PHOTOLUMINESCENCE METHODS FOR EVALUATION OF SOLUBILIZATION PARAMETERS AND DYNAMICS OF MICELLAR AGGREGATES. LIMITING CASES WHICH ALLOW ESTIMATION OF PARTITION COEFFICIENTS, AGGREGATION NUMBERS, ENTRANCE AND EXIT RATES

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Poisson statistics is a natural consequence of a distribution of solubilized molecules among micellar aggregates. With postulates of non-cooperativity and thermodynamic considerations, relationships for extent of solute fractionation between the micellar phase and the aqueous phase are derived, as are expressions for the handling of experimental data from the quenching of photoluminescence probes of micellar aggregates.

1. Introduction

Solubilization or association of organic and inorganic substrates by micellar aggregates is an essential feature of detergent solutions and plays an important role in biological and industrial processes [1,2]. The inclusion of substrates is often used to probe the nature of the microenvironment of micellar aggregates via spectroscopic methods [2]. Luminescent probes [3–15] have recently become widely used because photoluminescence techniques allow studies of dynamic phenomena in the range of 10^{1} to 10^{-12} s, luminescence spectroscopy is a sensitive analytical tool [3], and luminescence spectra provide a means of identifying transients and inferring the nature of their environments.

The quantitative treatment of spectral and dynamic data requires a knowledge of the probability distribution of the number of substrates among the micellar aggregates. The purpose of this work is to show that the Poisson statistics provide a natural extension of the simplest thermodynamic model and that the model is valid if the probe is only partially micellized.

2. Solubilization model

We wish to find, in terms of the known or measurable quantities of the total substrate concentration [S] and the total micelle concentration [M], the concentrations of the substrate in water [S_w], micelle bound [S_m] and the probability distribution P_s that a given micelle is associated with s substrate molecules[‡]. A stepwise association model can be written as

aqueous phase $M_0 + S_w \overset{K}{\rightleftharpoons} M_1$ micellar phase (1)

$$M_1 + S_w \rightleftharpoons M_2 \quad (2)$$

$$\vdots$$

$$M_{s-1} + S_w \rightleftharpoons M_s \quad (3)$$

where M_s is the micelle species associated with s substrate molecules. Thermodynamics of the simplest solubilization model leads to the following results:

[‡] The total micelle concentration, [M], can be related to the detergent concentration, [Det], and the critical micelle concentration, CMC, by $[M] = ([Det] - CMC)/\bar{n}$ where $\bar{n}$ is the mean micelle aggregation number. See ref. [2] for more detail.

$$[S_w] = [S]/(1 + K[M]), \quad (4)$$

$$[S_m] = K[M][S]/(1 + K[M]), \quad (5)$$

$$P_s = (\langle s\rangle^s/s!)\,e^{-\langle s\rangle}, \quad (6)$$

$$\langle s\rangle = K[S]/(1 + K[M]). \quad (7)$$

Here, K is the binding constant of the first association step and $\langle s\rangle$ is the mean number of substrates bound per micelle. Combination of eqs. (4) and (5) leads to

$$K = [S_m]/[S_w][M], \quad (8)$$

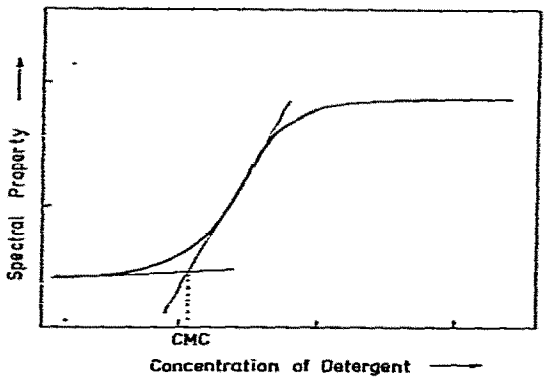
so that the stepwise solubilization model that leads to Poisson statistics of eq. (6) can, "symbolically", be represented by the following equilibrium ‡:

$$\text{aqueous phase} \quad S_w + M \overset{K}{\rightleftharpoons} S_m \quad \text{micellar phase} \quad (9)$$

3. Application to probe partitioning

If the spectral properties of a probe change on micellization, then eq. (9) can be used to predict the commonly observed [1,2,16,17] detergent concentration dependence of substrate spectral properties. Fig. 1 shows a typical detergent concentration dependence of substrate spectral property. Such data can be analyzed on the basis of eqs. (4) and (5) to provide the binding constant K of the substrate, and if coupled with dynamic measurements, the residence time of the substrate in micellar aggregates. This method will not be useful for accurate determination of the CMC if the probe possesses such a large affinity for micelles that measurable association effects occur with the low concentration of micelles that exist just below CMC.

