

Question 1. (18 marks)

For each of the observations below, write a statement that could explain these results, based on what the data shows and on your knowledge of cellular structure/ function (3 marks each)

- A.** If taxol is added to cells at metaphase, the chromosomes do not move towards the spindle poles in anaphase.
- B.** Electron micrographs show that mitochondria in heart muscle have a much higher density of cristae than mitochondria in skin cells.
- C.** Microtubules formed *in vitro* from tubulin that is bound to a non-hydrolyzable form of GTP were found to be exceptionally stable.
- D.** If mRNA for a secretory protein is translated *in vitro* in the absence of any cell membranes, the protein formed was slightly larger than the protein from the same mRNA translated in an intact cell.
- E.** Some membrane proteins can be readily extracted with 1M NaCl, whereas others require the use of an organic solvent or detergent.
- F.** The pH of the chloroplast thylakoid space (or lumen) decreases in light.

Question 2. (8 marks)

When the gene that encodes for Green Fluorescent Protein (GFP) is introduced into cells, GFP is expressed and localized in the cytoplasm of the cell. What specific sorting signals would you have to add to the GFP protein genetically to produce a protein that is targeted to the following final cellular destinations?

| | Sorting signal(s) to target the GFP to this location: |
|---|---|
| A. The nucleus | |
| B. The lysosome | |
| C. The lumen of the ER | |
| D. The stroma of the chloroplast | |

Question 3. (6 marks)

In this course we have used the term polarity in many different contexts; three examples are given below. For each context in the table, explain the meaning of the word 'polar' or 'polarity', and explain how polarity contributes to the function of each example.

| Context/example | Definition of 'polarity' in this context (1 mark each) | How this polarity contributes to the function of this example (1 mark each) |
|--|--|---|
| A. Polarity in a macromolecular polymer (such as a protein or nucleic acid) | | |
| B. Polar amino acid side chains | | |
| C. Polar microtubules | | |

Question 4. (10 marks)

You have discovered a new toxin, toxin X, which kills parasitic fungi.

A. You hypothesize that this toxin affects mitosis (M-phase). How would you test this hypothesis? (2 marks)

B. You find that the cells were not blocked in M-phase. In addition, you discover that this toxic chemical prevents phosphorylation of the fungi's M-cyclin dependent kinase (M-CDK). At what stage of the cell cycle would the fungal cells arrest if toxin X prevents M-CDK phosphorylation? (1 mark)

C. Design an experiment to test at what stage of the cell cycle the fungi arrest. State your hypothesis, experimental design and expected results. (7 marks)

Question 5. (7 marks)

The transferrin receptor is a transmembrane protein involved in the uptake of iron by receptor-mediated endocytosis.

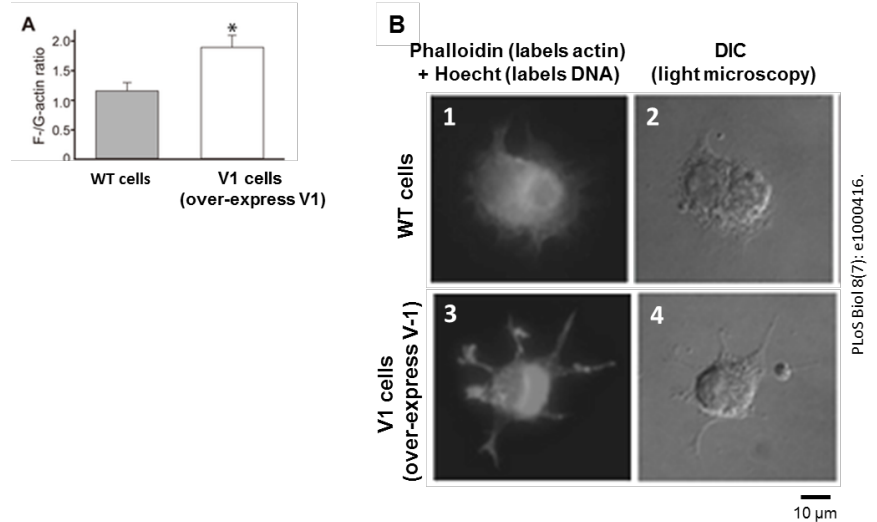
A. Describe the localization pattern you would expect to see in a normal cell by immunofluorescence microscopy if you use a fluorescent antibody against the transferrin receptor.

