

**Lesson 1 (31 October 2000)**

Welcome and introduction to class.

Attendance sheet.

Overview of syllabus topics.

Discuss what we need to know about essential and toxic metal ions in biological systems.

Inorganic Biochemistry: Transport and storage, introduction (Chapter 3, pp. 133-136).

Discuss xerox table of metal ions.

Discuss solubility and redox properties.

Discuss how microbes interact with metal ions, metal binding and storage (sequestration and precipitation), membrane transport (import and export), and chemical transformation (volatilization).

Role of chromium in glucose-tolerance factor and insulin-binding protein.

Methanogens and nickel enzymes.

Discuss archaea, eubacteria, and eucarya.

Discuss thermophiles, halophiles, and auto- and heterotrophy.

Discuss methane production (IB Chapter 5, pp. 216-219, 226-227; papers).

Discuss nickel metalloproteins and enzymes, active uptake of  $\text{Ni}^{2+}$  and incorporation into factor F430.

Post lecture comments - I was introduced again by one of the administrators, to a small group of four participants. We then waited until 6:25 p.m. to begin class, which had 5 people in attendance. Within 20 minutes there were approximately 10 people in class. I proceeded slowly covering important aspects of metal solution and redox chemistry. Most of the participants were in the first course and they did not remember most of the information that was presented during those sessions based on the questions that I asked in class tonight. I do not need to give exams to determine that fact. We took a 20 minute break at 7:45 p.m. and then started up again for another hour of lecture. I will finish the topic of methanogens next time and begin a discussion of electron transfer and cytochrome *c* oxidase and proton pumping. I have real doubts about the intellectual abilities of most of the participants.

**Lesson 2 (3 November 2000)**

Attendance sheet.

Review previous lecture (IB Chapter 3, pp. 133-136; Chapter 5, pp. 216-219, 226-227).

Role of chromium in glucose-tolerance factor and insulin-binding protein.

Inorganic Biochemistry: Metalloproteins and metalloenzymes: (II) Redox chemistry, methanogenesis, protein-protein electron transfer (Chapter 5, pp. 216-219, 226-227, 242-247).

Discuss autotrophy and nickel enzymes (slides and papers).

Discuss the role of metal- and ligand-centered redox with hydrogenase enzyme (papers).

Discuss paradox of metal ion uptake and sulfide requirement by methanogens (metal sulfide production).

Discuss proton-coupled electron transfer in ferredoxin I system.

Post lecture comments - Class went well tonight with both slides and multimedia presentation. The professor from Cajamarca was there and will come each Friday for class. There were approximately 10 people in attendance including one of the administrator/faculty who spent time in the United States. I finished what I intended to cover tonight and postpone cytochrome *c* oxidase until Tuesday's class. We will have multimedia once per week as a result of one of the projectors having been stolen. I will use a computer screen on the other nights. The course is shaping up with the use of recent literature citations to supplement the textbook.

**Lesson 3 (7 November 2000)**

Attendance sheet.

Review previous lecture (IB Chapter 5, pp. 216-219, 226-227, 242-247)

Inorganic Biochemistry: Case studies, cytochrome *c* oxidase (Chapter 10, pp. 389-400).

Fundamentals of Biochemistry: Electron transport and oxidative phosphorylation, electron transport (Chapter 17, pp. 497-511).

Discuss proton-coupled electron transfer in ferredoxin I system (X-ray structure paper).

Cytochrome *c* oxidase - low-potential sites (heme *a* and Cu<sub>A</sub>), high-potential sites (heme *a*<sub>3</sub> and Cu<sub>B</sub>), how the enzyme works, and proton pumping.

Discuss two proton-pumping models (X-ray structure and liposome reconstitution papers).

Post lecture comments - The class was well attended tonight and everyone stayed for both sessions, even Soloman. I was able to cover everything that I had outlined for the lecture. When I asked questions tht required the participants to use what I had taught in previous classes, only one person, the physician, was able to provide an answer. I am now convinced that the people simply listen to what I say but the information is not processed. These lectures are pure performance on my part.

**Lesson 4 (10 November 2000)**

Attendance sheet.

Review previous lecture (IB Chapter 10, pp. 389-400; FB Chapter 17, pp.497-511).

Discuss Problems 1, 2, and 3 (IB Chapter 5, pp. 250-251).

Fundamental of Biochemistry: Nucleotides and nucleic acids, nucleotide structure and function, nucleic acid structure (Chapter 3, pp. 41-53).

Fundamentals of Biochemistry: Nucleic acid structure. the DNA helix (Chapter 23, pp. 725-738).

