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General Kinetic Equation for Reactions Governed by Dual Florescence Absorption and Emission Spectroscopy

Neji P. A[1]*. & Ayuk A. A[1]

Department of Chemistry University of Cross River State, Calabar, Nigerias
*Corresponding to peterneji@unicross.edu.ng, ambapee2007@yahoo.com

Abstract

Two conformers are assumed to exist in the Franck-Condon excited state species of dual fluorescing molecules. Kinetic equations for the steady state condition of equilibrium of the S^*_A and S^*_B excited state conformers have been derived. The equations for the quantum yield and the quantum yields ratio have also been mathematically formulated. This derivation of the equations for the existence of dual fluorescence conformers following Franck Condon's Theory, has further explained in mathematical terms the basis for the assumptions used in the development of equipment and methods applied in Spectroscopy for dual fluorescent molecules or conformers in spectroscopic detection and measurements of the equilibrium concentrations of the dual fluorescing molecules e.g. the developments of probes (sensors) and ratio metric methods. These mathematical equations also explain the versatility of the ratio metric method in the measurement of the equilibrium concentrations of conformers as shown in the mathematical expressions for the equilibrium concentrations of the steady state quantum yields Φ_A and Φ_B, and the quantum yield ratio Φ_A / Φ_B.

Keywords: Dual fluorescence, conformers, kinetic equations, steady state equilibrium, quantum yields.

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Introduction

The mathematically derived equation in this work enables simplistic understanding of the existence of dual fluorescing conformers or molecules in dual fluorescent absorption and emission spectroscopy. And the assumptions and principles applied in the development of probes (sensors), fluorescent thermometers for thermal imaging dual-view microscopes, and other instruments for detection and measurement of equilibrium concentrations of the fluorescing molecules in solutions Barilaro

et al. (2009). Gösch & Rigler (2004). Other areas of applications of dual fluorescence spectroscopy include, the analysis of molecular noise from the Hz- to the GHz-region, facilitating measurements over a large dynamic range covering photo-physics, conformational transitions and interactions as well as transport properties of fluorescent biomolecules. The mathematical equations further explain the agreement that the fluorescing molecules exist in a ratio at equilibrium. Hence the mathematical expression of the equilibrium yield ratio. The significance and scope of this work is in the simplistic or seamless understanding of dual fluorescent spectroscopic detection and measurement of conformers concentrations at equilibrium mathematically expressed, which further leads to the simplistic understanding of the ratio metric analysis of the fluorescence emission at two different wavelengths, thermal imaging and fluorescence imaging in spectroscopic applications. Which is rarely explained in most publications in this area.

A simple example of the existence of dual fluorescence absorption and emission spectroscopy is that the conformers can be compared or liken to the two engines in an aero-bus in aerodynamics which must perform in equilibrium capacity and release equal power or energy in other to be able to power and stabilize the aero-bus flight. Thus, before any flight the aero mechanics or aeronautic engineers must test the functionality of the two engines and ascertain that they are functioning in equilibrium or synchronized capacity and are capable of powering and stabilizing the flight. Also, in dual fluorescence absorption and emission spectroscopy the existence of the absorbing and radiating molecules must be ascertain and their equilibrium concentration ratios determined on a steady state absorption and emission equilibrium. This ensures the steady state fluorescence absorption and emission of dual molecules. This has been explained and proven mathematically in this work. When a molecule has been irradiated with light, its electrons are promoted from the ground state, S_0 to an excited state of the same multiplicity (Yang *et al.*, 2018; Samuel *et* & Dan. 2019). Promotion of electrons to a higher singlet excited state is rare, though it does occur. The excited molecules do not remain in that state for long. The excited molecules eventually depopulate the excited state to fall back to the ground state in a process termed fluorescence (Yang. *et al.,* 2014; Yang. *et al.*, 2022). In the case of dual fluorescence, the decay follows a pathway as shown in figure 1.

