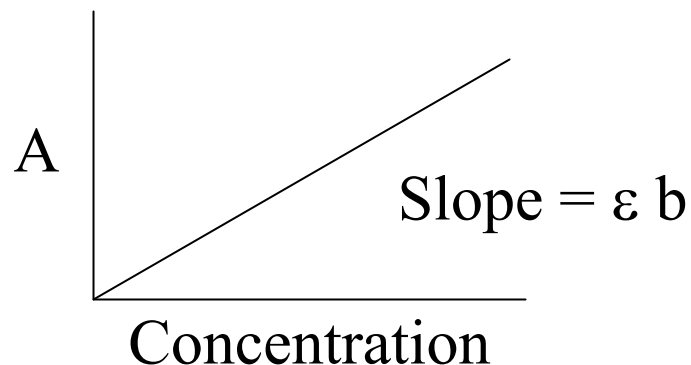


Chapter 14 – Applications of Molecular Absorption Spectrometry

~~Read: pp 367-380~~

~~Problems: 14-1,2,8~~

$$A(\lambda) = \epsilon(\lambda)bc$$



ϵ values in UV/Vis molecular absorption spectrometry range from 0 to 10^5 !

$$\epsilon = 8.7 \times 10^{19} \text{ PA}$$

Capture cross-section area $\sim 10^{-15} \text{ cm}^2$

Probability of energy-absorbing transition 0-1

Absorbance Measurements in Gas vs. Liquid Phase

Solvent matters!!!

Polar solvents tend to obliterate the fine structure.

As a rule, same solvent system should be used when comparing absorption spectra for identification purposes.

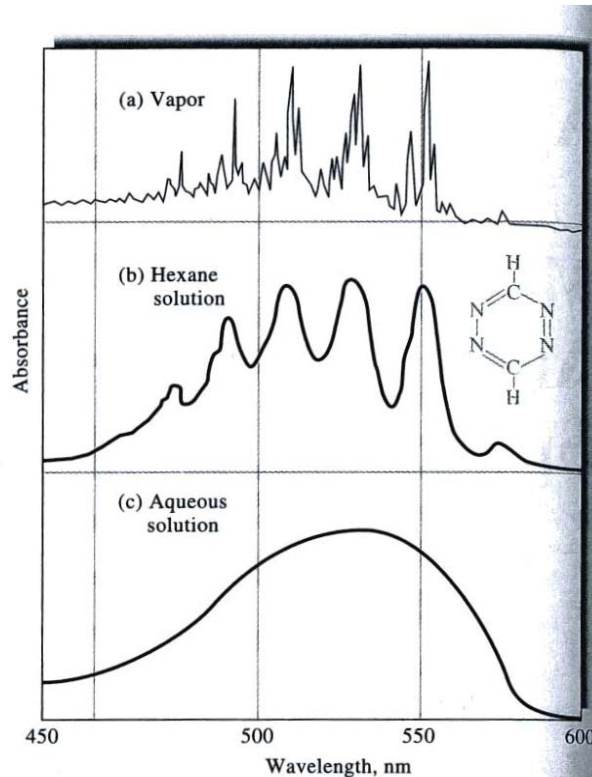


FIGURE 14-1 Ultraviolet absorption spectra for 1,2,4,5-tetrazine. In (a), the spectrum is shown in the gas phase, where many lines due to electronic, vibrational, and rotational transitions can be seen. In a nonpolar solvent (b), the electronic transitions can be observed, but the vibrational and rotational structure has been lost. In a polar solvent (c), the strong intermolecular forces cause the electronic peaks to blend, giving only a single smooth absorption band. (From S. F. Mason, *J. Chem. Soc.*, 1959, 1265.)

Absorption spectra for vapor shows much fine structure (e.g., numerous rotational and vibrational states associated with excited electronic state are visible.

In condensed state, less rotational freedom so rotational states not observed.

When chromophore is surrounded by solvent molecules, energies of vibrational levels are modified in a nonuniform way. Energy of a given state appears as a broad peak.

Effect of λ_{eff}

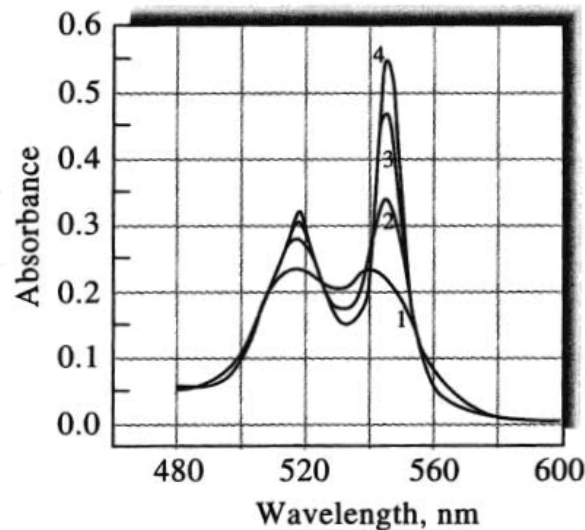


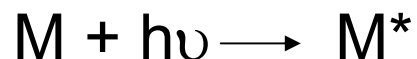
FIGURE 14-7 Spectra for reduced cytochrome *c* at four spectral bandwidths. (1) 20 nm, (2) 10 nm, (3) 5 nm, and (4) 1 nm. (Courtesy of Varian, Inc., Palo Alto, CA.)

Peak heights and peak separation are distorted at wider bandwidths.

Loss of resolution accompanies wider slit widths.

Spectra for qualitative applications should be measured with minimum slit width.

