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Characterization of the interaction between two Anti-viral drugs and Egg albumin

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ABSTRACT

The binding of two anti-viral drugs (1. Quercetin [Q] and 2. Amantadine [A]) to Egg albumin [EA] was investigated by fluorescence spectroscopy under simulation of physiological conditions. The quenching mechanism was suggested according to the fluorescence measurement. The Stern-Volmer quenching constants were determined. In addition, binding constants were also calculated at room temperature.

Key words: Quercetin, Amantadine, Egg albumin (EA), Fluorescence Spectroscopy.

INTRODUCTION

Flavonoids occupy an important position in chemistry and pharmacology. Flavonoids are a group of polyphenolic compounds extensively distributed in the medicinal plants, vegetables, fruit juices and a variety of beverages [tea, coffee, wines and fruit drinks]. Flavonoids particularly quercetin derivatives, have received more attention as dietary constituents during the last few years [1]. Many studies showed that flavonoids have a wide range of biological activities, such as anticancer, antiviral, antibacterial, antioxidants and anti-inflammatory effects [2-5].

Flavonoids are best known as radical scavengers. While these valuable effects generally are due to their abilities to accept free radicals, complexation properties with metal ions have also been recognized to contribute to the total biological activity [1].

Quercetin is known to a complex with various metal cations to form stable compounds, which have demonstrable antibacterial properties and anti-tumour activity [6-8].

Amantadine is an antiviral drug that has been used to treat influenza and Parkinson disease [9-14]. Amantadine (1-aminoadamantine) is used against infection with influenza type A virus and to ameliorate symptoms when administered during the early stages of infection [12], as well as in the management of herpes zoster [9]. Amantadine is usually given by mouth as the hydrochloride salt [9].

Albumin is a class of simple, water-soluble proteins that are found in egg white, blood serum, milk, and many other animal and plant fluids and tissues. Albumin has been used as the subject of many investigations because of its important roles in maintaining normal biochemical functions.

MATERIALS AND METHOD

Egg Albumin, Amantadine and Quercetin were purchased from Sigma Aldrich Company, Bangalore. Steady-state fluorescence spectra were taken CARRY ECLIPSE VARIAN FLUORESCENCE SPECTROPHOTOMETER.

RESULTS AND DISCUSSION

The steady-state fluorescence spectra of Egg Albumin with different concentrations of Amantadine and Quercetin are shown in figs. 1&2 respectively. From the figures it can be noted that the fluorescence intensities decrease when the concentration of Amantadine and Quercetin increase. The fluorescence maximum peak occurs at 337 nm.

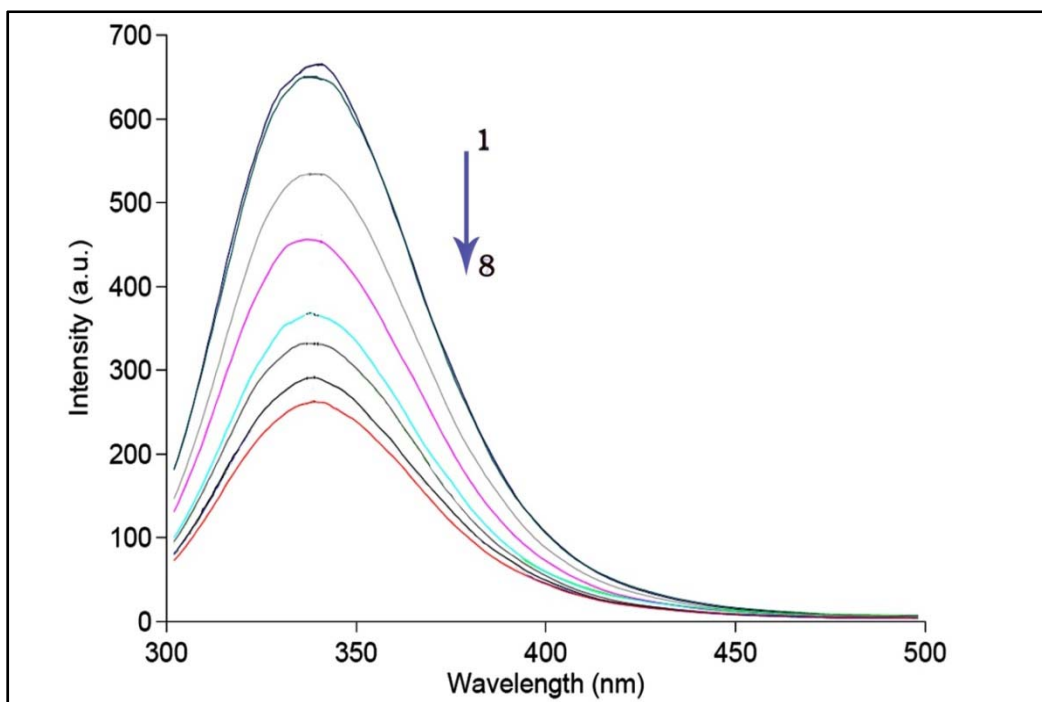


Fig.1. Steady state fluorescence spectra of Egg Albumin with different concentration of Amantadine mol dm⁻¹ (1. (0), 2. (1), 3.(2), 4.(3), 5.(4), 6.(5), 7.(6), 8. (7))

