TWO-PHOTON FLUORESCENCE EXCITATION IN FLUORESCEIN

D. S. Corrêa, S. C. Zilio, L. Misoguti, C. R. Mendonça

Instituto de Física de São Carlos - USP - São Carlos, SP, Brazil

Abstract

We report the implementation of the two-photon fluorescence excitation technique to study the two-photon absorption cross-section in organic materials. This method was applied to a fluorescein sample, which is a calibration material for this kind of experiment.

Experimental Setup

Two-photon excitation is created when, by focussing an intense light source, the density of photons per unit of volume and per unit of time becomes high enough for two photons to be absorbed simultaneously into a chromophore. In this work we report the implementation of a technique to study a two-photon fluorescence excitation process using femtosecond pulses.

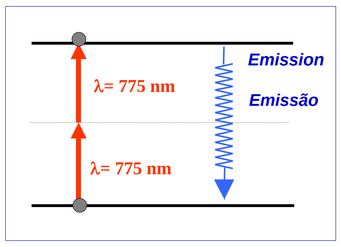


Figure 1 – Illustration of the two-photon excitation and subsequent emission.

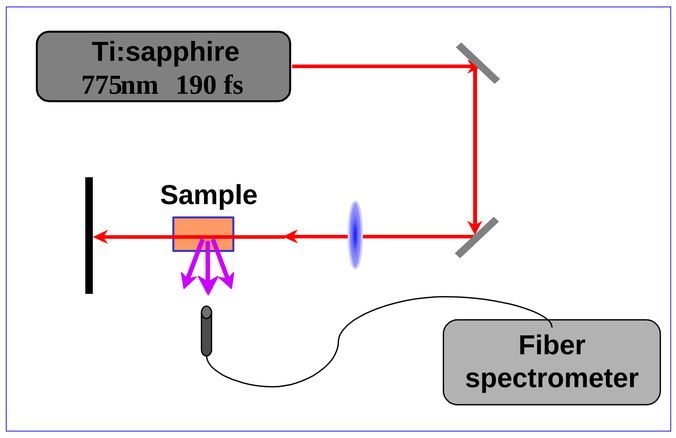
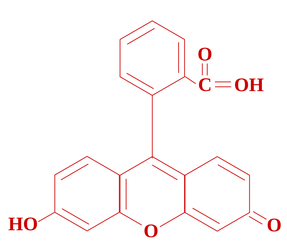


Figure 2 – Two photon excitation fluorescence experimental setup.

Sample Studied

A fluorescein methanol solution was used as a testing material for the two-photon fluorescence excitation (2PFE) method, since it has well known two-photon cross section.



- The fluorescein solution present an absorption peak around 490 nm, being completely transparent at 775 nm, the wavelength used in the two-photon experiment, as shown Fig.4.

- Figure 5 shows the fluorescence spectrum for the fluorescein solution when two-photon excitation was used (excitation at 775 nm), obtained with a pump irradiance of 5 GW/cm^2. As expected, this spectrum is identical to the one found in the literature for excitation at 388 nm.

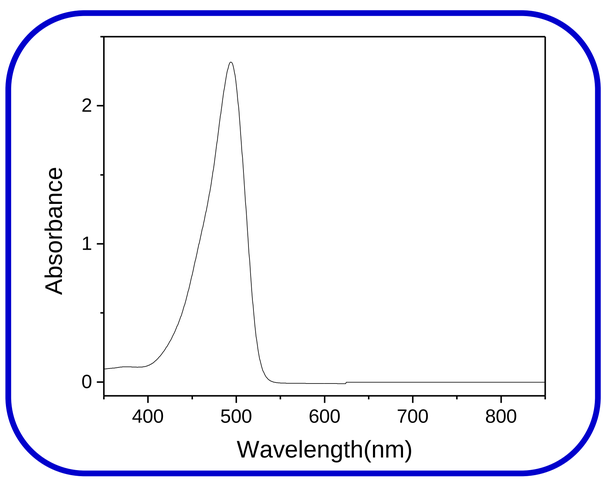


Figure 4– Absorbance spectrum of fluorescein methanol solution.

