Concentration Quenching in Chlorophyll

By JOHN DALTON

(*Department of Chemistry, City of London Polytechnic, 31 Jewry Street, London* EC3N 2EY)

Summary A modification of Forster's equation for the quantum yield of heterogeneous radiationless energy transfer improves its description of homogeneous concentration quenching, at the expense of a new parameter

which can be related to the number of steps taken by the excitation in its random walk and which seems to be independent of concentration

---

HOMOGENEOUS energy transfer processes in concentrated solutions of chlorophylls are of interest for the information they yield on the natural light-harvesting systems of photosynthesis Quantitative descriptions of chlorophyll concentration-quenching curves (Figure) have been attempted with the aid of Forster's equation[1] (1) for the

$$\Phi_t = \pi\gamma\exp(\gamma^2)\mathrm{erfc}(\gamma) \quad (1)$$

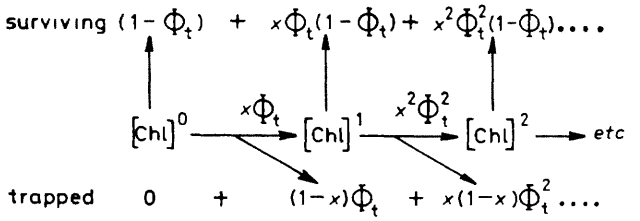
quantum yield of heterogeneous energy transfer (Φ_t) in the very weak coupling case,[2] with the empirical equation[3] (2) whose concentration parameter (γ') differs, in general,

$$\Phi/\Phi_f = 1/(1 + \gamma'^2) \quad (2)$$

from that of equation (1), and by large scale computer simulation[4] The best fits seem to be obtained with equation (2), whose single variable parameter is not directly interpretable in theoretical terms Equation (1), on the other hand, has a secure theoretical basis[1,2,5,6] and is experimentally well obeyed[6,7] in cases of heterogeneous transfer Its failure in the homogeneous case is presumably due to the neglected secondary fluorescence emission following radiationless transfer from the primary sites of excitation There is evidence from emission decay profiles,[8] fluorescence depolarisation measurements,[9] and computer simulation[4] that such secondary emission contributes significantly to the measured fluorescence intensity at high concentrations

Accordingly, those chlorophyll molecules contributing to the measured fluorescence intensity may be divided into populations labelled $[Chl]^n$ where n (= 0, 1, 2) indicates the number of radiationless transfers preceding excitation Thus $[Chl]^0$ represents the directly (*i e* radiatively) excited population, $[Chl]^1$ that excited by a single radiationless transfer from $[Chl]^0$, *etc* However, the observation of concentration quenching suggests that the radiationless transfer processes are not completely efficient In other words, each radiationless transfer step has a small but non-zero probability of terminating in a trap from which fluorescence intensity will not further contribute to the measured intensity It is convenient to define this trapping probability as $1 - \bar{x}$, so that $\bar{x}$ ($0 < \bar{x} < 1$) represents an average value of the survival probability per transfer

With this notation, the distribution of 'surviving' and 'trapped' excitation in each population $[Chl]^n$ is given by the Scheme The apparent fluorescence quantum yield (Φ) is then obtained by summing the contributions from the



SCHEME Energy partition during multiple homogeneous transfer

surviving excitation in each population They form an infinite but convergent geometric series with an exact sum [equation (3), where Φ_f represents the fluorescence quantum yield at infinite dilution]

$$\Phi/\Phi_f = (1 - \Phi_t) + \bar{x}\Phi_t(1 - \Phi_t) + (\bar{x}\Phi_t)^2(1 - \Phi_t) = (1 - \Phi_t)/(1 - \bar{x}\Phi_t) \quad (3)$$

The main assumption involved in writing equation (3) is that the quantities $\bar{x}$ and Φ_t may be separately averaged, the latter according to equation (1) With this proviso, $\bar{x}$ can be treated as an experimentally determined parameter, interpreted *via* its defining equation (3) In particular, the functional dependence of $\bar{x}$, the average survival probability per transfer upon solute concentration is not directly specified by equation (3) Surprisingly, however, excellent fits to the classical[4] data[3] of Watson and Livingston at realistic values of the Forster critical transfer distance[1] (Table) are obtained by taking $\bar{x}$ to be independent of concentration Standard estimates of error[10] are chlorophyll a in ether (Figure) 0 0235 from equation (2)

