

Chemistry 565, 665/Biochemistry 665: Fall, 2018

Instructor: Tom Record (3214 BSB) Email: mtrecord@wisc.edu Tel: 262-5332

Office Hrs (2221 BSB): Mondays 1:15 PM. Also after lecture or by appointment

Biophysical Chemistry

565/665 develops the principles of chemical thermodynamics and chemical kinetics and applies these principles to understand the energetics and mechanisms of biochemical processes in aqueous solution. Processes include chemical reactions (enzyme catalyzed) and noncovalent interactions, including the self-assembly interactions which form functional structures of proteins, nucleic acids, carbohydrates, lipids and their complexes, and the ligand-binding interactions which regulate them.

This material is useful both at a practical level (in understanding the chemical basis of methods for the separation, purification, and characterization of biological molecules) and at a conceptual level (in understanding the thermodynamic and mechanistic principles of biopolymer processes). Although starting with the basics of thermodynamics and kinetics, 565/665 progresses to a level suitable as background for more advanced graduate courses or literature work in biophysical areas including protein and nucleic acid stability and interactions, enzyme catalysis and bioenergetics.

565/665 is taught from handouts (chapters, papers, problem sets and examples; a nominal charge is made to cover the cost of reproducing these handouts). No textbook is required.

Supplementary Readings (on reserve in Chemistry and Steenbock Libraries)

A) Reference Biophysical Text for September (Not Required for Purchase)

I. Klotz and R. Rosenberg, *Chemical Thermodynamics* (Benjamin-Cummings). (Numerous editions; any of them is OK.) Chapters 1-11 and 14 provide a detailed and very readable treatment of fundamentals of thermodynamics, at the math level of 565/665, and are very useful outside reading for the first month of the class.

B) Other Reference Books (Not Required for Purchase)

J. Kuriyan, B. Konforti, D. Wemmer, *The Molecules of Life; Physical and Chemical Principles* (Garland, 2013). KKW is an advanced undergrad-beginning grad text from UC Berkeley that combines structural biology with chemical thermodynamics and kinetics at the level of 565/665. (The cover is a nice illustration of the attempt to combine these topics.)

K. Dill and S. Bromberg (2010 or 2003) *Molecular Driving Forces* (Garland). Excellent coverage of biophysical and polymer physical chemistry, from a statistical thermodynamic perspective. Moderately high math level.

T. Creighton, *Proteins: Structure, Molecular Properties* (2011; Freeman)

V. Bloomfield, D. Crothers and I. Tinoco, (2000) *Nucleic Acids* (USB);
also (1974) *Physical Chemistry of Nucleic Acids* (Harper Row)

C. Tanford (1980) *Hydrophobic Effect*; John Wiley & Sons.

K. van Holde, W. C. Johnson, P. S. Ho, *Physical Biochemistry*, Prentice-Hall

Course organization:

1. **Lectures, handout materials and the weekly problem sets** define the course content. You should prepare for class by working through the relevant parts of the handout chapters, sample problems and/or papers. Questions and discussion are encouraged in all class meetings (lecture and discussion).
2. **Discussion section meetings** on Thursdays discuss methods of problem solving, work sample problems, and discuss current lecture material. It is important to prepare for discussion section and to participate actively in discussion; the "optional" listing for these sections means only that no quizzes are given.
3. **Problem sets** are assigned on a weekly basis, and usually are due on Friday. **Doing conscientious work on all the problem sets, and submitting them on or before the date due, is very important for many reasons** (e.g. understanding the material, getting a good grade; see #5 below). Many assigned problems are taken from previous years' exams. It is a very good idea to **form a study group** to discuss approaches to solving problems with others in the class, but do not copy answers from others or from previous years' handouts. **Demonstration of effort on all problem sets is expected.** No credit will be given for work which is not your own.
4. **Exams:** The three exams are given as proctored problem sets. You may use the printed chapters and handouts, as well as your handwritten notes and problem set answers, but not other textbooks, reference materials or photocopied parts thereof. Any single-purpose calculator can be used, but no other electronic devices are permitted and must be put away during the exam. Makeup exams are not given.

Exam dates are **Thursday Oct. 11, Thursday Nov. 15, and Friday Dec. 14**. The first two exams will be from **7:15-9:15 PM**; the third exam will be from **7:45 - 9:45 AM**. Exams are cumulative, but will cover primarily the material since the cut-off date for the previous exam. Please let me know of any academic conflicts with these exam dates during the first week of the course.

5. **Grading:** Each exam counts 33% of the course grade.

The assigned problem sets contribute in several ways toward your grade:

- a) if in any third of the course, your problem set average is higher than your grade on the corresponding exam, the problem sets will count 25% and the exam 75% of your grade for that portion of the course;
- b) if the raw exam scores are curved upwards, you must have shown conscientious effort on all 4 of the corresponding problem sets to receive the curved score, and
- c) **conscientious effort on the problem sets and active participation in all aspects of the course** are taken into account in borderline final grade situations, while lack of effort on the problem sets will reduce your grade.

6. **Advice on the Problem Sets:** It is important that you attempt to work all the problem sets and turn in your work on time. It is equally important that you be able to do this efficiently, without spending too much time on false starts. Consequently you should plan to start working the problems early in the week in which they are due. Ask for help after class or in office hours if you are confused. Bring your attempts with you. **We will not discuss solutions of problems with people who have not thought about them and attempted to work them.**

Approximate exam grade conversion scale:

87 and above:	AB, A
75-86:	BC, B
65-74:	C
55-64:	D

Additions to 565/665 Syllabus

Fall, 2018

There is no required textbook for 565/665, which is taught from handouts (including handout problem sets) for which there is a one-time charge of about \$30. Klotz and Rosenberg (a "recommended" book) is a very readable introduction to Chemical Thermodynamics at the level of this course, and a useful reference for the first month of 565/665. Klotz and Rosenberg is on reserve in Steenbock and Chemistry libraries, so there is no reason to purchase it unless you find it so useful that you want a copy of your own. Other reference books are also on reserve; a list will be provided at the first class meeting.

Updated Catalog Statement for 565/665:

Chemical thermodynamics and chemical kinetics, applied to biochemical processes in aqueous solution. Expected background: Chem 327 or 329; Math 222; Physics 201 or 207; Biocore 303 or Biochem 501 or 507 (or concurrent registration) or consent of instructor.

Learning Outcomes:

Specific: 565/665 develops the principles of chemical thermodynamics and chemical kinetics and applies these principles to understand the energetics and mechanisms of biochemical processes in aqueous solution. Processes include chemical reactions (enzyme catalyzed) and noncovalent interactions, including the self-assembly interactions which form functional structures of proteins, nucleic acids, carbohydrates, lipids and their complexes, and the ligand-binding interactions which regulate them.

This material is useful both at a practical level (in understanding the chemical basis of methods for the separation, purification, and characterization of biological molecules) and at a conceptual level (in understanding the thermodynamic and mechanistic principles of biopolymer processes). Although starting with the basics of thermodynamics and kinetics, 565/665 progresses to a level suitable as background for more advanced graduate courses or literature work in biophysical areas including protein and nucleic acid stability and interactions, enzyme catalysis and bioenergetics.

General: Development of Core Competencies Sought by Graduate Schools, Medical Schools and Employers

Science Competency: Apply knowledge and skill in the natural sciences to solve problems related to molecular or macroscopic systems.

Thinking, Reasoning Competencies:

Critical Thinking: Use logic and reasoning to identify the strengths and weaknesses of alternative solutions, conclusions, or approaches to problems.

Quantitative Reasoning: Apply quantitative reasoning and appropriate mathematics to describe or explain phenomena in the natural world.

Scientific Inquiry: Apply knowledge of the scientific process to integrate and synthesize information, solve problems and formulate research questions and hypotheses; be facile in the language of the sciences and use it to participate in the discourse of science and explain how scientific knowledge is discovered and validated.

Written Communication: Effectively convey information to others using written words and sentences.

Interpersonal Competencies

Social Skills: Demonstrate awareness of others' needs, goals, feelings, and the ways social and behavioral cues affect peoples' interactions and behaviors; adjust behaviors appropriately in response to these cues; and treats others with respect.

Teamwork: Work collaboratively with others to achieve shared goals; share information & knowledge with others and provide feedback; put team goals ahead of individual goals.

Oral Communication: Effectively convey information to others using spoken words and sentences; listen effectively; recognize potential communication barriers and adjust approach or clarify information as needed.

Intrapersonal Competencies

Ethical Responsibility to Self and Others: Behave in an honest and ethical manner; cultivate personal and academic integrity; adhere to ethical principles and follow rules and procedures; resist peer pressure to engage in unethical behavior and encourage others to behave in honest and ethical ways; and develop and demonstrate ethical and moral reasoning.

Reliability and Dependability: Consistently fulfill obligations in a timely and satisfactory manner; take responsibility for personal actions and performance.

Resilience and Adaptability: Demonstrate tolerance of stressful or changing environments or situations and adapts effectively to them; be persistent, even under difficult situations; recover from setbacks.

Capacity for Improvement: Set goals for continuous improvement and for learning new concepts and skills; engage in reflective practice for improvement; solicit and respond appropriately to feedback.

Fall, 2018 Syllabus

Instructor: Tom Record

Biophysical Chemistry 565/665

Thermodynamics, Kinetics of Biochemical Processes

A) Introduction to Biopolymer Processes and to Applications of Chemical Thermodynamics and Chemical Kinetics to Biopolymer Processes (1-2 lectures: Read Overview, Introduction, Ch. 1 of Handouts)

B) Fundamentals of Chemical Thermodynamics of Systems and Processes (~12 lectures) We start with the Laws of Thermodynamics and progress rapidly to free energy and chemical potential. Chemical potential differences are the driving force for moving molecules in processes, both from one solution environment or phase to another and from reactant to product in a chemical reaction or noncovalent interaction.

1) Preliminaries:

Math of State Functions, Exact, Inexact Differentials, Partial Derivatives

(Ch. 1 Appendix; Klotz & Rosenberg (KR) Ch. 2);

Work, Heat; Reversible, Irreversible Processes (Ch. 2; KR Ch. 3).

2) First Law:

Energy, Enthalpy, Heat Capacity of Systems; Changes in these Functions in Processes (Ch. 2; KR Ch. 3-6).

3) Second Law:

Physical and Mathematical Properties of Entropy (Ch. 3; KR Ch. 7). \

4) Free Energy and Work:

Criteria for a Spontaneous Process and for Equilibrium at Constant T and P; Summary of State Functions; Maxwell Relations; Calculations of Entropy, Free Energy Changes for Simple Processes (Ch. 4,5; KR Ch 8)

5) Mixing as a Spontaneous Process; Ideal Gas Mixtures (Ch 5; KR Ch 14).

6) Thermodynamics of Mixtures:

Extensive, Intensive Quantities; Molar and Partial Molar Quantities; Multi-Component Systems, Mixtures; Characterizing Contributions of Different Components to the Thermodynamic Properties of a Mixture (Ch. 5, 6).

7) Chemical Potential (Partial Molar Gibbs Free Energy; Ch. 6, 7)

8) Applications of Chemical Potential to Analyze Fundamental Processes: Chemical Reaction and Chemical Equilibrium in a Gas-Phase Molecular Mixture; Phase Change and Phase Equilibrium in a Pure System where Molecular Mixing is Not Involved (Ch. 7)

C) Thermodynamics of Solutions (~5 lectures; Ch. 8)

1) Ideal Solutions (Random Liquid Mixtures), Mole Fraction Concentration Scale; Nonideal (i.e. Nonrandom) Solutions, Activity Coefficients. Nonrandomness arises from unequal interactions and/or unequal volumes of the species in the mixture; both are described by activity coefficients.

2) Very Dilute (also called "Ideal Dilute") Solutions; Thermodynamic Characterization of Solute-Solvent Interactions and Solute-Solute Interactions. Here the only nonrandomness is from hydration of the solute; the hydrated solutes form a random mixture with the solvent.

3) Solvent, Solute Chemical Potentials; Molar and Molal Concentration Scales.

4) Electrolyte Solutions (Dissociation; Hydration of Ions; Local Deviations from Electroneutrality (Non-random Mixing) in Salt and Biopolymer Solutions; Nonideality from Long-range Coulombic Interactions of ions observed at very low concentrations, especially for polyions).

Exam 1 on this material (first 4 problems sets): Thursday, October 11, 7:15 PM

D) Applications of Chemical Potential to Analyze Biochemical Processes and Equilibria in Solution (~10 lectures; Ch 9-12; KR 9, 10)

1) Processes and Equilibria Involving Solvent in Solution. Origin of Colligative Properties of Solutions (FP depression, BP elevation, VP lowering, Osmotic Pressure). Analysis of Osmosis and Reverse Osmosis. Living Cells as Osmotic Systems (Ch. 9).

2) Processes and Equilibria Involving Solutes in Solution:

a) Biochemical Covalent Reactions (ATP hydrolysis is the best characterized example) and Noncovalent Binding/Association Interactions (Ch. 10): One remarkable feature of these processes is how sensitive they are to solution variables like pH and concentrations of ligands, solutes and salt. Hence their equilibrium “constants” aren’t constant when these solution variables change.

b) Transfer of Solutes Between Environments, Phases (Ch. 11). These transfer processes are a useful way of modeling the transfer of protein functional groups from water to the protein interior when the protein folds or assembles with other proteins.

c) Transfer of Hydrocarbon Solutes to Water from a Nonaqueous Phase as a Model for the Hydrophobic Effect (Ch. 11). Burying hydrocarbon surface is a major driving force for protein processes including folding, subunit assembly, and binding.

d) Thermodynamics of Forming Ions from Uncharged Solutes in Water (Ch. 10, 11).

