
Preface

A mixture of two polymers, or one polymer and a salt, in an aqueous medium separates into two phases: this phenomenon is useful in biotechnology for product separations. Separation of biological molecules and particles in these *aqueous two-phase systems* (ATPS) was initiated over 40 years ago by P.-Å. Albertsson, and later proved to be of immense utility in biochemical and cell biological research. A boost in the application of ATPS was seen when problems of separations in biotechnology processes were encountered. Its simplicity, biocompatibility, and amenability to easy scaleup operations make the use of ATPS very attractive for large-scale bioseparations. Despite the advantages ATPS enjoys over other separation techniques, the application of two-phase systems has for a long time been confined to selected laboratories. Recent years have, however, shown a trend in which increasing numbers of researchers employ two-phase partitioning techniques in both basic and applied research.

Aqueous Two-Phase Systems: Methods and Protocols is a collection of cutting-edge methods intended to provide practical guidelines for those who are new to the area of separations in two-phase systems. Besides the established methods, many newly developed techniques with potential applications in biotechnology are also described. As an introduction, the first chapter provides a brief general overview of ATPS and its applications. The remainder of the volume is broadly divided into five sections. The first two sections are basic, describing methods for ATPS preparation and characterization, and the various partitioning techniques that may be employed. Multistage partitioning increases the resolving power of ATPS, allowing separation of materials differing only very slightly in physicochemical properties.

Partitioning applied to soluble molecules and particulates has been dealt with in the third section, where examples of different categories of materials are presented. Once the reader is acquainted with the methodology and the “tricks” to be used to obtain the desired partitioning, the separation technique may then be applied to any material of interest. Separation of particulates—including whole cells, membranes, and organelles—has been a major achievement of ATPS, one that greatly facilitates studies on cells and their properties. Purification of viruses is another successful example. With regard to soluble

molecules, partitioning has been most commonly applied to the separation of macromolecules, since their distribution between the two phases is influenced to a greater extent by a system variation than is the distribution of small molecules. This has enabled the application of ATPS even as an analytical tool to determine, e.g., the concentration and isoelectric point of proteins, molecular interactions, conformational changes of biomolecules, and so on. Lately, applications of ATPS in the separation of small molecules have also emerged. Molecules with defined properties are proving useful for understanding the interactions involved during partitioning, which would be helpful in the selection of appropriate phase systems for specific separation problems.

The main application of ATPS in biotechnology has been the isolation and purification of proteins; hence a significant part of *Aqueous Two-Phase Systems: Methods and Protocols*, compiled as Part IV, is devoted to this subject, including a glimpse of the large-scale handling of the two-phase separations. The real success of this technique has been in the extraction of proteins directly from crude feedstocks, where it has provided clarification, concentration, and even some purification in a single step. The extraction of proteins by spontaneous partitioning alone necessitates optimization of various parameters. The need to improve the selectivity of extractions has also led to exploitation of charge–charge, hydrophobic, and affinity interactions, in which specific binding groups are located in the phase used as the extractant. Integration of ATPS with other separation techniques provides scope for facilitating such selective extractions. Limiting the material costs for large-scale purposes still remains a challenge. The recycling of phase components is thus essential, which is easily done for some phase chemicals, but not for others. New phase materials with easy recyclability are being studied.

There has been interest in using aqueous two-phase systems in another area of biotechnology, i.e., *in situ* product recovery during biocatalytic processes. This concept has been presented in the last section of the volume. Analogous to aqueous systems are the newly developed polymer–polymer systems in organic solvents, which are useful with synthetic reactions.

My hope is that *Aqueous Two-Phase Systems: Methods and Protocols* will not only prove helpful in your research, but will also lead to discovery of the surprises and pleasures of aqueous two-phase systems separations. I wish to thank all the contributors to this volume for sharing their knowledge and practical experience with the reader. Special thanks are due to Associate Professor Göte Johansson, Emeritus Professor P.-Å. Albertsson, and Professor Bo Mattiasson for their useful suggestions.

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