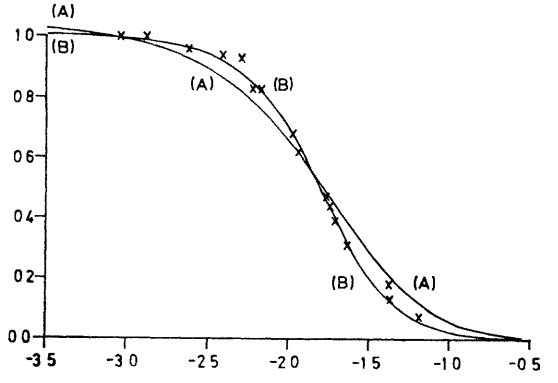


FIGURE Relative fluorescence emission intensity (ordinate, arbitrary units) of chlorophyll a in ether *vs* decadic logarithm of concentration/mol l^{-1} experimental data from ref 3 (A), best fitting unmodified Forster function [equation (3) with $\bar{x} = 0$], (B), best fitting modified Forster function [equation (3) with $\bar{x} = 0\ 960$]

TABLE Critical energy transfer distances (R_0/nm) for chlorophylls a and b

	Theory[a]	Expt	Experimental method
chl a (ether)	5 17	4 6	Equation (3), $\bar{x} = 0\ 960$
chl a (lecithin)	5 45	—	
chl a (castor oil)	—	8 7, 4 85	Fluorescence depolarisation, ref 9
chl b (ether)	—	6 9	Equation (3), $\bar{x} = 0\ 995$
chl b (lecithin)	5 55	—	
chl b (castor oil)	—	4 85, 3 5	Fluorescence depolarisation, ref 9
chl b → chl a (lecithin)	5 45	6 5	Equation (1), ref 7

[a] Theoretical values from ref 7 by the method of ref 1

with $C_{1/2} = 15{\cdot}61 \times 10^{-3}$ mol l^{-1} *vs.* 0·0187 from equation (3) with $C_0 = 4{\cdot}63 \times 10^{-3}$ mol l^{-1} and $\bar{x} = 0{\cdot}960$; and chlorophyll b in ether, 0·0253 from equation (2) with $C_{1/2} = 13{\cdot}63 \times 10^{-3}$ mol l^{-1} *vs.* 0·0247 from equation (3) with $C_0 = 1{\cdot}36 \times 10^{-3}$ mol l^{-1} and $\bar{x} = 0{\cdot}995$.

Thus equation (3) provides a new and apparently satisfactory way of determining the Förster critical transfer distance for homogeneous energy transfer from either fluorescence intensity or lifetime[8] data. In conjunction with Watson and Livingston's data,[3] it further suggests that the excitation trapping probability per transfer might be independent of concentration. A mechanism with this feature, based upon the Stokes' energy loss attending each transfer step in the Scheme, is under investigation.

(*Received, 15th August* 1979; *Com.* 877.)

[1] Th. Förster, *Z. Naturforsch.*, 1949, **4a**, 321; *Discuss. Faraday Soc.*, 1959, **27**, 7; erfc(γ) = 1 − erf(γ); for $\gamma > 3$, $\Phi_t = 1 - 1/Z + 1.3/Z^2 - 1.3.5/Z^3 \ldots$ where $Z = 2\gamma^2$.
[2] Th. Förster, 'Modern Quantum Chemistry, Part III,' ed. O. Sinanoglu, Academic Press, London, 1965, pp. 93—137.
[3] W. F. Watson and R. Livingston, *J. Chem. Phys.*, 1950, **18**, 802.
[4] G. S. Beddard and G. Porter, *Nature*, 1976, **260**, 366.
[5] D. L. Dexter, *J. Chem. Phys.*, 1953, **21**, 836.
[6] R. G. Bennett and R. E. Kellogg, 'Progress in Reaction Kinetics, vol. 4,' ed. G. Porter, Pergamon, London, 1967, ch. 6.
[7] A. R. Kelly and G. Porter, *Proc. Roy. Soc.*, 1970, A**315**, 149.
[8] G. S. Beddard, S. E. Carlin, and G. Porter, *Chem. Phys. Letters*, 1976, **43**, 27.
[9] J. C. Goedheer, thesis (Utrecht), 1957 and A. P. Losev and E. I. Zen'kevich, *Zhur. priklad. Spektroskopii*, 1968, **9**, 144: quoted in S. M. de B. Costa, J. R. Froines, J. M. Harris, R. M. Leblanc, B. H. Orger, and G. Porter, *Proc. Roy. Soc.*, 1972, A**326**, 503.
[10] 'Handbook of Physics and Chemistry,' 49th edn., Chemical Rubber Co., Ohio, 1968, sect. A254.