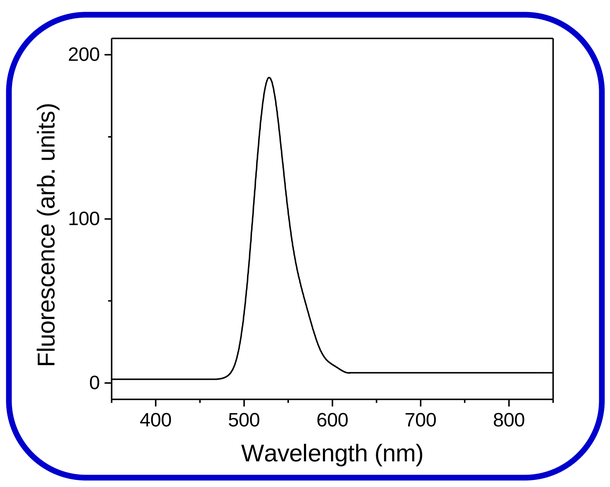


Figure 5– Two-photon fluorescence spectrum in the fluorescein methanol solution, for excitation at 775 nm.

Results

Two-photon fluorescence excitation measurements, similar to the one showed in Fig. 2, were carried out for different pump beam irradiances, in order to study the emitted light dependence on the excitation intensity. Figure 3 shows that the fluorescence signal exhibit a quadratic dependence on the irradiance, as expected for pure 2PFE processes. The solid line in this figure represents the theoretical fitting obtained with a quadratic polynomial function.

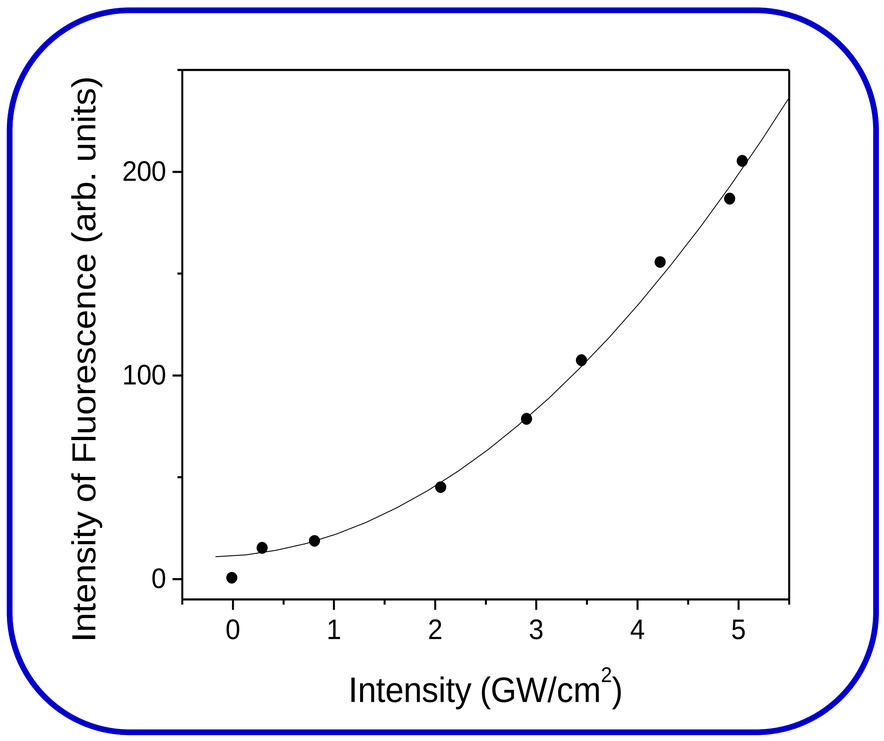


Figure 3– Two-photon fluorescence spectrum, for excitation at 775 nm, in the fluorescein methanol solution.

Conclusion

We have implemented the 2PFE experimental setup to investigate the two-photon absorption cross-section in organic materials. This technique was applied to a fluorescein methanol solution, that is a standard material for this type of experiment. The obtained results are in agreement with those presented in the literature, showing the reliability of this method for future works.

Acknowledgements

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