‡ Notice, however, that eq. (9) is symbolic of the micellization equilibria since in a mass action representation [M] would be the concentration of micelles free of substrates and not the total micelle concentration as is in eq. (8).

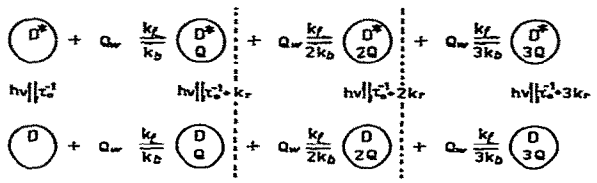


4. Application to quenching of luminescent probes

In such experiments one measures the luminescence intensity following steady state of pulsed excitation. With the aid of eqs. (4)–(7) one can predict the behavior of the general system where both donor and quencher species are partitioned between the water and micelle phases. However, as in practice, it will be more revealing to consider the simplified case where donor species, D, are almost exclusively micellized and at sufficiently low concentration such that $[D]/[M] \ll 1$ to ensure no self-interaction.

There exist a large number of general situations which will be a function of probe residence time (exit rate), probe entrance rate, quenching rate within the micellar aggregate, decay rate of the donor, etc. We consider here only situations for which the decay of the micellized donor is fast relative to its exit rate from the micelle. This restriction ensures that the excited donor will remain within the micelle during its lifetime. In practice aromatic compounds appear to possess exit rates of the order of 10^6 s^{-1} or less [14, 15]; thus, our discussion will refer most usefully to *fluorescent probes*. The recent discovery of a wide range of *phosphorescent probes* [18–20] for micellar aggregates requires consideration of an extended kinetic scheme which allows for escape of the excited donor into the aqueous phase.

◀ Fig. 1. Schematic representation of the commonly observed dependence of an observed spectroscopic property (photoluminescence quantum yield, luminescence lifetime, luminescence maximum, luminescence quenching constant, degree of luminescence polarization, etc.) on concentration of detergent. The "break" in the spectral property usually is related to the onset of formation of micellar aggregates at the "critical micelle concentration" or CMC.



Scheme 1. A generalized scheme for analysis of photoluminescence data obtained from detergent solution containing micelles. See text for discussion.

The kinetic scheme for which D and D* are completely micellized is given in scheme 1 where the circles symbolize micelles, τ_0 is the luminescence lifetime of a luminescent probe molecule D in the absence of quenchers, Q_w is the water solubilized quencher, k_f and k_b are the respective forward and backward rate constants of micellization and k_r is the unimolecular reactive rate constant of quenching in the micelle. The above scheme assumes that for a given micelle the quencher escape or reaction probability is proportional to the number of quenchers present, the former assumption being one that leads to the Poisson statistics. It is expected, and it will be shown, that the above scheme leads to a complicated function for the description of donor luminescence intensity reducing the reliability in the fit of data to theory. We can, however, break the above kinetic scheme into theoretically distinct and experimentally realizable cases by introducing the concept of *static quenching*. By "static" [4] or "active" quenching we mean the condition where the micelles containing D* and Q are completely quenched. In terms of the above kinetic scheme this means $k_r \ll \tau_0^{-1} + k_b$. Static quenching is expected when the mode of quenching is very fast such as in energy transfer processes. It will be shown that static quenching implies observation of single exponential decay of luminescence and that such measurements are crucial to an understanding of the system under study. $I(t)$ is measured following pulsed excitation and when in steady state irradiation the ratio of luminescence intensity in the presence and absence of quencher, I/I^0 is measured.

Case 1: Static quenching, quencher is totally micellized

This is the simplest case and one that leads only to information about the mean micelle aggregation number. Here emission occurs *only from that fraction of D* containing micelles that are free of quencher molecules*, that is, $P_0 = e^{-\langle q \rangle}$ where $\langle q \rangle = [Q]/[M]$. Though the overall luminescence intensity is reduced, *the measured lifetime of D* remains constant upon addition of Q* (see fig. 2).

For continuous irradiation we have

$$\ln(I^0/I) = [Q]/[M]. \qquad (10)$$

This equation has the same functional form as the Perrin's model [21] but the *constant* of "active sphere volume" has been replaced by the total micelle concentration, an experimental *variable*. A knowledge of the total micelle concentration would, of course, lead to the mean micelle aggregation number. In contrast to other methods [21] of aggregation number measurement, *the above method offers the possibility of simple experimentation and is not limited to detergent concentrations near the critical micelle concentration, CMC.* Analysis of the above type has been used [4] to evaluate the mean aggregation number of sodium dodecyl sulfate successfully. The reported [22] quenching of pyrene fluorescence by amines probably can also be categorized as case 1.

