PREFACE

This book reviews a range of modern methods for the study of the dynamic processes of individual biological molecules and of molecules in integrated macromolecular and membrane systems. Interest in the dynamic properties of biological structures has grown substantially in the past decade and is well exemplified by the fundamental progress recently made in understanding the relationship between protein structure and protein function. X-ray crystallography has established the details of the three-dimensional structure of proteins to unparalleled resolution, resolving the components of secondary structure, the helical and sheet conformations recognised from previous studies of synthetic polypeptides, and the specific but irregular conformation of the tertiary structural components. The detailed description of molecular interactions achieved in the resulting molecular structure is directly related to the large information content of the diffraction pattern and the precision in measuring it over a considerable period of time. Necessarily, the final models correspond to the time-average of the structure, and the possible time-dependent component of the information is largely suppressed. Parallel solution studies have for many years been interpreted on the basis of the potentiality for conformational change being an essential property of the protein structure, both as regards mechanism of action and also the potentiality for control of this action by the binding of additional "effector" molecules. The resolution of individual steps in the catalytic pathways of enzymes by analysis of steady-state kinetics, and more directly by the study of individual reaction steps using the methods of fast reaction techniques, has emphasized the time-dependent transformations of the typical enzyme during the time-span of a few milliseconds duration of the catalytic process.

There is therefore general agreement that proteins should be considered dynamic structures. Similar arguments can be advanced for all the other groups of biological macromolecules, for model biological lipid bilayer systems and for molecules and macromolecules in their attachment to, or being embedded in, the cell surface. Some motions, for example the rotations of individual groups or the vibrations of certain side chains, may be without direct biological significance, but may indeed be analogues of functionally mobile structures. In order to assess the relevance of the dynamic properties, highly sophisticated experimental methods are clearly required for investigations over a wide range of the time domain. Ideally, measurements should be performed on molecules which are in their truly native state, even devoid of "labels," using monitoring signals of high specificity, and reflecting properties which are amenable to exact theoretical formulation and detailed molecular interpretation. At the molecular level, such observations should be able to produce experimental information on the shortest time-scale which will inspire attempts at simulation by the powerful techniques of molecular dynamics calculations. At the supramolecular level, the formulation of the experimental approach and the interpretation of the observations depends upon the availability of theoretical treatments of the hydrodynamics of real molecules. This is an area where, despite major conceptual advances made in recent years, much remains to be done in extending the treatment from classical models.

In addition to a proper theoretical background, we also require the associated experimental capability for making time-resolved measurements of dynamic properties over a wide time range where the system under study is contained within an environment as close as possible to the natural biological context. Given the diversity of biological systems, this implies the existence of a wide range of problems for investigation. Recent developments include lateral and rotational diffusional motion of molecules in isotropic or anisotropic solution, as well as two-dimensional diffusional motion, lateral and rotational, for molecules constrained by a biological membrane. A long-term goal is the description of the dynamic properties of large assemblies of molecules, organised into the supramolecular structures which are involved in the time-dependent structural and functional evolution of that most complicated molecular machine—the biological cell. Our lack of knowledge of the physical nature of the environment encountered in a true biological system is, at present, a major deficiency in the interpretation of *in vivo* systems.

A wide range of physical methods has become available for the study of the dynamic properties of molecules. Many of these have the common property of being dependent upon the interaction of electromagnetic radiation with matter, and the potential area of concern is extremely large. Some selectivity in subject matter has therefore been necessary in composing this volume; in particular, two important areas at the extremes of the electromagnetic spectrum—X-ray diffraction and nuclear magnetic resonance—are not treated here. A recent volume "Mobility and Function in Proteins and Nucleic Acids" (CIBA Symposium 93; CIBA Foundation, London, 1983) deals with recent advances in applications of both these techniques to dynamic studies of biological molecules.

This volume describes recent studies of the dynamic properties of biological macromolecules, macromolecular assemblies and supramolecular structures. In particular, emphasis is given to the application of modern fast time-resolved optical spectroscopic techniques involving lasers and sychrotron sources, since their unusual power and time-structure provide major advantages for biophysical investigations. The volume is composed of a series of reviews, based initially on lectures given in April 1983 at a meeting in London of the British Biophysical Society. The techniques and topics presented here include the hydrodynamics of complex macromolecules, laser correlation spectroscopy, triplet-state probes of molecular motions and chemical kinetics, fluorescence and phosphorescence depolarization, fluorescence photobleaching techniques, electron paramagnetic resonance techniques, time-resolved fluorescence spectroscopy, synchrotron-based studies of time-resolved optical spectroscopy and time-resolved X-ray scattering in macromolecular self-assembly, and laser techniques for nanosecond temperature jump relaxation and picosecond optical spectroscopy.

Even with such diversity of interests and techniques, it is not possible to present a fully comprehensive account of this rapidly developing area of biophysical research. However, the reader will detect a common purpose in these contributions, and a common methodological approach based ultimately upon our theoretical understanding of the behaviour of ideal molecules in well-defined systems. In real experimental investigations, the important areas of biophysical research involve observations of nonideal molecules in imperfectly defined systems. Moreover the observations, by their very nature, can provide only an incomplete picture of actual behaviour. This fact enforces the need for the careful refinement of methods and techniques, and the development of the associated theory, plus the application of several experimental techniques to the same problem. In this regard, it is perhaps important to point out that apparently contradictory evidence as to the nature and degree of motion within an assembly may not in fact be incompatible, when the time-ranges of the different methods are taken into account. The methods described here allow investigations over the range of picoseconds to hours—some sixteen orders of magnitude. This suggest that dynamics, particularly in the fastest ranges now opened to investigation with the new technology, will continue to provide an enormously fruitful area of research for a considerable time to come. The editors hope that this volume will assist in introducing those unfamiliar with this area to recent developments in the techniques and their biophysical applications. For those already involved with some of these techniques, we hope that there will be additional stimulus in this fuller illustration of the potentiality of the present methods and inspiration for the development of even more powerful methods for future research.

We should like to record our thanks primarily to the contributors to this volume whose work this represents. Also our thanks go to the staff of Academic Press, London, for their considerable patience and expert assistance in the preparation of this publication and to David Clark and Edward Manser for their work in compiling the index.

P. M. Bayley*
R. E. Dale

*Dr. Bayley's mailing address is as follows:
National Institute for Medical Research
The Ridgeway
Mill Hill
London MW7 1AA, England