developing embryos, dipeptidases from *Escherichia coli* B and from Ehrlich Lettré mouse ascites-tumour cells, and a selection of endo- and exo-peptidases, many of which were not dealt with in Volume 19.

It will be a matter of regret to some people that, presumably owing to pressure on space, it has not been possible to bring up to date many of the much-studied enzymes that were discussed in Volume 19. For example, whereas there are sections on bromelain in both volumes, papain and ficin do not feature in Volume 45. Indeed, this must be a general problem that the Editors need to look at. Their understandable desire to include some basic material on more and more enzymes from more and more sources seems inevitably to deny them the opportunity to keep the reader up to date with the relatively well-studied enzymes on which mechanistic advances are so often made. But perhaps we expect too much. The excellence of some contributions to *Methods in Enzymology* may have led some of us to expect not only information about methods of purification and assay but also what amounts to a summary of a relevant chapter (if one exists) of *The Enzymes*. We may even have to resign ourselves to reading about a particular enzyme in both of these invaluable series!

In addition to the specialized sections on particular enzymes, Volume 45 contains three subsections on 'General Aspects': (a) an account of sensitive methods for the assay of trypsin-like enzymes, notably those using the fluorogenic active-site titrant methylumbelliferyl p-guanidinobenzoate, developed by Elmore's group, and the fluorogenic substrate the methylumbelliferyl ester of N-benzyloxy carbonyl-L-lysine, developed by Shaw's group; (b) a method of determining proteolytic activities by using casein substrates that relies on determination of released amino groups by the ninhydrin method; (c) active-site titration of the cysteine proteinases.

The reviewer felt obliged to overcome a natural reluctance to refer to work from his own laboratory and point out that there is a more convenient method of specifically titrating active sites in the cysteine proteinases, even in the presence of other thiols, than that discussed extensively by Kézdy & Kaiser in Section I(1). These authors suggest the use of a-bromo-4-hydroxy-3-nitroacetophenone and give details of a two-step synthesis. For those who can bring themselves to forego the undoubted delights of synthetic organic chemistry, it may be of interest that there is a commercially available reagent that will do the job just as well and perhaps better. The reagent, 2,2'-dipyridyl disulphide, is at first sight similar to the widely used Ellman's reagent 5,5'-dithiobis-(2-nitrobenzoic acid) but differs from it in two important respects. First, as was pointed out by Grassetti & Murray, its reactions with thiols below pH9 are essentially stoicheiometric. Secondly, it is a ‘two-protonic state electrophile’ and, when activated by protonation or hydrogen-bonding, reacts orders of magnitude more rapidly with the low-pKₐ thiol groups of inactive centres than with simple thiols or with denatured enzyme. This ability to react much more rapidly with intact active centres than with simple thiols in acidic media is shared by a-bromo-4-hydroxy-3-nitroacetophenone, but the better spectroscopic characteristics of the 2,2'-dipyridyl disulphide reaction facilitate titration of thiol groups in proteins.

K. BROCKLEHURST

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**Handbook of Enzymatic Methods of Analysis**

GEORGE G. GUILBAULT

*Marcel Dekker, New York and Basel, 1976, pp. 752, Sw.Fr. 126.00*

Admirers of Dr. Guilbault's numerous important contributions to analytical enzymology will be delighted to learn that his earlier book has been rewritten and brought up to date. In his preface he points out that in 1970 some 150 purified enzymes were available commercially, but this number has now more than doubled and in most cases
higher standards of purity have been achieved. Moreover about 50 enzymes are now available in stable insolubilized forms suitable for re-use over and over again.

The book consists of six chapters devoted to: general principles; the assay of enzyme activities; analysis of substrates; assay of purines, nucleotides, coenzymes, activators and inhibitors; the immobilized enzyme and its use in enzymic analysis; the automation of enzymic methods of analysis. These are followed by appendices summarizing symbols and abbreviations, the classification of enzymes, and a list of commercial sources of enzymes and reagents, and finally there are author and subject indexes.

In the chapter on the assay of enzyme activities, 40 of the commonest enzymes are discussed. Brief summaries of the properties of each enzyme, the reactions catalysed and its significance in disease, general biochemistry or industry are followed by reviews of assay procedures. Practical details are not included, but references to the original literature and to such works as Bergmeyer’s *Methods of Enzymatic Analysis* are given. The use of enzyme reactions in the determination of substrate and coenzyme concentrations is similarly treated.

Perhaps the most intriguing part of the book is that devoted to immobilized enzymes. After a general discussion of the various methods used, e.g. micro-encapsulations, adsorption, covalent cross-linking, inclusion in gel lattices and covalent binding to water-insoluble matrices, Dr. Guilbault gives an excellent account of the construction and applications of enzyme electrodes, which are extensively used in the determination of glucose and other substances in body fluids.

This book contains much information of value to biochemists, especially those concerned with analytical problems in the clinical laboratory, the food industry and industrial processes generally. The treatment is comprehensive, but in parts rather uncritical. For instance, hydrazone methods for lactate dehydrogenase, aspartate transaminase and alanine transaminase are mentioned without clear indications of their obsolescence. The clinical interpretations are generally sound, but workers in the diagnostic field will require much more than the brief summaries provided. There is no more than passing mention of reference methods, which have greatly exercised clinical biochemists during the past decade. A minor source of confusion arises from errors in the initials of certain authors: thus in the index the contributions of the late E. J. King appear mixed with those of J. King. The reviewer would also have liked to see more expression of the author’s personal opinions, based on his vast experience.

These, however, are relatively minor blemishes on an otherwise well-prepared book, which can be recommended as basic reading for analytical enzymologists and as a reference source.

J. H. WILKINSON

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**Analytical Applications of Complex Equilibria**

J. INCZÉDY (JULIAN TYSON, Translation Editor)

*Ellis Horwood, Chichester, 1976, pp. 328, £17.50*

The blurb on the jacket tells us that the introductory part of this book reviews the chemistry of complexes, complex-formation equilibria and those functions with the help of which the composition of the solutions containing complexes as well as the concentrations of the individual components can be calculated simply. Indeed the first 181 pages are devoted to these topics. However, the main burden is that problems can usually be solved by the application of a few well-known ideas: the Law of Mass Action, the concept of $\bar{n}$, the average ligand number, the law of conservation of mass and the electroneutrality condition. Good second-year stuff.

The next 135 pages deal with analytical applications: gravimetric analysis, precipitation titrations, redox titrations, polarography, spectrophotometry, liquid/liquid