Biochimica et Biophysica Acta, 357 (1974) 429–438

BBA 46799

THERMOLUMINESCENCE STUDIES ON PHOTOSYNTHETIC ENERGY CONVERSION

II. ACTIVATION ENERGIES FOR THREE ENERGY STORAGE STATES ASSOCIATED WITH PHOTOREACTION II OF HIGHER PLANTS

SUSAN LURIE* and WALTER BERTSCH

Department of Biological Sciences, Hunter College of the City University of New York, 695 Park Avenue, New York, N.Y. 10021 (U.S.A.)

(Received March 13th, 1974)

SUMMARY

Thermoluminescent glow curves of isolated chloroplasts were analyzed to determine activation energy, frequency factor and lifetime of the three glow peaks associated with Photoreaction II. Peak 1 had an activation energy of 0.8 eV, a frequency factor of $1.5 \cdot 10^{14}$/s and a lifetime of 0.02 s. This glow peak appeared to be rather different from peaks 2 and 3, which were quite similar to each other. Peaks 2 and 3 gave activation energies respectively of 0.48 eV and 0.57 eV, frequency factors of $7 \cdot 10^{7}$/s and $1 \cdot 10^{9}$/s, and lifetimes of about 1.5 s. A second, less reliable method of analysis gave activation energies of 0.72 eV for peak 1 and 0.44 eV for peak 2. Our values are compared with those obtained by other workers, and the possible metastable states reflected by the three glow peaks are discussed.

INTRODUCTION

Green plants, algae and photosynthetic bacteria can store some of the energy of absorbed light, even when they are illuminated at temperatures so low that enzymatic processes do not occur. Arnold and others [1–5] have employed the method of thermoluminescence to study this phenomenon. A sample of plant material is illuminated and cooled, then warmed in darkness at a constant rate. The luminescence is recorded as a function of temperature, and peaks in the resulting glow curve represent energy storage by Photoreaction II [6].

The energy of the first excited singlet of chlorophyll is about 1.8 eV (from the peak wavelength of emission spectra for fluorescence and delayed light, in vivo [7]). On the other hand, the redox poise of early steps in the photosynthetic electron transport chain at Photoreaction II is estimated to conserve only about 0.82 eV [8, 9].

* Present address: Faculty of Agriculture, The Hebrew University of Jersusalem, Rehovot, P.O. Box 12, Israel, inquiries should be sent to Walter Bertsch.

Therefore, up to 0.98 eV is available for stabilization of the metastable rates involved in charge separation by photosynthetic quantum conversion. This enormous stabilization energy is reflected in the low yield of delayed light, a back reaction in which chlorophyll of Photoreaction II is re-excited by early photoproducts.

Energy stored as metastable photoproducts eventually is stabilized as oxidation and reduction potential of carriers in the photosynthetic electron transport chain. By utilizing the method of thermoluminescence, metastable states can be "frozen in". In our previous paper [6] we reported an emission profile from chloroplasts consisting of three peaks. This suggests that three types of energy storage are involved in stabilization of early metastable photoproducts at Photoreaction II. In the present paper, we calculate the activation energies for re-excitation of the chlorophyll singlet by these three energy storage states. The activation energies for these three back reactions presumably reflect stabilization energies for three steps in photosynthetic quantum conversion at Photoreaction II of higher plants.

THEORY

Activation energy and frequency factor

It was shown by Randall and Wilkins [10] that the equation for thermoluminescence from inorganic crystals had the form:

$$I = -C(\mathrm{d}n/\mathrm{d}t) = Cn_0 s e^{-\int (s/B) e^{-E/kT} \mathrm{d}T} e^{-E/kT} \tag{1}$$

This expression is based on the decay process

$$I = -C(\mathrm{d}n/\mathrm{d}t) = Cnse^{-E/kT} \tag{2}$$

where I is the instantaneous emission intensity, n is the number of electrons in traps of depth E at time t, s is a frequency factor, B is the rate of warming, C is a geometry constant and T is absolute temperature. These equations have two unknowns of interest, E and s.

