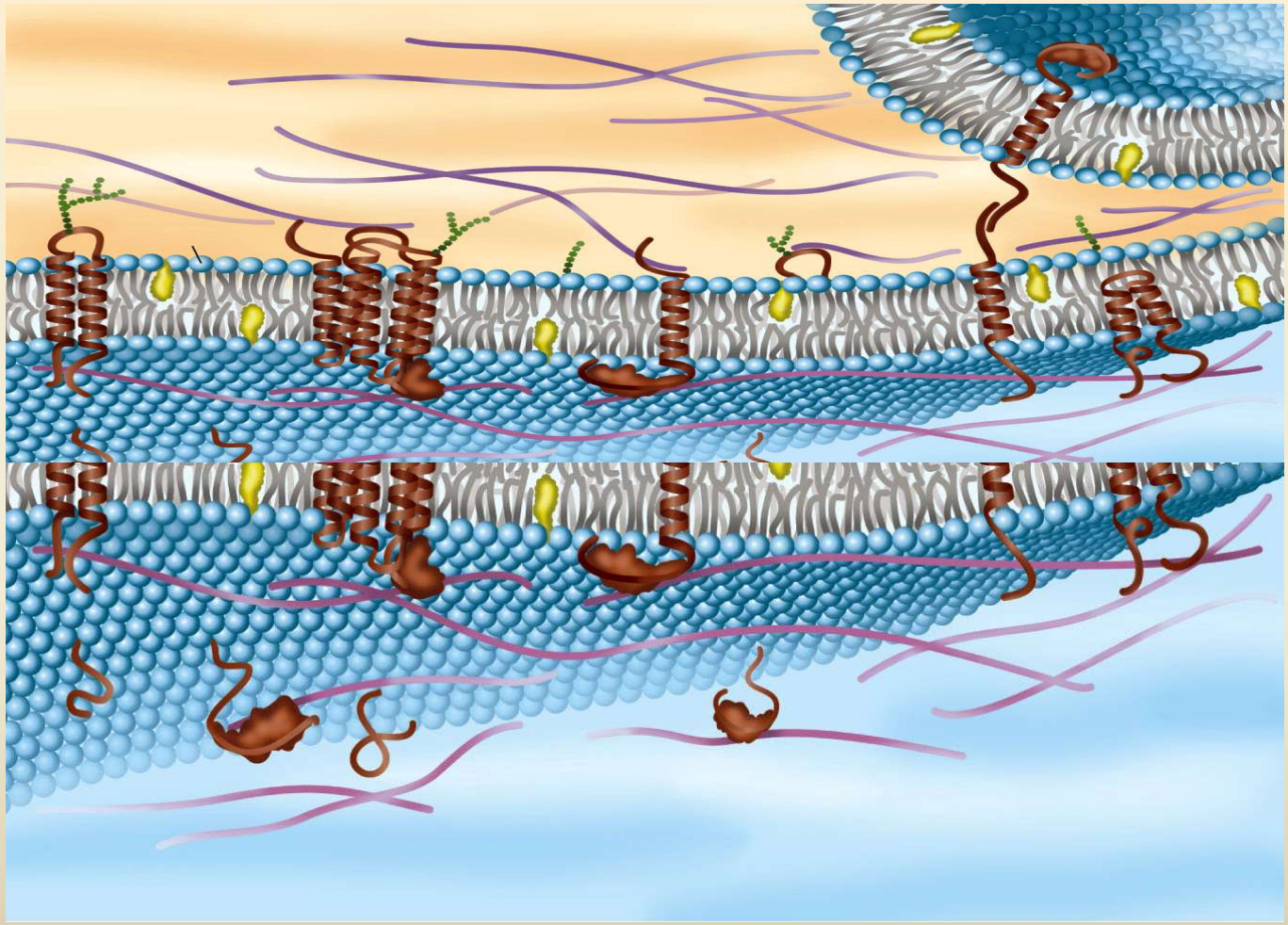
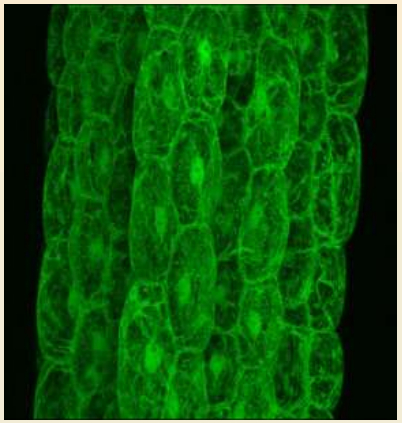