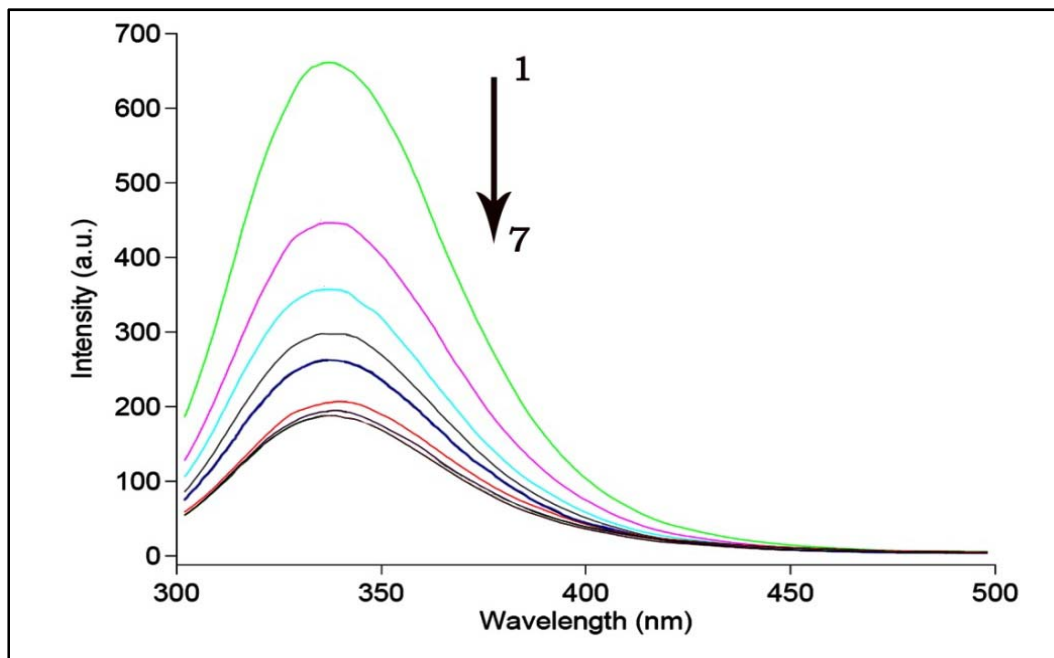


Fig.2. Steady state fluorescence spectra of Egg Albumin with different concentration of Quercetin mol dm⁻¹(1. (0), 2.(1), 3.(2), 4.(3), 5.(4), 6.(5), 7.(6), 8. (7))

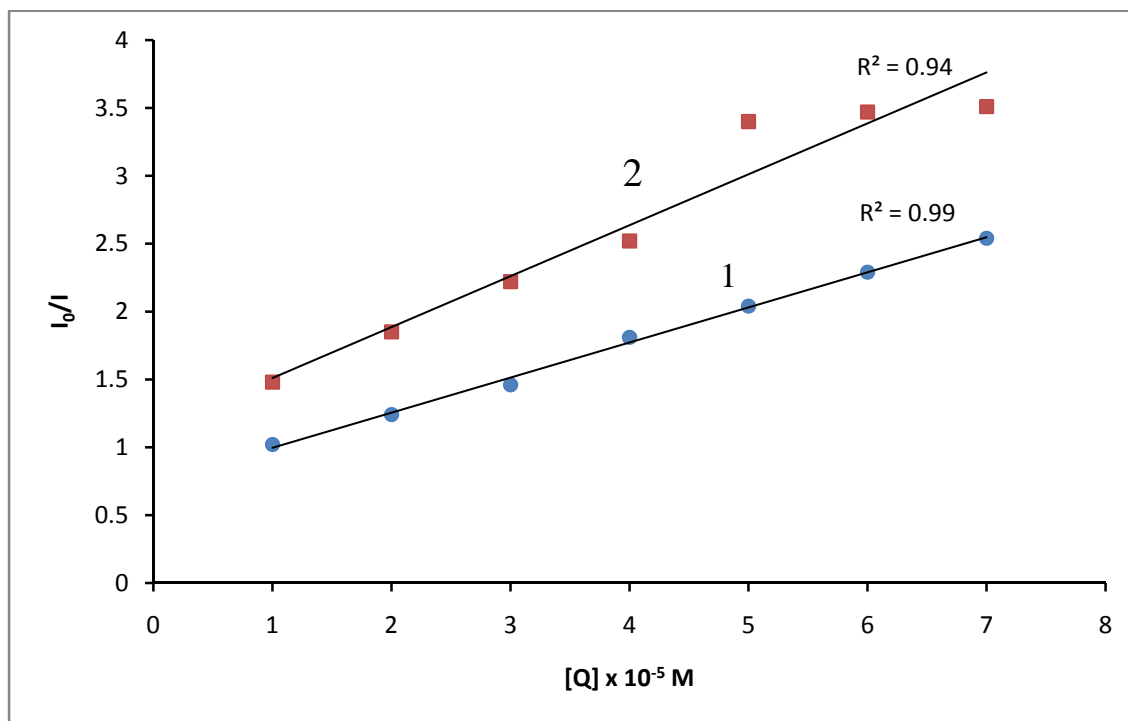


Fig.3 Stern-Volmer plot of I_0/I vs $[Q]$ with Egg Albumin with two different quenchers (1. Amantadine, 2. Quercetin)