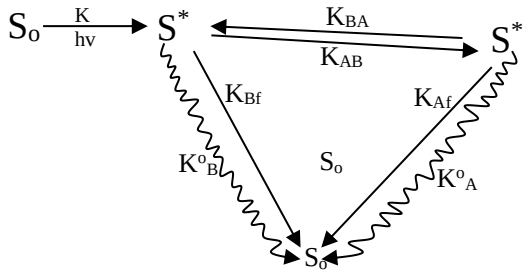

Figure 1 Diagram of the transition equilibrium and S^*_B states

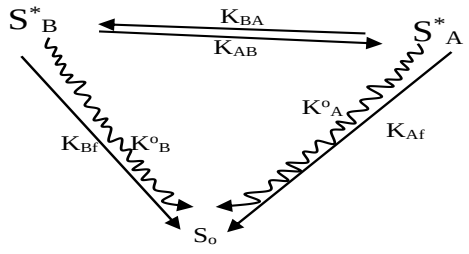

Figure 2 Equilibrium conformers of the S^*_A

At constant irradiation with light energy, hv, the process takes the form of the differential equation

$\frac{dcs_o}{dt} = kcs_o = I_{Abs}$ …equation 1

The term on the left-hand side of the equation represents the change in concentration of the molecules in the ground state, S_o with time; t. 1_{Abs} is the absorbed intensity of the radiation, which is equal to kcs_o the product of the rate constant and the concentration of the ground state, S_o, Also,

$\frac{dcs^*B}{dt} = 1_{ABS} + K_{AB}CS^*A - (K_{BA} + K_{BF} + K_B^0)CS^*{}_B$…equation 2

Here, the term on the left side of the equation represents the rate of change of the $S^X{}_B$ excited state. The $S^*{}_B$ state is one of the exciting singlet states formed. The term in parenthesis on the right-hand side of equation 2 represents the sum of the rate constant on both the radioactive and non-radiative processes involved in the depopulation of the $S^*{}_B$ state. Similarly,

$\frac{dcs^*A}{dt} = (1_{Abs}) + K_{BA}CS^*{}_B - (K_{AB} + K_{Af} + K^o{}_A)CS^*{}_A$…equation 3

Considering the steady state condition of the equilibrium conformers of $S^*{}_A$ and $S^*{}_B$ states as shown in figure 2:

$\frac{dcs^*{}_A}{dt} = I_{ABS} + KAB\ CS^*{}_B - (K_{AB} + K_{AF} + K_A^0)CS^*{}_A$ … equation 4

At constant light intensity, stationary state concentration of the $S^*{}_A$ and $S^*{}_B$ is rapidly attained and the sum of the change in concentration is equal to zero.

That is: $\frac{dcs^*{}_B}{dt} = 0 = \frac{dcs^*{}_A}{dt}$

Therefore, $\frac{dcs^*{}_A}{dt} = \frac{dcs^*{}_B}{dt} = K_{BA}CS^*{}_B - (K_{AB} + K_{Af} + K^*{}_A)CS^*{}_A = 0$…equation 5

It therefore follows from equation 5 that we get equation 6

$K_{BA}CS^*{}_B = (K_{AB} + K_{Af} + K^o{}_A)CS^*{}_A$…equation 6a.

And by rearranging, we get equation 6b.

i.e $CS^*{}_BK_{BA} = (K_{AB} + K_{Af} + K^o{}_A)\ CS^*{}_A$ and $CS^*{}_A\ K_{AB} = (K_{BA} + K_{Bf} + K^O{}_{B)}\ CS^*{}_B$… equation 6b

It also implies that: $CS^*{}_AK_{AB} = (K_{BA} + K_{Bf} + K^O{}_B)\ CS^*{}_B$ and $CS^*{}_B\ K_{BA} = (K_{AB} + K_{Af} + K^O{}_A)$ …. Equation 6c.

