

13. Curricular Unit

Advanced Physics Topics 1

Module

Photophysical Techniques in Biophysics (PTB)

Type

Lecture course

Contact hours

18

Professor/Researcher in charge

Paulo José Gomes Coutinho

Summary of Contents

Molecular photophysics

Concepts of molecular physics : rotational, vibrational and electronic structure

Types of molecular orbitals and electronic transitions

UV-Vis absorption spectra : selection rules, Franck-Condon factors

UV-Vis emission spectra: radiative and non-radiative processes, Jablonski diagram, Kasha and Stokes rules, Franck-Condon factors

Molecule solvent interactions and its effect on absorption/fluorescence spectra: orientational polarizability and Lippert-Mataga equations

Molecular aggregation and its effect on absorption/fluorescence spectra: H and J aggregates

Standard Photophysical Techniques

Absorption spectroscopy

Circular and linear dichroism

Fluorescence spectroscopy

Time-resolved fluorescence decays and spectra

Steady state and time-resolved fluorescence anisotropy

Time-resolved transient absorption spectroscopy

Excited state dynamics

Kinetics of photophysical processes: steady state and transient state, fluorescence lifetimes



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and quantum yield, the Strickler-Berg relation

Fluorescence quenching photophysical/photochemical processes: excimers and exciplexes, photoinduced electron transfer (PET), Forster resonance energy transfer (FRET), photoinduced proton transfer (PPT), photoisomerization (the vision process)

Dynamic quenching : Stern-Volmer equation and transient effects

Static quenching : distance distribution, Perrin active sphere, complex formation

Heterogeneous quenching: fluorophore medium non-homogeneity and variations in quencher accessibility

Application examples in biological systems and some advanced fluorescence techniques

Interaction of Surfactants with phospholipid vesicles.

Intercalation of fluorescent probes with DNA

Energy transfer in proteins

Confocal fluorescence microscopy: GFP ("green fluorescence protein") and semiconductor nanocrystals (quantum dots)

Fluorescence recovery after photobleaching (FRAP)

Single molecule spectroscopy

Fluorescence correlation spectroscopy (FCS)

Evaluation

Written test (60%)

Presentation of Laboratory work (40%)

Jury

Paulo Coutinho, Joaquim Agostinho, Orlando Frazão