

Visible Spectroscopy of Flower Pigments

Visible spectroscopy investigates the absorption of visible light by a sample. Students are often interested in visible-region spectroscopy because of the correlation to color vision. You can guess the visible spectrum of an object by looking at its color.

The Nature of Light

Light is composed of tiny, massless particles called photons. The properties of photons are very wavelike, so photons can be characterized by their frequency (ν) and wavelength (λ). The wavelength is the distance from one peak to the other peak in the oscillating wave (see below). The frequency is the number of times the “peak” passes a stationary point every second. Wavelength is usually expressed in nm (10^{-9} m) and frequency is expressed in Hz (1/s) or MHz (10^6 Hz). Wavelength and frequency are related by the equation

$$\lambda \nu = c \quad (c = 3.00 \times 10^8 \text{ m/s})$$

Every time one peak passes the stationary point, it moves one wavelength, therefore $\lambda \nu$ is equal to the velocity of the photon (the speed of light, c).

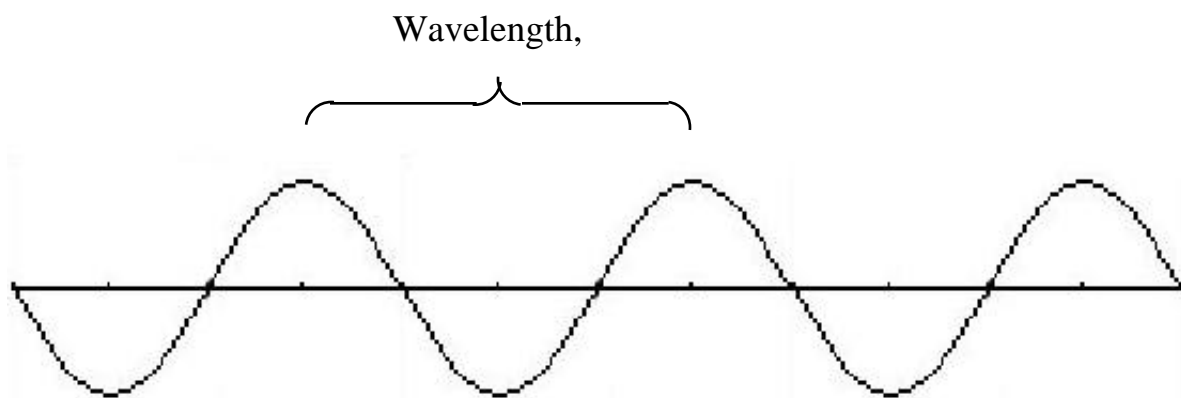


Figure 1. Depiction of a wave and the definition of the wavelength.

As stated before, light exists as particles called photons. Since photons have no mass, it might be better to describe them as energy packets. A photon with frequency, ν , has the same amount of energy as any other photon with the same frequency. The energy for a photon is given below and is dependent on the wavelength :

$$h \nu = E$$

h = Planck's constant, 6.626×10^{-34} Joule•s

E = energy

The higher the frequency, the more energy each photon has. Therefore, the two ways to increase the amount of energy from a beam of light are to increase the frequency (change the color) of the light while keeping the number of photons the same, or increase the number of photons (make

the light brighter).

The Visible Spectrum

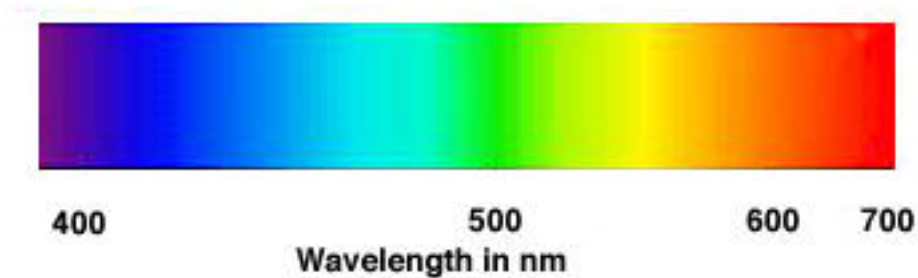


Figure 2. The visible spectrum

The visible spectrum includes photons with wavelengths from ~400 to 780 nm. The blue region has the shortest wavelengths and therefore, the highest energy. The region with wavelengths from ~200 - 400 nm is the ultraviolet (above violet). With shorter wavelengths than the visible, ultraviolet light is more energetic than visible light and this accounts for its damaging effects on skin and DNA. Light having wavelengths from ~780 nm to 300,000 nm is in the infrared region (below the red). Light in the infrared has less energy than visible light.