Grossweiner [11] suggested a method by which E and s could be found separately, and thus be determined experimentally. The method involves three steps. 1. Taking a ratio of the intensity at any point to the maximum intensity which is found at the peak of the glow curve, using Eqn (1). 2. Holding the heating rate, B, constant so that (s/B) can be factored out of the integral. The resulting integral can be reduced by integrating by parts or substitution. 3. Integrating the remaining integral, which is of the form

$$\int \frac{e^x}{x} \mathrm{d}x$$

to a series

$$-e^{-u}(1/u - 1/u^2 + 2/u^3 - 3/u^4 + \ldots)$$

where $u = -x$. For values of $E/kT > 20$ (e.g. $E > 0.42$ eV), less than 10 % error is made by dropping all terms after the second in the expansion.

Then, having performed the three steps outlined above, it is found that

$$y+\ln\frac{I}{I_{max}}=1-\left(\frac{T}{T_{max}}\right)^2 e^{-y} \tag{3}$$

where $y=(E/k)\,(T_{max}-T)/TT_{max}$, I_{max} is the maximum emission intensity (at the glow peak), and T_{max} is the temperature at which the intensity is maximum. For temperatures that are close together, $(T/T_{max})^2$ is approximately 1, so Eqn (3) becomes with rearrangement:

$$y+e^{-y}=1-\ln f \tag{4}$$

where f is the intensity ratio: I/I_{max}. From the definition of y, we may also write:

$$E=ykTT_{max}/(T_{max}-T) \tag{5}$$

Once y has been determined from Eqn (4), it is possible to determine E by substituting the appropriate T and T_{max} into Eqn (5). Although Eqn (4) does not lend itself to analytic solution, it can be solved numerically by interpolation to any desired degree of accuracy. The right side of Eqn (4) is a constant for any f (e.g. for any I in a given glow curve). The left side then has two values of y satisfying the equation for a given f: a positive y and a negative y. The physical condition that the activation energy be positive imposes the mathematical condition that y be positive when the temperature difference is positive (rising side of glow curve) and that y be negative when the temperature difference is negative (falling side of glow curve). Thus, on the rising side of any simple first order glow curve, at the point of half maximum intensity, y equals approximately 1.461, and

$$E=\frac{1.461\,kT_{\frac{1}{2}}T_{max}}{T_{max}-T_{\frac{1}{2}}} \tag{6}$$

where $T_{\frac{1}{2}}$ is the temperature at which the half maximum intensity occurs on the rising side of the glow curve. E may thus be determined experimentally using Eqn (6)*.

An expression for s as a function of E can be found by differentiation of Eqn (2) with respect to T, so that at the glow peak maximum

$$E/k=(s/B)\,e^{-E/kT_{max}}T^2_{max} \tag{7}$$

and

$$s=\frac{BEe^{E/kT_{max}}}{kT^2_{max}} \tag{8}$$

Having found E from Eqn (6), s may be determined experimentally from Eqn (8).

Rising side of peak 3

From Fig. 1 it is clear that Eqn (6) can not be used for calculating the E of the shoulder which represents peak 3 of our chloroplast glow curves. The rising side of peak 3 is completely obliterated by the falling side of peak 2, so that neither $I_{\frac{1}{2}}$ nor $T_{\frac{1}{2}}$

* The numerical coefficient of our Eqn (6) differs from Grossweiner's value of 1.51, in his Eqn (10). We attribute this to an arithmetical error by Grossweiner.

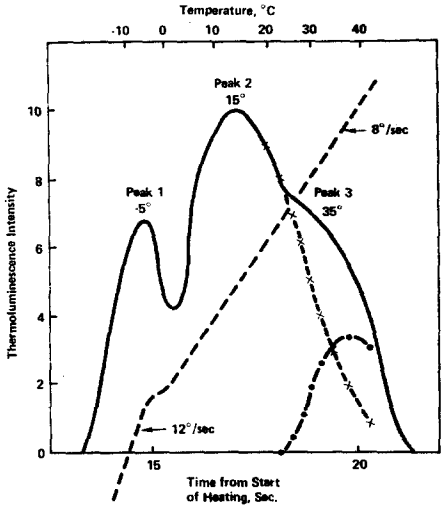

Fig. 1. Thermoluminescence of isolated chloroplasts showing the calculated falling side of peak 2 and determination of peak 3 by subtraction. The calculation is given in Table I. The dotted line is the heating curve of the sample. Reaction mix: 0.025 M Tricine (pH 7.8), 0.02 M NaCl, 0.005 M $MgCl_2$, 65 μg Chl/ml.

