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Research Article

Energy transfer between egg albumin and amantadine a fluoresence bioflavonoid

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Abstract
Flavonoids occupy an important position in chemistry and pharmacology. Various falvonoids, particularly amantadine have potential to form molecular complexes with nucleic acid structure and have attracted recent attention for their prospective clinical and pharmacological utility. Absorption spectrum of Egg albumin and fluorescence spectrum of amantadine in SPAN 40 solution have been recorded and discussed in detail.

Keywords: Energy transfer, Egg albumin, amantadine, SPAN 40.

Introduction:
Flavonoids possess pronounced biological activities against coronary heart disease, cancer, inflammation (1-3) etc. Several studies have been performed on the reaction of flavonoids with active free radicals [N_3, Ho, etc.] by pulse radiolysis (4,5) and high performance liquid chromatography (HPLC) (6,7).

Flavonoids exert their protective effective to different extents with different efficiencies depending upon the structures of the molecules. Recently O-H bond dissociation enthalpies (BDEs) have been successfully used to express the free radical scavenging ability of Polyphenolic antioxdiants (8,9).
Fluorescence as a detection method provides high sensitivity down to the single molecule, ease of application, and a subnanosecond temporal and submicrometer spatial resolution. A high reactivity combined with selectivity toward antioxidant the possibility to generate radical mimicking probes in situ., by an instantaneous generation and a sensitive detection method are other essential prerequisites (10).

Materials and Methods
Egg albumin, SPAN 40, and amantadine were purchased from sigma Aldrich company, Banglore and were used without further purification. UV/Vis., absorption spectra were taken using Shimadzu 1650 P4 UV-Visible Spectrophotometer and fluorescence measurements were made by Shimadzu RF 5301 PC Spectrofluorophotometer.

Results and Discussion
Fluorescence resonance energy transfer occurs when the emission spectrum of the donor overlaps the absorption spectrum of the acceptor. The dependence of the energy transfer rate on the interaction distance has been widely used to measure the distance between the donor and the acceptor.
Generally, the maximum distance is in the range of 1-10nm (11). According to Forster non-radiation energy transfer theory (11,12), energy transfer is related not only to the distance between the acceptor and donor, but also to the critical energy transfer distance, (R_0)

$$E = \left(\frac{R_0^6}{R_0^6 + r^6} \right) \qquad (1)$$

where R_0 is the critical transfer distance when the transfer efficiency is 50% and r, the mean distance between the centres of the donor and acceptor dipoles. The donor and acceptor here a EA and amantadine respectively. E is the energy transfer efficiency calculated with equation (2)

$$E = \left(1 - \frac{I}{I_0} \right) \qquad (2)$$

where I and I_0 are the fluorescence intensity of EA

with and without amantadine respectively. R_0 can be given by,

$$R_0^6 = 8.8 \times 10^{-25} K^2 N^{-4} \phi J \qquad (3)$$

where K^2 is the spatial orientation factor of the dipole, N, the refractive index of the medium, ϕ the fluorescence quantum of yield of the donor in the absence of the acceptors, J expresses the overlap integral of the fluorescence emission spectrum of the donor and the absorption spectrum of the acceptor. J is given by,

$$J = \frac{\Sigma F(\lambda)\ \varepsilon(\lambda)\lambda^4 \Delta\lambda}{\Sigma F(\lambda)\Delta\lambda} \qquad (4)$$

where F (λ) is the fluorescence intensity of donor at wavelength range λ and Σ(λ), the molar absorption coefficient of the acceptor at wavelength λ with unit of $Cm^{-1}mol^{-1}$. In this work, J was obtained as 8.32×10^{-14} Cm^3 M^{-1} integrating the overlap of the UV absorption spectrum of amantadine and the fluorescence emission spectrum of EA.

The calculated values are compiled in Table 1 and the overlap spectrum is shown in Fig.1.

Fig. 1: The overlap of UV absorption of Amantadine (solid line) with the fluorescence Emission Spectrum of EA (Dotted line) with SPAN 40

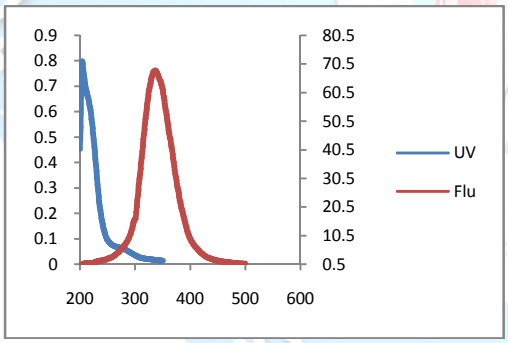

Table-1 Efficiency transfer energy (E), Critical energy transfer distance (R_0) of EA with Amantadine

Quencher	Energy	R_0 Å	J (cm^3M^-	rÅ
Amantadine	0.3173	22.03	2.73X10$^-$	25.04

Conclusion

The distance between the donor and acceptor has been calculated according to Forster's theory of dipole-dipole energy transfer. The result indicates that the energy transfer from EA to amantadine occurs with high probability, for r is in the scope of 2-8, it implies that the energy transfer from EA to amantadine with high possibilities.

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