Study guide 3

Additional Resources:

Readings from Madigan and Martinko, 2006, Biology of Microorganisms
pp. 160-164, 371-383, 426-434 and 557-577.

Readings from Staley et al. 2007, Microbial Life
Chapter 8 and other sections as appropriate.

The Prokaryotes, vol. 1; chapter 2 (Stackebrandt)

<u>Questions from Study Guide 2:</u>

17. Describe the nitrogen cycle on earth, including the microbial groups important at each step and the oxidation state of N.
18. What are the major phylogenetic groups of nitrifying bacteria? What about the anaerobic nitrifying bacteria?
19. Compare the energetics of ammonia and nitrite oxidation. How do nitrifying bacteria reduce NAD^+?
20. What are three rationales for the poor growth of the nitrifying bacteria?
21. How much nitrite must be oxidized to reduce NAD+? Why is so much required?
22. Where is the Anammox reaction found? Be able to describe the pathway of ammonia oxidation, the electron donor and electron acceptor.

Define, distinquish, and/or describe the following terms:
nitrification
nitrogen cylce
nitric oxide
nitrous oxide
nitrate
nitrite
ammonia
nitrifying bacteria
ammonia-oxidizing bacteria
Nitrospira
ammonia monooxygenase
hydroxylamine oxidoreductase
nitrite-oxidizing bacteria
nitrite oxidoreductase
K_s
Anammox
candidatus
hydrazine-cytochrome c oxidoreductase

Study Guide: H_2 and CO oxidizing bacteria using O_2 as an electron acceptor

1. The bioenergetics under standard conditions for the oxidation of H_2 and carbohydrates are very similar. Why do most H_2-oxidizing bacteria utilize reverse electron transport for NAD^+ reduction?
2. What are the standard conditions for a reaction? How is the free energy of a reaction calculated under conditions that are not the standard conditions?
3. How is selenocystiene incorporated into proteins?
4. What is the ecology of H_2-oxidizing bacteria? How is the physiology dependent on the ecology?

Define, distinquish, and/or describe the following terms:

knallgas bacteria
hydrogenase
selenocystiene
SelB
carbon monoxide dehydrogenase
molybdenium-pterin coenzyme
membrane-bound hydrogenase
Calvin cycle
reverse citric acid (TCA) cycle
hydroxypropionate cycle
Winogradsky

Microaerophiles and mechanisms of O_2 toxicity

1. Two organisms grow in the presence of air. One is an aerotolerant anaerobe and the other is a facultative aerobe. How might you tell which is which?
2. What is the rationale for magnetism in the magnetotactic bacteria?
3. Describe the magnetosome, what is it composed of and where it is found in the cell.
4. What is the difference between N polar and S polar magnetotactic bacteria?
5. Describe the reactive intermediates formed during the reduction of O_2. In the cell, what initiates the formation of these compounds?
6. What are the mechanisms of O_2 toxicity? How do superoxide dismutase, catalase, and peroxidase protect against O_2 toxicity?
7. Describe the methods bacteria protect themselves against O_2 toxicity. Why might anaerobes be especially O_2 sensitive?

General anaerobic respirations

1. Compare the type of microbial interactions found in the anaerobic and aerobic environments.
2. Why are some electron acceptors used before others in the anaerobic environment? What is the order in which these acceptors are utilized.
3. Why must there be fundamental differences in how energy is made during anaerobic respiration in the sulfate-reducing bacteria, methanogens, and homoacetogens on one hand and the denitrifiers and iron-reducing bacteria on the other hand.
4. Given the electron acceptors, know the reduced product: Mn^{+4}, Fe^{+2}, NO_3^-, SeO_4^{-2}, dimethylsulfoxide, trimethyl-N-oxide, AsO_4^{-3}, fumarate, S^0, SO_4^{-2}, and CO_2.
5. How can anaerobic respiration be distinguished from facilitated fermentation?
6. Compare the phylogenetic diversity of denitrifying bacteria, iron-reducing bacteria, sulfate-reducing bacteria, homoacetogens and methanogens. Can this diversity be explained by the biochemical specializations necessary for each of these types of metabolism? Support your answer, yes or no.
7. Compare the carbon and energy sources utilized by the sulfate-reducing bacteria, the homoacetogens, and the methanogens. Which of these are generalists and which are specialists.
8. In general terms, compare fermentations to anaerobic respiration in terms of the transfer of electrons and the biosynthesis of ATP.

Iron-reducing bacteria

1. Describe the diversity of iron-reducing bacteria. Are they common in more than one bacterial phylum? What kinds of electron donors are used? Do they use other electron acceptors?

Denitrifying bacteria

1. Compare the pathways of denitrification, sensu stricto and sensu lato, to nitrate assimilation.
2. Compare the midpoint potential of the NO_3/N_2 couple to the O_2/H_2O couple and discuss how it affects the bioenergetics of denitrification and the electron transport chain in denitrifying bacteria. How do denitrifying bacteria utilized multiple electron acceptors at the same time?
3. Why are these bacteria important for the treatment of wastewater?

Sulfate-reducing bacteria

1. Describe the distribution of sulfate-reducing activity, SO_4^{-2}, H_2S, FeS, and Fe_2S in marine sediments.
2. Compare the pathways of assimilatory and dissimilatory sulfate reduction. How do these organisms reduce sulfate when that reaction is very unfavorable? What is a biochemical rationale for the differences?
3. Why is pyrophosphate hydrolysis important in sulfate reduction?

