

Systems Microbiology 1.084J/20.106J PROBLEM SET #1 – Solutions

Problem 1

a. Describe the different sorts of evidence for life's appearance on the early Earth ~ 3.5 bya. The evidence for this phenomenon is isotopic record, rocks and microfossils, organic geochemical records, and molecular evolution. Microfossils exist that look as though they could be from cyanobacteria. Stromatolites have been formed from cyanobacteria and other algae binding sediment together forming 'bio-layers'. Carbon isotope signatures from 3.4 billion year old mat-like structures suggest carbon dioxide fixation. The presence of stable carbon and oxygen isotopes also suggest bacteria were producing large quantities well before other oxygen-consuming organisms were present. Molecular signatures, such as steranes and 2-methyl hopanes, only known to be produced by eukaryotes and cyanobacteria, respectively, have been isolated from extremely old rock. The presence of iron beds is another clue to the presence of bacteria changing the atmosphere over from mainly carbon dioxide to more nitrogen and oxygen.

b. Which evidence seems most compelling to you, and why?

Answers will vary.

c. What dramatic change ~ 2.5 billion years ago has influenced biology on Earth ever since, and how did it happen?

Great Oxidation event. Bacteria evolved that were capable of producing oxygen via photosynthesis, cyanobacteria. They pumped (and continue to pump) large amounts of oxygen into the atmosphere.

Problem 2

a. Describe how you might determine the stable isotope compositions of different compounds in inorganic, as well as organic or living materials.

Isotope ratio description: compare ratios of heavy and light isotopes of compounds.

b. What elements might be useful for investigating biological processes using stable isotope ratios?

Hydrogen, carbon, nitrogen, oxygen and sulfur.

c. Describe in detail the metric that's used to differentiate stable isotope content of a specific element in different compounds.

Isotope ratios, R. $R = \text{heavy isotope} / \text{light isotope}$. Mass spectrometry is used to compare the isotopic ratio of a sample to a known standard:

QuickTime™ and a
TIFF (LZW) decompressor
are needed to see this picture.

Problem 3

a. Draw the general structure of gram negative and gram positive bacterial cell walls.

b. What gives these cell walls their structural integrity, how, and why is that important.

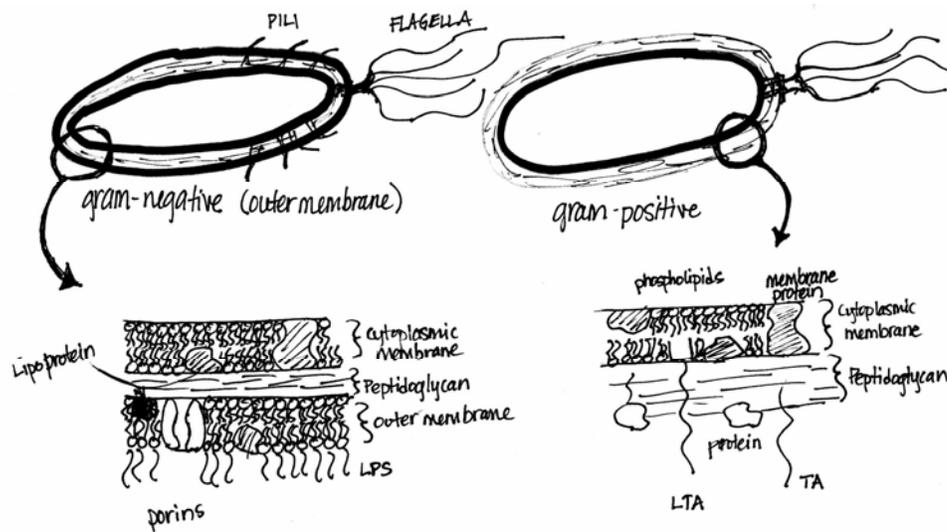
Structural integrity of bacterial cell walls is provided by peptidoglycan crosslinking, which is important for stability and flexibility of the cell to withstand stress/force.

c. What are the major differences, and their biological implications?

Gram-positive: no outer-membrane thus cell wall consists of only wall-anchored proteins, teichoic acids & lipoteichoic acid in the peptidoglycan layer.

Gram-negative: outer-membrane + peptidoglycan layer (periplasm); the outer-membrane consists of lipopolysaccharide, porins, lipoproteins, and phospholipids.

Biological implications: (reasonable answers accepted) The outer-membrane of gram-negative organisms allows space for many enzymes to reside outside the cell membrane such as chemo receptors and transporters, as well as porins on through the outer-membrane. At the same time, endotoxins due to lipid A of LPS are present in gram-negative organisms.



Problem 4

- Describe the various functions of the cell membrane in bacteria. The cytoplasmic membrane is important for anchoring proteins, acting as a permeability barrier, and for energy conservation.
- Name three different classes of transport systems. Uniporters, symporters, and antiporters
- Describe the three different functional types of transport systems, and their general similarities and differences.
 - Simple transporters: involve only a membrane-spanning component (ie Lac permease transporter)
 - Phosphotransferase-type transporters: involve series of proteins that cooperate in the transport event (ie phosphotransferase system)
 - ABC systems: involve a membrane-spanning component, a periplasmic-binding component and ATP-hydrolyzing proteins (kinases).

Problem 5

There exists a motile marine cyanobacterium that can swim at speeds of 80 $\mu\text{m}/\text{sec}$, but no one has ever identified its flagella, despite repeated attempts. Its mode of motility still remains a mystery, but its Reynolds (R) number is not. Assuming a swimming speed of 80 $\mu\text{m}/\text{sec}$, and a cell radius of 2000 nm, calculate R for this bacterium. How does this compare to *E. coli*?

$R = a \cdot v \cdot \rho / \eta = a \cdot v / \nu$; where a is the length, ν is kinematic viscosity, ρ is fluid density, v is velocity, and η is fluid viscosity.

$$R = 2000\text{nm} \cdot 80,000\text{nm}/\text{sec} \cdot 1/10^{12}\text{nm}^2/\text{sec} = 1.6 \times 10^{-4}$$

R is comparable to *E. coli*.

Problem 6

- Draw and describe the structural and functional components and properties of bacterial flagella, and their general mode of synthesis. Flagella are long, spiral-shaped rods anchored in the cytoplasmic membrane, and are attached to the cell surface by a complex of flagellar motor proteins, which include L & P rings anchoring the hook to the outer membrane and peptidoglycan, Mot proteins, MS and C rings that surround the Fli proteins (motor switch) located in the cytoplasmic membrane. There is a hook in the flagellum filament that adjoins the flagellum and flagellar motor. Flagellum is a multimer of the protein flagellin. Synthesis begins with the motor proteins, then the hook ending with flagellin monomers to grow the flagellum.
- If you were to follow the synthesis of flagella with a "pulse chase" experiment, by adding first unlabelled amino acids followed by radio-labeled amino acids, what pattern of labeling during flagellar biosynthesis would you expect to see? (A, B, or C) **A**, flagellin monomers added to tip, not base.