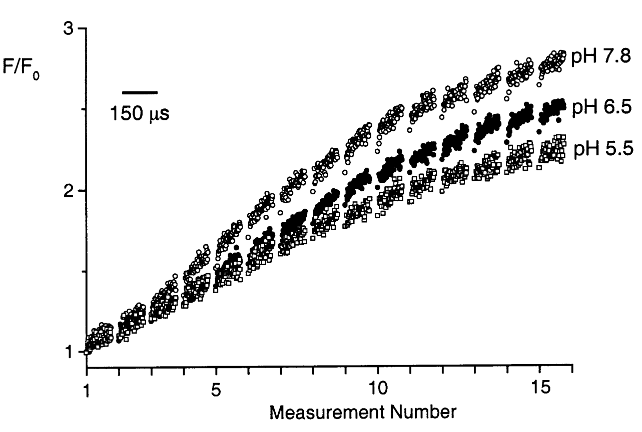

Fig. 3. Fluorescence induction by the measuring light at pH 5.5, 6.5, and 7.8 (as indicated). The experiments of Fig. 2 were repeated without firing the saturating laser flash.

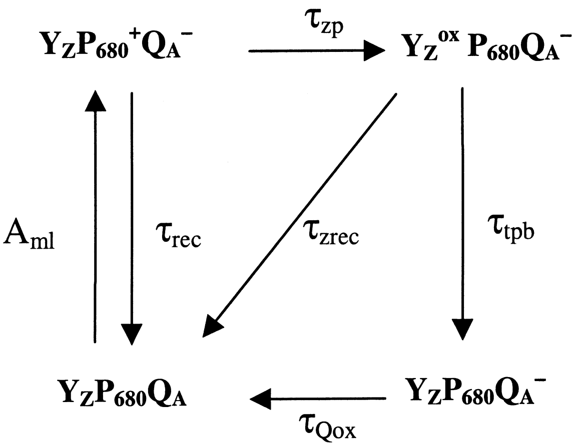

Fig. 4. Kinetic scheme used to describe the data in Fig. 2. The τs indicate time constants for the separate processes. τ_{zp}, P_{680}^{+} reduction by Y_Z; τ_{rec}, $P_{680}^{+}Q_A^{-}$ recombination; τ_{tpb}, quasi-first-order time constant for Y_Z^{ox} reduction by TPB; τ_{zrec}, $Y_Z^{ox}Q_A^{-}$ recombination; τ_{Qox}, oxidation of Q_A^{-} (in the presence of DCMU). A_{ml} denotes a proportionality constant for the excitation rate induced by the measuring light (see text).

3.1. Description of the analysis

The fluorescence kinetics was calculated by simulating the population dynamics within the scheme shown in Fig. 4. The kinetics of the system is described by a set of differential equations for the population of the states X_n:

$$\frac{dX_n}{dt} = -X_n\sum_m k_{nm} + \sum_m k_{mn}X_m \tag{1}$$

where k_{nm} is the (pseudo) first-order rate constant for the reaction $X_n \rightarrow X_m$. A special case is the charge separation induced by the actinic effect of the measuring light (k_{ml}), which may occur after charge recombination has taken place. Its rate will be affected by the competing quenching by $Y_ZP_{680}^{+}Q_A^{-}$. For the rate constant k_{ml} in Eq. 1 we use [16]:

$$k_{ml} = \frac{A_{ml}\kappa}{1+\kappa X_q} \tag{2}$$

A_{ml} is a constant proportional to the measuring light intensity, $\kappa = F_M/F_0 - 1$, and $X_q = Y_ZP_{680}Q_A + Y_ZP_{680}^{+}Q_A^{-}$. A_{ml} was kept fixed at a value estimated on the basis of the rapid phase of fluorescence induction by the measuring light (Fig. 3). After fitting the flash-induced kinetics of Fig. 2, it was checked that the parameter values obtained also produced a reasonable description of Fig. 3.

The time course of the populations X_n was calculated by integrating the system of Eq. 1 (with Eq. 2 inserted) using a suitable integration routine internal to MATLAB starting from initial conditions X_n $(t=0)$. For the first flash the initial condition was $Y_ZP_{680}^{+}Q_A^{-}=1$ (or $Y_ZP_{680}Q_A=1$ for the measurements without flash excitation). The system was integrated during the measuring period with A_{ml} given by Eq. 2 and after the measuring period with $A_{ml}=0$ (measuring light out). The initial conditions for the second flash were given by the outcome of the integration at $t=100$ ms after the first flash (see below) and conversion of all $P_{680}Q_A$ into $P_{680}^{+}Q_A^{-}$. In this way the simulation progressed until the desired number of flashes was reached.