Note that it is from equation 6b and 6c that we obtained equation 7a and 7b, respectively.

i.e. $\frac{CS^*{}_A}{CS^*{}_B} = \frac{K_{BA}}{K_{AB} + K_{Af} + (K^oA)}$ … equation 7a

and $\frac{CS^*{}_B}{CS^*{}_A} = \frac{K_{AB}}{K_{BA} + K_{Bf} + (K^oB)}$ … equation 7b

From the law of mass action, the equilibrium constant, K_c can be obtained from the equilibrium reaction as depicted in the excited states in Figure 2 and written as: $K_c = \frac{CS^*{}_A}{CS^*{}_B} = \frac{K_{BA}}{(K_{AB} + K_{Af} + K^O{}_A)}$ …equation 8a

Or $K_c = \frac{CS^*{}_B}{CS^*{}_A} = \frac{K_{AB}}{K_{BA} + K_{Bf} + K^O{}_B}$ … equation 8b

The interesting part of this kinetic is the comparison of the measurable quantum yields of the excited states and the various rate constants as indicated in equation 8.

Determination of the quantum yields $\phi_{A\ and\ \phi_B}$ for the respective $S^*{}_A$ and $S^*{}_B$ states. The quantum yield for the $S^*{}_B$ State is the ratio of the number of molecules depopulating the $S^*{}_B$ to the number of photons absorbed. This involves spectrophotometrically measurable quantities of absorption and emission.

i.e; $\phi_B = \frac{K_{BF}\ CS^*{}_B}{KCS_O}$ … Equation 9

From equation 5,

$\frac{dcs^*{}_B}{df} = 0$ … equation 10

Therefore, combining equation 1 and 4 and rationalizing we get the expression for KCS_o

$KCS_o + K_{AB}CS^*_A - (K_{BA} + K_{Bf} + K^O_B)\ CS^*_B$ … equation 11

We get equation12 to obtain the expression of KCSo

$KCS_o = (K_{BA} + K_{Bf} + K^O_B)\ CS^*_B - K_{AB}CS^*A$ …equation 12

$KCS_o = (K_{AB} + K_{Af} + K^o_A)\ CS^*_A - K_{BA}CS^*_B$ …equation 12b

Therefore, substituting for KCS_o in equation 9 we get equation 13

$$\phi_B = \frac{K_{Bf}CS^*_B}{(K_{BA}+K_{Bf}+K^O_B)CS^*_B - K_{AB}CS^*_A} \text{…equation 13}$$

But from eqution 8,

$CS^*_A = K_cCS^*_B$

Substituting for CS^*_A in equation 13 we get 14.

Therefore,

$$\phi_B = \frac{K_{Bf}CS^*_B}{(K_{BA}+K_{Bf}+K^O_B)CS^O_B - K_{AB}CS^*_BK_c} \text{…equation 14}$$

Substituting for K_c in equation 14 we get equation 15,

Recall, $K_c = \frac{K_{BA}}{K_{AB}+K_{Af}+K^O_A}$ …From equation 8

$$= \frac{K_{Bf}CS^*_B}{(K_{BA}+K_{Bf}+K^O_B)CS^*_B - K_{AB}\ CS^*\ B\frac{K_{BA}}{K_{AB}+K_{AF}+K^O_A}}$$

…equation 15a

$$= \frac{K_{Bf}CS^*_B}{(K_{BA}+K_{Bf}+K^O_A)CS^*_B - \frac{K_{BA}\ CS^*_B\ K_{BA}}{K_{AB}+K_{AF}+K^O_A}}$$

…equation 15b

Recall

$CS^*_B = K_{AB} + K_{AF} + K^o_A$, from equation 6b

$CS^*_A = K_{BA} + K_{BF} + K^o_B$, from equation 6c

Substituting the above from equation 6b in equation 15b and rationalized, we get 16

$$\phi_B = \frac{K_{Bf}\ (K_{AB}+K_{Af}+K^O_A)}{(K_{BA}+K_{Bf}+K^O_B)(K_{AB}+K_{Af}+K^O_A) - K_{AB}\ K_{BA}}$$