Absorbing Species



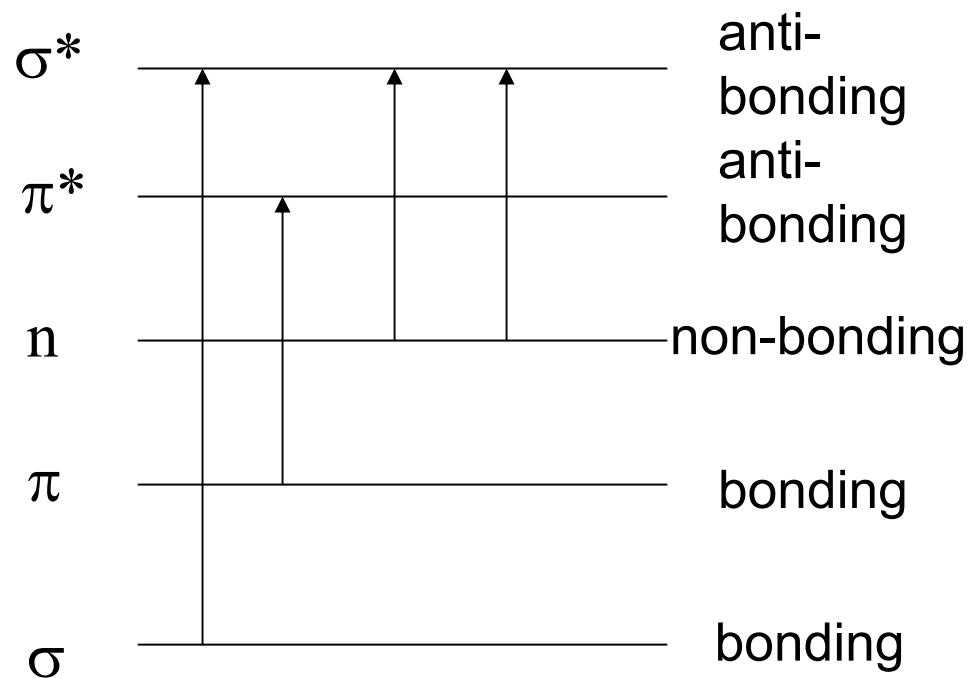
Excitation event (10^{-9} s)



Relaxation event

UV/Vis – excitation of bonding electrons!

Can be used for quantitative purposes and for functional group (type of bonding) information.



Absorbing Organic Molecules Containing σ , π and n Electrons

TABLE 14-2 Absorption Characteristics of Some Common Chromophores

Chromophore	Example	Solvent	λ_{\max} (nm)	ϵ_{\max}	Type of Transition
Alkene	$C_6H_{13}CH=CH_2$	<i>n</i> -Heptane	177	13,000	$\pi \rightarrow \pi^*$
Alkyne	$C_5H_{11}C \equiv C-CH_3$	<i>n</i> -Heptane	178	10,000	$\pi \rightarrow \pi^*$
			196	2,000	—
			225	160	—
Carbonyl	$\begin{array}{c} O \\ \\ CH_3CCH_3 \end{array}$	<i>n</i> -Hexane	186	1,000	$n \rightarrow \sigma^*$
			280	16	$n \rightarrow \pi^*$
Carboxyl	$\begin{array}{c} O \\ \\ CH_3CH \end{array}$	<i>n</i> -Hexane	180	large	$n \rightarrow \sigma^*$
			293	12	$n \rightarrow \pi^*$
Carboxyl	$\begin{array}{c} O \\ \\ CH_3COH \end{array}$	Ethanol	204	41	$n \rightarrow \pi^*$
Amido	$\begin{array}{c} O \\ \\ CH_3CNH_2 \end{array}$	Water	214	60	$n \rightarrow \pi^*$
Azo	$CH_3N=NCH_3$	Ethanol	339	5	$n \rightarrow \pi^*$
Nitro	CH_3NO_2	Isooctane	280	22	$n \rightarrow \pi^*$
Nitroso	C_4H_9NO	Ethyl ether	300	100	—
			665	20	$n \rightarrow \pi^*$
Nitrate	$C_2H_5ONO_2$	Dioxane	270	12	$n \rightarrow \pi^*$

Absorbing functional groups = *chromophores*

Olefins and aromatics

$\sigma \rightarrow \sigma^* < 185 \text{ nm}$

$n \rightarrow \sigma^* 150\text{-}250 \text{ nm}$

$\pi \rightarrow \pi^*$

$n \rightarrow \pi^* 200\text{-}700 \text{ nm}$

Conjugation = delocalization lowers energy level of π^* orbital

Typical Absorption Spectra for Organic Molecules

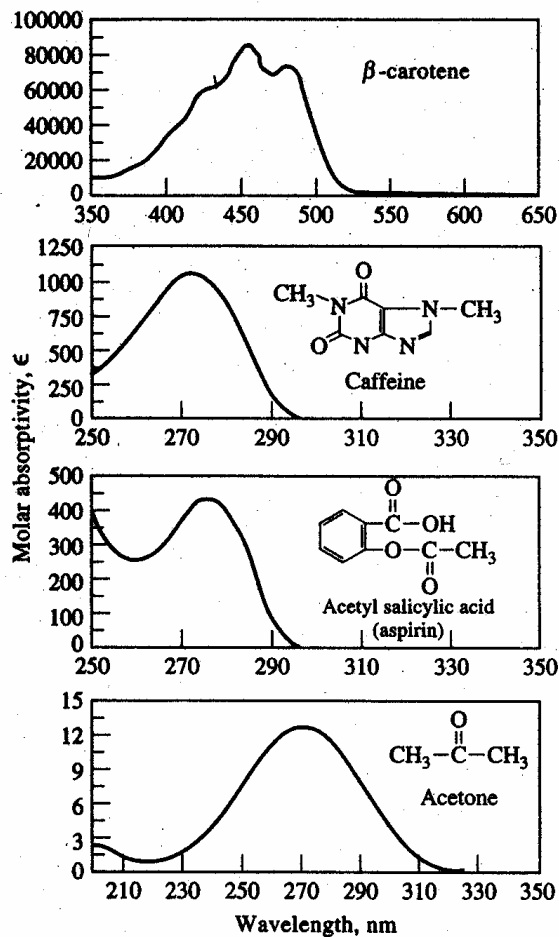


Figure 14-4 Ultraviolet spectra for typical organic compounds.