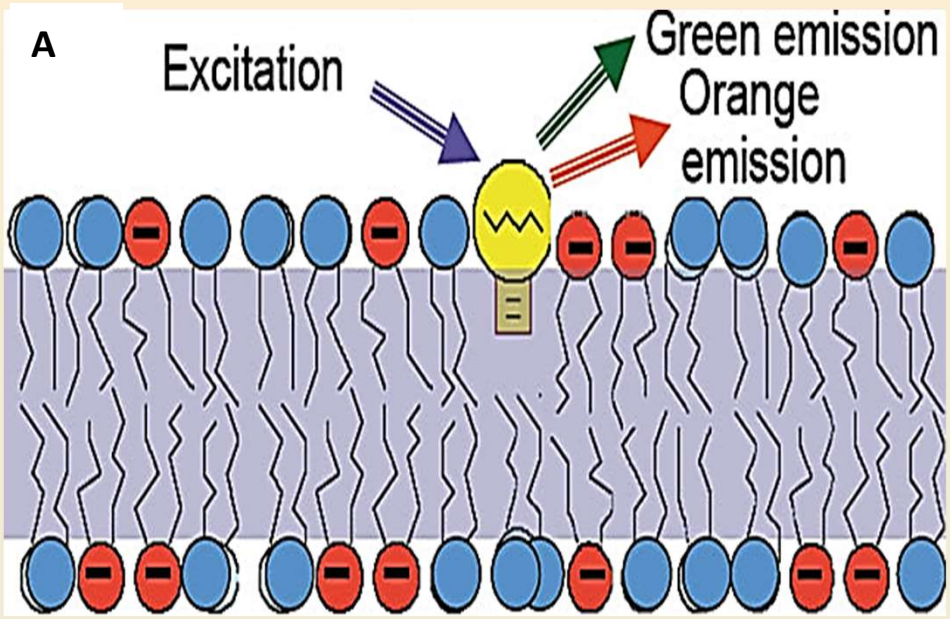
Cell Membranes
Class Activity