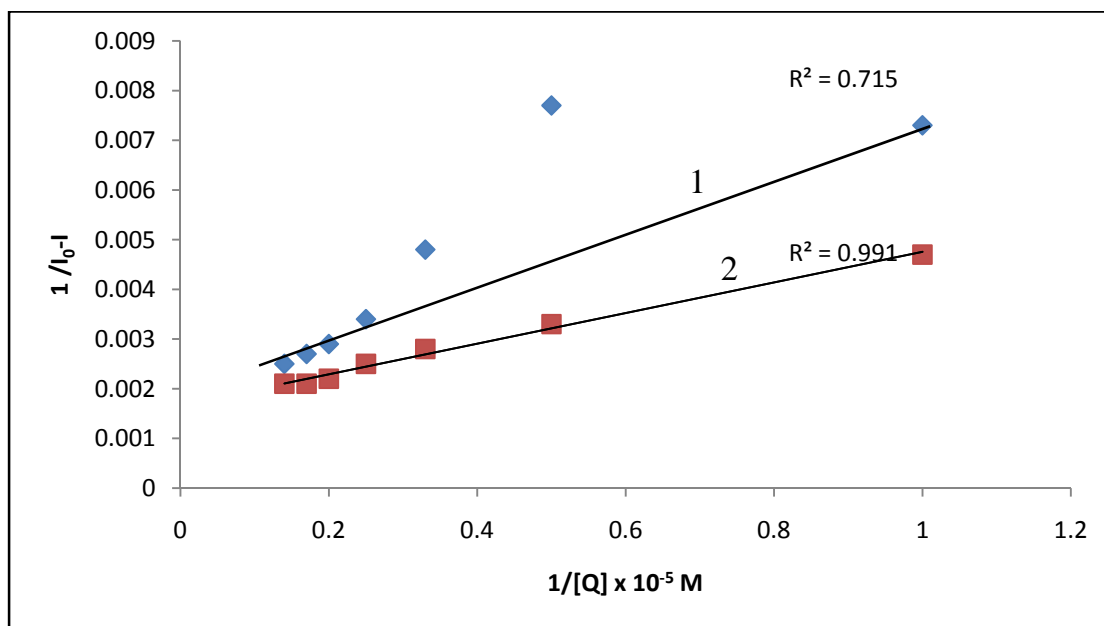


Fig.4 Plot of $\frac{1}{I_0 - I}$ Vs $\frac{1}{[Q]}$ Egg albumin with two different quencher(1.Amantadine, 2. Quercetin)

Fig.3 shows the Stern-Volmer plot $[I_0/I \text{ versus } \frac{1}{[Q]}]$ of Egg Albumin with two different quenchers Amantadine and Quercetin. Stern-Volmer constant, regression coefficient, quenching rate constant and standard Deviation have also been calculated and are presented in table 2.

Fig.4 shows the plot $[\frac{1}{I_0 - I} \text{ versus } \frac{1}{[Q]}]$ of Egg Albumin with two different quenchers Amantadine and Quercetin. From this plot the excited state formation constants (K_e) have been calculated. From these calculated values, the change in free energy (ΔG_e) values have also been calculated and are presented in table 3.

CONCLUSION

In this paper, the interaction of the antiviral drugs (Quercetin and Amantadine) with EA has been investigated by fluorescence spectroscopic technique under simulated physiological conditions. This study shows that Quercetin and Amantadine binds to Egg albumin and quenches the intrinsic fluorescence of Egg albumin efficiently. Binding constants are evaluated and tabulated.

Table. 1 Energy, Ionization potential, electron affinity, molar extinction, coefficient, solvent parameter and stroke's shift

Quenchers	λ_{abs}	$\lambda_{\text{emission}}$	Energy (ev)	ID (ev)	EA (ev)	Log $\epsilon M^{-1} \text{cm}^{-1}$	Z (nm)	Stoke's shift
Amantadine								
Quercetin	280	337	4.44	10.79	-0.34	-1.55	1.02×10^{11}	6.41×10^{12}

No change was observed for λ_{abs} and λ_{emi} by the use of Quercetin and Amantadine. So, same values were obtained for these constants.

Table. 2 Stern-Volmer constant, regression coefficient, quenching rate constant and standard Deviation

Quenchers	K_{sv}	R^2	K_q	S.D
Amantadine	0.35	0.99	0.07	0.60
Quercetin	0.40	0.94	0.08	0.84

Table.3 Excited state Formation Constants (K_e) and the Change in Free Energy (ΔG_e).

Quenchers	K_e	ΔG_e
Amantadine	0.13	5139.05
Quercetin	0.03	8833.62

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