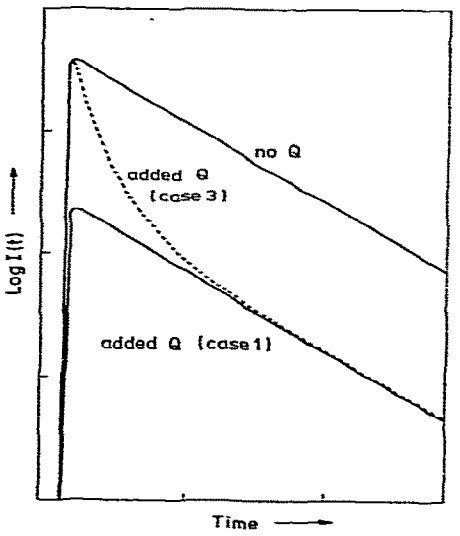


Fig. 2. Schematic representations of case 1 (limit of complete static quenching of D* when a micelle contains at least one Q and no quenching of D* when a micelle contains no Q), and case 3 (limit of nonstatic quenching). Decay of D* is strictly exponential for case 1 and non-exponential for case 3. In both cases Q is completely micellized. See text for further discussion.

Case 2: Static quenching, quencher is partially micellized

Experimental results that fall in this category can lead to information about the mean micelle aggregation number, the quencher binding constant and mean residence time in the micelle aggregate. Because quenching is static, as in case 1, emission still occurs only from that fraction of D^* containing micelles that are free of quencher molecules, that is, $P_0 = e^{-\langle q\rangle}$, where now according to eq. (7) the average number of quenchers per micelle is $\langle q\rangle = K[Q]/(1 + K[M])$. The lifetime τ of the emitting states is, however, reduced by dynamic diffusional quenching by water solubilized quencher molecules Q_w. This is shown in the equation

$$\tau^{-1} = \tau_0^{-1} + k_f[Q]_w, \qquad (11)$$

where the rate constant of quenching equals the forward rate constant of binding k_f since every association results in complete quenching‡‡. Fig. 3 shows the expected luminescence decay shape corresponding to case 2.

Substituting eq. (4) in (11) we have

$$\tau^{-1} = \tau_0^{-1} + k_f[Q]/(1 + K[M]), \qquad (12)$$

which leads to the interesting prediction that *the observed luminescence lifetime increases as a function of increasing detergent concentration*. This is simply a result of the fact that increased detergent concentration reduces the quantity of water solubilized quenchers. For continuous irradiation we have

$$I^0/I = (1 + k_f\tau_0[Q_w])\exp\{K[Q]/(1 + K[M])\}, \qquad (13)$$

where the first and second terms are contributions to intensity quenching from dynamic and static quenching, respectively. A series of investigations [23] of the quenching of naphthalene fluorescence by cyclic azo compounds has been shown to be of the case 2 type and from the data mean micelle aggregation number, quencher binding constant and residence time ($k_b^{-1} = K/k_f$) have been derived. It should be noticed that eq. (13) reduces to eq. (10) when the quencher becomes totally micellized, that is, $K[M] \gg 1$.

‡‡ The observed rate constant of quenching need not be diffusion controlled since it is limited to the value of forward binding constant k_f.

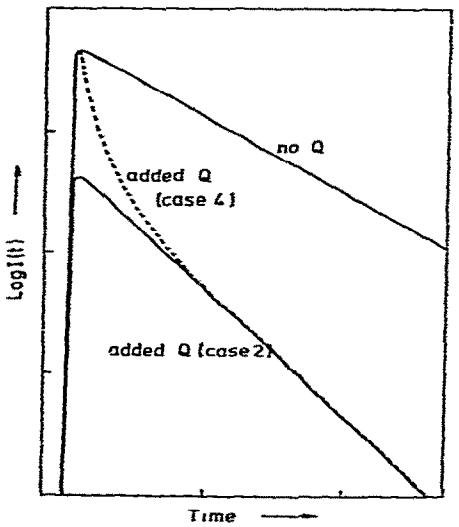


Fig. 3. Schematic representation of case 2 (limit of static quenching with partially micellized quencher) and case 4 (limit of nonstatic quenching with quencher partially micellized). See text for discussion.