Post lecture comments - The class was well attended after starting at 6:30 p.m. with only 5 people, but soon there were 10 to 12 participants. I used the computer and monitor this evening to illustrate nucleotide and nucleic acid structures. There was a question about the tetranucleotide that I drew on the board as being incorrect with 5'-OH and 3'-phosphate groups, but this is correct. I asked some questions and did some problems from the book during the first half of the lecture. Few people understand the material or they do not understand the questions that I am asking.

**Lesson 5 (14 November 2000)**

Attendance sheet.

Review previous lecture (FB Chapter 3, pp. 41-53; Chapter 23, pp. 725-738).

Discuss the structure in FB Figure 3.6, it is correct as written, and shorthand notation for RNA and DNA, 5' and 3'.

Discuss A-DNA in dsRNA, and RNA-DNA hybrids and Z-DNA forms in high salt and organic solvents.

Discuss Problems 2 and 3 (FB Chapter 3, p. 75).

Discuss Problem 1 (FB Chapter 23, p. 771).

Fundamentals of Biochemistry: Nucleic acid structure. the DNA helix, forces stabilizing nucleic acid structures, fractionation of nucleic acids, DNA-protein interactions (Chapter 23, pp. 739-756).

Inorganic Biochemistry: Fundamentals of inorganic biochemistry, biological ligands, the relationship between nucleotide and protein sequence (Chapter 1, pp. 43-49).

Inorganic Biochemistry: Alkali and alkaline earth metals, complexes with nucleic acids (Chapter 6, pp. 273-278).

Post lecture comments - Class was well attended again but people are showing up at 6:20 p.m. We needed to go over last lecture again because a number of people were absent during the discussions of DNA structure. I found that most of the participant were unaware of many of the standard techniques used in molecular biology, i.e., pulsed field gel electrophoresis, abortive transcription. I was able to get to some of the basic protein motifs found in protein-DNA structures and will need to continue the discussion on Friday. We have probably two more lectures before getting into the details of metal ion uptake and gene regulation and then superoxide dismutase gene regulation.

**Lesson 6 (17 November 2000)**

Attendance sheet.

Review previous lecture (FB Chapter 23, pp. 739-742, 747-756; Chapter 6, pp. 273-278).

Discuss Problems 6 and 7 (FB Chapter 23, p. 771).

Discuss effect of adding metal ions to DNA solutions,  $T_m$  increases.

Fundamentals of Biochemistry: Nucleic acid structure. DNA-protein interactions (Chapter 23, pp. 753-760).

Fundamentals of Biochemistry: Proteins: primary sequence, protein purification, protein sequencing (Chapter 5, pp. 96-114).

Discuss phage display technique.

Post lecture comments - Class started late at 6:30 p.m. but was well attended for both halves. The computer display of transcription factors was well received and a big help in explaining how proteins interact with DNA. I was able to cover protein purification and sequencing topics tonight so that I can concentrate on DNA-protein techniques and transcription in Tuesday's lecture.

**Lesson 7 (21 November 2000)**

Attendance sheet.

Review previous lecture (FB Chapter 23, pp. 753-760; Chapter 5, pp. 96-114).

Discuss sedimentation velocity and sedimentation equilibrium centrifuge methods.

Discuss Chou-Fasman secondary structure predictions in Table 6.1 (FB p. 139).

Discuss Problems 1, 3, 4, and 6 (FB Chapter 5, p. 122).

Discuss Problem 8 (IB Chapter 1, pp. 62-63).

Fundamentals of Biochemistry: Nucleic acid structure. DNA-protein interactions (Chapter 23, pp. 756-758).

Discuss zinc fingers (paper).

Inorganic Biochemistry: Experimental methods, biochemical methods, measuring the molecular mass of a protein, measuring the molecular mass and length of polynucleotides, measurement of macromolecular-ligand binding affinities, protein isolation and purification (Chapter 2, pp. 114-118).

Post lecture comments - There were only 5 people to begin class and 6 at the end of the evening's lecture. I was able to get through Zn fingers and cover some methods for analyzing DNA-protein interactions. I will have my choice of what to cover during the next four lectures.

**Lesson 8 (24 November 2000)**

Attendance sheet.

Review previous lecture (FB Chapter 23, pp. 756-758; IB Chapter 2, pp. 114-118).

Discuss electrophoretic mobility shift assays with cold protein, cold label, and protein-protein or protein-DNA crosslinking.

Discuss Problem 3 (IB Chapter 7, pp. 316-317).

Inorganic Biochemistry: Metal complexes as probes of structure and reactivity, nucleic acids (Chapter 9, pp. 357-364).

Discuss DNase protection, methyl sulfate, and restriction endonuclease protection assays for protein-DNA interactions.

