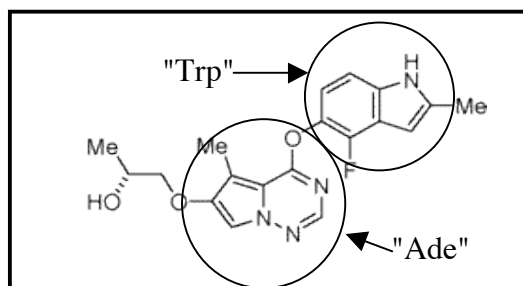


Answer Key - Quantitative Question from Lecture #9

(a) Ring systems with conjugated double bonds absorb UV in the 260 nm range. One portion of the kinase inhibitor (upper right) looks somewhat like tryptophan, and another (bottom middle) looks vaguely like a purine, such as adenine. Trp absorbs maximally at 278 nm (molar extinction coefficient at 278 nm = $\epsilon_{278 \text{ nm}} = 5,500$ liters/mole-cm); but, if you look at the absorption spectrum of Trp (which is relatively insensitive to pH), Trp absorbs pretty well at 260 nm too (approx. $\epsilon_{260 \text{ nm}} = 3,600$ liters/mole-cm). Adenine absorbs maximally at 260 nm and, at pH 7, has an $\epsilon_{260 \text{ nm}} = 13,300$ liters/mole-cm. In general, absorbances of two separated chromophores on the same molecule are additive. Therefore, a rough guess-timate would be that the kinase inhibitor has an $\epsilon_{260 \text{ nm}} = \sim 16,900$ liters/mole-cm.



(b) The Beer-Lambert Law states that $A = \epsilon cl$, where ϵ is the molar extinction coefficient (in liters/mole-cm), c is the concentration (in moles/liter), and l is the path length of the cuvette (in cm). You are given that 100-fold diluted solution of the kinase inhibitor displays an $A_{260 \text{ nm}} = 0.3$.^{*} Hence, the concentration of the original stock solution that you were sent by the company is:

$$c = 30 / \sim 16,900 \text{ liters/mole-cm (1 cm)} = 0.00178 \text{ moles/liter} = \boxed{\sim 1.8 \text{ mM}}$$

^{*}This value (0.3) is in a good range to get a reliable measurement of A in a spectrophotometer. When molecules in solution are too concentrated, they self-shield each other, and this masking yields an absorbance value that less than the actual concentration. A reasonable rule of thumb is that to get a reliable measure of absorbance, you want to be below an $A = 1$ and you want to check that you are in the linear range, i.e. a solution that is twice as concentrated yields an A that is twice as high and a solution that is twice as dilute yields an A that is half as great.