

# Fluorescence Spectrophotometry

## IN BRIEF...

- Powerful technique for sensitivity and selectivity of molecules
- Non-destructive quantitative and qualitative analysis
- Sample handling for the analysis of liquid and solid materials
- Analysis volume: < 1  $\mu\text{L}$  for liquids and ~ 1 mm for solids
- Heavily used in research laboratories
- Heavily used in Life Science research laboratories
- Common technique used for the analysis of inorganic metal complexes

## Thermo Scientific Products:

- Quantech filter fluorometer
- NanoDrop 3300 micro-volume fluorospectrometer
- Lumina fluorescence spectrometer



## Applications

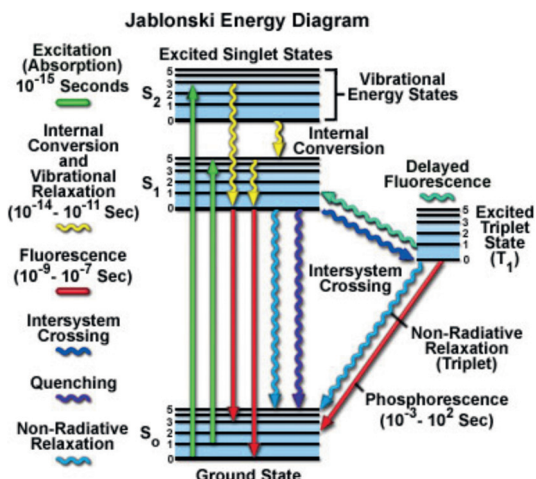
Fluorescence spectrophotometry is a common research technique used in the Life and Material Sciences. It can be applied to a wide range of applications ranging from quantitative analysis to structure analysis of molecules such as proteins.

Some well known and common applications include:

- Researching LED/OLED materials
- Researching Quantum dots (ZnSe, CdS, CdSe, CdTe, PbSe)
- Researching inorganic phosphor materials used for liquid crystal displays (LCD)
- Forensic analysis of environmental contaminants
- Researching Fluorescence Energy Transfer (FRET) for detection and distance measurements
- Intercellular ion measurements such as Ca and Mn
- Polarization measurements for quantification and diffusion
- Protein and DNA structure determination
- Nucleic acids using Fluorescence probes

## Basic Theory

Some molecules when they absorb ultraviolet or visible light emit light when their electrons return from an excited state back down to the ground state. Luminescence is the term that is used to describe the emission of light that occurs from electronically excited states. The term Luminescence can refer to several techniques including Fluorescence, Phosphorescence, Chemiluminescence and Bioluminescence. The Jablonski diagram illustrates the various processes that occur between absorption and emission of light by the various Luminescence techniques. A typical diagram is shown below:



## Technology

Fluorescence is a mature technique. Historically fluorescence instrumentation fundamentally has followed one of two designs. A cost effective approach is illumination by a broad wavelength light source with discrimination of both the exciting light and resultant emission light that reaches the detector by the use of optical filters. A typical research grade fluorescence instrument is designed with two independently controlled monochromators. The user has complete control of each monochromator for wavelength and bandwidth. Recent advances in narrow and broad-band LED light sources from the far UV to the NIR wavelength regions have also opened up the flexibility for additional designs. With the addition of new fluorescence reagents or probes, advancements in the technology has continued. These include technologies such as Fluorescence Energy Transfer (FRET), time-resolved fluorescence (TRF), fluorescence polarization (FP) and fluorescence correlation spectroscopy (FCS).



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