4. Compare how sulfate-reducing and sulfur-reducing bacteria generate a PMF. How do these compare to how a denitrifier does the same thing.
5. Learn the TCA cycle! Describe the modifications observed in some sulfate-reducing and sulfur-reducing bacteria. For each modification, give some biochemical rationale. Know the unique enzymes to the modified pathways.
6. How do some sulfur-reducing bacteria oxidize succinate?
7. Describe how sulfate-reducing bacteria promote steel corrosion.

Homoacetogenic bacteria

1. How do homoacetogens make a living during growth on H_2 + CO_2?
2. Describe the tetrahydrofolate pathway of acetate oxidation and synthesis. Include the reactions catalyzed by acetyl-CoA decarbonylase/synthase and formate dehydrogenase, reactions dependent on ATP, and reactions that might be coupled to the PMF. You do not have to know every step in the pathway, but you should know the oxidation state of the carbons from acetate through the pathway.
3. Compare how the tetrahydrofolate pathway is utilized during growth on H_2 + CO_2, sugars, and aromatic compounds (such as vanillin).
4. Describe the reactions that are common in anaerobes leading from pyruvate to acetate.
5. Describe the processes that lead to acetate biosynthesis in a soil particle.

Methanogenic archaea

1. Contrast the three types of methanogenesis.
2. In general terms, compare the pathways of methanogenesis and acetogenesis from H_2 + CO_2.
3. For each of methanogenic coenzymes, know their general function (electron carrier, C-1 carrier, etc.) and something about their structure (deazaflavin, Ni-containing tetrapyrrole).
4. How does the threshold concept explain why certain electron acceptors are used first in anaerobic environments.
5. What is the biochemical rationale for the lower threshold in *Methanothrix* than *Methanosarcina*?
6. Write something educated about the distribution of homoacetogens and methanogens in the guts of termites.
7. Are methanogens generalist or specialists when compared to the homoacetogens? Why?

Fermentations

1. During the fermentation of glucose, know how many ATPs and reducing equivalents are produced from the biosynthesis of the various fermentation products: lactate, acetate, ethanol, formate, CO_2+H_2 , succinate, butyrate, butanol, acetone, 2-propanol, acetoin, diacetyl, and butanediol.
2. In glycolysis, what steps are involved in coupling to the consumpt ion or production of ATP or NAD^+ reduction?
3. Know the major types of high energy bonds and some representative compounds involved in

substrate level phosphorylation.

4. For the following fermentations, describe the ‘oxidative’ and ‘reductive branches (you do not have to know the pathway but should be familiar with the key intermediates, products, and steps involved in energy generation): propionate fermentation of lactate, propanediol fermentation of glycerol, fermentation of glutamate to propionate and acetate.
5. Be able to calculate the carbon recovery and oxidation/reduction balance for a fermentation given the moles of substrate consumed, moles of product formed, and the structural formulas for the substrates and products.
6. For the fermentation table below, fill in the missing values indicated by a *.

substrate/ product	formula	t_0 (umol/ml)	t_f (umol/ml)	delta t (umol/ml)	C-mol	redox #	mol amt R/O
substrate:							
glucose	$C_6H_{12}0_6$	110	10	*	*	*	*
products:							
ethanol	C_2H_6O	10	210	*	*	*	*
total CO_2	CO_2	25	225	*	*	*	*
biomass	CH_2O	50	50	*	*	*	*
		substrate/product=			*		*

<u>Interspecies H_2 transfer</u>

1. How does H_2 production and consumption in the anaerobic environment affect the pathway of degradation of organic matter? How does the H_2 concentration affect the fermentation pathway of *Clostridium thermocellum* and *Selenomonas ruminantium*?
2. Why is coculture with a H_2-consuming organism necessary for anaerobic growth of a syntrophic bacterium on butyrate and propionate? How does this affect the amount of free energy available to each organism?
3. How does *Syntrophomonas wolfei* oxidize butyrate? Why can it grow in the absence of a H_2-utilizing bacterium on crotonate?

Define, distinquish, and/or define the following terms: [for the enzymes: know the reaction catalyzed, the organism in which they are found, their physiological role, and anything of special interest-- like an unusual cofactor; for the coenzymes: you do not have to know the chemical structure, but you should be able to describe the structure in words, describe the types of reactions in which it participates, and describe the organisms that possess them]

microaerophile
capnophile
aerobe
anaerobe
aerotolerant anaerobe
obligate anaerobe
strict anaerobe
facultative aerobe
magnetotactic
magnetosome
magnetite
greigite
Fe_3O_4
MamB
triplet O_2
singlet O_2
superoxide
O_2^-
hydrogen peroxide
H_2O_2
hydroxyl radical
$OH^{\cdot}$
free radical
superoxide dismutase
catalase
peroxidase
carotenoids
glutathione
methoxylated compounds
vanillin
methanofuran
coenzyme F_{420}
coenzyme F_{430}
coenzyme M
HS-HTP
H_2 threshold
acetate threshold
fermentation
oxidative branch
reductive branch
respiratory protection
nitrate reductase
nitrite reductase
nitric oxide reductase
nitrous oxide reductase
food web
food chain
primary consumers
intermetabolic group
anaerobic respiration
facilitated fermentation
dissimilatory sulfate reduction
assimilatory sulfate reduction
APS
PAPS
Desulfotomaculum
Thermodesulfobacterium
Archaeoglobus
citrate synthase
ATP-citrate lyase
dye-linked malate dehydrogenase
tetrahydrofolate
tetrahydromethanopterin
acetyl-CoA decarbonylase/synthase
formate dehydrogenase
Mo-pterin
W-pterin
lignin
Stickland reaction
acyl thioester
phosphoenol ester
phosphoanhydride
phosphoacyl anhydride
pyrophosphate
phosphotransferase
interspecies H_2-transfer
syntroph