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INTRODUCTION TO PAPERS ON POLYACRYLAMIDE GEL ELECTROPHORESIS

The field of polyacrylamide gel electrophoresis (PAGE) has undergone explosive development. Today it stands as one of the most popular methods for fractionation of proteins (and other charged macromolecules) and for testing homogeneity. The use of PAGE as a quantitative physical-chemical tool, for example, in the estimation of molecular weight, rather than as a qualitiative method, is already well established. Other applications, such as for measurement of free mobility, molecular net charge, isoelectric point, and apparent diffusion coefficient, are just beginning to be exploited. A recent article (*1*) and two monographs (*2,3*) review the progress in this field. However, before we can make further progress in this direction and evaluate the precision and accuracy of the results obtained, it is necessary to examine several basic assumptions, and ask:

1. Is the gel completely polymerized (e.g., is a 10% gel really a 10% gel)?
2. What is the reproducibility of polymerization?
3. What are the effects of polymerization catalysts on the buffer (pH, ionic strength, boundary displacements), on the pore size of the gel, and on the protein?
4. What is the effect of gel cross-linking on the nature of the gel, the pore size, and on observed mobilities, retardation coefficient, and extrapolated free mobilities?
5. What is the oxidation-reduction potential of polyacrylamide gels, how does this affect proteins, and how can this be altered?
6. How valid are the predicted properties of T. M. Jovin's computer output of 4269 multiphasic buffer systems, which permit steady-state stacking (SSS), unstacking, and restacking over the entire pH spec-

trum? What are the effects of polyacrylamide gel on the operative properties of the stacking and separation gel buffers (pH, specific conductance, boundary displacements, etc.)?

7. Can SSS and isotachophoresis [SSS with spacer ions, (ITP)] be exploited for both analytical and preparative-scale protein fractionation, using polyacrylamide gels as an anticonvective supporting medium?

In addition to these "basic" questions, certain technical problems involving apparatus and procedures have been investigated: (1) development of a slab apparatus for multiple samples, for use with multiphasic buffer systems, at any pH, at either 0 or 25°C; (2) development of a gradient-maker designed specifically for use in PAGE.

The papers in this issue of *Separation Science* deal with these questions and problems. The answers obtained are still preliminary and often incomplete in several respects. Hopefully, these "pilot" studies will be supplanted by more definitive studies: directions for further study are outlined specifically in each paper. Also, it is hoped that the present results will be useful to others as they have been in the authors' laboratory—by indicating the numerous assumptions and limitations inherent to PAGE, and ways to surmount, or at least minimize, the effects of these limitations.

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