$n \rightarrow \pi^*$ are often shifted to shorter wavelengths (hypsochromic or blue shift) with increasing solvent polarity.

$\pi \rightarrow \pi^*$ are often shifted to longer wavelengths (bathochromic or red shift) with increasing solvent polarity.

Absorption Involving d and f Electrons

Crystal-Field Theory

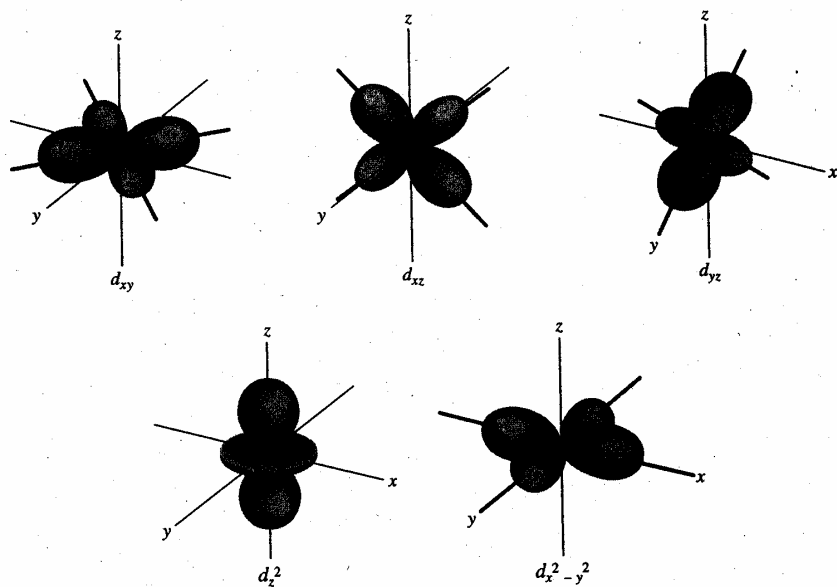


Figure 14-8 Electron-density distribution in the five d orbitals.

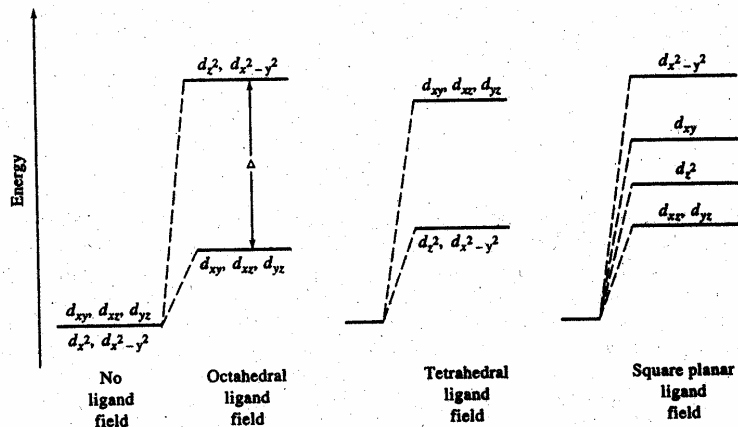


Figure 14-9 Effect of ligand field on d -orbital energies.

First and second transition-metal series. Compounds are colored as absorption occurs at visible wavelengths (300-700 nm).

Chapter 15 - Molecular Luminescence Spectrometry

Read: pp 399-417

Problems:15-1,3,7

Light emission process!

Radiative or non-radiative decay/relaxation.

Fluorescence and phosphorescence vs. chemiluminescence

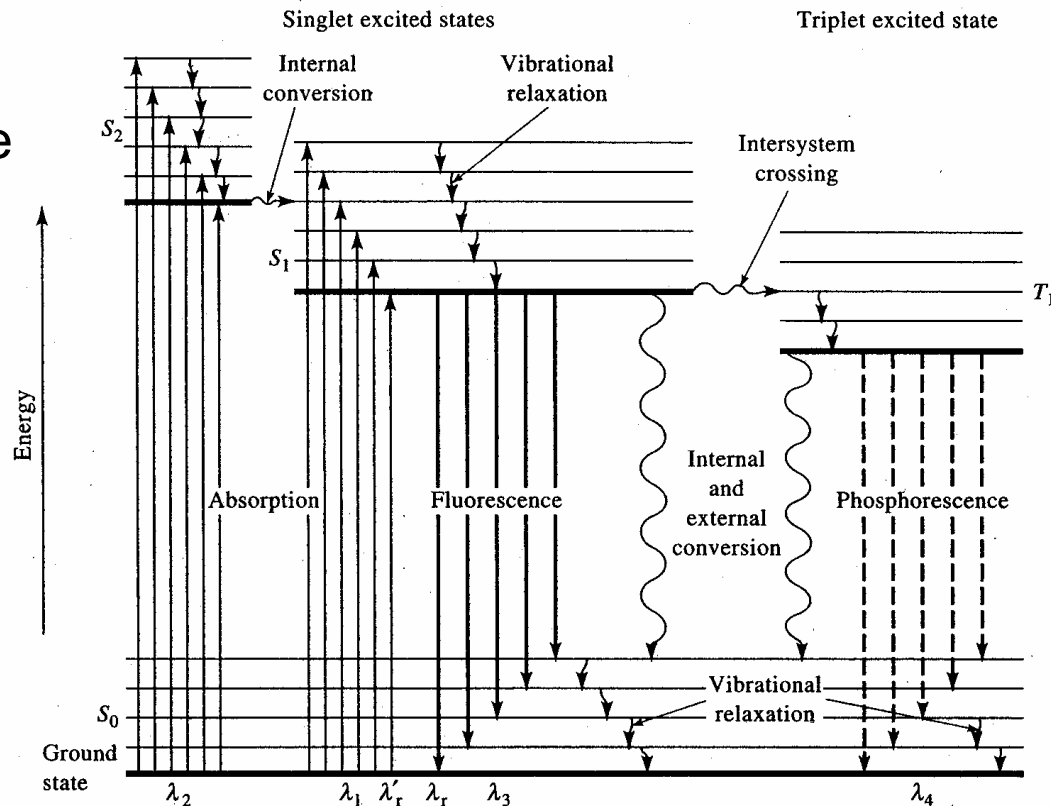


Figure 15-1 Partial energy diagram for a photoluminescent system.