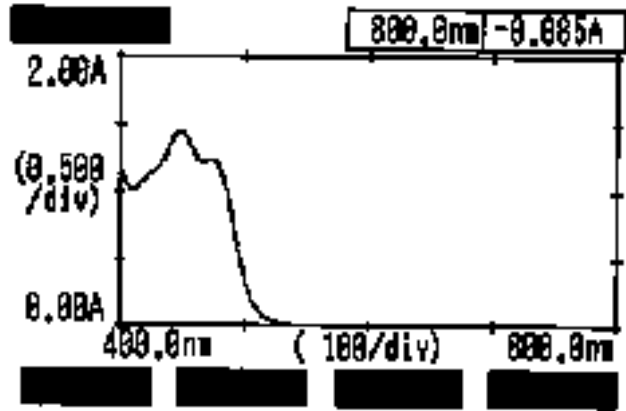
Short wavelength = high frequency = high energy
Long wavelength = low frequency = low energy

Color

Many objects around us are colored. The light we see as color originates as sunlight. Sunlight contains all the colors of the visible spectrum and therefore appears to be white (incandescent light bulbs act the same way). When the light reflects off of a surface or passes through a solid, some of the light is absorbed (removed). When photons of specific wavelengths are removed, we see the remaining colors (the ones not absorbed). If we see a blue object, it is blue because it absorbs the red, yellow, orange, and green light. After white light hits the object, only the blue light is not removed and we see the object as blue.

The Absorption Spectrum of Flower Pigments

Flowers are a convenient source of colored compounds. Preferential absorption results in the colors seen in the flower petal. The amount of light of a certain frequency that is absorbed by a compound can be measured by an instrument called a spectrometer. By measuring the amount of light absorbed at different frequencies of visible light an absorption spectrum can be constructed. Such a spectrum describes which frequencies of light are absorbed and to what extent. An example of an absorption spectrum for a yellow flower is shown below.



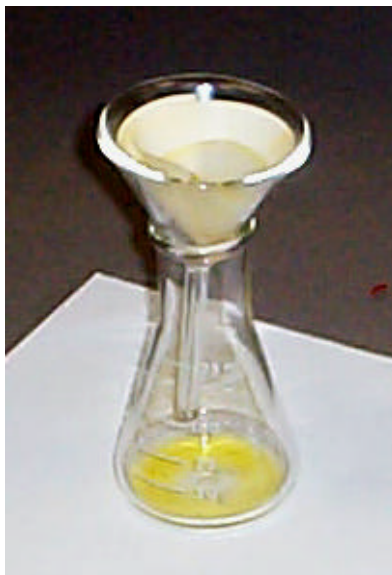
The pigments in the flower absorb the blue and green light between 400 and 500 nm (the y-axis is absorption) leaving the yellow, orange, and red light to be reflected. This gives the flower its color.

Experimental:

- (1) Gather some flower petals
- (2) Grind several petals with a mortar and pestle, adding ethanol to dissolve the pigments



