The Teaching of Biochemistry: An Innovative Course Sequence Based on the Logic of Chemistry

Henry V. Jakubowski*
Department of Chemistry, College of Saint Benedict, Saint John's University, 37 South College Avenue, St. Joseph, MN 56374-2099

Whyte G. Owen
Department of Biochemistry and Molecular Biology, Mayo Clinic Foundation, 200 1st Street S.W., Rochester, MN 55905

Organizing and presenting an introductory course in biochemistry can be a daunting task. Two major issues must be confronted in teaching this or any introductory course. First, how should topics be selected that represent the depth and breadth of modern biochemistry without overwhelming students (and instructor)? Second, in what order should the topics be covered? Presumably, the chosen order should create a coherent and sequential understanding of biochemistry, not a fragmented one without logical connections among topics. Textbook authors offer assistance in addressing these concerns in two ways. They implicitly suggest an order of presentation by how chapters are arranged and they offer philosophical interpretations to describe biochemistry. Scrutiny of the philosophy statements and chapter organization of textbooks reveals commonalties among textbooks. Garret and Grisham write that "Chemistry is the logic of biological phenomena" (*1*). Stryer suggests that "common molecular patterns and principles underlie the diverse expression of life" (*2*). Lehninger, Nelson, and Cox describe the "molecular logic of life" (*3*). These and other modern authors find consensus in their analyses of the nature of biochemistry and offer startlingly similar organization of chapters.

This consensus, however, does not lead to a linkage between philosophy and content. The present organization of texts is not derived from the central dogma of biology, since in most books protein structure precedes significant discussions of nucleic acid structure and function. Rather, it seems to reflect evolving tradition based on historical trends in biochemistry research, as evidenced by chapter organization of major biochemistry texts, starting from the 1935 edition of Harrow's *Textbook of Biochemistry* (*4, 5*). Early texts commenced with discussions of carbohydrate chemistry, followed by lipids and then proteins. Texts from the late '60s onward invariably led with protein chemistry and deferred carbohydrate and nucleic acid chemistry until much later (*5–7*). Although modern authors speak of a "chemical logic", it is not evident in textbook organization. This manuscript describes a higher-order organizing principle, based in chemical logic and understanding, from which topics and order of presentation derive. New topics can then be introduced in a fashion which the students perceive not as random but as a logical extension based on a developing understanding.

*Corresponding author. Email: HJAKUBOWSKI@CSBSJU.EDU.

Course Description

Four preeminent biological questions of our day serve as a framework for the course:

- Can the structure and function of a protein be explained and predicted from its primary sequence?
- How is gene expression regulated?
- How can extracellular information regulate intracellular processes?
- How can enzymes catalyze biochemical reactions with such speed, selectivity, and with regulation?

These questions provide the context for the study of chemistry as applied to biological phenomena. Throughout the course, three major recurring chemical principles become evident: structure determines function/activity; binding reactions initiate all biological events; and chemical principles, such as dynamic equilibria (mass action) and reaction kinetics and mechanisms, derived from the study of small molecules, can be applied to the behavior of macromolecules.

The course sequence is divided into seven parts. In each sequence, students study chemistry in the framework of specific biological questions, which fit under the umbrella of the preeminent biological questions mentioned earlier. The order of the topics is based on evolving chemical logic. The first topic is lipid and lipid aggregate structure/function instead of amino acids and proteins, as is typical. Before taking a biochemistry course, students have little significant exposure to the chemical properties of macromolecules, so beginning with the study of small molecules makes sense. Since most lipids are amphiphiles, their structural diversity can be simplified by considering them as simple structures with spatially distinct polar and nonpolar ends.

Single-chain and double-chain amphiphiles aggregate in a thermodynamically spontaneous manner to form micelle and bilayer structures, respectively, with the nonpolar parts sequestered from water and associated with themselves and the polar parts solvent-accessible. This simple model introduces students to the notion that structure mediates properties, to the important concept of intermolecular forces, and to the thermodynamics of the hydrophobic effect—all critical elements ultimately required to understand the much more complicated topic of protein folding and stability. The concepts of mass conservation, dynamic equilibria and kinetics, and chemical potential are used to understand how aggregation at equilibrium depends on amphiphile concentration. Lipids serve as useful models to introduce stereochemistry and prochirality

as well. Likewise, it is easier to understand how torsion-angle changes in the aliphatic side chains of phospholipid molecules alter acyl chain packing than it is to understand the complexities of a Ramachandran plot.

From a chemical perspective, it is more logical to introduce the spontaneous self-assembly of small amphiphilic molecules into large multimolecular aggregates than to start with the physiochemical properties of 20 different amino acid that vary in size and hydrophobicity and proceed to the complexities of intramolecular protein-folding reactions. The biological questions related to lipid structure and function in course sequence 1 and the associated chemical principles that are introduced and reviewed in the process are shown in Table 1.