Process of Excitation and Emission

- **Absorption of light** - 10^{-15} s and related to ϵ
- **Vibrational relaxation** – excess vibrational energy in solution immediately lost in solution due to collisional deactivation, 10^{-12} s.
- **Internal conversion** – intermolecular process by which a molecule passes to a lower energy electronic state *without* emission of light. Overlap of vibrational energy levels in two electronic energy levels.
- **External conversion** – deactivation of an excited electronic state by interaction and energy transfer between the excited molecule and solvent or other solutes.
- **Intersystem crossing** – process in which spin of an excited electron is reversed and change in multiplicity results. Most common when vibrational manifold overlap exists and when the molecule has a heavy atom substituent (e.g., Br, I).
- **Fluorescence and Phosphorescence** – relaxation of an excited state via light emission. Time scales range from 10^{-6} s to 100's s.

Quantitative Aspects of Fluorescence

Measurements

$$F = 2.3\Phi\varepsilon bCP_o$$

- Φ = quantum efficiency = # molecules emitting/total # molecules excited
- ε (L/mol-cm) and b (cm) have their usual meanings
- P_o in incident radiant power density (watts/cm²)

- Linear relationship, $F = KC$

- *Self-absorption* and *self-quenching* cause negative deviations from linearity (i.e., reduced fluorescence intensity).

- Φ increases with lower temperature, increased structural rigidity, $\pi \rightarrow \pi^*$ transition, and can be affected by solvent type and pH.

- Electron donating groups (NH₂, OH) tend to enhance fluorescence while electron withdrawing groups (Cl, COOH) tend to inhibit it.

Excitation and Emission Spectra

Resonance vs. non-Resonance Fluorescence

Non-Radiant losses result in *red shift* in fluorescence.

Excitation at fixed wavelength and recording the emission spectra.

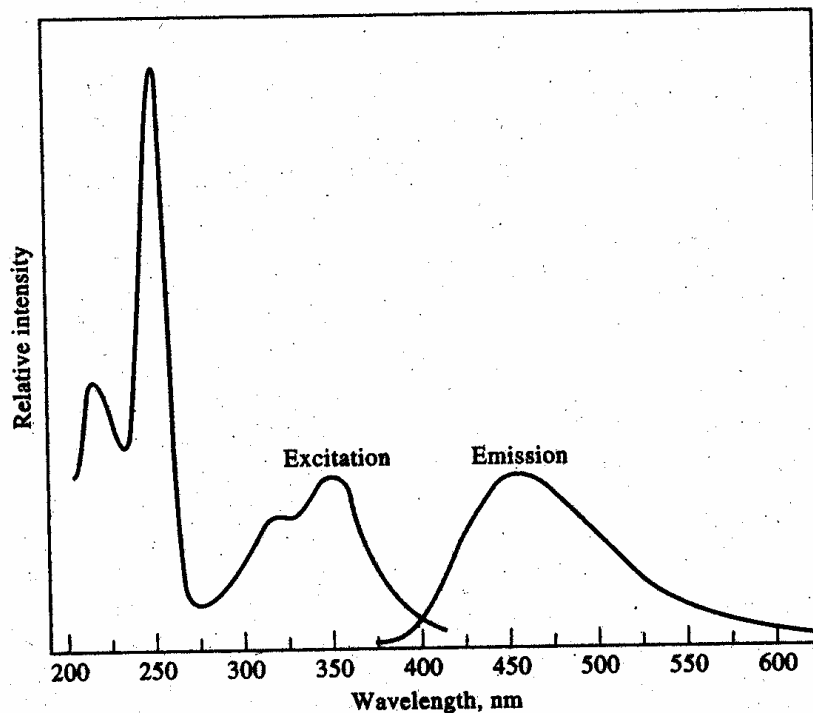


Figure 15-2 Fluorescence excitation and emission spectra for a solution of quinine.

Basic Design of a Simple Fluorometer

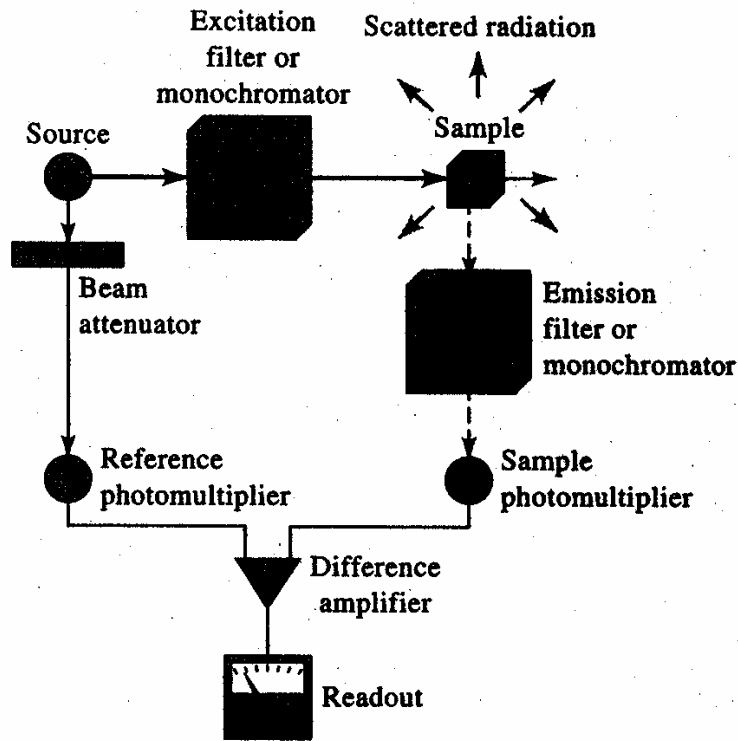


Figure 15-4 Components of a fluorometer or a spectrofluorometer.

