C H A P T E R 1

WHAT BIOCHEMISTS STUDY

The goal of biochemistry is to understand the chemical basis of biological phenomena. This introductory chapter begins our presentation of the current state of that understanding by covering three main subjects.

We first give a perspective on the field of biochemistry, including some information on the methodologies used. The realm of biochemistry is the biosphere, which consists of organisms of all types and the environments in which they live. Our intent is to describe the vantage point from which biochemists view the biosphere as they attempt to explain the operation of organisms and their interactions with their environments in chemical terms.

Second, we summarize the contents of this book and its organization into sections that present structural, functional, and informational aspects of biologically important molecules.

Finally, we focus on one important compound. Of all the compounds in the biosphere, water is the most abundant. Because of its abundance and the fact that water is essential to cells and their environments and accounts for 70 to 90% of the mass of most cells, we devote the end of this chapter to the properties of this amazing substance, the solvent for biochemical reactions.

1.1 CELL AND ORGANISM AS BIOCHEMICAL ENTITIES

Biology in its broadest sense may be regarded simply as the study of the biosphere. What are the most characteristic properties of the organisms that populate the biosphere? A particular organism may be characterized by its form and essential processes such as reproduction, development, and energy transduction. That organisms take particular forms and mediate specific processes reflects and requires order. Reducing disorder, or entropy, inevitably requires energy. Organisms excel in utilizing energy to create order, and complex organisms do so on a continuous basis. What we know about how organisms use energy to create orderly structures and carry out orderly processes comes primarily from years of effort on the part of thousands of biochemists. One of the most important contributions of biochemistry to biology is the understanding that has been attained about how the chemical processes can create order.

The process that most definitely sets organisms apart from other objects is reproduction. Both cellular and noncellular organisms give rise to more organisms of similar design as part of a life cycle that may be simple or elaborate. For cellular organisms, cell division is, of course, an essential event in reproduction. Energy is used to manufacture new molecules that are incorporated into new structures in a precise developmental process that leads to the formation of two new cells. In contrast to most cells, the viruses and other even less complex noncellular organisms all are parasitic or symbiotic; they depend on specific host cells to provide an environment in which replication can occur by processes that do not overtly resemble cell division. For all organisms then, cellular and noncellular, the site of action is the cell, and the cell is the entity that is essential to reproduction.

The biochemist views the cell as an exceedingly complex and refined machine. These machines have capabilities that are far beyond those of current man-made machines. A given cell may have a very large repertoire of responses to environmental changes. The cells of a multicellular organism must be able to interact to give the even greater repertoire of responses that characterize higher organisms. "Adaptability" is a characteristic of the cell-machine. The complex organism detects and processes stimuli from the environment and from within the organism with sophistication. Movement, secretion, cell division, and so on are accomplished in a precisely controlled fashion to allow the entire organism to grow and reproduce itself.

The biochemist must be aware of the abilities of organisms to change and adapt, not only in the time frame of an individual organism but also on an evolutionary time scale. It is not enough to consider any noncellular, single cell, or multicellular organism in isolation or only as it exists today. Organisms can be classified as belonging to species or similar taxonomic or functional groupings. Organisms exchange, or at least transmit, symbolically encoded, controlling information. That is, they have genetic systems. Capabilities of the progeny reflect the capabilities of the parent(s), and the progeny follow definite plans of development. Those plans, though definite, are not rigid. They are modified by the environment and time. Organisms cannot be understood without a consideration of what their antecedents must have been like. Organisms have evolved and adapted to changing conditions on a geological time scale and continue to do so. Thus, biochemists seek chemical explanations of how organisms adapt to their environment both in the short term and over the eons.

The cell carries out the most elaborate of chemical transformations while maintaining its internal environment within definitely controlled limits. Cells have boundaries and, within those boundaries, compartments. There, thousands of types of small molecules and macromolecules, in an aqueous (water) environment, undergo chemical transformations and exchange materials and heat with the exterior of the cell. The elaborate macroscopic behavior of living systems anticipates a great variety of microscopic catalytic and regulatory mechanisms. Only a small fraction of these are now understood in molecular detail.

Fortunately, the results currently available show a startling unity of biology from a biochemist's point of view. Organisms, from the simplest noncellular entity to the most complex vertebrates and higher plants, are seen to have in common many chemical structures and reactions. Thus, the biochemist studying a particular organism may be able to anticipate its biochemical reactions from what is already known from other, even distantly related, organisms. This grand guideline, that organisms show a unity at the chemical level that is unsuspected when one views their biological diversity, strongly influences the thinking of biochemists.

