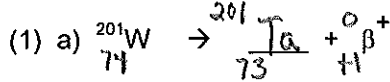


1. You have visited your doctor about a "lump" on your back. She runs a genomics marker test using a DNA microarray to compare "normal" cells vs. "lump" cells. After 24 hours exposure, mRNA is harvested, cDNA prepared using red-dye markers for the "normal" cell sample and green-dye markers for "your lump" cells. Any gene product that shows **no difference** in expression between the two cell lines would be indicated by a yellow colored spot.

(1)

2. Balance the following radioactive decay equation by filling in the blank with the missing item.



(1) b) A radioisotope has a **rate constant** of 0.037 / yr. Calculate the **half-life** of the radioisotope.

Half-life = 18.7 yr

$$t_{1/2} = \frac{\ln 2}{k} = \frac{0.693}{0.037/\text{yr}} = 18.7 \text{ yr}$$

(1) c) How many **years** will it take for a sample of this radioisotope rated at 20 microCuries to undergo radioactive decay to the point where it loses 98% of its current activity? 106 years.

$$A = A_0 e^{-kt} ; 0.4 = 20 e^{-kt} \Rightarrow \ln \frac{20}{.4} = kt \text{ or } t = \frac{\ln 50}{k} = 106 \text{ yr}$$

3. SDS gels are greatly improved in resolution by running a "stacking" gel and a "resolving" or "running" gel.

a) Name two key property differences between the "stacking" gel and the "resolving" gel that contribute to the improved resolution of running DJSC PAGE.

(1) a) pH - lower pH in stacking gel (Gly ~ 0 charge)

b) % gel - lower % acrylamide in stacking gel

What is the role of each of the following in performing SDS-PAGE?

(1) a) pH - vary pH so glycine in buffer goes from ~ 0 charge to ⊖ charge in running gel.
b) Coomassie Blue: - general stain for proteins

4. The equation of motion for a small, spherical particle of mass (m) and frictional coefficient (f) that is initially at rest, and then acted on by a constant force (F) at time t = 0 is $F - fv = ma$.

(From calculus recall that $F - fv = m(dv/dt)$ solves to $v = (F/f) [1 - \exp(-ft/m)]$.)

a) Show that such a particle will initially accelerate but over time will approach a "maximal" velocity.

(1) $v = \frac{F}{f} (1 - e^{-f \cdot t / m}) ; \text{ as } t \rightarrow \infty, e^{-f \cdot t / m} \rightarrow 0 \Rightarrow v_{\infty} = \frac{F}{f}$

(2) b) The diffusion constant for a protein is determined to be $0.258 \times 10^{-6} \text{ cm}^2/\text{s}$ at with $T = 20^\circ \text{C}$, and $\eta = 0.01 \text{ (g/cm-s)}$. It has a diameter of 80Å, a density of 1.3 g/cm^3 and a $v\text{-bar}$ of $0.73 \text{ cm}^3/\text{g}$ protein.

Calculate the frictional coefficient ratio (f/f_{min}) for this protein and comment on the expected shape of the molecule (spherical or not).

$$f = 6\pi\eta R_0 = 6(3.1416) \frac{0.01 \text{ g}}{\text{cm-s}} (40 \cdot 10^{-8} \text{ cm}) = 7.5 \cdot 10^{-8} \text{ g/s}$$

$$f_{\text{exp}} = \frac{R \cdot T}{N \cdot D} = \frac{(8.314 \cdot 10^7 \text{ g-cm}^2 / \text{m-s}^2 \cdot \text{K}) (293 \text{ K})}{6.02 \cdot 10^{23} / \text{m} (0.258 \cdot 10^{-6} \text{ cm}^2 / \text{s})} = 1.57 \cdot 10^{-7} \text{ g/s}$$

$$\frac{f}{f_0} = \frac{1.57 \cdot 10^{-7}}{7.5 \cdot 10^{-8}} = 2.1$$

5. What is typically measured by dynamic light scattering (LS)? $D \rightarrow F \rightarrow R_h$

(2)

What wavelengths are normally employed in making circular dichroism (CD) spectra?