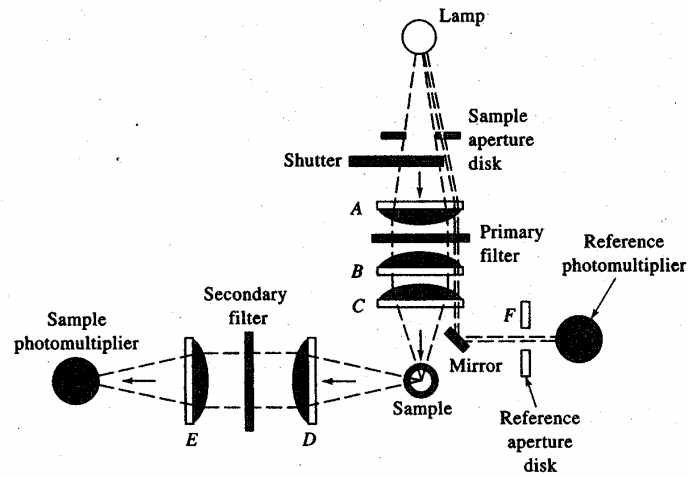


Figure 15-6 A typical fluorometer. (Courtesy of Farrand Optical Co., Inc.)

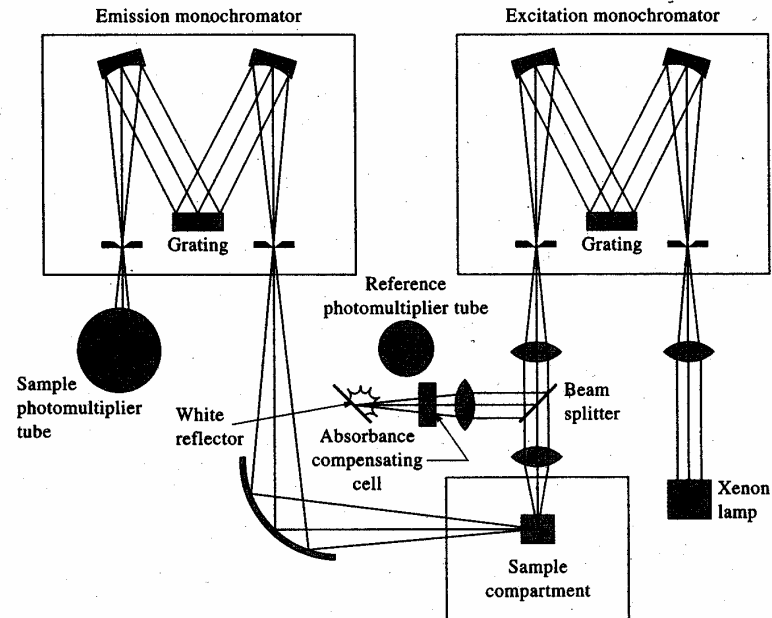


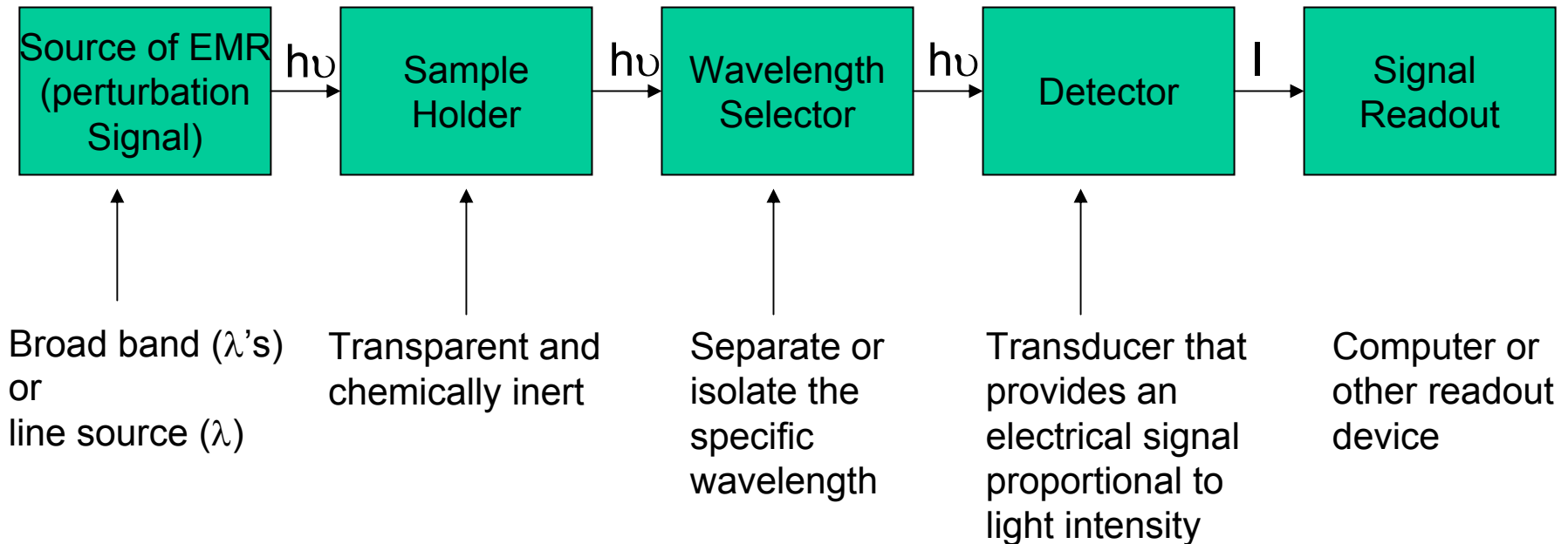
Figure 15-7 A spectrofluorometer. (Courtesy of SLM Instruments, Inc., Urbana, IL.)