(3) Gravity filter the mixture to remove the solid

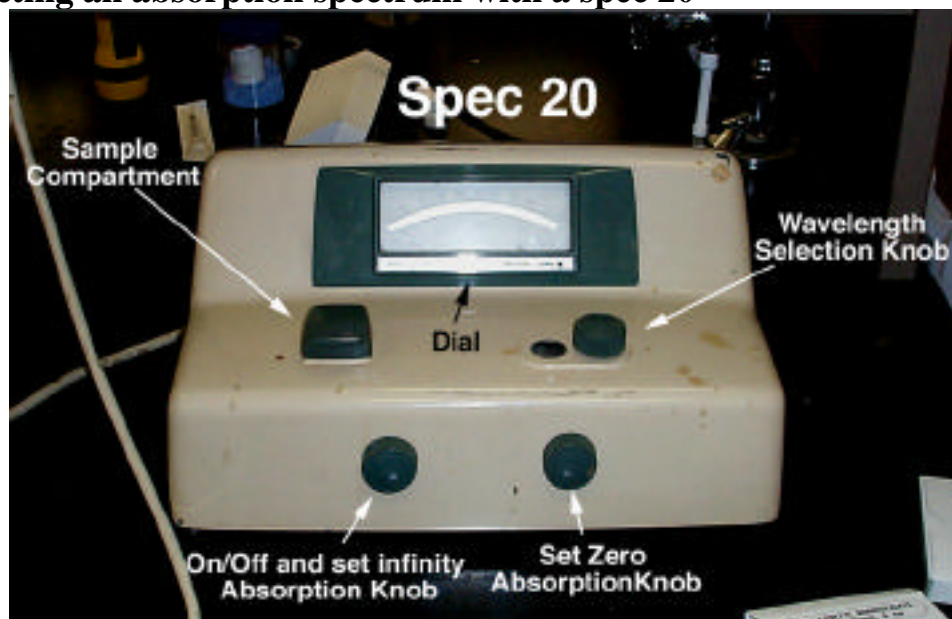


(4) Place the liquid in a cuvette

The liquid should not be highly colored. If the absorption reading exceeds 1.5 at any point, the liquid is too concentrated and should be diluted by half.

(5) Construct a spectrum of the pigment(s) using the spec 20 as instructed below

Constructing an absorption spectrum with a spec 20



(1) Turn the spec 20 on and allow it to warm up for 15 minutes

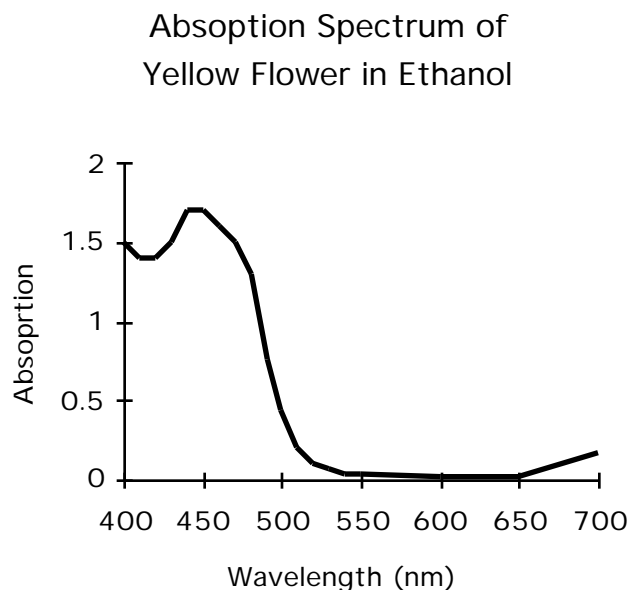
(2) Use the left knob to set the absorption reading to infinity with no sample in the sample compartment

(3) Place ethanol in one cuvette (your blank) and the sample in another cuvette.

- (4) Dial in the longest wavelength spectrum you want to measure
- (5) Place the blank in the sample compartment and close the lid. Using the right knob, set the absorption to 0.
If you can't set the right knob to 0, lower the wavelength until you can. If the lamp is not very strong, you will not be able to measure all the way to 800 nm. You may have to start at 700.
- (6) Replace the blank tube with the sample tube and record the wavelength and the absorption.
- (7) Lower the wavelength and repeat steps 5 and 6. Take measurements every 50 nm when the absorption is low (under 0.1) and every 10 nm when the absorption is above 0.1.
- (8) Create a spreadsheet (using Excel, Quattro Pro, etc) with wavelength in one column and absorption in another. Create a scatter plot (x-y scatter plot) with wavelength as the x-axis and absorption as the y-axis.
- (9) Put the appropriate labels on the graph.

Sample Data

Wavelength	Absorption
700	0.165
650	0.01
600	0.015
550	0.025
540	0.035
530	0.06
520	0.1
510	0.2
500	0.44
490	0.76
480	1.3
470	1.5
460	1.6
450	1.7
440	1.7
430	1.5
420	1.4
410	1.4
400	1.5



This experiment was developed by Michael R. Jordan at Oklahoma Baptist University . Reproduction for academic use is permitted.