TABLE I

SAMPLE CALCULATION OF FALLING SIDE OF PEAK 2

The data are those of Fig. 1, and the results are plotted in Fig. 1. The constant y was calculated numerically from Eqn (4) as a function of f, the ratio of emission intensities. These y values hold for the falling side of any first order glow curve. The T values were calculated from Eqn (9) for a peak 2 activation energy of 0.456 eV and a peak 2 T_{max} of 297 °K.

$f = I/I_{max}$	y	T
0.9	−0.426	304
0.8	−0.601	307
0.7	−0.741	310
0.6	−0.866	312
0.5	−0.985	314
0.4	−1.105	317
0.3	−1.235	319
0.2	−1.385	322
0.1	−1.587	326

can be directly measured for peak 3. However, if intensities on the falling side of peak 2 could be calculated, then the rising side of peak 3 could be obtained by subtraction of the calculated peak 2 intensities from the total emission. Having estimated E for peak 2, this can be done by using Eqns (4) and (5) to calculate back the T at which a particular ratio of I/I_{max} would occur. Table I gives numerically estimated y values for a series of intensities ratios, f, from 1/10–9/10 of peak emission on the falling side of any simple first order glow curve, using Eqn (4). The temperature at which each intensity will occur is found by re-arrangement of Eqn (5):

$$T = \frac{ET_{max}}{E - kyT_{max}} \tag{9}$$

and utilization of the appropriate y value. Eqn (9) is then used to determine the shape of the falling side of peak 2, and this is subtracted from the total measured glow curve to give the rising side and position of maximum emission for peak 3. Table I gives calculated T values for each intensity ratio of the peak 2 falling side of Fig. 1, using this method. We were thus able to estimate T_{max} and $T_{\frac{1}{2}}$ for peak 3, and using Eqs (6) and (8) we made estimates for E and s of peak 3.

Activation energy from Arrhenius equation

Activation energies of glow curves can be estimated by making Arrhenius plots of the emission intensities for the initial portion of a glow peak. This assumes that the energy lost in the initial portion is small compared to the total stored energy of that state. Since emission intensity (photons/s) is proportional to the rate of the back reaction which re-excites chlorophyll, we may write:

$$I \alpha dn/dt = nse^{-E/kT}$$

then,

$$\ln I = -E/kT + \ln Cns \tag{10}$$

By plotting $\log_{10} I$ on the ordinate and $1/T$ on the abscissa, the slope of the exponential rise in thermoluminescence is $E/2.3k$.

We have used this method to check our activation energy determinations by the Grossweiner method. We consider the Arrhenius method to be more inaccurate than the Grossweiner method because only the initial part of the glow can be used. This is the portion with greatest experimental error. Fig. 2 gives our results for peak 1 of three different glow curves. No additions were made to the chloroplasts for these determinations. Fig. 2 also shows our results for peak 2, which was measured in the