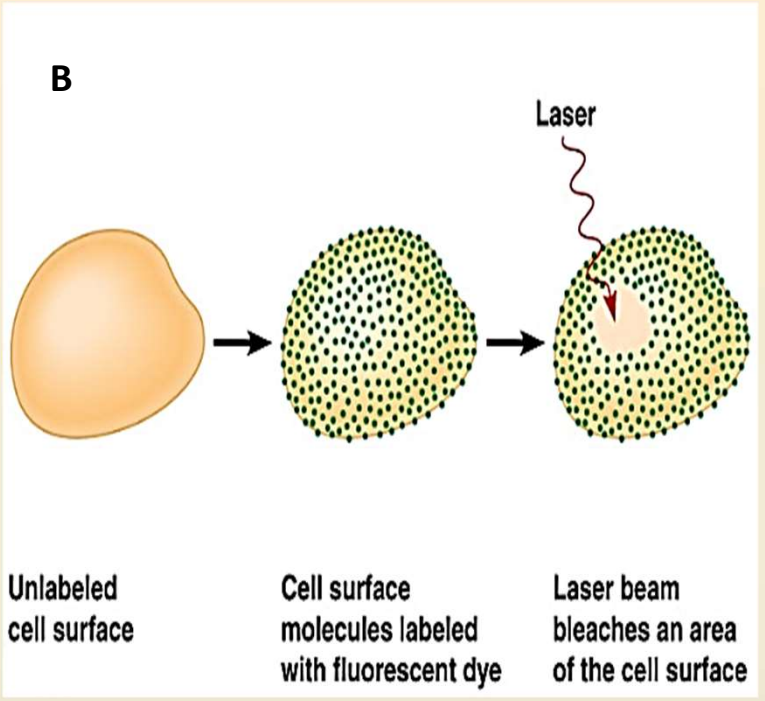


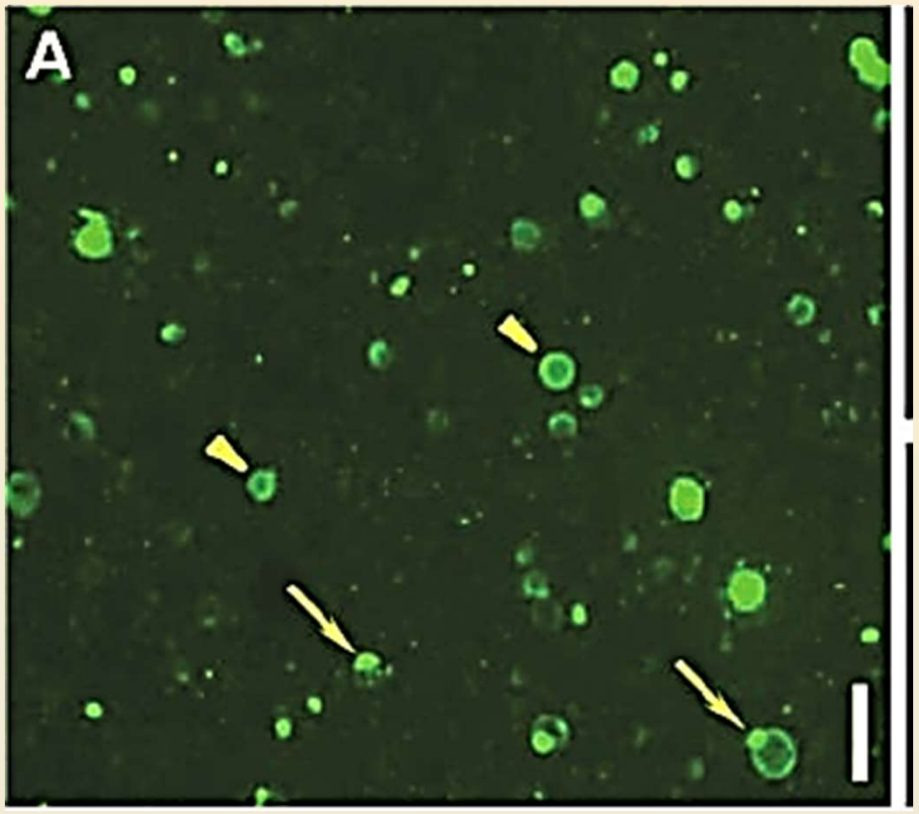
Membrane Fluorescent tags and Fluidity



Membrane lipids can be labelled with fluorescent tags by chemically attaching fluorescent compounds to phospholipids. When subjected to UV light, these fluorescent compounds absorb energy and then release it as light within the visible spectrum of the electromagnetic spectrum. When membranes that are tagged with fluorescent compounds are subjected to UV light, the entire surface of the cell will glow evenly. When shone on a tiny region of the cell, a strong focused laser light causes the denaturing of fluorescent tags and therefore loss of fluorescent activity in the subjected membrane region. Under UV light, this region looks dark giving the impression that there is a ``hole`` in the membrane or as if this region of the membrane has been ``bleached.``


A
Excitation
Green emission
Orange
emission


B
Laser
Unlabeled
cell surface
Cell surface
molecules labeled
with fluorescent dye
Laser beam
bleaches an area
of the cell surface