Chapter 7 – Components of Optical Instruments

~~Read pp. 164-173; 180-190; 191-200~~

~~Problems: 1,2,3,6,16,19~~

Configuration of an instrument for an **absorption** measurement.



Remember: All light intensity loss must be due to *absorbance* by the analyte. Therefore, two measurements are always necessary: one with the analyte present and a background (without the analyte).

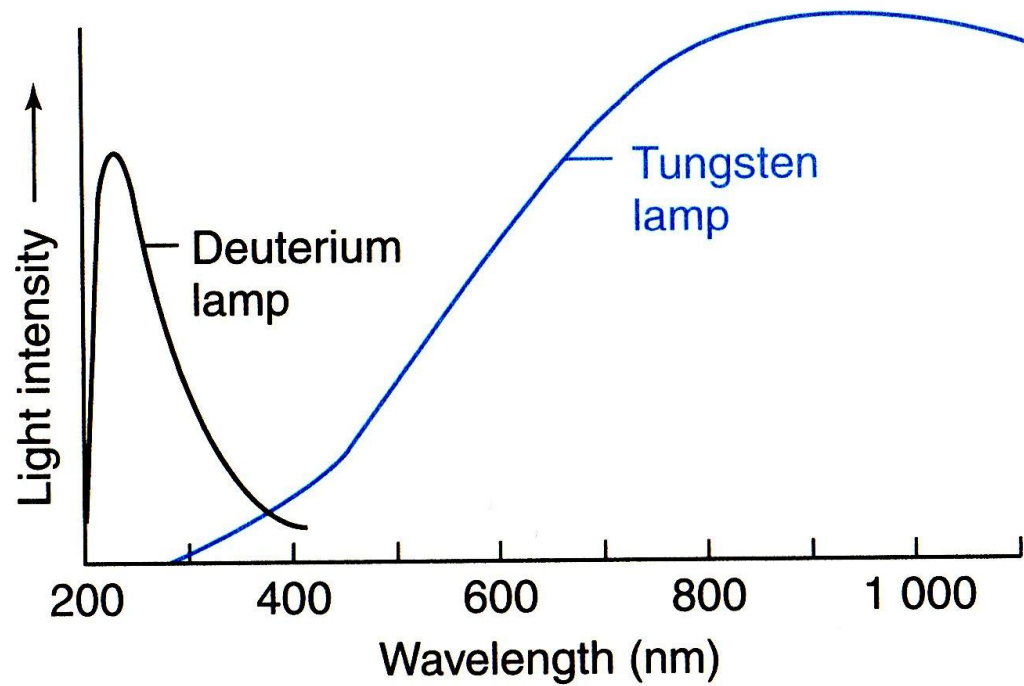


Figure 20-3 Intensity of a tungsten filament at 3 200 K and a deuterium arc lamp.

Detectors or Transducers

- Devices that record intensity changes in the incident light and convert these intensity changes to a proportional electrical signal.
- $I_{ph} \sim \text{light intensity}$ $S = kP + k_d$
- Single channel or multichannel types.
- Sensitivity, stability, dark current, can it respond to more than one wavelength simultaneously, etc.
- Phototubes, photodiodes vs. photomultiplier tubes vs. charge transfer devices (CCD's).

Types of Detectors

Single Channel

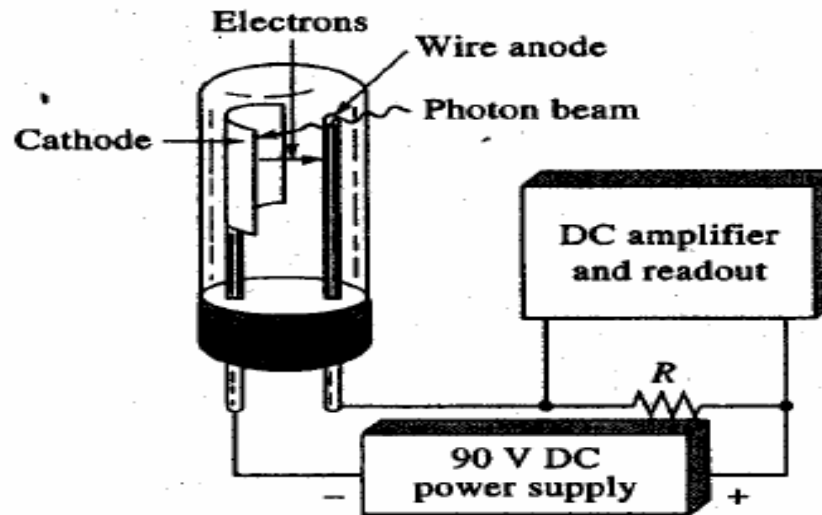


Figure 7-27 A phototube and accessory circuit. The photocurrent induced by the radiation causes a potential drop across R , which is then amplified to drive a meter or recorder.

