system applied to the resolution of histones. Aggregate formation of some of the histone fractions may be one of the major factors responsible for some of the numerous bands reported in these electrophoresis systems. Employment of urea, as suggested here and in the recently reported preparative electrophoresis of proteins on polyacrylamide gels[5], may be necessary to eliminate the anomalous behavior of histones in PAGE systems.

Acknowledgement

This work was supported by Grant AM-05169 from the National Institutes of Arthritis and Metabolic Diseases, U.S. Public Health Service.

Department of Chemistry, California State College at Long Beach, Long Beach, Calif. (U.S.A.)

R. D. BAUER
W. A. THOMASSON

1 H. C. MCALLISTER, Y. C. WAN AND J. L. IRVIN, *Anal. Biochem.*, 5 (1963) 321.
2 A. DREIDGET, L. D. JOHNSON AND A. M. MARKO, *Can. J. Biochem. Physiol.*, 41 (1963) 2507.
3 K. MURRAY, *Ann. Rev. Biochem.*, 34 (1965) 209.
4 G. F. VANDE WOUDE AND F. F. DAVIS, *Anal. Biochem.*, 12 (1965) 444.
5 P. H. DUESBERG AND R. R. RUECKERT, *Anal. Biochem.*, 11 (1965) 342.

Received November 5th, 1965

J. Chromatog., 22 (1966) 496–498

Partition chromatography of some organic acids on a cation exchange resin

It is known that air dry 10% divinylbenzene cross-linked sulfonated polystyrene resin, when equilibrated with 70% aqueous acetone, absorbs water preferentially, so that the water content of the liquid absorbed is found to be higher than that of the external solution[1]. The distribution of an organic substrate which is sparingly soluble in water between these two phases will be greatly in favour of the outer solution and thus provides an explanation both for the negative adsorption and for the low catalytic activities of sulfonated polystyrene resin in the hydrolysis of various aliphatic esters in 70% acetone in water[2].

RUCKERT AND SAMUELSON[3] have also shown that strongly polar nonelectrolytes, such as sugars, can be taken up effectively from mixed solvents by means of ion exchange resins. On the basis of this, they were able to separate sugars chromatographically on anion exchange resin. Dowex 1-X8 in the sulfate form was used as the stationary phase and the eluent was 74% aqueous ethanol[4].

Separation of various organic acids using similar chromatographic systems has been attempted, in some cases successfully. In the present work cross-linked sulfonated polystyrene resin, Amberlite CG-120, in the hydrogen ion form was used. Citric acid, malic acid and tartaric acid could be separated using the mixture acetone–dichloromethane–water (160:100:9, v/v) as eluent, while an eluent containing more dichloromethane (acetone–dichloromethane–water (20:15:1, v/v), made possible the separation of fumaric acid, glutaric acid and succinic acid.

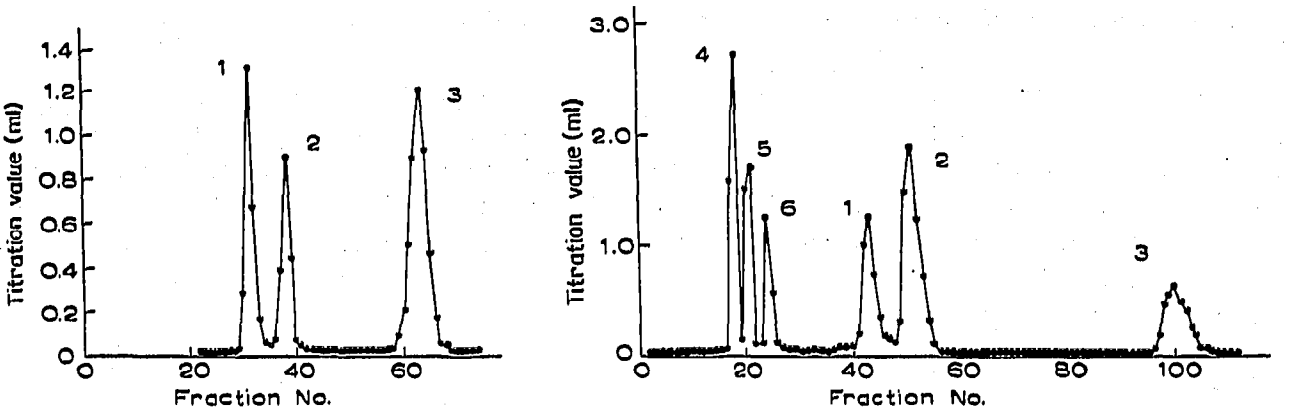

Fig. 1. Elution of organic acids. The compounds in the order of their elution from the column are: citric acid (1), malic acid (2) and tartaric acid (3). Column size: 0.75 × 75 cm. Eluent: acetone–dichloromethane–water (160:100:9, v/v). Fraction size: 40 drops.

Fig. 2. Elution of organic acids. The compounds in the order of their elution from the column are: fumaric acid (4), glutaric acid (5), succinic acid (6), citric acid (1), malic acid (2) and tartaric acid (3). Column size: 0.78 × 81 cm. Eluent: acetone–dichloromethane–water (20:15:1, v/v). Fraction size: 42 drops.

Amberlite CG-120 (200–300 mesh, screened wet in the sodium ion form) was washed successively on a glass filter with 6 *N* hydrochloric acid (10 volumes), water, 2 *N* sodium hydroxide (10 volumes), water, 2 *N* hydrochloric acid (10 volumes), water and then 95 % acetone in water (10 volumes). The washed resin was finally washed and equilibrated with the solvent to be used for chromatography, and suspended in two volumes of this solvent. The suspension was poured into the chromatographic tube and allowed to settle under gravity. When about 200 ml of the solvent had passed through the column, it was ready for use. Samples were dissolved in the solvent to be used for chromatography and 1 ml of the solution was introduced on to the column. Elution was performed at 15° to 20° and the effluent was collected in fractions of 40 or 42 drops. The flow rate was about 7 fractions per hour. Each fraction was titrated with 0.01 *N* sodium hydroxide and the titration values were plotted against the fraction number. As shown in Figs. 1 and 2, the elution sequence of the organic acids studied was similar to that observed on partition chromatography on hydrated silica gel. Recovery of these acids ranged from 90 to 100 %.

The column could be used repeatedly. The chromatographic system described above seems to be useful for the separation of acidic and neutral polar substances. Purification of sulfoxides on Dowex 50 in the hydrogen ion form, using benzene and ethanol as solvents[5], may be regarded as an example of this.

Department of Genetics, Osaka University Medical School, Osaka (Japan) TOKUICHIRO SEKI

1 C. W. DAVIES AND G. G. THOMAS, *J. Chem. Soc.*, (1952) 1607.
2 V. C. HASKEL AND L. P. HAMMETT, *J. Am. Chem. Soc.*, 71 (1949) 1284.
3 H. RUCKERT AND O. SAMUELSON, *Svensk Kem. Tidskr.*, 66 (1954) 337.
4 O. SAMUELSON AND B. SWENSON, *Anal. Chim. Acta*, 28 (1963) 426.
5 V. HORÁK AND J. PECKA, *J. Chromatog.*, 14 (1964) 97.

Received November 1st, 1965