Case 3: Nonstatic quenching, quencher is totally micellized

It was seen that static quenching (cases 1 and 2) was distinguished by the fact that the luminescence decay is single exponential. Conversely, nonstatic quenching is distinguished by nonexponential decay of luminescence. This is because the lifetime of D^* in a given micelle depends on the number of cohabitant quencher molecules. With the assumption that for a given micelle the quenching probability is proportional to the number of cohabitant quencher molecules, it can be shown that the luminescence decay function is given by

$$I(t)/I(0) = \exp\{-[t/\tau_0 + \langle q\rangle(1 - e^{-k_r t})]\} \qquad (14)$$

and for continuous irradiation

$$\frac{I}{I^0} = e^{-\langle q\rangle}\sum_{q=0}\frac{\langle q\rangle^q}{[1 + q(k_r\tau_0)]\,q!}, \qquad (15)$$

where for totally micellized quenchers $\langle q\rangle = [Q]/[M]$. The dashed curve of fig. 2 shows typical decay pattern expected from eq. (14). The quenching [11] of pyrene fluorescence by Cu^{2+} ions in sodium dodecyl sulfate seems categorizable to case 3 and data has yielded the

mean interaction time, k_r^{-1}, of micelle bound substrates for this case. It is to be noted that eq. (15) reduces to (10) of case 1 when $k_r\tau_0 \to 0$, that is, when quenching becomes static.

Case 4: Nonstatic quenching, quencher is partially micellized

This is the most general case and the total picture of scheme 1 must be considered. If $[M_q^*]$ represents the concentration of D^* containing micelles bound to q quencher molecules, then the general rate equations are

$$d[M_0^*]/dt = -(\tau_0^{-1} + k_f[Q_w])[M_0^*] + k_b[M_1^*], \quad (16)$$

$$d[M_q^*]/dt = k_f[Q_w][M_{q-1}^*] - \{\tau_0^{-1} + k_f[Q_w] + q(k_r + k_b)\}[M_q^*] + (q+1)k_b[M_{q+1}^*]. \quad (17)$$

The general solution to this difference-differential equation has already been reported [24] and is reproduced here. For the luminescence decay function, one obtains

$$\frac{I(t)}{I(0)} = \exp\{-[t/\tau + \langle q\rangle \alpha_r^2(1 - e^{-(k_r+k_b)t})]\}, \quad (18)$$

where

$$\tau^{-1} = \tau_0^{-1} + k_Q[Q_w], \quad (19)$$

$$k_Q = \alpha_r k_f, \quad \alpha_r = k_r/(k_r + k_b), \quad (20)$$

where $[Q_w]$ and $\langle q\rangle$ are given by eqs. (4) and (7), respectively. For continuous irradiation the luminescence intensity can be found by integration of (18) over all time. The result is

$$\frac{I}{I^0} = \frac{\tau}{\tau_0} e^{-\langle q\rangle\alpha_r^2} \sum_{q=0}^{\infty} \frac{(\langle q\rangle\alpha_r^2)^q}{[1 + q(k_b + k_r)\tau_0]q!}. \quad (21)$$

The dashed curve of fig. 3 shows typical decay of luminescence expected of eq. (18). Ideally, analysis of data on the basis of eq. (18) would yield all parameters obtainable in cases 1–3. In practice, the fit of data to so many parameters is complicated. Notice that eq. (18) is reduceable to those applicable to cases 1–3 when assumptions pertaining to those cases are imposed on it. Furthermore, the dynamic quenching rate constant k_Q can be much smaller than the forward association constant k_f if the residence time is much shorter than the interaction time constant (eq. (20)). The quenching of pyrene fluorescence [6] by methylene iodide and nitromethane was analyzed on the basis of an equation similar to (18) and seems to be categorizable to case 4. Experiments with the quenching of 1,5-dimethyl naphthalene fluorescence in detergent by oxygen under high pressure also appear to follow case 4.

5. Situations involving excited donor escape and quenching in the aqueous phase

Situations other than those considered here have been encountered in the literature, particularly with triplet probes. For example, a study of the sensitization of rare earth ion luminescence by energy transfer from aromatic hydrocarbon triplet donors in SDS solution has been reported. The results are consistent with nearly complete micellization of the probe but suggest a rapid exchange of the probe with aqueous phase relative to quenching [14]. In this case, $k_r \ll k_b$ and eq. (18) becomes

$$I(t)/I(0) = \exp[-(t/\tau + k_r\langle q\rangle t)]. \quad (22)$$

In such a situation, k_r the interaction constant for quenching, may be evaluated.