These understandings can then be applied to the more complex subject of intramolecular protein-folding reactions and protein stability in sequence 2 (Table 1). Included in an expanded discussion is a more modern view of the hydrophobic effect, with its associated heat capacity changes, and the denaturing effect of chain conformational entropy. The role of the hydrophobic effect and secondary structure in protein stability are extrapolated from the behavior of benzene in water and thermodynamic cycles involving the transfer of *N*-methylacetamide from water to a nonpolar solvent. Dynamic and linked equilibria considerations, along with reaction kinetics, are used to describe the varying effects of denaturing (urea, guanidinium chloride) and stabilizing (ammonium sulfate, glycerol) solutes on protein stability, as well as the competing processes of protein folding and aggregation in vitro and in vivo. The same principles that determine protein structure and function can be applied to the study of the structure and stability of nucleic acids, complex carbohydrates, and glycoproteins.

Function now necessarily follows. Since all biological events are initiated by binding, a purely physical process, the logic of chemistry suggests it should be studied as sequence 3 (Table 1). Indeed, in most textbooks, introductory chapters on protein function focus on the binding of dioxygen, a simple ligand, to myoglobin and hemoglobin. Macromolecule–drug interactions and cell–cell adhesion can be discussed as additional relevant examples. The control of gene expression, a topic of preeminent importance to modern biologists, can be discussed from the logic of chemistry as an essential outcome of the binding of transcription factors and appropriate enzymes to each other and DNA in the active transcription complex. It is particularly important to stress how equilibrium and mass conservation principles, along with reaction kinetics, effectively determine the concentration-dependent behavior of all molecules, including the processes of binding and spontaneous structure formation.

Binding is an antecedent to the expression of biological activity. The simplest expression of activity that involves a simple physical, not covalent, process is binding and transport of solute molecules across a biological membrane, which comprises sequence 4 (Table 1). Mathematical analyses of the flux of solute across a membrane catalyzed by a transport protein involves the same assumptions (rapid equilibrium, steady state binding) and leads to the same equations (hyperbolic dependence of flux with outer solute concentration, effect of competitive inhibitors) as when Michaelis–Menten enzyme kinetics mechanisms are modeled. The study of enzyme kinetics follows in sequence 5 (Table 1) as a logical extension of the expression of molecular function involving the addition of a more complex step, namely, a chemical transformation. Through the study of enzyme kinetics, students learn how to obtain a low-resolution understanding of the structure–activity relationships of enzymes and of their chemical mechanisms.

Course sequence 6 (Table 1) enhances this resolution by discussing the detailed mechanism of specific enzymes whose structures are known. Preceding this, the basis for catalysis by small molecules is discussed. Following the chemical logic that the properties of macromolecules can be inferred from small molecules, students learn that with respect to catalysis, enzymes are "not different, just better" than small-molecule

Table 1. Summary of the Course Sequences

Sequence	Biological Questions	Chemical Principles
1. Lipids: Structure–function	For what reasons do single-chain amphiphiles form micelles, while double chain amphiphiles form bilayers? Could cells with bilayers form spontaneously?	Intermolecular forces (IMF), attractive/repulsive forces, free energy, chemical potential, dynamic equilibria, reaction kinetics, free-energy transfer, hydrophobic effect, entropy, diffusion, phase transition, conformational changes
2. Amino acid and protein structure; carbohydrate/ nucleic acid structure	What rules govern protein folding and stability? Can the 3-D structure of a protein be predicted from the primary sequence? What is the basis for the existence of secondary structure? What rules govern nucleic acid structure?	IMF, attractive/repulsive forces, free energy, chemical potential, dynamic equilibria, reaction kinetics, free-energy transfer, hydrophobic effect, entropy, diffusion, phase transition, conformational changes
3. Protein function: Binding and stability	What is the origin of protein function/activity and specificity? How are binding, function, and activity regulated? How is gene expression controlled? How do drugs interact with macromolecules? What rules govern cell–cell interactions?	IMF, equilibria, reaction kinetics, binding/titration curves, rate constants, data analysis, cooperative interactions, conformational changes, diffusion/dynamics
4. Protein function: Transport	How can molecules cross a lipid bilayer with the specificity required of biological processes?	IMF, binding, equilibria, rapid equilibrium/steady state, rate constants, data analysis, cooperative interactions, conformational changes, diffusion/dynamics, chemical potential
5. Protein function: Kinetics	Without a 3-D structure, how can the structure and function of an enzyme be studied?	IMF, binding, equilibria, rapid equilibrium/steady state, data analysis, cooperative interactions, conformation changes, diffusion/dynamics, chemical kinetics
6. Protein function: Chemical transformation	How can enzymes catalyze biochemical reactions with such speed, selectivity, and sensitivity, and with regulation? Do enzymes use mechanisms different from other catalyzed reactions?	IMF, binding, equilibria, conformational changes, diffusion/dynamics, acid/base catalysis, electrostatic catalysis, covalent catalysis, transition state stabilization, activation energy, reaction coordinates and mechanisms, free energy
7. Protein function: Energy and signal transduction	How can extracellular information regulate intracellular processes? How can energy in the form of light and chemical energy be utilized?	IMF, binding, equilibria, catalysis, free energy, conformational changes, electrical and chemical potentials

catalysts, as previously described by Knowles (*8*).

