

Statistics of the photon distribution in the set of photosynthetic antenna domains

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The distribution of the excitations caused by the pump laser pulse in the domains of light-harvesting antenna is shown to be the hypergeometrical one. The experimental curves of the photo-oxidation of reaction centers versus the light intensity of pico-second pulses measured for two distinct samples with monocentral antenna domains are in excellent agreement with the statistical distribution predicted.

Antenna domain; Statistical distribution; Quantum yield

1. INTRODUCTION

The study of the fluorescence yield of antenna is known to be a useful technique for the investigation of primary photosynthetic events [1–3]. To explain the decrease in the fluorescence yield with pulse intensity two distinct approaches have been proposed. Mauzerall [4,5] has assumed that the excitations caused by actinic pulse are Poisson distributed in the domains of antenna. Another point of view [3,6,7] implies that fluorescence quenching occurs due to bi-excitation processes such as singlet-singlet annihilation. This latter approach has also been used to explain [8,9] the absorption picosecond experiments with purple bacteria [10–12]. In particular it has been declared that the oxidation of reaction centers as a function of pulse intensity could be described by using the same set of parameters as that obtained from fitting the fluorescence data within the bi-excitation model without recourse to statistics [9].

Nevertheless it is quite evident that in the case of

restricted migration of excitations between domains, the statistical distribution ought to affect the quantum yield even in the absence of any bi-excitation effects. At the same time the Poisson distribution postulated by Mauzerall [4,5] does not seem to be applicable a priori to the system under consideration since it implies 'sampling with replacement' (see e.g. [13]), while a once-excited chlorophyll molecule is supposed to become transparent for other photons of the same energy (sampling without replacement).

In this paper we deduce that excitations are hypergeometrically distributed in the set of antenna domains. We also present the experimental curves of oxidation of reaction centers versus the pulse intensity for two pigment-protein complexes composed of monocentral domains. The comparison of experimental data with the theoretical model provides the estimation of the number of chlorophyll antenna units forming a sole domain.

2. MATERIALS AND METHODS

The pigment-protein complexes B890 were isolated from the purple bacterium *Chromatium minutissimum* according to the modified method of Moskalenko and Yerokhin [14]. These com-

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plexes contain 30 light-harvesting Chl *a* molecules per one reaction center (RC) and have the monocentral organization. The pigment-protein complexes of photosystem I from bean leaves were prepared as described elsewhere [15]. These complexes contain 60 Chl *a* molecules per 1 RC (P-700) and are also monocentral [16].

The picosecond measurements were carried out with a difference spectrometer described in [10,17]. The homogeneity of light intensity over beam cross-section was achieved by inserting of a clouded glass. To vary the intensity of the pump beam the cell with a tunable path length containing a mixture of water with indian ink was used.

3. RESULTS AND DISCUSSION

3.1. Theoretical model

The distribution of the excitations in the set of antenna domains could be deduced in the following way. Let the sample be composed of N molecules which are divided into k domains each of which consists of n molecules ($N = kn$). The probability, W_i , that from the total number of absorbed photons, p , any given domain absorbs i photons is equal to (number of possibilities to place i photons in n molecules) $\times$ (number of possibilities to place the remainder of photons,

$p - i$, in the remainder of molecules, $N - n$): (total number of possibilities to place p photons in N molecules) or

$$W_i = \frac{\binom{n}{i} \binom{N-n}{p-i}}{\binom{N}{p}}, \quad (1)$$

where $\binom{x}{y}$ stands for binomial coefficient.

Eqn 1 is known as hypergeometric distribution. In accordance with real conditions of the experiment one can conclude that $p \gg i$ and $(N - p) \gg (n - i)$. Then eqn 1 reduces to binomial distribution (see e.g. [13]):

$$W_i = \binom{n}{i} \cdot (1 - p/N)^{n-i} \cdot (p/N)^i. \quad (2)$$

Eqn 2 permits one to calculate the probability W_i as a function of a 'mole fraction' of excited molecules, $p/N \equiv z$.

