

## Solvent dependence of the photochemistry of the voltage-sensitive fluorescent membrane probe RH421

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Voltage-sensitive fluorescent styryl dyes, such as RH421 and di-8-ANEPPS, have attracted great interest as a means of optical imaging of electrical field strength within living cells, visualizing electrical transients in neurons and for the investigation of the reaction mechanisms of ion pumps, for example, the Na<sup>+</sup>,K<sup>+</sup>-ATPase. In spite of the significant advances that they have no doubt allowed, they do have some drawbacks. The most notable ones are their phototoxicity and their photochemical instability. In order to overcome these drawbacks and synthesize improved dyes, fundamental studies of their photochemical behaviour are required. This is the purpose of our study.

On illumination of solutions of RH421 with 577 nm light from a mercury lamp we have found that the dye undergoes two separate photochemical reactions, one producing a fluorescence increase and the other producing a fluorescence decrease. The amplitude of the fluorescence decrease is very dependent on the polarity of the environment, becoming increasingly less pronounced as the solvent polarity decreases and almost disappearing when the dye is bound to a lipid membrane. The fluorescence increase, on the other hand, is virtually independent of solvent polarity. This reaction has been attributed to a single-photon excited state photoisomerisation of the dye (Amoroso *et al.*, 2006). The origin of the fluorescence decrease is unclear at this stage, but the fact that it is more prominent in polar solvents such as water and that its amplitude increases with time following addition to water suggest that it may be related to dye aggregation.

Amoroso S, Agon VV, Starke-Peterkovic T, McLeod MD, Apell H-J, Sebban P & Clarke RJ. (2006) *Photochemistry Photobiology*, **82**: 495-502.