$$I_{\text{ph}} \text{ (photocurrent)} = kP \text{ (radiant power)}$$

Types of Detectors

Single
Channel

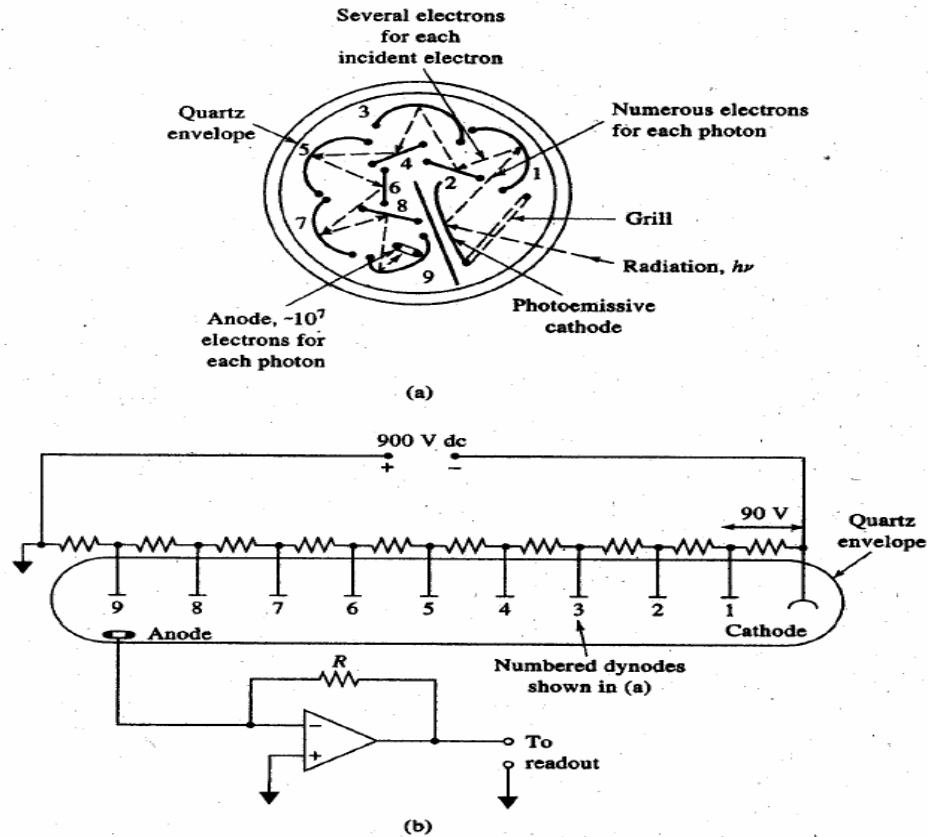
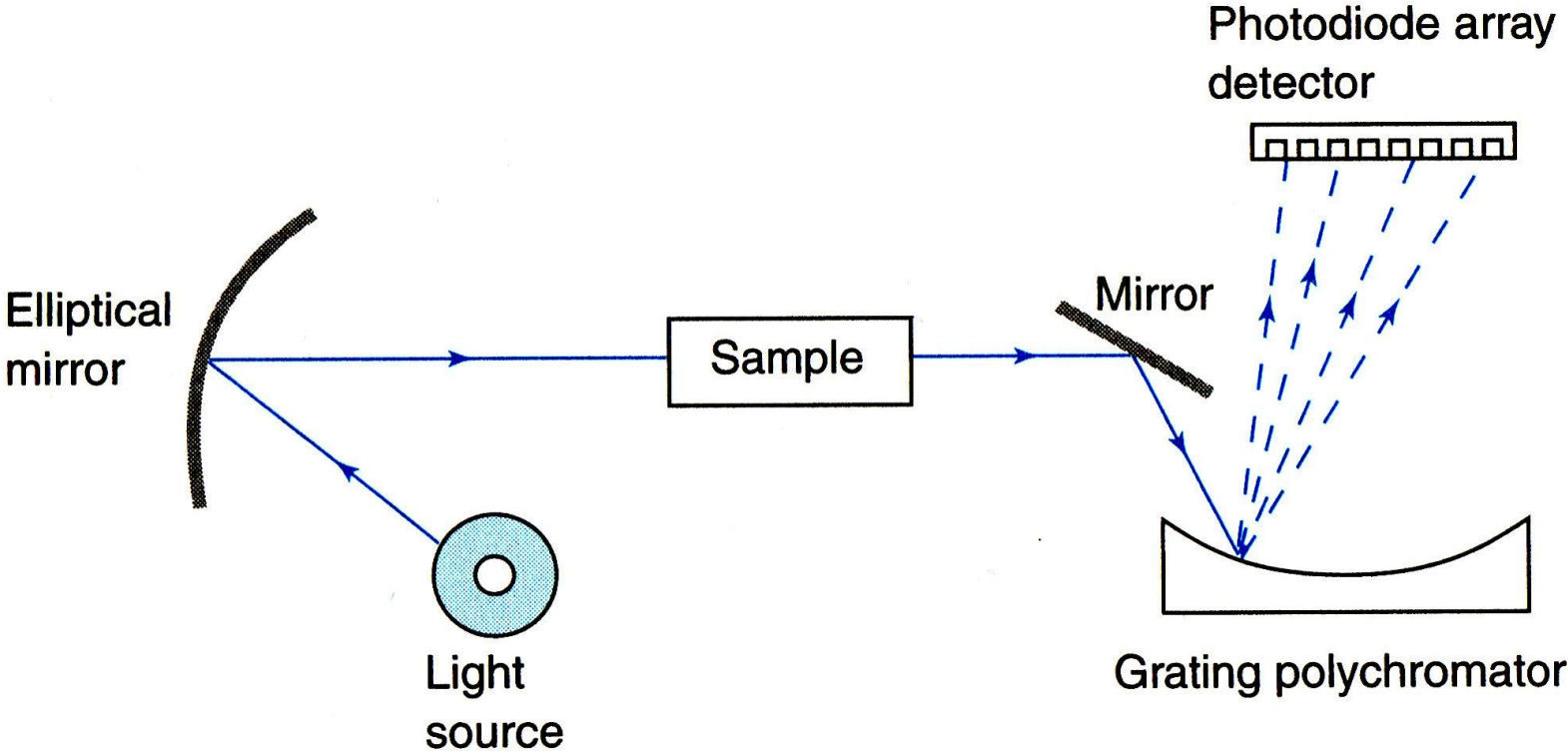
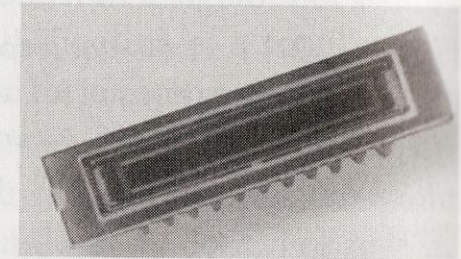
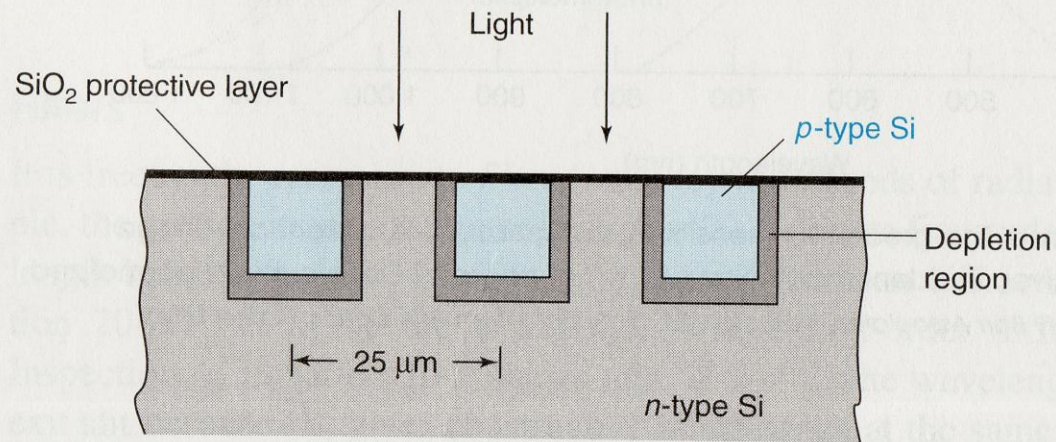