180 - 240 nm

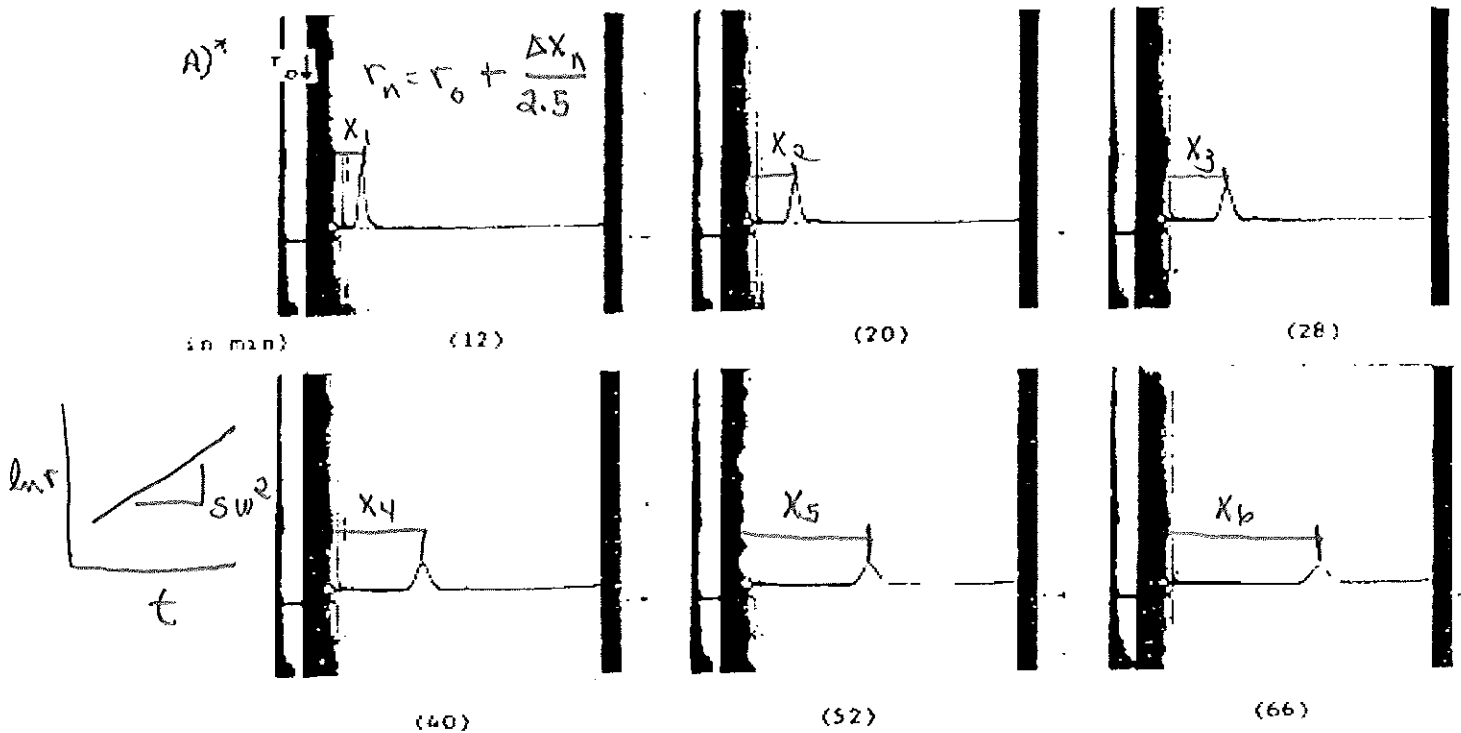
Not so spherical

6. Determine the sedimentation coefficient (s) and molecular weight (M) for the sample that gave the following data when subjected to: A) a sedimentation velocity run using Schlieren optics, and B) a sedimentation equilibrium run using interference optics.