B. How would this labeling pattern be changed in a mutant cell in which the transferrin receptor fails to bind to adaptin/clathrin complexes? (2 marks) Why? (1mark)

Question 6 (11 marks)

Actin Capping Protein (ACP) binds to the plus end of actin filaments, preventing the actin filaments from gaining or losing monomers. Its activity is blocked by regulatory proteins such as V-1.

In this experiment, Takeda et al. (2010) examine the role of V-1 and ACP in the regulation of actin polymerization. In Panel A, the ratio of F-actin (actin filaments) to G-actin (actin monomers) were measured for WT (normal) and V1 (over-expresses V-1) cells. In Panel B, actin filaments and nuclei were stained with fluorescent dyes and cells were examined through fluorescence and light microscopy. Assume that both cell lines express the same amount of ACP.



A. Describe the data shown in A (1 mark) and explain what they mean (2 marks).

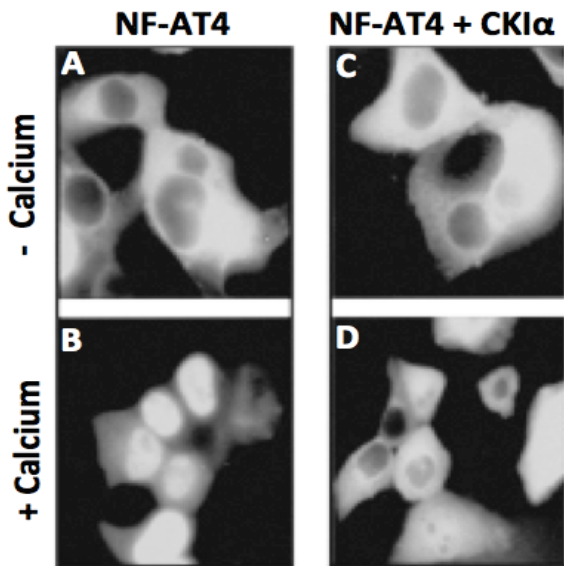
B. Describe the data for each panel shown in B (2 marks) and explain what they mean (2 marks).

C. Based on the data shown in A and B, what accounts for the changes in cell shape that are seen between the WT and V1-over-expressing cells? (2 marks)

D. What can you conclude about the role V-1 has in actin polymerization? (2 marks)

Question 7 (10 marks)

NF-AT4 is a transcription factor that is regulated by calcium. Here, Zhu et al. (1998) use GFP-tagged NF-AT4 to establish that NF-AT4 function is also regulated by CKI α kinase, a protein that adds phosphate groups directly to NF-AT4.



Adapted from Cell 93:851

A. Consider panels A and B. Describe what the data show and explain the effect that the addition of calcium has on the NF-AT4 protein. (3 marks)

B. Consider panels C and D. Describe what the data show and explain how the presence of CKI α impacts the effect of calcium on calcium-dependent NF-AT4. (3 marks)

C. Based on the data above propose a model for how calcium and CKI α can influence the localization of NF-AT4 in these cells? (2 marks)

D. Why would addition of a phosphate group to NF-AT4 alter its function? (2 marks)

Question 8 (Outline. 20 marks)

Each organelle in the cell is a unique microenvironment where certain cellular functions are performed. Write an essay outline to critically assess the following statement: "The intracellular areas of low pH are essential to the function of some organelles." Your arguments (and associated supporting evidence) should provide examples from 3 different organelles.

Thesis statement:

Argument 1 and evidence:

Argument 2 and evidence:

Argument 3 and evidence: