Biology 5357

Chemistry & Physics of Biomolecules

Examination #3

Glycobiology, Membranes
& Membrane Proteins Module

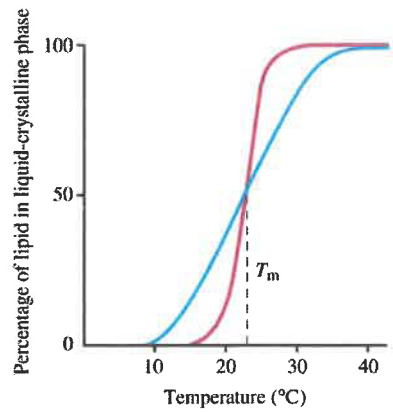
December 9, 2019

Answer Key

6. Cholesterol is an important component of biological membranes (8 points total):

(a) What class of lipid is cholesterol (1 point)?

sterol lipid



(b) What is the proportion of cholesterol in the endoplasmic reticulum vs. plasma membrane (2 points)?

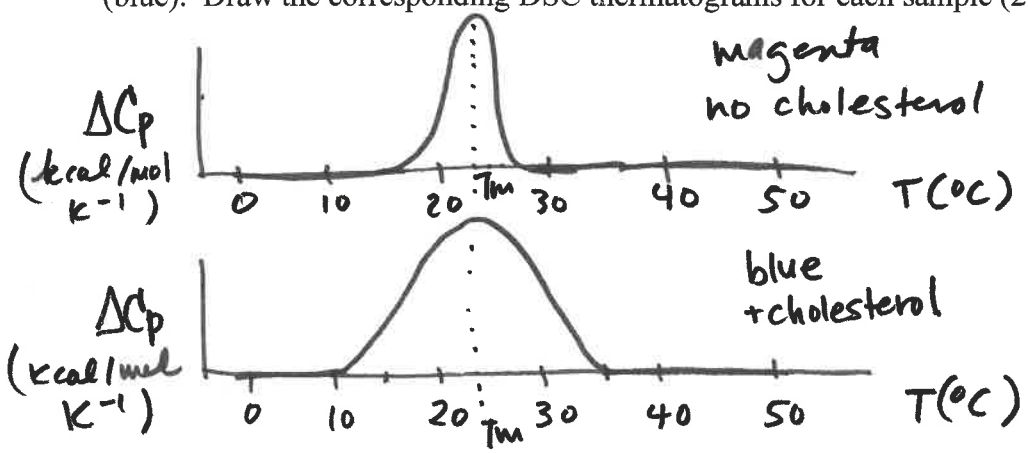
ER: chol/PL = 0.15 (chol is ~10%)

PM: chol/PL = 1.0 (chol is 50%)

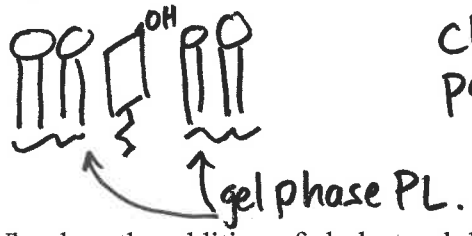
(c) What does cholesterol do to the membrane thickness (1 point)?

increases membrane thickness

(d) Figure 1 shows results from a differential scanning calorimetry (DSC) experiment of membranes with pure phospholipids (magenta) and phospholipids + cholesterol (blue). Draw the corresponding DSC thermatograms for each sample (2 points).



(e) Why does the addition of cholesterol increase membrane fluidity at 15 °C (1 point)?



cholesterol disrupts chain packing & VDW interactions in gel phase.

(f) Why does the addition of cholesterol decrease membrane fluidity at 30 °C (1 point)?

cholesterol orders chains and elongates neighbors promoting VDW interactions in fluid phase

7. Describe how the following factors affect the free energy of micelle formation, and provide an explanation why (6 points total):

(a) Increasing the chain length (2 points)

- increases micelle size and tail interactions
- decreases CMC and free energy (stabilizing)

(b) Increasing the salt concentration (2 points)

For ionic detergents & amphiphiles:

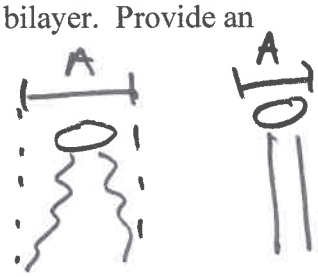
- reduces headgroup repulsion
- decreases CMC and free energy (stabilizing)

(c) Increasing the temperature (2 points)

For nonionic detergents/amphiphiles up to cloud point:

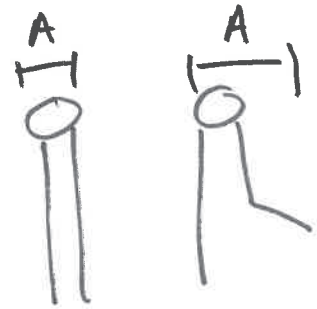
- increases micellar size
- decrease CMC and free energy (stabilizing).

8. Describe how the following factors affect the lipid area in a fluid bilayer. Provide an explanation why (8 points total):



(a) Increasing chain length for saturated chains (2 points)

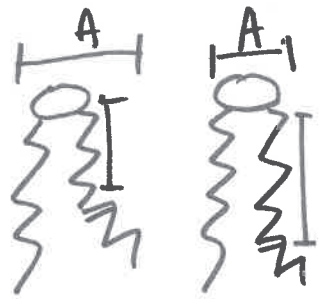
- decreases lipid area.
- due to increas in chain interactions favoring elongation



(b) Saturated vs. unsaturated (2 points)

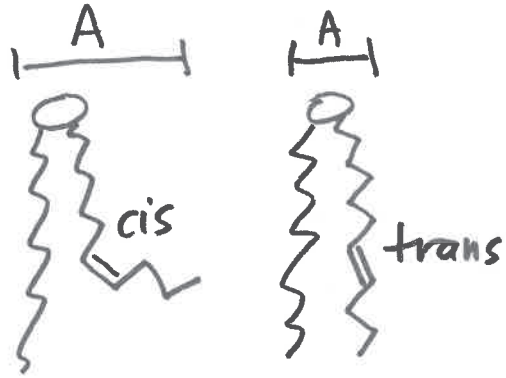
- unsaturation increases area compared to saturated chains
- double bond fixes the chain angle and prevents elongated chains.

(c) For unsaturated chains, moving the position of the double bond (2 points)



- moving the double bond position towards the end of the chain reduces lipid area.
- increases chain contacts favoring elongation

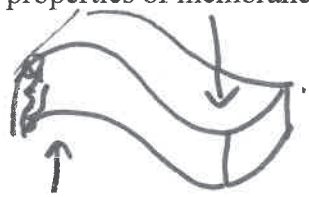
(d) Cis vs. trans unsaturation (2 points)



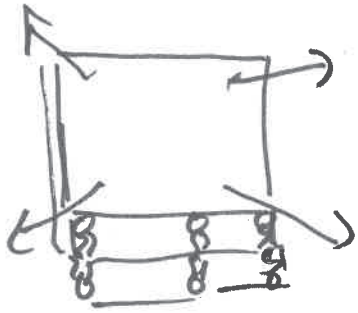
- cis double bonds increase lipid area compared to trans.
- trans locks chain into extended configuration promoting packing

9. What are four material properties of membranes (4 points total)?

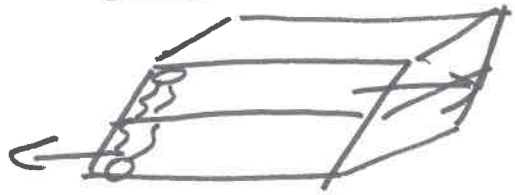
bend



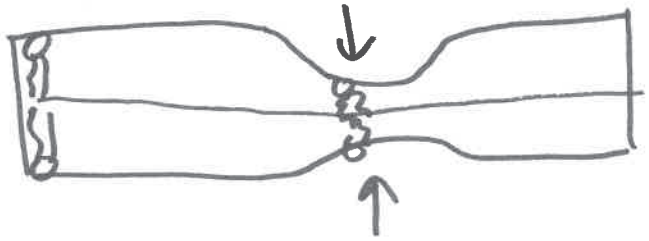
stretch



shear



thichness change.



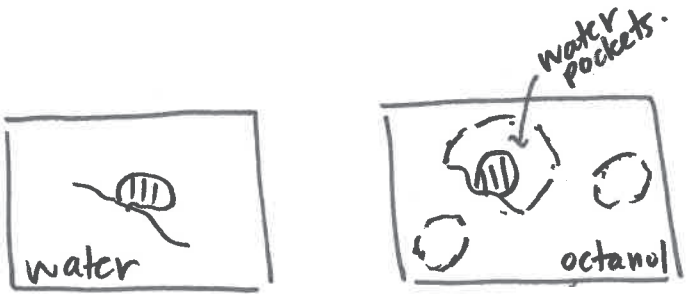
10. Consider the 5 different amino acid partitioning scales that were discussed in class: (i) Radzika, (ii) Wimley, (iii) Moon, (iv) Hessa and (v) MacCallum (20 points total).

(a) Describe the setup of each experiments/calculation. Draw a cartoon depicting the change in states from water to the "lipid" model (10 points).

Radzicka

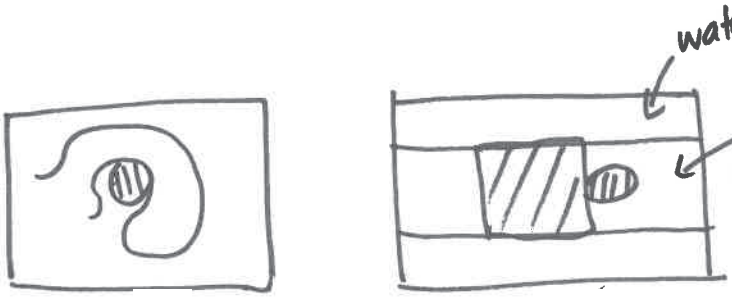
- experiment
- partitioning from isotropic water to isotropic non-polar solvent.

Wimley (octanol) pentapeptide



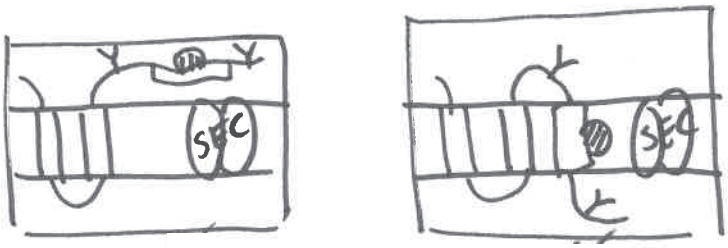
- experiment
- partitioning from water to hydrophobic environment w/ hydrophillic pockets.

Moon & Fleming



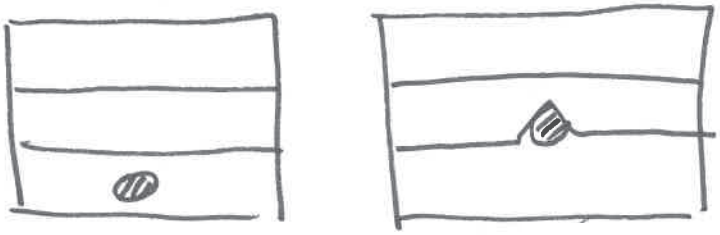
- experiment
- from free energy of folding
- partitioning into membrane core.

Hessa (translocon)



- experiment
- partitioning in cells via translocon
- glycosylation marks water vs. transmembrane.

MacCullum & Tieleman



- computational
- free energy calculation of partitioning from water to lipid bilayer
- molecular defects in membrane structure stabilize charged aas.

(b) The results of each of these methods give different values for the partitioning free energies of side chains from water to lipid bilayers, with the Hessa scale providing the lowest free energies. Provide a ranking of the other 4 methods relative to the Hessa partitioning scale (4 points).

Highest	(1): Radzicka
	(2): MacCallum
	(3): Moon
	(4): Wimley
Lowest	(5): Hessa

(c) Provide an explanation of why the Radzicka method measures larger free energies than the Hessa method (4 points).

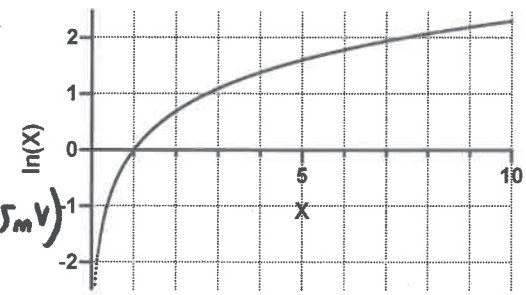
- Radzicka method measures partitioning from water to isotropic, cyclohexane. nonpolar solvent environment
this gives a maximum of the free energy electrostatic penalty
- Hessa measures partitioning/folding via translocon in cells. → environment is not isotropic hydrophobic core.
→ aqueous defects via translocon or bilayer
→ folded protein is present
→ other proteins are present

(d) Often times, hydropathy analysis based on these these partitioning scales incorrectly predicts transmembrane helices in membrane proteins. Provide one reason why and explain (2 points).