…equation 16

Expanding equation 16 to eliminate – K_{AB} K_{BA} and get equation 17

$$\phi_B = \frac{K_{Bf}\ (K_{AB}+K_{Af}+K^O_A)}{K_{BA}K_{AB}+K_{BA}+K_{Af}+K_{BA}\ K^O_A+K_{Bf}K_{AB}+K_{Bf}K_{Af}+K_{Bf}K^O_A+K^O_BK_{AB}+K^O_BK_{Af}K^O_AK^O_B-K_{AB}K_{BA}} \text{ equation 17}$$

Rationalizing equation 17 to get 17b

$$\phi_B = \frac{K_{Bf}\ (K_{AB}+K_{Af}+K^O_A)}{K_{BA}K_{Af}+K_{BA}K^O_A+K_{Bf}K_{AB}+K_{Bf}K_{Af}+K_{Bf}K^O_A+K^O_BK_{AB}+K^O_BK_{Af}K^O_BK^O_A+K^O_BK_{AB}+K^O_BK_{Af}+K^O_BK^O_A} \text{ ….}$$

equation 17b

Factorizing equation 17b to get equation 17c

$$\phi_B = \frac{K_{Bf}\ (K_{AB}+K_{Af}+K^O_A)}{K_{Af}\ (K_{BA}+K_{Bf}+K^O_B)\ K^O_A\ (K_{BA}+K_{Bf}+K^O_B\ )K_{AB}(K_{Bf}+K^O_B)} \text{ … equation 17c}$$

Further factorizing equation 17c to get equation 18

$$\phi_B = \frac{K_{Bf}\ (K_{AB}+K_{Af}+K^O_A)}{(K_{AB}+K_{Af}+K^O_A)\ (K_{BA}+K_{Bf}+K^O_B\ )(K_{Bf}+K^O_B)} \text{…equation 18}$$

But $K'_A = (K_{Af} + K^O_A) and\ K'_B = (K_{Bf} + K^O_B)$

Substituting in equation 18 to get equation 19

$$\phi_B = \frac{K_{Bf}(K_{AB}+K'_A)}{(K_{AB}+K'_A)\ (K_{AB}+K'_B)K'_B} \text{ …equation 19}$$

Rearranging equation 19 to get equation 19b

$$\phi_B = \frac{K_{Bf}(K_{AB}+K'_A)}{K'_B(K_{AB}+K'_A)\ (K_{BA}+K'_B)} \text{ …equation 19b}$$

The quantum yield of S^*_A state can also be defined as the ratio of the number of molecules depopulating the S^*_A state to the number of photons absorbed. This also involves the spectrophotometrically measurable quantities of absorption and emission.

Thus; $\phi_A = \frac{K_{Af} CS^*_A}{(K_{BA} + K_{Bf} + K^0_B) CS^*_B - K_{AB} CS^*_A}$ …equation 20, similar to equation 13

Also call

$Kc = \frac{CS^*_A}{CS^*_B}$ and $CS^*_B = \frac{CS^*_A}{Kc}$ while $CS^*_A =$ $KcCS^*_B$ …equation 21

Also $Kc = \frac{CS^*_A}{CS^*_B} = \frac{K_{BA}}{K_{AB} + K_{AF} + K^O_A}$ …from equation 8

Substituting for CS^*_B in equation 20 we get equation 22

$$\Phi_A = \frac{K_{Af} CS^*_A}{(K_{BA} + K_{Bf} + K^O_B)\frac{CS^*_A}{KC} - K_{AB} CS^*_A}$$

…equation 22

$$\Phi_A = \frac{K_{Af} CS^*_A K_c}{(K_{BA} + K_{Bf} + K^O_B) CS^*_A - K_{AB} CS^*_A}$$

…equation 22b

Substituting $\frac{K_{BA}}{K_{BA} + K_{Bf} K^O_B}$ for Kc in equation 22b and dividing top and bottom by CS^*_A

$$\Phi_A = \frac{K_{Af}\, CS^*_A K_{AB}}{\frac{K_{BA} + K_{Bf} + K^O_B}{(K_{BA} + K_{Bf} + K^O_B) CS^*_A - K_{AB} CS^*_A}}$$ …equation 23

$$\Phi_A = \frac{K_{Af} K_{AB}}{\frac{K_{BA} + K_{Bf} + K^O_A}{K_{AB} + K_{Bf} + K^O_A - K_{AB} CS^*_A}}$$ … equation 23b