For practical reasons the integration of the system was terminated at $t=4*\tau_{tpb}$ when all reactions except the slow Q_A^- oxidation between flashes were finished. The initial conditions for the next flash were then calculated by taking the resulting $X_n(t=4*\tau_{tpb})$ and assuming some probability for the reaction $Y_ZP_{680}Q_A^{-}\rightarrow Y_ZP_{680}Q_A$ to occur between this time and the next flash. For $Y_Z^{ox}P_{680}Q_A^{-}$ recombination the rates measured by Diner et al. [9] were used.

Finally, the quenching by carotenoid triplet states [17], which causes the 'undershoot' of the fluorescence yield below the level observed just before the flash, was accounted for. The amount of carotenoid triplet (^{3}Car) formed upon flash number N was assumed to be proportional to the fluorescence level just before this flash ($F(-0,N)$) and its decay time constant τ_q was kept fixed at 3 μs [18]:

$$^3\mathrm{Car}(t,N)=A_qF(-0,N)e^{-t/\tau_q} \tag{3}$$

The proportionality constant A_q was used as a fit parameter and found to be 0.3–0.5 population units/F_M.

The calculated population dynamics was translated into fluorescence yield kinetics as follows. The states $Y_ZP_{680}Q_A$ and $Y_ZP_{680}^{+}Q_A^{-}$ are assumed to be equally strong fluorescence quenchers. The fluorescence yield can then be related to the fraction of PS II RCs in these quenching states or, conversely, to the fraction of RCs in the non-quenching states $Y_Z^{ox}P_{680}Q_A^{-}$ and $Y_ZP_{680}Q_A^{-}$. This was done by taking into account the non-linear relationship between fluorescence yield F and fraction of non-quenching RCs, C, according to Paillotin [19]:

$$\frac{F}{F_M}=1-\frac{1-C}{\frac{1}{\varepsilon}-pC} \tag{4}$$

The trapping efficiency $\varepsilon=(F_M-F_0)/F_M$. F_0, the fluorescence yield corresponding to all RCs in the quenching states, was assumed to be the fluorescence yield after dark adaptation and normalized to 1. F_M, the fluorescence yield corresponding to all RCs in non-quenching states, is normally close to 5 in oxygen-evolving spinach thylakoids. The maximum fluorescence yield reached in Fig. 2 is lower and pH dependent. Similar effects of Tris-washing have been observed before [20]. We have used a F_M value of 3.7 to accommodate the maximum reached at pH 7.8. The lower levels reached at pH 6.5 and 5.5 were assumed to result from an increased fraction of RCs that could not produce a stable charge separation. This fraction was found to be 5–10% for both pH 6.5 and 5.5.

The connectivity parameter p is the probability of excitation transfer from a PS II with a non-quenching RC to another PS II, as originally defined by [21] and corresponds to ω in [19]. It has been shown previously that in Tris-washed PS II this probability was unity [16], indicating a situation where the Stern–Volmer relation for fluorescence quenching applies. We used $p=1$ in our calculations. Using a lower value results in a larger fraction of RCs that is not observed in the fluorescence after the first flash, i.e., in a faster recombination.

3.2. Fit results

The fluorescence kinetics that was calculated as outlined above was fitted to the data in Fig. 2. Expectedly, if only a single time constant for P_{680}^{+} reduction was allowed (in addition to the unresolved phase at pH 7.8), the slower fluorescence rise in the microseconds time domain had to be due to the actinic effect of the measuring light. All of the miss probability on the first flash must then be caused by charge recombination during the more rapid phase and very short, pH dependent recombination times are required. It was found that a consistent fit could indeed be obtained this way for all three pH

Table 1
Best-fitting time constants (in μs) of $Y_ZP_{680}^+Q_A^-$ recombination (τ_{rec}) and P_{680}^+ reduction by Y_Z (τ_{zp}) for the data of Fig. 2 and reaction scheme of Fig. 4

pH	Slow rise[a] due to measuring light		Slow rise due to slow phase in P_{680}^+ reduction by Y_Z	
	τ_{rec}	τ_{zp}	τ_{rec}	τ_{zp}
5.5	68	33	91	28 (0.81)[b] 150 (0.19)
6.5	51	25	118	16 (0.60) 102 (0.40)
7.8	26 26 (0.36)	1 f[c] (0.64)	100 f	1 f (0.64) 14 (0.15) 767 (0.11)

[a] The two model options, and the other parameters used in the fit, are described in the text.

[b] For the components of P_{680}^+ reduction by Y_Z, their relative weights are given in parentheses.

