Bio322: Biophysics Mid Semester Exam

Course coordinator: Chaitanya Athale

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Maximal score 20; Time: 2 hours

Attach the answer sheet to the answer table at the time of submission.
All questions are compulsory.

1 Short answers- each question carries *1 mark*

1. A normal human fibroblast has 46 chromosomes. What is the end-to-end length of all the DNA in such a cell in metres? Assume the chromosomal DNA has been separated from protein content, and each and every chromosome is line up end-to-end with the other.

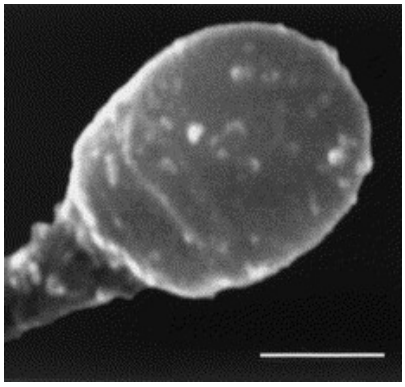

Figure 1: The head of a human sperm. The scale bar is 1 μm.

2. Provide an order of magnitude estimate of the volume of such a human fibroblast in litres?

3. For the human genome in a human fibroblast, what is the ratio $V_{nucleus} : V_{chromatin}$, where chromatin is DNA uniformly interspersed with nucleosomes. Assume for this estimate you can ignore higher order organization. Given: DNA is bundled in nucleosomes that extend every 200 bps. The histone-octamer combined with DNA forms a cylinder of diameter 7nm and length 6 nm.

4. What is the ratio of $V_{nucleus} : V_{chromatin}$ in a human sperm nucleus? Given: Sperm head with scale bar in Fig. 1. Assume the entire head is contains the nucleus.

5. What are the volume and surface area of single human immunodeficiency virus (HIV) particles? Refer to Fig. 2.

6. The matrix of the HIV particle (Fig 2) is made up of a complex polyprotein called Gag. How many Gag proteins are present per virus particle? Given: The diameter of an HIV particle is ≈ 130 nm.

7. How many lipid molecules are associated with virus particle (Fig 2)?

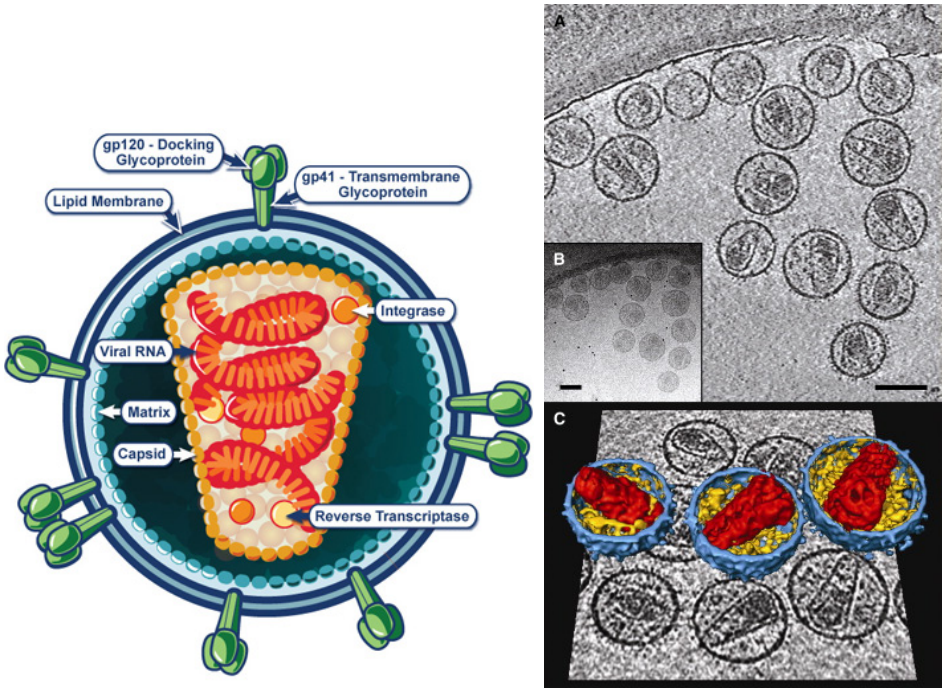

Figure 2: *(left)* A cartoon representation of the HIV virus with its outer lipid envelope. *(right top)* An electron micrograph of HIV particles and *(right bottom)* 3D rendered sample virions cut into half with membrane (blue) space between membrane and the core (yellow) and the viral capsid (red) (taken from Briggs et al. (2006) Structure. 14(1) p15-20). The scale bar is 100 nm.

8. 5ml of oil can cover a lake with an area of 200 m^2. What is the thickness of the oil film?

9. During protein folding in an aqueous environment, when the amino group leucine minimizes its contact with water during protein folding, which energetic term changes, to reduce the free energy of folding?

10. *E. coli* has a typical length of 2 μm and width of 1 μm. When grown on lactose as the only sugar, the bacterium synthesizes β-galatosidase ($M_r = 450,000$ Da), which catalyzes

the breakdown of lactose. How many molecules of β-galatosidase are likely to be found in a single bacterium? Given: the bacterial density $\rho = 1.2gm/cm^3$, and 14% of the total cell mass is soluble cytosolic protein, of which 1% is β-galatosidase.

11. For an *E. coli* bacterium with weight 1 pg, the number of molecules that are proteins is approximately $2.4 \cdot 10^6$. What is the percent of dry weight of the bacterium?

12. Stretching a DNA filament using an optical tweezer results in extension (x) over its original length (L) due to an applied force (F). Given dsDNA has a spring constant of $k_{DNA} = 10^{-5}pN - nm^{-1}$, what should the force applied be in order to achieve 25% extension of DNA?

13. At the time of early divisions in the fruit fly *Drosophila*, only nuclear divisions occur. They take 8-10 minutes per division. How many nuclei will have formed after 100 minutes, if the embryo started with 1 nucleus?

14. In an *E. coli* cell grown on glucose as the only sugar source, lacI (lactose inhibitor) proteins are expressed, which repress the expression of the *lac-operon*. In order for repression to occur the *lacI* needs to form tetramers. What is the average time in milliseconds it takes for two *lacI* proteins to encounter each other?

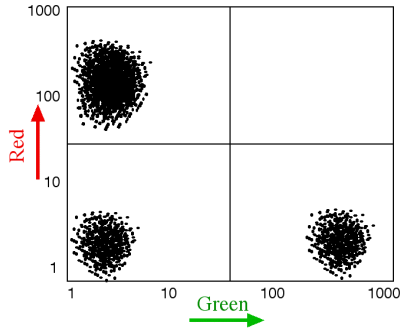

Figure 3: A population of cells expressing two markers which were antibody stained using two unique dye-complexed antibodies with colors red and green respectively were sorted using a fluorescence activated cell sorter (FACS) machine. The scores indicate frequency counts of cells on both X- and Y-axes.

15. What is the average frequency of cells that are labelled with both red (R) and green (G) antibodies based on a FACS analysis the result of which is illustrated in Fig. 3?

16. Given the tip of an atomic force microscope (AFM) cantilever is approximated as a sphere of radius R makes an indentation of depth h the force on the cantilever is given by the Hertz Model as $F(h) = \frac{4\sqrt{R}}{3} \cdot E^* \cdot h^{3/2}$. What is E?

17. What are the units (SI) and dimensions of E^* in the Hertz Model?

18. If *E. coli* cells are subjected to osmotic shock with 500 mM NaCl, will the diffusive mobility of intra-cellular molecules increase or decrease? If diffusive mobility of a fluorescently tagged molecule was estimated by FRAP, will the half-time of recovery of a FRAP curve change in the osmotically shocked cell and will it be greater or less than the untreated cell?

19. What is the quantity measured in patch clamp recordings of ion channels and what are the typical units used for eukaryotic cells?

20. Calculate the entropy (S) of a lac repressor protein of copy number 10 and number of binding sites $5 \cdot 10^6$.

Name:
Roll no.:
Date:

2 ANSWERS

Question no.	Answer
1	$\sim$2 m
2	$2 \cdot 10^{-12}$ L = 2 pL
3	$V_{nuc} : V_{chromatin} = 75.58$
4	$V_{nucsperm} : V_{chromatin} = 1.2$
5	$V_{HIV} = 1.15 \cdot 10^{-3} \mu m^3 = 1.15 \cdot 10^{6} nm^3 = 1.15 \cdot 10^{-3} fl = 1.15 \cdot 10^{-18} L = 1.15 attolitre$, $A_{HIV} = 0.053 \mu m^2 = 5309.3 nm^2$
6	$N_{GAG} = 4225$ since $A_{GAG} = \pi \cdot (2nm)^2$ and virus surface area is 5309 nm^2
7	$A_{lipid} = 0.5 nm^2$, $N_{lipid} \approx 2 \cdot 10^5$
8	Thickness of the lipid layer $h = 25nm$
9	Entropy
10	$N_{\beta-Gal} \approx 1800$
11	$N_{protein} = 2.4 \cdot 10^6$, $M_{protein} \approx 30000 Da$, dry weight of E. coli 25%, thus proteins form 30% of the dry weight of *E. coli*. Between 20-30% will be accepted as an answer, depending on steps in the calculation.
12	$Force = 2.5 \cdot 10^{-6} \cdot LpN$ where L is the length of DNA. If a specific value for L is chosen, the answer is accepted.
13	Assuming time per division (t_d) to be 10 mins, $N = 2^{t/t_d}$ where t is the time, $N \approx 5793$.
14	Spacing distance between molecules (d) is $d = c^{-1/3}$ where c is the concentration in molecules per μm^3. $N_{lacI} \approx 1000$ molecules assuming 2 μM concentration. $d = 100nm$. Assuming typical diffusion coefficient $D = 100 \mu m^2/s$, the time for encounter is $t_{diff} = 10^{-4} s = 0.1 ms$
15	FACS frequency of cells with BOTH dyes $\approx 0 - 2$.
16	Young's modulus
17	Units: $N - m^{-2}$ or $kg/m - s^2$, dimensions $[M^1 L{-}1 T^{-2}]$
18	$D_{NaCl} < D_{control}$ and $t_{1/2}^{NaCl} > t_{1/2}^{control}$
19	Current is measured in patch clamp while voltage is constant. Typically cellular currents are measured in pico-Amperes (pA).
20	$S = 1.9 \cdot 10^{-21} J/K$