SEPARATION SCIENCE APPLIED TO ANALYSES ON BIOLOGICAL SAMPLES

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The historical development, present 'state-of-the-art' and future projections of separation science in the analytical context are presented, using specific examples to illustrate various aspects. Despite notable advances in the separation technology that must precede the application of a specific measurement procedure and/or detection device, many problems and difficulties remain. The difficulties are compounded by the development of instrumentation often presenting an apparently high degree of automation but possibly with unsuspected jeopardy to the reliability of analytical results.

Human beings tend to think in terms of the development of virtually anything as a temporal sequence of events. Man progresses in life from infancy, through youth, to adulthood, then on to old age; books are often written with a prologue at the beginning and an epilogue at the end; scientific papers have a progression from introduction to discussion. Indeed, in preparing to write a scientific manuscript or to make a presentation at a scientific meeting, one is often torn between reporting the results of a series of experiments in the temporal sequence in which they were performed, or re-ordering them to make more 'sense' to the audience.

This temporal frame of development has also existed for analytical chemistry in general and for separation science in particular (Fig. 1). Less than 50 years ago, the analytical chemist was virtually restricted to two technologies for separation of substances from the complexity that characterizes most biological materials. Distillation procedures date back to the days of alchemy: if the
desired substance was sufficiently volatile, it could be isolated by heating the entire mass, then cooling the vapours to produce a condensate. Sophisticated versions of such procedures are still in use today: micro-distillation apparatus, cold-finger procedures, Kjeldahl procedures remain in use for specialized purposes.

Along with distillation procedures that took advantage of the differing vapour pressures or boiling points of chemicals, the early separation scientists made use of various precipitation procedures to separate and isolate substances on the basis of differing solubilities. Such techniques were versatile: they served well in both research and teaching. I can still remember my first chemistry laboratory experiment as an undergraduate. We were given an unknown mixture of NaCl, naphthalene and sand. The only equipment issued consisted of an analytical balance, beakers, a Bunsen burner, a porcelain crucible, a filter funnel with papers, and a supply of benzene and water. The recommended initial procedure was to weigh the unknown, then add a modest amount of benzene and stir the entire mass for 15 min. After pouring this through a filter paper, the filtrate was collected in a beaker and allowed to evaporate in a fume hood overnight; the naphthalene crystals were then weighed. The residue remaining in the first filter paper cone was then dissolved in water. The process of filtration and evaporation of that filtrate overnight led to crystals of NaCl that could be weighed. Finally, the material remaining in the second filter paper cone was transferred to a crucible and heated with the burner; this procedure left dry sand to be weighed.