If the number of domains is sufficiently large the probability W_i is known to be treated as the fraction of the domains absorbing i photons. Assuming that open reaction centers remain only in the domains which absorb no photons we can write the number of closed reaction centers in the sample as

$$M_{RC+} = k(1 - W_0), \quad (3)$$

where W_0 is the fraction of unexcited domains. Calculating W_0 in accordance with eqn 2 and substituting it as well as $k = N/n$ into eqn 3 we obtain the expression for the yield of photooxidation of reaction centers in monocentral domains:

$$\phi \equiv \frac{M_{RC+}}{p} = \frac{1 - (1 - z)^n}{nz} \quad (4)$$

3.2. Comparison with experimental data and discussion

The curves calculated from eqn 4 for different values of n as well as the experimental points measured for two distinct samples composed of monocentral domains are shown in figs 1 and 2. It is seen from fig.1 that the experiments performed with the pigment-protein complex B890 are well described by a theoretical curve with $n = 30$ in a good agreement with biochemical data. At the

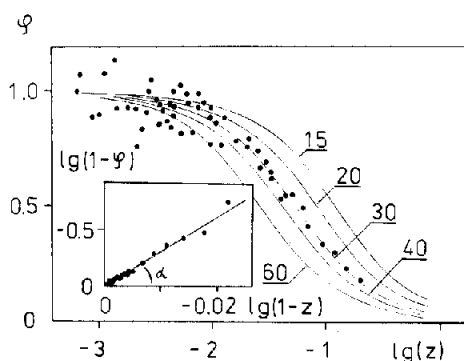


Fig.1. The yield of photooxidation of reaction centers in the pigment-protein complex B890 from *C. minutissimum* vs the common logarithm of the mole fraction of excited molecules. Theoretical curves are plotted in accordance with eqn 4 for different n -values. The measurements were carried out in the presence of sodium ascorbate in the 1 mm cell ($A_{885} = 1.64$, $A_{930} = 0.18$, $T = 65\%$) with $\lambda_{ex} = 930$ nm, $\lambda_{pr} = 898$ nm. The probe pulse was 1000 ps after the pump pulse. See text also.

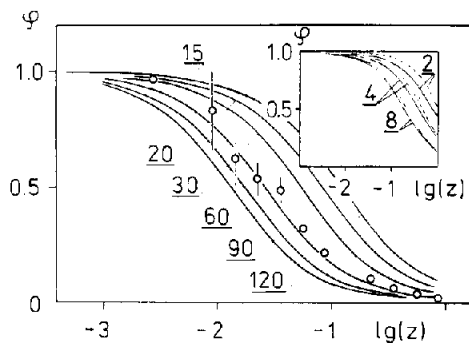


Fig.2. The yield of photooxidation of reaction center P-700 from photosystem I of bean leaves vs the common logarithm of the mole fraction of excited antenna molecules. Theoretical curves are plotted in accordance with eqn 4 for different n -values. The measurements were carried out in the presence of 5 M TMPD in 2 mm cell ($A_{667} = 1.30$) with $\lambda_{ex} = 667$ nm, $\lambda_{pr} = 700$ nm. The probe pulse was 420 ps after the pump pulse. The quantum yield was calculated taking into account both the damping decrement of light intensity with path length and the presence of a fraction of photoinactive P-700. The dashed lines in the inset correspond to a Poisson distribution. See text also.

same time the theoretical curve with $n = 60$ gives a good fit to experimental values obtained for the pigment-protein complex from bean leaves (fig.2). This also coincides with biochemical conclusions [16]. In the inset of fig.1 the experimental points are plotted in logarithmic coordinate in which according to eqn 4 the observable dependence should be a straight-line with a slope equal to n . It is seen from the inset that this does occur and the slope is equal to 30.

The attractive fit of theoretical curves with the experimental data in the region where $z \geq 1/n$ was obtained for both pigment-protein complexes permits one to conclude that the bi-excitation processes at least of the type of $S_1 + S_1 \rightarrow S_0 + S_0 + \text{heat}$ are negligible in these complexes. As far as the singlet-singlet annihilations of another type are concerned, e.g. $S_1 + S_1 \rightarrow S_j + S_0 \rightarrow S_1 + S_0 + \text{heat}$, they apparently could not affect the dependence $\phi(z)$ for monocentral domains. Indeed even if processes of this type are much more rapid than trapping excitations by reaction centers there would be at least one excitation in a domain which will close the reaction center.

As we have already mentioned the statistical ap-

proach to the fluorescence yield problem was proposed by Mauzerall [4,5], who has assumed a Poisson distribution of excitations in the domains. It is known that the binomial distribution [2] reduces to a Poisson distribution if $z \ll 1$ and $n \rightarrow \infty$, and so that n/N is constant [13]. In the case of monocentral domains of sufficiently large size ($n \gg 1$), when we practically confined ourselves to the calculation of the probability W_0 , such a limiting process seems to be reasonable. However in the case of either small domains [18,19] or, for example, bacteriorhodopsin triads [20], a Poisson distribution becomes unsuitable (see inset of fig.2).

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