- assumes independent amino acid partitioning free energy
- cooperativity exists on partitioning energy because first amino acid does work to create defect in membrane reducing the energy for additional charges.
- does not consider interactions with neighboring protein segments

11. Consider a cell in seawater ([K^+] ~ 10 mM, [Na^+] ~ 500 mM, [Cl^-] ~ 500 mM). What would be the membrane potential of a cell (internal ion concentrations [K^+] ~ 4 mM, [Na^+] ~ 100 mM, [Cl^-] ~ 100mM) if the membrane contained open Na^+ selective leak channels (2 points)?

$$V_M = \frac{RT}{zF} \ln \frac{[Na^+]_o}{[Na^+]_i} = E_{Na^+}$$



assume room temp ($\frac{RT}{F} \approx 25mV$)

$$V_m \simeq \frac{25mV}{+1} \ln \frac{500mM}{100mM}$$

$$\boxed{V_m \simeq +40mV}$$

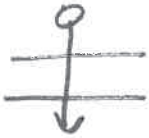
1.61

∴ the membrane potential would be +40 mV if only Na^+ channels were open.

12. Explain why the cell membrane is physical barrier to charged species (2 points total)?

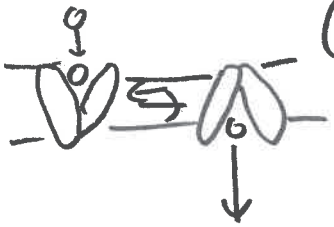
- cell membrane is an electrostatic barrier.
- it is a low dielectric medium
- it cannot solvate charges well compared to water.

13. What are the three main methods of passive transport across membranes? Describe the physical mechanism of each. (6 points total).



① passive diffusion
- diffusion of small, nonpolar neutral species driven by concentration gradients.

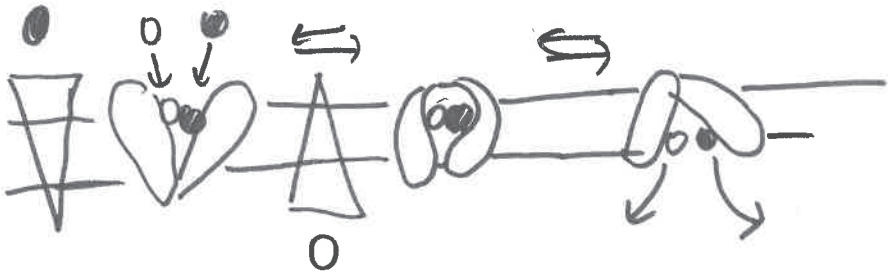
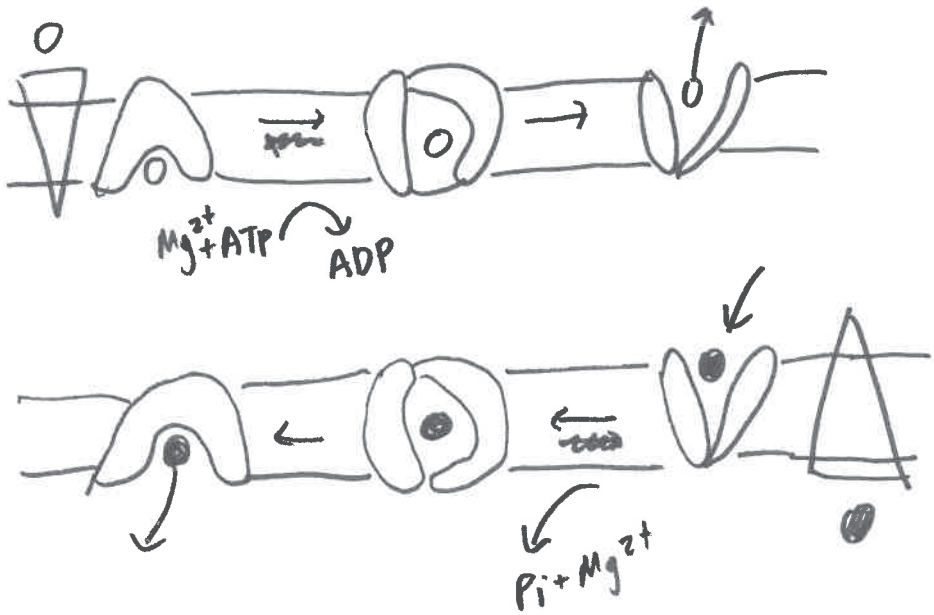
② ion channels
- diffusion of ions driven by electrochemical gradient.



③ facilitated transport/diffusion
- uniporters.
- driven by concentration gradient

14. What are the two main methods of active transport across membranes? Explain what drives the uphill movement of species in each situation (4 points total).

① secondary active transport
facilitated transport.
- cotransport or antitransport.
- uphill movement of one species is driven by downhill movement of other.



② active ATP driven pumps.
- conformational changes driven by ATP hydrolysis or phosphatase activity.
- both species move uphill.