Sidney P. Colowick and Nathan O. Kaplan

Methods in ENZYMOLOGY

Volume XXXIV

AFFINITY TECHNIQUES

Enzyme Purification: Part B

Edited by

William B. Jakoby and Meir Wilchek

Methods in Enzymology

Volume XXXIV

Affinity Techniques

Enzyme Purification: Part B

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William B. Jakoby

SECTION ON ENZYMES AND CELLULAR BIOCHEMISTRY
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Preface

The last few years have brought progress in affinity chromatography to what must surely be the log phase of development. The interest and enthusiasm for affinity methods is understandable since all of us faced with the problem of purifying macromolecules would prefer an "easy way." The way of affinity chromatography literally seizes on the specificity of the macromolecule to bring about adsorption of only that population with such specificity. Ideally, specificity in the form of a competing ligand is again used to desorb the macromolecule. With good fortune, a tissue extract may be purified to a degree approaching homogeneity for one protein by a single pass through an appropriate affinity column. That the goal is seldom attained does not preclude aiming for it; the fact that it is occasionally reached serves as a spur.

We would have been delighted to be able to present definitive studies which could be used as a clear outline for the design of new systems. Unfortunately, this is not possible. In this explosive phase of development our choice has been one of waiting until the point of definition arrives or of presenting the developments of affinity methodology, as applied to purification, in its present imperfect form. Obviously, we have chosen the latter course. Despite the limitations, we believe that we are now at a stage in which the simplistic notions of affinity chromatography can be examined; there is just sufficient experience to allow beginning guidelines to be formed and suggestions for correction to be advanced.

The result of assembling such experience is a volume in which the organization is imperfect and in which there is much repetition. Methods of purifying dehydrogenases, for example, are presented in several separate sections, and certain individual enzymes claim similar distribution. We chose not to mention the number of descriptions for the best means of adding cyanogen bromide. Yet we included all this material and allowed the repetition so that the investigator contemplating the affinity approach can both obtain an idea of what may be best for the individual system and what the expectation may be as to the limits to which a method can be stretched. Since the titles of the articles cannot be completely informative, we have added separate lists of ligands and of the macromolecules which have been purified with them. In addition, much of the literature on purification methods which was not included is referred to in the first article.

Although our approach and concentration have been oriented toward enzyme purification, affinity methods are presented for such diverse sys-

tems as cells, antibodies, and specifically modified proteins. The material on antibody serves mainly as a guide to the enzymologist since methods specific to working with these species of protein have been examined in great detail elsewhere.

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MEIR WILCHEK

METHODS IN ENZYMOLOGY

EDITED BY

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