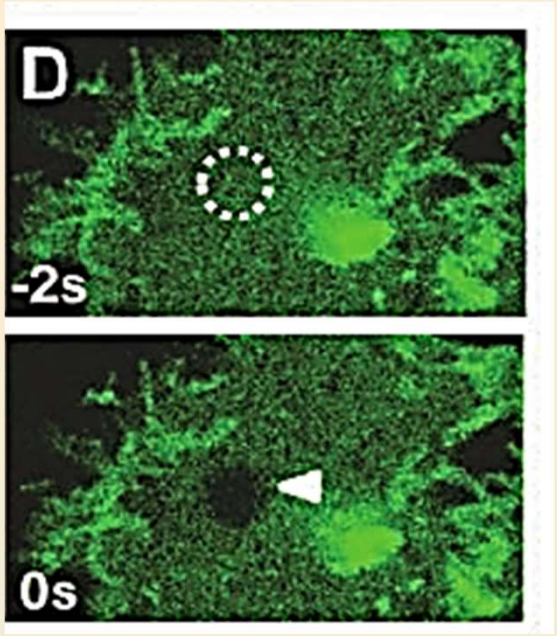
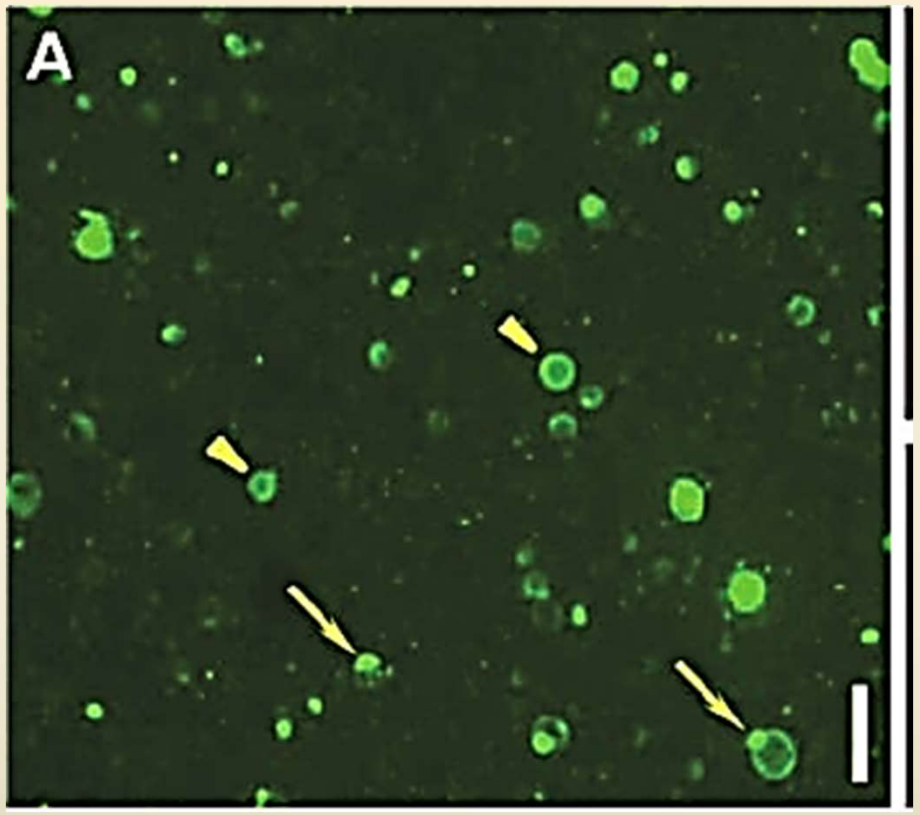
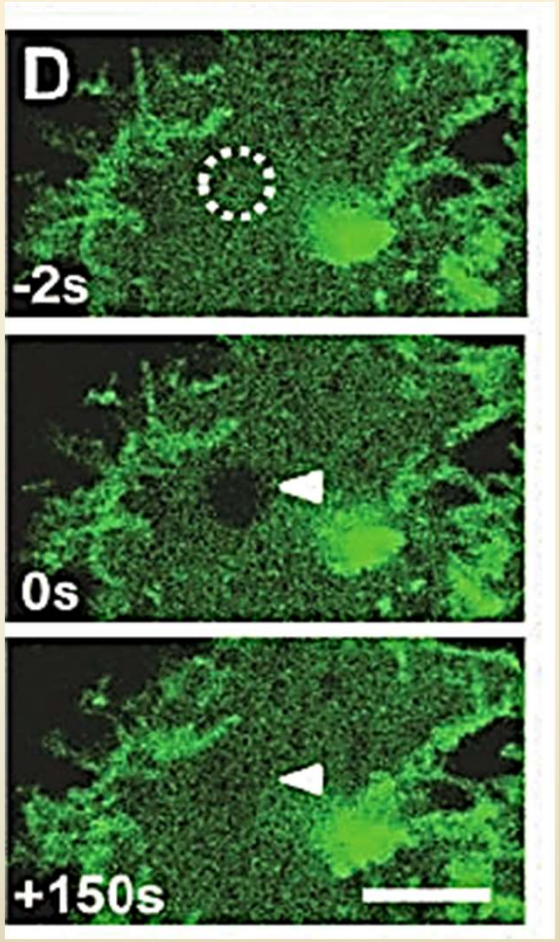