1.2 THE BIOCHEMIST'S POINT OF VIEW

Biochemistry has its roots in medicine, nutrition, agriculture, fermentation, and natural products chemistry. Today, it is principally concerned with the chemistry of molecules found in and associated with living systems, especially the chemistry of the interactions of these molecules. Most biochemists operate in a particular philosophical framework. The usually unspoken assumption is that the activities of cells, for all of their complexity, must be susceptible to explanation as chemical and physical phenomena. Developing this understanding has required and will continue to require the careful application of chemical and physical laws and methods *in combination* with the careful biological manipulation of the systems under study.

Biochemistry is in large measure, but not wholly, a reductionist science. Biochemists routinely disrupt cells and fractionate what has been released into the extract for the purpose of obtaining one or another component of the cell in isolation. The biochemist's intent in preparing extracts and fractions is to reduce the number of variables that remain uncontrolled and to obtain a more clear-cut and reliable result. The cell has capabilities beyond those of its subsystems, but what we know about the cell has come largely from studying isolated parts of the cell, even individual molecules.

A common example of the reductionist approach is an *in vitro* enzyme assay. Enzymes are the principal *in vivo* catalysts of the cell. An enzyme will effectively facilitate, in the test tube, complex chemical transformations that would occur at only an imperceptible rate in the absence of the enzyme. The investigator selects conditions of temperature, concentration, and so on that can be achieved in the test tube and allow the enzyme to transform biologically authentic substrates into biologically authentic products. However, the conditions that are convenient for the investigator may resemble only slightly the conditions of the intact cell. Interpretation of the results in the context of the intact cell requires care and considerable attention to the detailed physiology of the system. Often, the results must be correlated with other biochemical, physiological, and/or genetic data before the puzzle can be solved.

Several modern biochemical approaches are not reductionist but take advantage of technological advances to study intact systems. We complete this subsection with a discussion of four such approaches. Each allows the investigator to deal directly with the intact organism and yet learn with great precision the course of a particular reaction, the consequences of introducing a specific change into a particular structure, and so forth.

Radioisotopes are tools that allow biochemists to observe metabolic transformations in the intact organism. The radioactive forms of specific molecules, often with only the atoms at a specific position in the molecules labeled, allow the radioactive molecules to be located against a background of almost unlimited numbers of other molecules. This sensitivity and specificity are achieved because atomic events, such as radioactive decay, generate energetic, readily detected subatomic particles. Atomic events typically have 10^5 or more times the energy of chemical events. Thus, acetic acid may be prepared with one or both of its carbon atoms radioactive, that is, with unstable, radioactive ^{14}C atoms replacing the far more abundant, nonradioactive ^{12}C atoms. Some of the radioactive acetate, when submitted to an intact organism, will be incorporated into compounds, such as lipids, that are being synthesized by the organism at that time (see, e.g., Chapter 13). The chemical difference between acetate with ^{14}C atoms and acetate with ^{12}C atoms is so small that the two forms of acetate are interchangeable in biochemical reactions. Subsequent analysis of extracts prepared from the organism or its secretions will reveal which compounds, those that are radioactive, have been formed from the radioactive acetate. Although this analysis, like the reductionist approaches described above, requires disruption of cells, the incorporation itself occurred in the intact organism. Thus, if the experiment is carried out properly, the results should reflect the metabolism of the intact organism.

A second approach is the use of nuclear magnetic resonance spectroscopy, abbreviated NMR. NMR allows specific chemical reactions to be monitored in undisrupted cells. For example, the nuclear magnetic resonance signals from inorganic phosphate ions in a cell allow the hydrogen ion concentration (see Section 1.6) in the cell to be estimated.

Recombinant DNA technology (see Chapter 22) allows genes to be moved, *in vitro,* from one location to another in a chromosome. Genes even may be synthesized and/or rearranged *in vitro.* These can then be used to transform organisms and produce new genotypes. The biochemical consequences to the intact organism, of changing only a single gene, then can be assessed.

The fourth example of a nonreductionist, minimally intrusive approach is the transfer of macromolecules or even subcellular organelles from one organism to another. Microinjection and other technologies for accomplishing this now are well developed, at least for transfer to single cells. Often, the macromolecules or organelles will function in the foreign cell, allowing the effects of different, and nearly undisturbed, cellular backgrounds on the functioning of a particular enzyme or protein, for example, to be assessed.

1.3 ARRANGEMENT OF THIS BOOK

This fifth edition has three sections.

Part 1, Structures and Functions of Biological Molecules, is concerned with one of the oldest endeavors of biochemistry: chemical characterization of the macromolecules and the low molecular weight molecules of living systems. Although the cell is the premiere structure of living systems, we reserve an in-depth discussion of it for Chapter 8, the first chapter of Part 2. Chapter 8 is intended to build on preceding descriptions of the components of cells so that the cell, its membranes and compartments, can be presented in biochemical terms.

Part 2 is Energy Metabolism and Biosynthesis of Small Molecules. The highly specific and controlled chemical reactions by which small molecules