MOLECULAR ABSORBANCE SPECTROSCOPY SDSU CHEM 251

MOLECULAR ABSORBANCE

- As molecules contain many electrons, a wide range of electrons they can absorb light of many different wavelengths.
- For quantitative absorbance measurements we need to focus on a single wavelength related to the compound of interest.
- For the cranberry juice, the optimal wavelength to monitor would be at ~525 nm.



MONOCHROMATOR COMPONENTS

- As most detectors do not differentiate wavelengths, so monochromators are used to select the wavelength of analysis.
- A diffraction grating separates the light into separate wavelengths and with proper angling, only the desired wavelength reaches the detector.



ABSORBANCE QUANTIFICATION

- Absorbance (or transmittance) measurements can be used to quantify the amount of an analyte.
- Beer's Law is used to correlated concentration to absorbance measurements.
- $\boldsymbol{\varepsilon}$ is the molar absorptivity
- **b** is the path length
- C is the molar concentration

(a)	P ₀	s a m p l e	P _T	-	
(b)		b l a k	P ₀		

$$A = -\log T = -\log \frac{P_T}{P_o}$$

	Transmittance	Absorbance	
Beer's Law:	100%	0.00	
A CLC	60%	0.22	
$A = \mathcal{E}\mathcal{B}\mathcal{C}$	30%	0.52	
	10%	1.00	
]	8	2.00	

LIMITATIONS TO BEER'S LAW

- Beer's law is effective, but it does encounter limitations that cause non-linear deviations in the absorbance signal.
- <u>Limitations</u>:
 - Too high analyte concentration
 - Refractive index change
 - Equilibria variations
 - Instrumental limitations