Dividing top and bottom by CS^*_A, of equation 23

Substituting Kc CS^*_B for CS^*_A in equation 23b we get 24

$$\Phi_A = \frac{K_{Af} K_{BA}}{\frac{K_{AB} + K_{Af} + K^O_A}{K_{BA} + K_{Bf} + K^O_B - K_{AB}\, CS^*_B\, K_c}}$$ …equation 24

Substituting for Kc in equation 24 to get equation 25

$$\Phi_A = \frac{K_{Af}\, K_{BA}}{\frac{K_{AB} + K_{Af} + K^O_A}{K_{BA} + K_{Bf} + K^O_B - \frac{K_{AB} CS^*_B K_{BA}}{K_{AB} + K_{AF} + K^O_A}}}$$ … equation 25

Recalling $CS^*_B = (K_{AB} + K_{AF} + K^O_A)$ and rationalizing to eliminate CS^*_B

$$\Phi_A = \frac{K_{Af}\, K_{BA}}{\frac{K_{AB} + K_{Af} + K^O_A}{K_{BA} + K_{Bf} + K^O_B - K_{AB} K_{BA}}}$$ … equation 26

Multiply top and bottom of equation 26 by $K_{AB} + K_{Af} + K^O_A$

$$\Phi_A = \frac{K_{Af}\, K_{BA}\, (K_{AB} + K_{Af} + K^O_A)}{\frac{K_{AB} + K_{Af} + K^O_A}{K_{BA} + K_{Bf} + K^O_B\, (K_{AB} + K_{Af} + K^O_A) - K_{AB} K_{BA}}}$$

$$\Phi_A = \frac{K_{Af}\, K_{BA}}{(K_{BA} + K_{Bf} + K^O_B)\, (K_{AB} + K_{AF} + K^O_A) - K_{AB} K_{BA}}$$ … equation 26b

Expanding the denominator of equation 26 we get 27

$$\Phi_A = \frac{K_{Af} K_{BA}}{K_{BA} K_{AB} + K_{BA} K_{Af} + K_{BA}\, K^O_A + K_{Bf} + K_{AB} + K_{Bf} K_{Af} + K_{Bf} + K^O_A + K^O_B K_{AB} + K^O_B + K_{AB} + K^O_B K_{Af} + K^O_B + K^O_A - K_{BA} K_{AB}}$$ equation 27

Rationalizing or eliminating $K_{AB} K_{BA}$ in equation 27 we get equation 28

$$\Phi_A = \frac{K_{Af} K_{BA}}{K_{BA} K_{AF} + K_{BA} K^O_A + K_{Bf} + K_{AB} + K_{Bf} K_{Af} + K_{Bf} + K^O_A + K^O_B K_{AB} + K^O_B K_{AB} + K^O_B K_{Af} + K^O_B + K^O_A}$$ equation 28

Factorizing equation 28 we get equation 29

$$\Phi_A = \frac{K_{Af} K_{BA}}{K_{Af}(K_{BA} + K_{Af} + K^O_A) K^O_A (K_{BA} + K_{Bf} + K^O_B)\, K_{AB} (K_{Bf} + K^O_B)}$$ …equation 29

Further factorizing equation 29 to get equation 29b

$$\Phi_A = \frac{K_{Af} K_{BA}}{(K_{AB} + K_{Af} + K^O_A)(K_{BA} + K_{Bf} + K^O_B)\ (K_{Bf} + K^O_B)}$$ … equation 29b

But $K'_A = K_{Af} + K^O_A$ and $K'_B = K_{Bf} + K^O_B$ and substituting in equation 29b to get equation 30