The final sequence, 7 (Table 1), involves specific examples of how enzymes can transduce both energy and information signals into usable outputs. Energy transduction, involving the conversion of light, electrochemical gradients, or chemical energy into phosphoanhydride bonds, is discussed. Special attention is paid to biological oxidation reactions. Several questions are introduced to provoke discussion and challenge students' knowledge of oxidation reactions. Students propose explanations for the fact that oxidation reactions of organic molecules using dioxygen are thermodynamically but not kinetically favored and to explain the need for different types of biological oxidizing agents for energy transduction. Signal transduction at the cell membrane serves as an excellent capstone area of study, since it incorporates ideas from each sequence.

Conclusion

The approach for a biochemistry course described in this manuscript has its limitations and is, therefore, subject to some compromises. Topics still must be omitted and traditional textbooks cannot be followed in a sequential fashion. The content is weighted toward physical biochemistry and protein structure–function and de-emphasizes carbohydrate chemistry. Finally, it might appear to be deficient in "biological logic". Many of these concerns, however, are not problematic. For example, the study of carbohydrates and lipids, with emphasis on their chemical transformations in metabolism, is more appropriately accomplished in a second semester course, whereas detailed study of nucleic acids is best accomplished in a separate course in molecular biology. Students who take biochemistry courses during their junior or senior year presumably have developed more mature learning skills that do not require following the book in a strictly linear fashion. Supplemental handouts can be distributed to address any deficiencies that may arise. With respect to biological logic, students still acquire an understanding of the main organizing principle of modern biology, the central dogma of biology. In addition, the entire course is framed in the context of significant biological questions. Although these ideas are described in terms of a one-semester biochemistry course, which often serves as a capstone to a chemistry major, they should work as well in the first semester of a two-semester course. Traditional labs can still be utilized, but a lab dealing with micelle and lipid bilayer formation (*9*) would ideally be positioned first.

Abeles et al. (*10*) have developed an alternative textbook, which reflects several of the organizing principles discussed here. In the preface to their recent text, *Biochemistry*, they state "biochemistry can now be taught with emphasis on a few chemical principles that rationalize large bodies of biochemical facts" by "presenting chemical principles and facts that pertain to a biochemistry system and then to explain the system itself". Their approach is different from what has been presented here in that their rigorous book is organized predominantly around principles of organic chemistry. Proteins are presented in the beginning of the text (after an overview of biochemistry and organic reactions) using case studies of chymotrypsin and trypsin. This early focus on covalent protein structure comes at the expense of the study of lipids and spontaneous lipid structure, which appear in the last three chapters of the book, as do transport phenomena.

Any real limitations derived from the proposed organization are small in comparison to the resulting gains. Clearly, there is no one correct way to teach any course, but what any successful course needs is a coherent and logical organizing structure that leads to an emerging understanding. Chemical logic, as reflected in unified and accepted descriptions of the nature and practice of biochemistry, provides this thread and dictates the course organization described here. It does so in ways that other organizing principles cannot.

Literature Cited

1. Garrett, R.; Grisham, C. *Biochemistry*; Saunders: Orlando, FL, 1995.
2. Stryer, L. *Biochemistry*, 4th ed.; Freeman: New York, 1995.
3. Lehninger, A.; Nelson, D.; Cox, M. *Principles of Biochemistry*, 2nd ed.; Worth: New York, 1993.
4. Harrow, B. *Textbook of Biochemistry*, 1st–5th eds.; Saunders: Philadelphia, 1935–1950.
5. Harrow, B.; Mazur, A. *Textbook of Biochemistry*, 6th–10th eds.; Saunders: Philadelphia, 1954–1971.
6. Lehninger, A. *Biochemistry*; Worth: New York, 1970.
7. Mahler, H.; Cordes, E. *Biological Chemistry*; Harper/Row: New York, 1971.
8. Knowles, J. *Nature* 1991, *350*, 121–124.
9. Jakubowski, H. V.; Penas, M.; Saunders, K. *J. Chem. Educ.* 1994, *71*, 347–349.
10. Abeles, R.; Frey, P. I.; Jencks, W. *Biochemistry*; Jones and Bartlett: Boston, 1992.