Note: the figures below have been magnified to allow you to make measurements from the figures. The "r" can be determined from the reference points (r₀) and the magnification factors. Assume T = 20° C, density of buffer = 0.9978 g/mL, and v-bar = 0.737 cm³/g for the protein, and η = 0.01 (g/cm-s) for both experiments.

A) Sed. Vel.: ω = 40,000 rpm, magnification factor (2.5X), r₀ = 5.72 cm. (times are given in minutes).

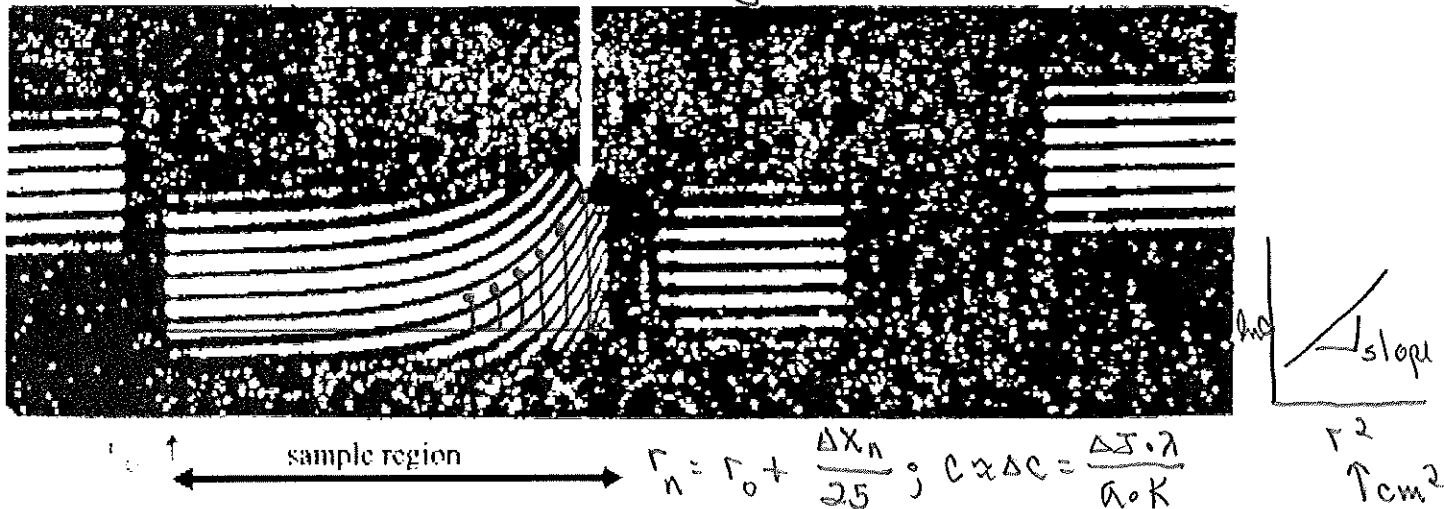
(4) Report "s" in proper units [s = 19 · 10⁻¹³ sec = 19 S] (Show work and attach plot).



B) Sed Equilibrium: ω = 5200 rpm, magnification factor (25X), r₀ = 6.75 cm. Calculate M in g/mol (4pts) and

(5) also estimate the concentration of the protein at the position with the white arrow (1 pt). Assume the cell path length to be 12.00 mm, λ = 546 nm, and (dn/dc = 0.186 (g/cm³)⁻¹).

[M = ~ 1.0 · 10⁶ g/mol ; []_{arrow} = ~ 1.4 mg/ml] (Show work and attach plot).



I hereby declare that I did this assignment independently:

$$\text{slope} = \frac{M(1 - \bar{v}_p)\omega^2}{2RT(K)} \cdot \frac{1}{\text{sec}^2}$$

(1/cm²)

↑ 8.314 · 10⁷ g·cm² / s²·m·K