Figure 7-29 Photomultiplier tube: (a) cross-section of the tube and (b) electrical circuit.

$$I_{ph} \text{ (photocurrent)} = kP \text{ (radiant power - amplified)}$$

Multichannel Detector (Multiple Wavelengths Simultaneously)





(a)

(b)

Figure 20-13 (a) Schematic cross-sectional view of photodiode array. (b) Photograph of array with 1 024 elements, each 25 μm wide and 2.5 mm high. The central black rectangle is the photosensitive area. The entire chip is 5 cm in length. [Courtesy Oriel Corporation, Stratford, CT.]

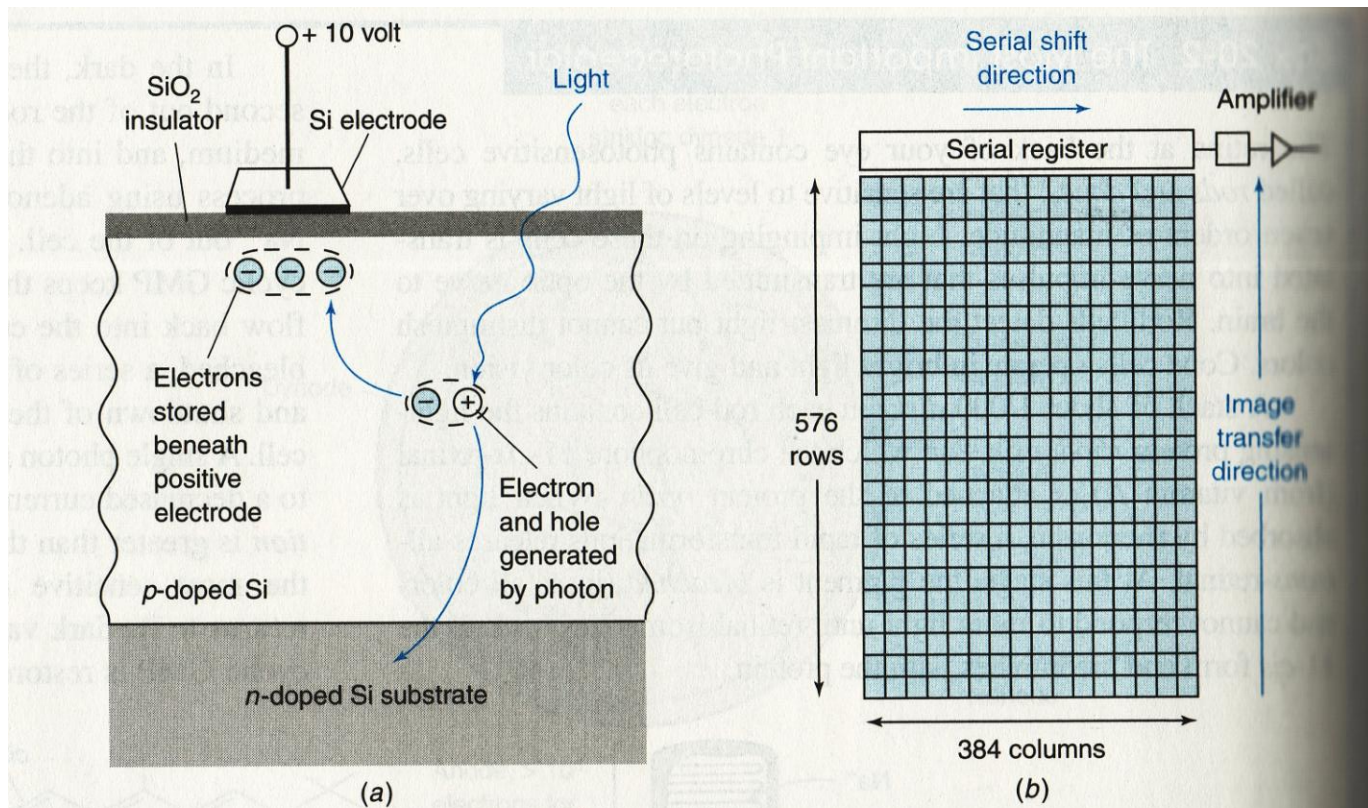


Figure 20-15 Schematic representation of a charge coupled device. (a) Cross-sectional view, indicating charge generation and storage in each pixel. (b) Top view, showing two-dimensional nature of an array. An actual array is about the size of a postage stamp.