E) Macromolecular Recognition, Self-assembly and Other Noncovalent Biopolymer Processes in Water Micelle Formation, Nucleic Acid Helix Formation and Folding, Protein Folding, Protein-Ligand and Protein-Protein Interactions; Roles of the Hydrophobic Effect, Base Stacking and other Noncovalent Interactions (Ch. 12)

F) Ligand Binding to Biopolymers; Regulation of Processes by Ligands

(~10 lectures; Ch. 13)

- 1) Noncooperative and Cooperative Binding Isotherms for Ligand-Biopolymer Interactions
- 2) Thermodynamic Characterization of Ligand Binding: Equilibrium Dialysis, Spectroscopic and Calorimetric Methods
- 3) Analysis of Ligand Binding and Ligand Effects using Binding Polynomials.
- 4) Regulation of Processes and Equilibria of Biopolymers by Ligands.
- 5) Applications: O₂ Binding to Mb and Hb, *Lac* Repressor-*lac* Operator Interactions: Well-characterized examples of binding interactions which are regulated by other (allosteric, competitive) binding interactions). Cooperativity of ligand binding from coupled conformational changes or other coupled processes.

After 4 problem sets, Exam 2 covers this material (up to Section F4): Thursday, November 15, 7:15 PM

G) Interactions of Proteins, Nucleic Acids with Solutes, Salts from the Hofmeister series and Polymers (Excluded volume agents); Effects of these Solutes, Salts and Polymers on Protein and Nucleic Acid Processes (Ch. 14, 15; ~4 lectures)

Solutes and Salts form Series in how they interact with proteins and nucleic acids and affect their processes: the progressions below are (left to right) from solutes that interact favorably with proteins and nucleobases and destabilize their assemblies to ones that don't.

Solute series: Urea, formamide, glycerol, proline, glycine betaine, TMAO

Hofmeister (noncoulombic) Salt Ion Series:

Anions: SCN⁻, I⁻, ClO₄⁻, Br⁻, Cl⁻, Acetate⁻, F⁻, Glutamate⁻, SO₄²⁻, HPO₄²⁻

Cations: GuH⁺, NH₄⁺, K⁺, Na⁺, alkylated NR₄

Monomer to Polymer Series: Ethylene glycol (HOCH₂CH₂OH) to Polyethylene Glycol (HOCH₂(CH₂OCH₂)_nCH₂OH)

H) Chemical Kinetics and Mechanisms of Biopolymer Processes (~16 lectures; Ch 16-19) Kinetics and thermodynamics are complementary disciplines; kinetics provides information about key aspects of processes that are not significant for thermodynamics (how fast they occur; their path or mechanism). Unlike thermodynamics, kinetics has few fundamental principles and little predictive power; in particular you can't predict the kinetics or mechanism of a process from its balanced chemical equation. An understanding of thermodynamics is needed to interpret kinetic data.

- 1) Rate Laws, Rate Equations for Elementary Reactions (First Order, Second Order; Irreversible, Reversible; Relaxation Analysis (Ch. 16).
- 2) Diffusion-Collision Step in Second Order Reactions in Solution (Diffusion Limit, Facilitated Diffusion; Ch. 17).
- 3) Chemical Limitations to Reaction Rates: Enthalpic and Entropic Contributions to Activation Free Energy Barriers of Elementary Reactions; Reactions with Intermediates (Ch. 17)
- 4) Mechanisms of Reactions with Intermediates and their Analysis using Steady State or Rapid Equilibrium Approximations (Ch. 18).
- 5) Kinetics and Mechanisms of Enzyme-Catalyzed Reactions: Biophysical Basis of Catalysis, Catalytic Antibodies, Cooperative Enzymes, Regulation of Catalytic Activity by Ligand Concentration (Ch.19).
- 6) Case Studies: Kinetics and Mechanisms of Protein Folding, Nucleic Acid Helix Formation, Protein-Protein and Protein-Nucleic Acid Interactions (RNA Polymerase-Promoter, *Lac* Repressor-*lac* Operator).

After 3 problem sets, Exam 3 covers this material (primarily from section F5 on): Friday, December 14, 7:45 – 9:45 AM