[c] f denotes a parameter that was fixed in the fit to the value shown.

values (Table 1, left column), but the result seems highly unlikely. It is well established that the rate of P_{680}^+ reduction by Y_Z in Tris-washed PS II depends on pH and that of $P_{680}^+Q_A^-$ recombination does not [5,7]. The fit would imply the opposite.

The model calculation outlined above was therefore expanded to allow biphasic reduction of P_{680}^+ by Y_Z, so that it could account for both phases of the fluorescence rise. Most of the miss probability then results from recombination during the slow phase. Since the relative weight of this phase was not increased on the second and later flashes, the biphasic kinetics cannot be due to a heterogeneity that is static on the time scale of measurement (100 ms). The best-fitting parameter values in this case are listed in the right column of Table 1. The time constant of $P_{680}^+Q_A^-$ recombination is found to be close to 100 μs at pH 5.5 and 6.5, and this value also produced a good fit at pH 7.8 (when it was left free in the fit a shorter time was found at pH 7.8). Due to cross correlation in the fit of τ_{rec} with the time constant and relative weight of the slow component of P_{680}^+ reduction, the value of τ_{rec} is not very precisely determined. However, values significantly larger than the 0.2 ms observed in the presence of Y_Z^{ox} were unlikely and values larger than 0.5 ms did not allow a fit with the model used.

4. Discussion

The results show that the presence of Y_Z^{ox} may slightly decrease the efficiency of $P_{680}^+Q_A^-$ formation, but does not significantly accelerate its recombination. The much slower $P_{680}^+Q_A^-$ recombination in cyanobacterial mutants lacking Y_Z [8–10] is probably not due to the absence of Y_Z^{ox} during its measurement. This also appears from the measurements on Tris-washed PS II preparations from cyanobacteria, presumably in the presence of Y_Z^{ox} [7,12].

We have no indication that in spinach thylakoids the recombination rate is much affected by destruction of the oxygen evolving complex by Tris-treatment. In oxygen-evolving thylakoids, we have shown earlier [11] that a 0.1 ms $P_{680}^+Q_A^-$ recombination would be required to explain the increased miss probability in D_2O by the slower P_{680}^+ reduction observed. In addition, a much slower $P_{680}^+Q_A^-$ recombination in intact PS II seems unlikely because it would be expected to decrease the efficiency of photosynthesis, as explained below.

The lifetime of $P_{680}^+Q_A^-$ and any product derived from it is ultimately limited not by the lifetime of the excited state [22,23] but by the thermally activated recombination via $P_{680}^+Pheo^-$ to the triplet state of P_{680} [2,3]. The maximum stabilization of the charge separation that can be obtained by the electron transfer equilibrium constant K_{HQ} between $P_{680}^+Pheo^-Q_A$ and $P_{680}^+Pheo\ Q_A^-$ is determined by loss of the charge-separated state via electron tunneling from Q_A^- directly to P_{680}^+. In plant PS II, this reaction occurs in approximately 2.5 ms [24]; it might be slower in cyanobacteria. A further increase of K_{HQ} would not increase the lifetime of $P_{680}^+Q_A^-$ but decrease the free energy available for photosynthesis. To avoid that, K_{HQ} is likely to be so small that the lifetime of $P_{680}^+Q_A^-$ is indeed much shorter than 2.5 ms and therefore not significantly reduced by direct electron tunneling from Q_A^- to P_{680}^+. This is not long enough to drive the reaction with the 'substrates' H_2O and plastoquinone. Further stabilization will have to come from a subsequent electron transfer step (the oxidation of Y_Z), increasing the distance between the charges and thereby decreasing the direct recombination rate.

The rate of activated charge recombination, k_{QHP}, is the rate of electron transfer from Q_A^- back to Pheo

(k_{QH}) times the probability that $P_{680}^{+}Pheo^{-}$ will lost by recombination (k_{HP}) rather than form $P_{680}^{+}Q_A^{-}$ again (k_{HQ}):

$$k_{QHP} = k_{QH}\frac{k_{HP}}{k_{HP}+k_{HQ}} \tag{5}$$

or, in terms of time constants $\tau(=1/k)$ and with the equilibrium constant for electron transfer between Pheo and Q_A, $K_{HQ} = \tau_{QH}/\tau_{HQ}$:

$$\tau_{QHP} = K_{HQ}(\tau_{HP}+\tau_{HQ}) \tag{6}$$

The condition that the direct recombination of $P_{680}^{+}Q_A^{-}$, with time constant τ_{QP}, is to be avoided means $\tau_{QHP} \ll \tau_{QP}$ and hence:

$$K_{HQ} \ll \frac{\tau_{QP}}{\tau_{HP}+\tau_{HQ}} \tag{7}$$

If τ_{QP} is 2.5 ms [24], $\tau_{HQ} = 0.3$ ns [25] and $\tau_{HP} = 0.3$ ns [26], K_{HQ} is probably less than 10^6. This implies a standard free energy difference between $P_{680}^{+}Pheo^{-}Q_A$ and $P_{680}^{+}PheoQ_A^{-}$ of less than 0.36 eV, in agreement with [2]. Perhaps in cyanobacteria a slightly larger value may apply, but values near 0.6 eV, as often assumed in the literature [1], are not compatible with efficient photosynthesis.

Acknowledgements

This research was supported by the Chemical Sciences section of the Netherlands Organization for Scientific Research (NWO-CW).

References

[1] B.A. Diner, G.T. Babcock, in: D.R. Ort, C.F. Yocum (Eds.), Oxygenic Photosynthesis: The Light Reactions, Kluwer Academic Publishers, Dordrecht, 1996, pp 213–247.

[2] H.J. van Gorkom, Photosynth. Res. 6 (1985) 97–112.

[3] H.J. van Gorkom, Bioelectrochem. Bioenerg. 16 (1986) 77–87.

[4] M.J. Schilstra, F. Rappaport, J.H.A. Nugent, C.J. Barnett, D.R. Klug, Biochemistry 37 (1998) 3974–3981.

[5] H. Conjeaud, P. Mathis, Biochim. Biophys. Acta 590 (1980) 353–359.

[6] J. Haveman, P. Mathis, Biochim. Biophys. Acta 440 (1976) 346–355.

[7] S. Reinmann, P. Mathis, H. Conjeaud, A. Stewart, Biochim. Biophys. Acta 635 (1981) 429–433.

[8] C.A. Buser, L.K. Thompson, B.A. Diner, G.W. Brudvig, Biochemistry 29 (1990) 8977–8985.

[9] B.A. Diner, D.A. Force, D.W. Randall, R.D. Britt, Biochemistry 37 (1998) 17931–17943.

[10] J.G. Metz, P.J. Nixon, M. Rögner, G.W. Brudvig, B.A. Diner, Biochemistry 28 (1989) 6960–6969.

[11] R. de Wijn, T. Schrama, H.J. van Gorkom, Biochemistry 40 (2001) 5821–5834.

[12] S. Gerken, J.P. Dekker, E. Schlodder, H.T. Witt, Biochim. Biophys. Acta 977 (1989) 52–61.

[13] H.H. Robinson, C.F. Yocum, Biochim. Biophys. Acta 590 (1980) 97–106.

[14] R. de Wijn, H.J. van Gorkom, Biochemistry 40 (2001) 11912–11922.

[15] R. Ahlbrink, M. Haumann, D. Cherepanov, O. Bögershausen, A. Mulkidjanian, W. Junge, Biochemistry 37 (1998) 1131–1142.

[16] H.J. van Gorkom, M.P.J. Pulles, A.L. Etienne, in: H. Metzner (Ed.), Photosynthetic Oxygen Evolution, Academic Press, London, 1978, pp. 135–145.

[17] L.N.M. Duysens, G.A. den Haan, J.A. van Best, in: M. Avron (Ed.), Proceedings of the Third International Congress on Photosynthesis I, Elsevier, Amsterdam, 1975, pp. 1–12.

[18] P. Mathis, W.L. Butler, K. Satoh, Photochem. Photobiol. 30 (1979) 603–614.

[19] G. Paillotin, J. Theor. Biol. 58 (1976) 219–235.

[20] F. Reifarth, G. Renger, G. Christen, Photosynth. Res. 51 (1997) 231–242.

[21] P. Joliot, A. Joliot, C. R. Acad. Sci. Paris 258 (1964) 4622–4625.

[22] L.N.M. Duysens, in: The Photochemical Apparatus, Its Structure and Function, Brookhaven Symp. Biol. 11 (1958) 10–25.

[23] R.T. Ross, M. Calvin, Biophys. J. 7 (1967) 595–614.

[24] S. Reinmann, P. Mathis, Biochim. Biophys. Acta 635 (1981) 249–258.

[25] A.M. Nuijs, H.J. van Gorkom, J.J. Plijter, L.N.M. Duysens, Biochim. Biophys. Acta 848 (1986) 167–175.

[26] M. Volk, M. Gilbert, G. Rousseau, M. Richter, A. Ogrodnik, M.E. Michel-Beyerle, FEBS Lett. 336 (1993) 357–362.