Figure 1. (A) Arrows indicate fluorescent cells whose membranes were treated with fluorescent tags. (D) A cell membrane surface. The dotted-line circle indicates an area that was subjected to a laser beam, before (-2s) and after the "bleaching" of the membrane (0s).

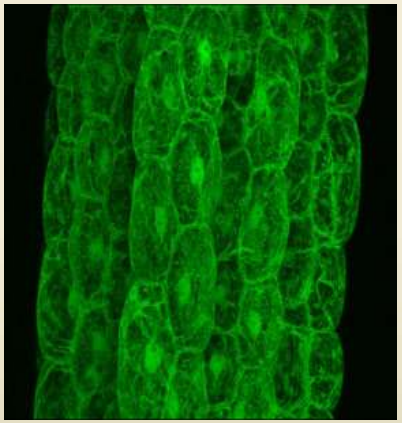
What do you think will happen to the bleached region when laser light is turned off?

A. It will get bigger.
B. It will get smaller, but will not disappear.
C. It will get smaller and completely disappear.
D. It will remain the same.


A


D
-2s
0s
+150s

How can this be used to measure membrane fluidity?



Membrane Fluorescent tags and Fluidity

This table shows data for cells with altered membrane composition. Cell membranes of the different cells were tagged with fluorescent compounds and then excited with UV light. The cells were then subjected to a localized focal laser beam in order to create a bleached region ("hole") that can be examined under the microscope. The time it took the bleached region to become fluorescent again was recorded for each cell type.

Condition	Time (s) for "hole" to become fluorescent
No alteration	**65±7**
↓ length of fatty acid chains	38±2
↑ cholesterol content	88±9
↑ unsaturation of fatty acid chains	42±2
↑ membrane protein content	90±9

Which condition resulted in a membrane with highest fluidity?

A. No alteration

B. ↓ length of fatty acid chains

C. ↑ cholesterol content

D. ↑ unsaturation of fatty acid chains

E. ↑ membrane protein content

Condition	Time (s) for "hole" to become fluorescent
No alteration	**65±7**
↓ length of fatty acid chains	38±2
↑ cholesterol content	88±9
↑ unsaturation of fatty acid chains	42±2
↑ membrane protein content	90±9