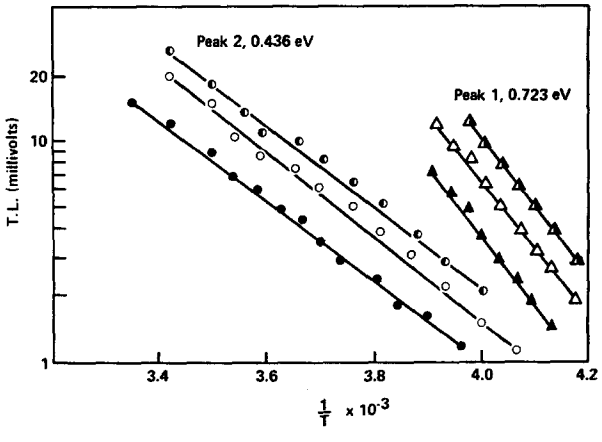

Fig. 2. Arrhenius plot of the initial rise of peak 1 and peak 2. Reaction mix as in Fig. 1 except the top curve for peak 1 was observed in the presence of $5 \cdot 10^{-4}$ M $K_3Fe(CN)_6$, and all the curves for peak 2 were observed in the presence of $2 \cdot 10^{-6}$ M DCMU.

TABLE II

CALCULATION OF THE ACTIVATION ENERGY, FREQUENCY FACTOR AND HALF-LIFE OF ENERGY STORAGE STATES 1, 2 AND 3

Calculations made from Eqns [6], [8] and [11]

Conditions	T* (° C)	E (eV)	s (s^{-1})	r (s)
storage state 1				
no additions	−8	0.878	$5.2 \cdot 10^{14}$	0.013
	−6	0.823	$2.0 \cdot 10^{14}$	0.020
	−5	0.760	$2.2 \cdot 10^{13}$	0.028
	−4	0.748	$5.0 \cdot 10^{13}$	0.021
	−6	0.810	$1.1 \cdot 10^{14}$	0.015
ferricyanide	−12	0.850	$2.9 \cdot 10^{14}$	0.010
	−8	0.726	$5.6 \cdot 10^{13}$	0.031
	−6	0.815	$2.3 \cdot 10^{13}$	0.025
	−5	0.778	$5.2 \cdot 10^{13}$	0.019
	−8	0.810	$2.0 \cdot 10^{14}$	0.027
Average		0.799	$1.5 \cdot 10^{14}$	0.020
storage state 2				
no additions	30	0.452	$1.8 \cdot 10^{7}$	2.045
	36	0.453	$1.3 \cdot 10^{7}$	2.946
	38	0.531	$1.8 \cdot 10^{8}$	1.050
	22	0.476	$6.1 \cdot 10^{7}$	1.552
	22	0.482	$8.6 \cdot 10^{7}$	1.432
ferricyanide	38	0.443	$5.7 \cdot 10^{7}$	2.533
	32	0.492	$1.0 \cdot 10^{8}$	1.700
DCMU	28	0.451	$1.5 \cdot 10^{7}$	2.360
	22	0.476	$6.1 \cdot 10^{7}$	1.552
	25	0.510	$1.1 \cdot 10^{8}$	1.453
Average		0.476	$7.0 \cdot 10^{7}$	1.851
storage state 3				
no additions	42	0.604	$2.6 \cdot 10^{9}$	1.231
	46	0.655	$4.8 \cdot 10^{8}$	2.303
	38	0.557	$3.2 \cdot 10^{8}$	2.150
	30	0.505	$1.1 \cdot 10^{9}$	1.621
	40	0.525	$7.2 \cdot 10^{8}$	1.725
ferricyanide	38	0.558	$5.9 \cdot 10^{8}$	1.294
	40	0.562	$5.2 \cdot 10^{8}$	1.413
DCMU	30	0.580	$1.8 \cdot 10^{9}$	1.451
	35	0.545	$5.2 \cdot 10^{8}$	1.652
	32	0.573	$1.3 \cdot 10^{9}$	1.722
Average		0.567	$9.9 \cdot 10^{8}$	1.655

presence of 3(3,4-dichlorophenyl)-1,1-dimethylurea (DCMU) in order to eliminate peak 1. Similar determinations could not be made for peak 3.

Life-time

Once E and s are determined, the life-time (or half-life), r, of the state can also be determined. Assuming the process to be first order:

$$r = 1/e = \frac{\ln 2}{p} = \frac{e^{E/kT} \ln 2}{s} \tag{11}$$

where p is the probability of emission.

MATERIALS AND METHODS

The apparatus and procedure have been described previously [6]. In brief, a 1 ml sample of spinach chloroplasts (approximately 50 μg chlorophyll) was cooled to −196 °C and illuminated during the cooling. Illumination time was approximately 2 min and incident light intensity was $5 \cdot 10^5$ erg $\cdot$ cm$^{-2} \cdot$ sec^{-1}, white light. After cooling, the sample was heated rapidly in the dark. Temperature and light emission of the sample were monitored simultaneously.

RESULTS

Table II gives the results of our analysis of several glow curves by the Grossweiner method. As previously reported [6], the electron acceptor ferricyanide enhanced peak 1 and decreased peaks 2 and 3. Conversely, the inhibitor of electron transport, DCMU, abolished peak 1 and enhanced peaks 2 and 3. Table II shows that these additions did not affect the three parameters which we calculated: E, s and r.

Fig. 2 gives our analysis of 6 different glow curves by the Arrhenius method. The two methods are in approximate agreement for the two activation energies which could be obtained from the Arrhenius method: 0.799 eV compared to 0.723 eV for peak 1 and 0.476 eV compared to 0.436 eV for peak 2.

DISCUSSION

There have been a number of attempts at calculating the activation energy connected with metastable states of Photoreaction II, using either thermoluminescence or delayed light. Arnold and Azzi [12] used Eqn (2) which contains two unknown parameters: activation energy and frequency factor. They obtained a frequency factor from a simple glow curve of dried chloroplasts and used this frequency factor for calculating the activation energy of glow curves from living *Chlorella* cells. They obtained activation energies of 0.53 eV and 0.62 eV for peaks which apparently correspond to our peaks 2 and 3. There is, however, little expectation that a frequency factor from dried chloroplasts should be applicable to thermoluminescence from living cells, especially as the dried system gave completely different glow curves from the living system. In any case, our analysis (Table II) shows that different glow peaks from Photoreaction II have widely different frequency factors.

Fleischmann [5] analyzed the thermoluminescence of photosynthetic bacteria by choosing frequency factors ranging from the 10^8 of Randall and Wilkins [10] to the $2.4 \cdot 10^9$ of Arnold [2]. Fleischman obtained activation energies of 0.45–0.55 eV. These values would be accurate only if the chosen frequency factors are close to the actual ones. The thermoluminescence of both bacteria and algae is observed in the same temperature range as our peaks 2 and 3 from chloroplasts. It seems possible that our mathematical treatment might yield results from these systems showing our peaks 2 and 3 to be identical to the algal, and similar to the bacterial systems.

Shuvalov and Litvin [4] combined an analysis of thermoluminescence and delayed light. They presented a detailed analysis of activation energies by dividing the decay curve of ms and s delayed light into five components and assuming first order decay kinetics. The resulting activation energies were compared to those obtained from Arrhenius plots of the initial portion of glow curves. This approach overlooks certain complexities in the delayed light decay. Delayed light decay from 0.01–1 s follows second order kinetics better than first order [13, 14]. Separation of components in the second time-range was done with excitation by red or far-red light, but this decay is known to be extremely complicated and to involve many components [15]. We have been unable to associate any single part of the delayed light decay with a specific peak (or peaks) of thermoluminescence [6]. Furthermore, Shuvalov and Litvin [4] found many discrepancies between the thermoluminescence peaks and the components of delayed light with which they were supposedly associated: opposite effects of DCMU, differing excitation spectra, differing emission spectra, activation energies differing by up to a factor of 2 and Arrhenius constants differing by up to 10^5. On balance, it seems possible that Shuvalov and Litvin's ability to correlate certain aspects of thermoluminescence and delayed light may have been fortuitous.

We are unable to account for the large discrepancies in the Arrhenius activation energies reported by Shuvalov and Litvin and those we have calculated by the same method. From qualitative data we associate their peak L_2 (emitted at -16 °C and "severely depressed" by DCMU) with our peak 1, and their peak L_3 (emitted at 20 °C and "intensified" by DCMU) with our peaks 2 plus 3 [6]. From Arrhenius plots they found 0.35 eV compared to our 0.72 eV for peak 1 (L_2), and 0.9 eV compared to our 0.44 eV for peak 2 (L_3).

Two groups have recently obtained activation energies from analysis of temperature-jump thermoluminescence. This measurement is done by illuminating chloroplasts and then subjecting them to a sudden temperature increase in the dark. The mechanisms of the measurement limit the time-resolution to 1 s or longer after irradiation. The temperature-induced stimulation is therefore of delayed light in the second time-range. Jurinsinic and Govindjee [16] found an activation energy of 0.32 eV. Malkin and Hardt [17] found an error of a factor of 2 in the first group's analysis, thus bringing Jurinsinic and Govindjee's activation energy into close agreement with that of Malkin and Hardt: 0.68 eV. We do not know whether this temperature-jump activation energy corresponds to any of our glow curve peaks, since it lies about midway in energy between our peaks 3 (0.57 eV) and our peak 1 (0.8 eV). The temperature-jump determination is quite close to our Arrhenius plot value of 0.72 eV for peak 1, but we consider the Arrhenius method to be less accurate than the Grossweiner method.

We are unable, as yet, to unambiguously relate the three glow peaks to stabilization of specific steps in quantum conversion by Photoreaction II. It is unlikely that the thermoluminescence represents long-lived electronic excited states: the lifetimes are too long. A more likely possibility is that the thermoluminescence is due to oxidized and reduced moieties formed by the first step of quantum conversion. If these moieties were the primary electron donor and the primary electron acceptor, as suggested by Lavorel [18], then only two metastable states would be expected:

$$Z\text{chl}Q + h\nu \rightarrow Z\text{chl}^*Q \rightarrow Z\text{chl}^+Q^- \rightarrow Z^+\text{chl}Q^-$$

since the light-producing back reaction from $Z\text{chl}^+Q^-$ would have a different activation energy from the back reaction beginning with $Z^+\text{chl}Q^-$, and both metastable states might be "frozen in". This possibilities can account for either one or two glow peaks, but not for three.

Rubin and Venediktov [3] have extended Lavorel's suggestion by assuming that different glow peaks might reflect activation energies of secondary acceptors which back-react with the primary donor. In this case, one might expect to observe more than three peaks in the glow curve, and the lowest activation energy would represent the basic stabilization energy of quantum conversion: 0.48 eV for our peak 2. This value does not fit well with the 0.98 eV which is available.

Another possibility is that thermoluminescence might arise from "freezing in" of transitory metastable states which normally lead to oxidation and reduction of the primary donor and acceptor. In this case, one might expect slightly different activation energies for opposite sides of a two-sided reaction center, and both energies together should add up to about 0.98 eV. This is approximately what we have observed for peaks 2 and 3 (0.48 plus 0.57 equals 1.05 eV). Furthermore, DCMU increases these two peaks, in agreement with the idea that DCMU blocks utilization of early photoproducts by the electron transport chain. Ferricyanide reduces peaks 2 and 3, in agreement with the idea that it activates utilization of early photoproducts by the electron transport chain. Thus, we may speculate that peaks 2 and 3 represent stabilization energies at opposite sides of Reaction Center II, but this idea must by confirmed by more precise determinations of the two activation energies.

How can we account for peak 1? At least two different explanations are possible: that of Lavorel [18] and that of Rubin and Venediktov [3]. If peak 1 represents the stabilization energy for the primary stabilized products of Reaction Center II, then the activation energy we observe should be 0.98 eV or larger, minus any proton motive force which has been built up across the membrane by electron transport. Crofts et al. [9] calculate that about 0.27–0.32 eV might be available from the proton motive force to lower the activation energy for the light-emitting back reaction. Thus, one might expect to observe a glow peak in the range of 0.7–0.8 eV under conditions where a large proton motive force might build up. Since our work was done in the absence of ADP and orthophosphate, we meet this requirement. The effects of addition of ferricyanide and DCMU on peak 1 are, however, somewhat difficult to explain on this hypothesis. Ferricyanide did not change the activation energy of peak 1, although presumably it would have increased the membrane proton motive force by activating electron transport. DCMU should have completely abolished the proton motive force, and we should have observed a new glow peak with activation energy of about 0.98 eV. Since DCMU abolished peak 1, we would be forced to conclude that the new peak was somehow missed in our observations. Furthermore, ferricyanide enhanced peak 1, but would not be expected to increase pools of the primary donor and the primary acceptor in oxidized and reduced forms.

