Book Reviews

Luminescent Spectroscopy of Proteins by Eugene A. Permyakov

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Various luminescence techniques are of unquestioned value and applicability for the study of the solution properties and interactions of proteins. The amino acids tryptophan, tyrosine, and phenylalanine, provide intrinsic reporter groups. Information can be obtained about the mobility, solvent accessibility, and polarity of the microenvironment of these groups. Because of overlap in the fluorescence and phosphorescence spectra of individual lumiphores, such information about specific residues requires the resolution of spectral contributions, which can be a major challenge. Even when such resolutions are not possible, luminescence methods can also be useful for monitoring the thermodynamics and kinetics of induced conformational changes, subunit association, or ligand binding reactions.

"Luminescent Spectroscopy of Proteins" by Eugene A. Permyakov is a modest-sized monograph that reviews a number of aspects and applications of fluorescence and phosphorescence methods to the study of proteins. The book begins with a treatment of the fundamental principles of luminescence spectroscopy, including discussions of wave functions, Jablonski diagrams, the Frank-Condon Principle, and various excited state reactions (radiative transitions, intersystem crossing, collision quenching, and resonance energy transfer). A second chapter focuses on the luminescence properties of the above three amino acids, including discussion of the quenching functional groups that comprise amino acid side chains. The final chapter discusses luminescence properties of tryptophan, tyrosine, and phenylalanine residues in proteins. The emphasis is almost entirely on steady-state fluorescence properties, and many of the examples are studies with calcium binding proteins α-lactalbumin and parvalbumin. A particular strategy presented in this chapter is the assignment of individual protein tryptophan residues into one of five classes (internal and unperturbed; internal and forming a 1:1 exciplex with a neighboring polar group; internal and forming a 2:1 exciplex with neighboring polar groups; partially solvent-exposed; and fully solvent-exposed). In using fluorescence methods to monitor protein conformational transitions and ligand binding, the author correctly points out that only certain fluorescence observables report the fractional conversion from one state to another. Also, the author describes "phase plots" (observing fluorescence intensity changes at two emission wavelengths to see if the changes occur in concert) to monitor such conversions and to test whether there is an intermediate state(s). The presentations on ligand and proton binding are among the more informative in the book.

The author states that the purpose of his book is to provide general information at a level that is intermediate between textbooks and specialized volumes on fluorescence spectroscopy. His emphasis is clearly on steady-state methods, because these are more available to most researchers. There are some merits to these purposes, but I find this book to be narrow in its treatment of the field, and I feel that it barely serves its intended purpose. The brief coverage of spectroscopic instrumentation can be accepted, as such information can be found in other books and review articles. However, the light treatment of time-resolved fluorescence studies and resonance energy transfer studies with proteins, and the complete absence of a description of fluorescence anisotropy methods and data, make this book of limited value for someone seeking an introduction to modern applications of fluorescence spectroscopy to proteins and other macromolecular systems. Almost all of the applications to specific protein systems are from previous publications by the author and his colleague, Dr. E. A. Burstein; very little data from other labs are presented. Actually, the last chapter is a collection of studies by these researchers on calcium binding proteins.

There are a couple of specific problems that I have with the interpretations and analysis of fluorescence data. I do not see much value in the attempt to classify individual tryptophan residues into one of five categories (see above). In my opinion, the range of possible microenvironments of tryptophans in proteins is too broad to be described by five categories. I also prefer that nonlinear least-squares methods be used to fit models to data, rather than relying on linear plots (such as the above mentioned "phase plot"). Finally, although the book is fairly easy to read, there are several minor mistakes, particularly involving the chemical structure of functional groups.