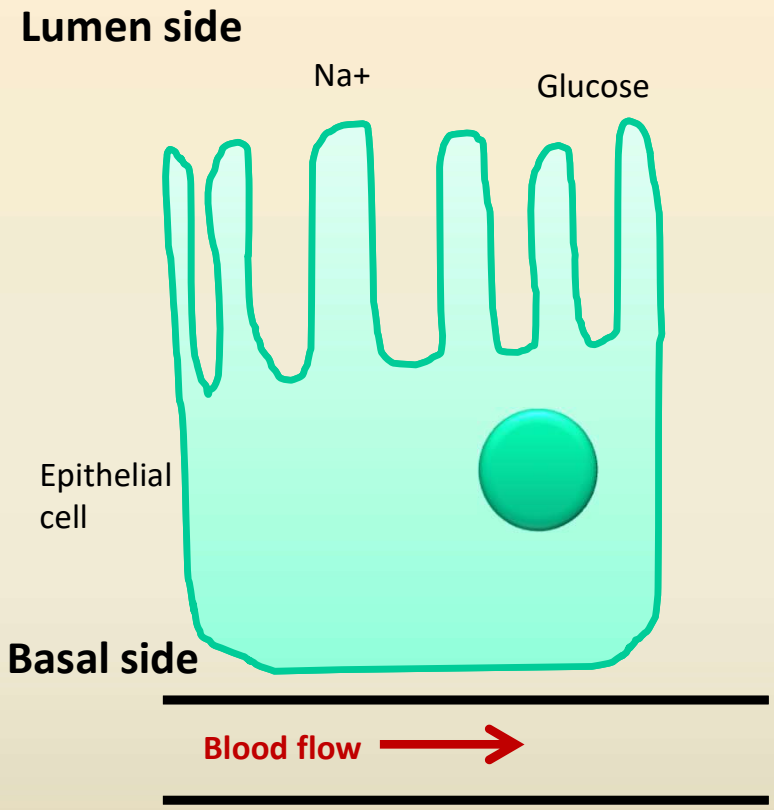
Using a graph, predict the relationship between cholesterol content and membrane fluidity (*use Smart Board if available*).

You've just discovered an organism whose cell membranes are dominated by saturated phospholipids with extremely long hydrocarbon chains. In what type of habitat did you most likely find it?

A. Salty
B. Isotonic
C. Hot
D. Cold
E. None of the above

Location of Membrane Proteins

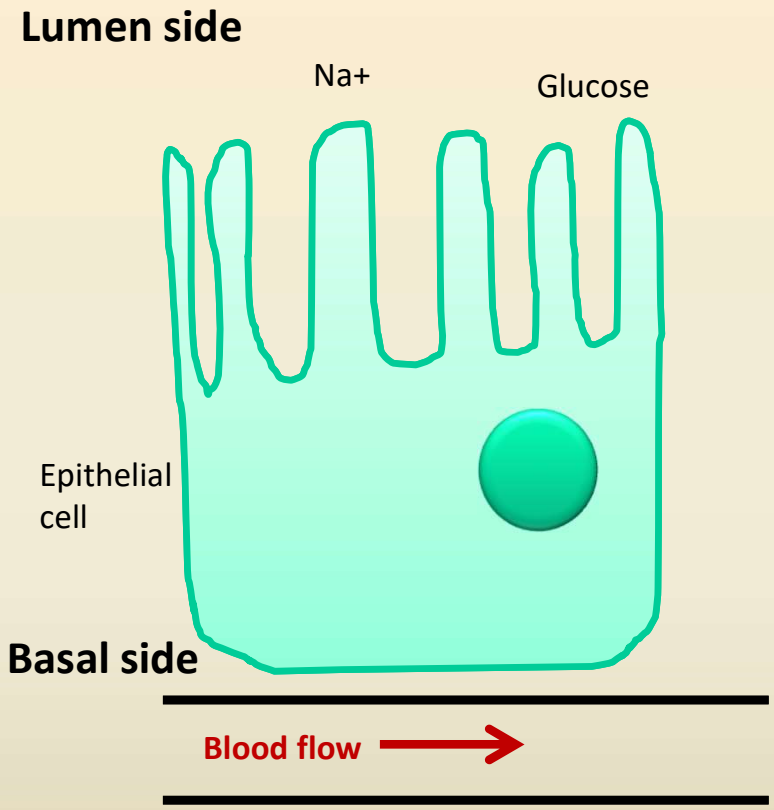
Consider the epithelial cells lining the lumen of your small intestines. Nutrients and dietary minerals, such as glucose and Na+, are absorbed from the lumen side of the intestines, through the epithelial cells, and into blood vessels. For these substances to cross the membranes of epithelial cells, special membrane proteins (pumps, transporters, channels, etc.) are needed to be located strategically on the luminal and/or basal sides of the epithelial cells.