$\Phi_A = \frac{K_{Af}K_{BA}}{(K_{AB}+K'_A)(K_{BA}+K'_B)K'_B}$ …equation 30

$\Phi_A = \frac{K_{Af}K_{BA}}{K'_B(K_{AB}+K'_A)(K_{BA}+K'_B)}$… equation 30b

From equation 19 and equation 30 which are the quantum yields i.e Φ_A and Φ_B and taking the ratio of the two quantum yields or dividing Φ_A / Φ_B

Hence: $\Phi_A = \frac{K_{Af}K_{BA}}{(K_{AB}+K'_A)(K_{BA}+K'_B)K'_B}$

$\Phi_B = \frac{K_{Bf}(K_{AB}+K'_A)}{K'_B(K_{BA}+K'_B)(K_{AB}+K'_A)}$

Dividing Φ_A / Φ_B

$\Phi_A / \Phi_B = \frac{\frac{K_{Af}K_{BA}}{K'_B(K_{AB}+K'_A)(K_{BA}+K'_B)}}{\frac{K_{Bf}(K_{AB}+K'_A)}{K'_B(K_{AB}+K'_B)(K_{AB}+K'_A)}}$

Therefore, rationalizing $\Phi_A / \Phi_B = \frac{K_{Af}K_{BA}}{K_{Bf}(K_{AB}+K'_A)}$

Discussion

The quantum yields Φ_A and Φ_B have been obtained purely from the rate constants. They can be calculated simply using the mathematical procedure, the Simpson rule. The area of the emission spectrum divided by the area of the absorption spectrum is the quantum yield. The ratio of Φ_A / Φ_B can only be obtained from dual fluorescing molecules, as they are molecules that dual fluoresce. Dual fluorescence has been studied by various authors (Lakowicz 2006; Thomas *et al.* 2017; Chrayteh *et al.,* 2020). Φ_A, is calculated from the S_A state which produces the S^*_A conformer and Φ_B from the S_B state which also produces the S^*_B – conformer. At room, temperature only the S^*_B fluoresces, as the temperature is lowered the population of the S^*_A state takes over up to a point where the population of the S^*_B state disappears or is no longer significant (Sanggeun *et al.,* 20 19; Trojanowiczm, 2020: Tomin *et al.*, 2015). The temperature dependence of the S_A and S_B states is what gives rise to S^*_A and S^*_B fluorescence bands. This can be seen in the works of Lippert for molecules of benzene derivative (Diego Frezzato, 2020; Diego, 2021) and for the naphthalene derivative (Delgadillo *et al.,* 2019). and Sun et al 2020) The ground state of each of these molecules is excited first to the S^*_B state usually at room temperature and with a decrease in temperature, the S^*_A state is formed and the molecule now fluoresces additionally from these states, see Figure 1 and 2. The rate constants K_{AB}, K_{BA} and the radiative rate constants K_{Bf} and K_{Af} are the ones used in the determination of the quantum yields of these fluorescent states, S^*_A and S^*_B. The two states are in equilibrium as depicted below:

$$S^*_B \underset{K_{AB}}{\overset{K_{BA}}{\rightleftharpoons}} S^*A$$

At room temperature, the S_B state only is in existence. As the temperature is lowered both states exist, until at a certain very low temperature the S^*_A state only exists. Fluorescence now takes place either from the S^*_B State at room temperature or the S^*_A state at very low temperature. But for fluorescence to occur, the S^*_B and the S^*_A states must be populated, as the temperature decreases the rate constant K_{AB} also increases.

K_{BA} on the other hand decreases and almost no longer takes place as the S^*_B fluorescence completely disappears. The appearance of the fluorescence band is mainly determined by the radiative rate constant K_{Bf} for the S_B state and K_{Af} for

the S_A state. We can therefore see that quantum yields can be determined from the equilibrium consideration of the S^*_B and S^*_A states.

Conclusion

The radiative fluorescence of S^*_B and the S^*_A states in equilibrium positively give rise to the Φ_A and Φ_B quantum yields which were mathematically derived with only the equilibrium and radiative rate constants.

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Conflict of Interest

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