Fundamentals of Biochemistry: Transcription and RNA processing, RNA polymerase (Chapter 25, pp. 813-822).

Post lecture comments - Lecture was well attended with 12 participants present for the entire night. I was able to cover some new techniques with them but Solomon as usual either doesn't pay attention or misunderstands what is being discussed because he asks off the wall questions. He appears to have questions already in mind and asks them without listening to the discussions. I will be able to finish transcription regulation next lecture and go into metal homeostasis.

**Lesson 9 (28 November 2000)**

Attendance sheet.

Review previous lecture (IB Chapter 9, pp. 357-364; FB Chapter 25, pp. 813-822).

Discuss summary figure of promoter and terminator regions.

Discuss structures of RNA Pol (unimportance of  $\omega$  (omega) subunit) and Rho.

Discuss *E. coli* and *Bacillus*  $\sigma$  factors.

Discuss Problems 1, 2, 3, and 5 (FB Chapter 25, p. 843).

Discuss Problem 10 (IB Chapter 1, p. 63).

Discuss Problem 3 (IB Chapter 9, p. 385).

Fundamentals of Biochemistry: Transcription and RNA processing, transcription in eukaryotes (Chapter 25, pp. 822-830).

Fundamentals of Biochemistry: Regulation of gene expression, regulation of prokaryotic gene expression, regulation of eukaryotic gene expression (Chapter 27, pp. 894-901, 910-916).

Discuss transcription factors,  $\sigma$  (alternate) factors, operons and regulons ( $\sigma^{70}/\sigma^{54}$  review).

Post lecture comments - Class started late again and was well attended after a few minutes of discussion. I was able to cover the rest of transcription and regulation of gene expression. There were many questions but some of the concepts I spoke of did not sink in right away. For example, I discussed how Lac repressor binds to distant operator sites to loop DNA and then when I presented the graph of sequence position versus activator or repressor status, Solomon asked how a protein that binds so far away from the start of transcription can repress transcription. I was told that some of the people enjoyed this class.

**Lesson 10 (1 December 2000)**

Attendance sheet.

Review previous lecture (FB Chapter 25, pp. 822-830; FB Chapter 27, pp. 894-901, 910-916).

Discuss Problems 3, 4, and 5 (FB Chapter 27, pp. 930-931).

Inorganic Biochemistry: Transport and storage, introduction, metal ion uptake and transmembrane ion transport, transport of storage of metal ions in vivo (Chapter 3, pp. 133-162).

Discuss structures of receptors and siderophores.

Discuss structures of ferritin and metallothionein.

Discuss structures of Shiga-like toxin and verotoxin.

Inorganic Biochemistry: Metals in the regulation of biochemical events, regulation of cellular concentrations of metal ions (Chapter 7, pp. 303-314).

Discuss metal ion uptake (Mg, Zn, Fe), siderophore and receptor structure, and gene regulation (Fur and DtxR papers).

Post lecture comments - The class started at 6:30 p.m. with two participants and then the rest trickled in over the next 20 minutes. I was able to cover metal ion uptake and regulation but not heavy metal detoxification mechanisms. The structures of the membrane siderophore receptors and toxins went over well with the class. I get questions from the same two people as usual.

**Lesson 11 (5 December 2000)**

Attendance sheet (hand out St. Louis key chains).

Review previous lecture (IB Chapter 3, pp. 133-162; IB Chapter 7, pp. 303-314).

Discuss regulation of ferritin and transferrin by IRE-BP and Fe.

Discuss regulation of *fur* by OxyR and SoxRS.

Inorganic Biochemistry: Cell toxicity and chemotherapeutics, metal toxicity (Chapter 8, pp. 338-341).

Inorganic Biochemistry: Case studies, mercuric reductase (Chapter 10, pp. 400-406).

Discuss heavy metal resistance and gene regulation (Cd and Hg papers).

Inorganic Biochemistry: Cell toxicity and chemotherapeutics, oxygen toxicity (Chapter 8, pp. 319-337).

Discuss superoxide dismutase catalytic mechanism and gene regulation (X-ray structure, kinetics, and gene regulation).

Copper chaperones

Discuss ccc family and ccs (ccs and CuZnSod papers).

Thank the class for their attendance and participation.

Express my gratitude for the warm and open hospitality of Trujillo and the University.

Post lecture comments - The class started at 6:30 p.m. and was well attended. There were 12 participants. The dean announced that there would be a special ceremony to present me as a honorary professor on Thursday at 6 p.m. I was able to cover all of the topics on my list for tonight, but not in the detail that I would have liked. The class was provided with an evaluation form to complete concerning my courses. The results will be tabulated and sent to the Fulbright Commission and a copy will be provided to me.