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TEMPERATURE DEPENDENCE OF THE DELAYED FLUORESCENCE FROM WHEAT LEAVES TREATED WITH 3-(3,4-DICHLOROPHENYL)-1-1,-DIMETHYLUREA

VASILIJ N. GOLTSEV <sup>a</sup>, PAVEL S. VENEDIKTOV <sup>a</sup>, DARENIC A. JANUMOV <sup>b</sup>

Biophysical Department, Biological Faculty, Moscow State University, Moscow, and
Plant Physiology Laboratory, Agriculture Institute, st. Nemchinovka, Moscow (U.S.S.R.)

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## Summary

Light saturation curves of the delayed fluorescence of wheat leaves treated with 3-(3,4-dichlorophenyl)-1,1-dimethylurea (DCMU) were measured at different temperatures. Calculated activation energies for a half-saturation actinic light intensity and saturated delayed fluorescence emission were 0.89 and 0.32 eV, respectively. On the basis of the kinetic model energy levels of Photosystem II reaction center components were estimated.

### Introduction

In the accompanying paper [1] a kinetic model was proposed for delayed fluorescence of Photosystem II (PS II) inhibited by DCMU. Using this model it is possible to obtain analytical relations between parameters of light curves of delayed fluorescence and electron transfer rate constants within PS II:

$$L = L_{\text{max}} \frac{I}{I + I_{1/2}}$$

where

$$L_{\text{max}} = N \cdot k_{\text{r}}^* \cdot \varphi_{\text{f1}} \cdot \frac{k_{-1}}{k_1}; I_{1/2} = \frac{k_{-1}}{k_1} \cdot k_{\text{r}}$$

where I is the actinic light intensity;  $L_{\max}$  is the delayed fluorescence intensity at saturating light;  $I_{1/2}$  is the constant equal to actinic light intensity for which  $L = L_{\max}/2$ ; N is the number of PS II reaction centers;  $k_r^*$  and  $k_r$  are the rate constants for radiation and radiationless recombination of primary charges in

the reaction center of PS II;  $k_{-1}/k_1$  is the equilibrium constant for electron transport reactions between electron donor of PS II (Z) and reaction center chlorophyll (P);  $\varphi_{f1}$  is the prompt fluorescence yield.

Keeping in mind that the temperature dependencies of  $k_1^*$  and  $k_{-1}/k_1$  are determined, respectively, by  $\Delta G_1$  and  $\Delta G_2$  ( $\Delta G_1$  is the activation energy required for an electron to rise from the level of the reduced primary acceptor to the first singlet level of P and  $\Delta G_2$  is the redox potential difference of Z and P, respectively) one can write:

$$k_{\rm r}^*(T) = \overline{k}_{\rm r}^* \cdot \exp\left(-\frac{\Delta G_1}{kT}\right); \frac{k_{-1}}{k_1} = \exp\left(-\frac{\Delta G_2}{kT}\right),$$

where  $\overline{k}_{r}^{*}$  is a frequency factor.

Since radiationless recombination does not require any activation energy [1], it is evident, that the activation energy measured from the  $L_{\text{max}}$  temperature dependence is equal to  $\Delta G_1 + \Delta G_2$ , and that measured from the temperature dependence of  $I_{1/2}$  is equal to  $\Delta G_2$ .

In the paper which is reported here we measured the temperature dependence of parameters  $L_{\text{max}}$  and  $I_{1/2}$  and estimated free energy changes associated with electron transfer from Z to P<sup>+</sup> and from the excited P to Q.

### Materials and Methods

Wheat plants were grown 10–12 days in a 'Sherer' growth chamber at  $18^{\circ}$ C under a periodic illumination (20 000 lux), 16 h/day. Prior to measurements, the leaves were infiltrated with a  $1 \cdot 10^{-4}$  M solution of DCMU, and left in the solution for 2 hs.

Delayed fluorescence intensity was measured in a convetional Bequerel-type phosphoroscope with a time interval of 2.5 ms between excitation and measurement.

Excitation was given from a tungsten-halogen lamp, with the beam passed through a water filter and a red cut-off glass filter ( $\lambda$ >620 nm). Attenuation was done by means of calibrated neutral filters.

The sample holder was cooled by a continuous flow of freon and sample temperatures were regulated with an electric heater mounted in the holder. Using an electronic device, the temperature was adjusted to be stable to within  $\pm 0.1$ °C.

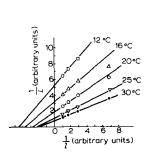
#### Results and Discussion

Experiments revealed influence of temperature on the light curves of the delayed fluorescence from DCMU-treated wheat leaves. The curves plotted as double reciprocal plots for different temperatures are presented in Fig. 1. Intersections with axes give values of  $L_{\rm max}$  and  $I_{1/2}$ .

The energies  $\Delta G_2$  and  $(\Delta G_2 + \Delta G_1)$  were defined from the Arrhenius plots for  $I_{1/2}$  and  $L_{\text{max}}$  (see Fig. 2).

The activation energies for  $L_{\text{max}}$  and  $I_{1/2}$  (mean of 12 separate experiments) were found to be 0.89  $\pm$  0.06 eV and 0.32  $\pm$  0.04 eV, respectively.

The experimental value of 0.32 eV for  $\Delta G_2$  corresponds to the equilibrium



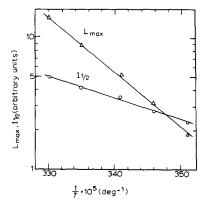


Fig. 1. Double reciprocal plots of DCMU-treated wheat leaves delayed fluorescence (L) as function of actinic light intensity (I). The numbers at curves correspond to the sample temperatures.

Fig. 2. Arrhenius plots of  $L_{\text{max}}$  and  $I_{1/2}$ .

constant  $k_{-1}/k_1 \approx 2 \cdot 10^{-5}$  for the donor side of PS II.  $\Delta G_1$  is 0.89 - 0.32 eV = 0.57 eV.

Using the value of 1.8 eV for the energy of a chlorophyll excited state and -0.035 eV as the redox potential of the primary acceptor of PS II [2], one can represent the energy levels of the PS II components on a redox potential scale (see Fig. 5 in Ref. 1). Simple calculations give a value of +1.2 eV for the redox potential of P.

The value for the activation energy, measured on the basis of  $L_{\rm max}(T)$ , is nearly the same as that reported in Ref. 3 (0.9 eV) for the delayed fluorescence intensity of DCMU-treated chloroplasts. This value is, however, smaller than the difference between the energy of red quantum (1.8 eV) and the energy stored by the electron transfer from water ( $E_{\rm m,pH7}$  = +0.8 V) to Q ( $E_{\rm m,pH7}$  = -35 mV), i.e., 0.965 eV. Perhaps, this is because in the presence of DCMU the  $H_2O/O_2$  couple does not act as an electron donor for P<sup>+</sup> and P is in equilibrium with Z ( $S_1/S_2$  states of the oxygen evolving system). Using our data, one finds an estimate for the redox potential of Z as +0.875 V.

The value of 0.57 eV obtained by us for the free energy loss for the electron transition from P to Q is not far from that calculated by Jursinic and Govindjee [4] from yields and life-times of prompt and microsecond-delayed fluorescence of pea chloroplasts.

# References

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