Another important situation is one for which all the important quenching of a strongly micellized probe occurs *in the aqueous phase*. In this case, the exit of the probe from the micelle may become the rate limiting process for donor decay, i.e., quenching occurs with unit efficiency at sufficiently high (aqueous) quencher concentration [15,25]. The quenching of triplet 1-bromonaphthalene by NO_2^- ions in SDS solution was found to conform to this case [15]. The kinetics for this situation cannot be deduced from our formulation, but may be readily derived from standard steady state considerations. The resulting expression allows evaluation of k_b.

6. Conclusion

The simple model of substrate solubilization by detergent solutions presented here is useful in that it

quantifies this important property in terms of only one constant, that is, the binding constant of the first association step. An understanding of the "solubilization power" of a detergent solution can, therefore, be reduced to a study of such constants under differing conditions. In addition, the model predicts that for a fixed concentration of detergent the quantity of substrate solubilized is limited by its solubility in pure water.

We caution the experimenter to remember that a variety of possible situations will exist in practice and that the main analysis here will be best applied to very hydrophobic probes with relatively short decay times[‡].

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‡ During the course of submission and refereeing of this paper a report has appeared which points out the statistical distribution of quencher molecules to fluorescence quenching experiments. Our results are in complete accord with this publication [26].

References

[1] K.L. Mittal, ed., Micellization, solubilization and microemulsions, Vols. 1 and 2 (Plenum Press, New York, 1977).
[2] J.H. Fendler and E.J. Fendler, Catalysis in micellar and macromolecular systems (Academic Press, New York, 1975).
[3] N.J. Turro et al., in: Micellization, solubilization and microemulsions, Vol. 1, ed. K.L. Mittal (Plenum Press, New York, 1977) pp. 75–86.
[4] N.J. Turro and A. Yekta, J. Am. Chem. Soc. 100 (1978) 5951;
F. Perrin, Compt. Rend. Acad. Sci. (Paris) 178 (1924) 1978.
[5] J.K. Thomas, Accounts Chem. Res. 10 (1977) 133.
[6] P.P. Infelta, M. Grätzel and J.K. Thomas, J. Phys. Chem. 78 (1974) 190.
[7] M. Almgren, Photochem. Photobiol. 15 (1972) 297.
[8] R.C. Dorrance and T.F. Hunter, J. Chem. Soc. Faraday Trans. I 70 (1974) 1572.
[9] U. Khuanga, B. Selinger and R. McDonald, Australian J. Chem. 29 (1976) 1.
[10] B.K. Selinger and A.R. Watkins, Chem. Phys. Letters 56 (1978) 99.
[11] M.A.J. Rodgers and M.F.D. Wheeler, Chem. Phys. Letters 53 (1978) 165.
[12] H.J. Pownall and L.C. Smith, J. Am. Chem. Soc. 95 (1973) 3136.
[13] M. Grätzel and J.K. Thomas, in: Modern fluorescence spectroscopy, Vol. 2, ed. E.L. Wehry (Plenum Press, New York, 1976).
[14] M. Almgren, F. Greiser and J.K. Thomas, J. Am. Chem. Soc. 101 (1979) 279.
[15] J.K. Thomas, F. Greiser and M. Wong, Ber. Bunsenges. Physik. Chem. 82 (1978) 937.
[16] K. Tsujii, J. Sunamoto, F. Nome and J.H. Fendler, J. Phys. Chem. 82 (1978) 423.
[17] K. Kalyanasundaram and J.K. Thomas, J. Am. Chem. Soc. 99 (1977) 2039.
[18] K. Kalyanasundaram, F. Grieser and J.K. Thomas, Chem. Phys. Letters 51 (1977) 501.
[19] N.J. Turro, K.C. Liu, M.F. Chow and P. Lee, Photochem. Photobiol. 27 (1977) 523.
[20] R. Humprey-Baker, Y. Moroi and M. Grätzel, Chem. Phys. Letters 58 (1978) 207.
[21] N.A. Mazer, G.B. Benedek and M.C. Carey, J. Phys. Chem. 80 (1976) 1075.
[22] Y. Waka, K. Hamamoto and N. Mataga, Chem. Phys. Letters 53 (1978) 242.
[23] A. Yekta and M. Aikawa, Columbia University, unpublished results.
[24] M. Tachiya, Chem. Phys. Letters 33 (1975) 289.
[25] F.H. Quina and V.G. Toscano, J. Phys. Chem. 81 (1977) 1750.
[26] P.P. Infelta, Chem. Phys. Letters 61 (1979) 88.