Deacon's Challenge No. 70 Answer

The transmittance of a solution of NADH at 340 nm is 45%. What is the absorbance at 340 nm of a 1 in 5 dilution of this solution?

Absorbance, but not transmittance, is linearly proportional to concentration. Therefore first convert 45% transmittance to absorbance.

If I_0 is the intensity of incident light and I the intensity of transmitted light, then:

$$\text{transmittance } (\%T) = \frac{I \times 100}{I_0} \quad \text{and} \quad \text{absorbance } (A) = \log_{10} \frac{I_0}{I}$$

The expression for %T can be arranged to: $\frac{I_0}{I} = \frac{100}{\%T}$

Which can be substituted into the expression for A to give:

$$A = \log_{10} \frac{100}{\%T} = \log_{10} 100 - \log_{10} \%T$$

Substituting 2 for $\log_{10}$ 100 gives the following useful expression:

$$\boxed{A = 2 - \log_{10} \%T}$$

Substitute %T = 45% to convert to absorbance:

$$A = 2 - \log_{10} 45 = 2 - 1.65 = 0.35$$

Assuming that Beer's Law is obeyed, division of this value by 5 gives the absorbance of a 1 in 5 dilution:

$$\text{Absorbance of 1 in 5 dilution} = \frac{0.35}{5} = \mathbf{0.07}$$

Question 71

Enzymologists recommend that whenever possible the substrate concentration in an enzyme assay should be at least ten times the Michaelis constant (Km). What is the rate of reaction achieved (expressed as multiples of the maximal velocity), for an enzyme reaction which obeys simple Michaelis-Menten kinetics, when the substrate concentration is exactly ten times the Km value?