Following the idea of Rubin and Venediktov [3], the peak 1 activation energy might represent a pool of oxidized photoproducts stabilized outside the reaction center. DCMU would abolish such a pool and ferricyanide might increase it by activating electron transport. We can not place this hypothetical pool on the reducing side of the reaction center because ferricyanide would then decrease it. In this case, the 0.8 eV of peak 1 would represent the stabilization energy for the oxidizing side of the reaction

center (about 0.5 eV) plus the activation energies required to reach the pool (0.2–0.3 eV). This hypothesis also gives an explanation for the absence of peak 1 in glow curves of whole algal cells: rate constants on the oxidizing side of Reaction Center II might not allow build up of this particular pool in algae. Such an explanation for the absence of peak 1 in whole algae is consistent with the observation of Joliot et al. [19] that the back reactions ("deactivation") of the S_2 and S_3 states of oxygen evolution are much faster in algae than in chloroplasts of vascular plants. The idea that peak 1 might represent an oxidizing pool is also consistent with our observation [6] that peak 1 is not excited by illumination at temperatures below −50 °C, since some electron transport steps might not function at low temperature.

In conclusion, we would stress that several interpretations of the glow curves are possible, and that further work must be done to clearly associate the three types of energy storage with stabilization of specific steps in quantum conversion.

ACKNOWLEDGEMENTS

This research was supported by a research grant from U.S. National Science Foundation to Professor Walter Bertsch. We wish to thank Dr Peter Stern of Tel Aviv University and Mr Robert Randall of Kennicott Copper Corp. for helpful discussions of the theory of glow curves.

REFERENCES

1 Arnold, W. and Sherwood, H. (1959) J. Phys. Chem. 63, 2–4
2 Arnold, W. (1966) Science 154, 1046–1049
3 Rubin, A. B. and Venediktov, P. S. (1969) Biofizika U.S.S.R. 14, 105–109
4 Shuvalov, V. A. and Litvin, F. F. (1969) Mol. Biol. U.S.S.R. 3, 59–73
5 Fleischman, D. E. (1971) Photochem. Photobiol. 14, 65–70
6 Lurie, S. and Bertsch, W. (1974) Biochim. Biophys. Acta 357, 420–428
7 Strehler, B. and Arnold, W. (1951) J. Gen. Physiol. 34, 309–311
8 Hill, R. and Bendall, F. (1960) Nature 186, 136–138
9 Crofts, A. R., Wraight, C. A. and Fleischman, D. E. (1971) FEBS Lett. 15, 89–100
10 Randall, J. F. and Wilkins, J. H. F. (1945) Proc. Royal Soc. 184, 366–389
11 Grossweiner, L. I. (1953) J. Appl. Phys. 24, 1306–1309
12 Arnold, W. and Azzi, J. R. (1968) Proc. Natl. Acad. Sci. U.S. 61, 29–36
13 Mayne, B. C. (1968) Photochem. Photobiol. 8, 107–113
14 Clayton, R. (1969) Biophys. J. 9, 60–76
15 Bertsch, W. F. and Azzi, J. R. (1965) Biochim. Biophys. Acta 91, 15–26
16 Jurinsinic, P. and Govindjee, (1972) Photochem. Photobiol. 15, 331–348
17 Malkin, S. and Hardt, H. (1973) Biochim. Biophys. Acta 305, 292–301
18 Lavorel, J. (1966) Current Topics in Photosynthesis (J. B. Thomas and J. C. Goedheer, eds), p. 39, Ad. Donker, Rotterdam
19 Joliot, P., Joliot, A., Bouges, B., and Barbieri, G., (1971) Photochem. Photobiol. 14, 287–305