Location of Membrane Proteins

Predict the location of the following membrane proteins on the luminal and basal sides of intestinal epithelial cells and place them on the drawing of the cell shown above (*use Smart Board*). Justify your answers.

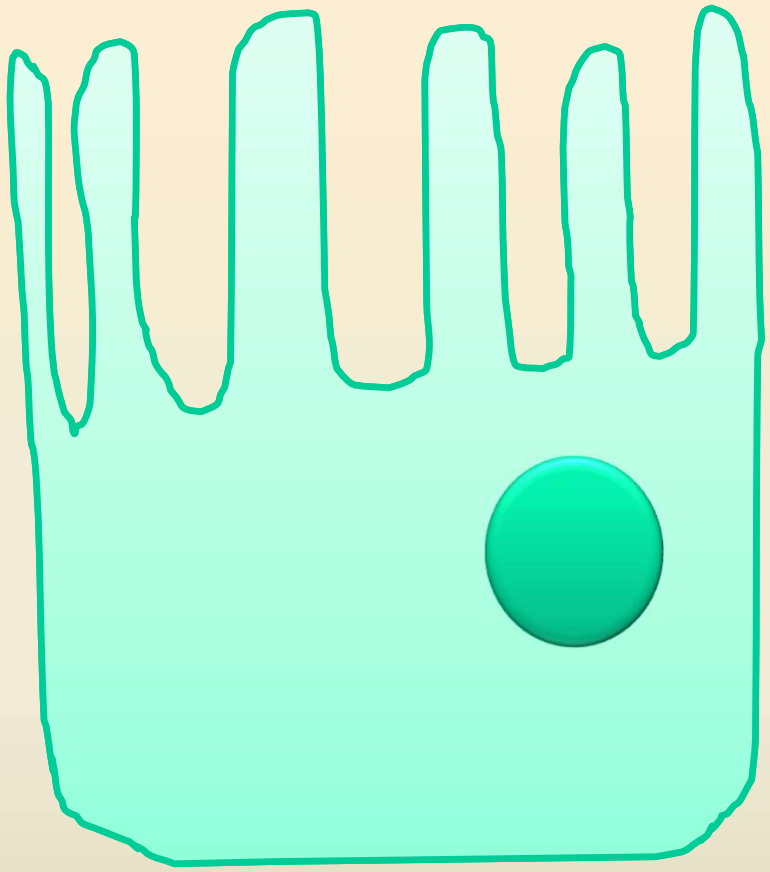
- Na/K pump
- Glucose/Na symporter
- GLUT- 1 transporter



Lumen side

Na+

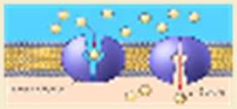
Glucose



Epithelial cell

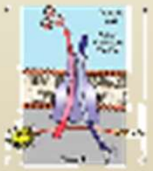
Basal side

Blood flow



GLUT-1

Na+/glucose symporter



Na+/K+ pump

Which of the following types of cell junctions is involved in determining the permanent location of membrane proteins in intestinal epithelial cells?

A. Tight junction
B. Desmosomes
C. Gap junctions
D. None of the above. Cell junctions do not determine the location of membrane proteins.