

Wir haben versucht, die Trennung von Betain und Cholin mit nur *einem* Austauscher zu erreichen. Für die Isolierung der in Zuckerrübensäften vorkommenden freien Aminosäuren verwenden wir eine Säule mit dem stark sauren Kationenaustauscher Lewatit S 100 (H-Form) (NIEMANN<sup>8</sup>), der die anorganischen Kationen, die Aminosäuren, Betain und Cholin absorbiert (SCHNEIDER *et al.*<sup>9</sup>). Aminosäuren und Betain werden mit 2 *N* Ammoniak eluiert. Nach dem Abdampfen des Ammoniaks wird Betain mit Reineckesalz in saurer Lösung gefällt und direkt gravimetrisch oder nach Auflösung in Aceton colorimetrisch<sup>2</sup> bestimmt. Der Austauscher wird nach Spülung mit Wasser mit 2 *N* Salzsäure gewaschen und so für den nächsten Versuch regeneriert. Dabei wird gleichzeitig das Cholin eluiert. Das Eluat wird eingeeengt und darin das Cholin mit Reineckesalz in saurer Lösung gefällt und gravimetrisch oder colorimetrisch<sup>2</sup> bestimmt.

Unsere Modellversuche mit Standardlösungen und Zuckerrübensäften ergaben eine quantitative Elution und eine eindeutige Trennung von Betain und Cholin. Wir konnten durch papierchromatische Untersuchungen der Reineckatfällungen die Befunde von BREGOFF *et al.*<sup>10</sup> und CARRUTHERS *et al.*<sup>6</sup> bestätigen, dass in Zuckerrüben neben Glycinbetain keine anderen Betaine vorkommen.

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<sup>3</sup> H. E. STREET, A. E. KENYON UND G. M. WATSON, *Biochem. J.*, 40 (1946) 869.

<sup>4</sup> D. D. CHRISTIANSON, J. S. WALL, R. J. DIMLER UND F. R. SENTI, *Anal. Chem.*, 32 (1960) 874.

<sup>5</sup> O. HRDÝ UND S. LOCHMANOVÁ, *Českoslov. farm.*, 9 (1960) 335; *ref. Z. anal. Chem.*, 180 (1961) 310.

<sup>6</sup> A. CARRUTHERS, J. F. T. OLDFIELD UND H. J. TEAGUE, *Analyst*, 85 (1960) 272.

<sup>7</sup> H. G. WALKER UND R. ERLANDSEN, *Anal. Chem.*, 23 (1951) 1309.

<sup>8</sup> A. NIEMANN, *Naturwiss.*, 47 (1960) 514; *Z. anal. Chem.*, 181 (1961) 543.

<sup>9</sup> F. SCHNEIDER, E. REINEFELD, F. AMDING UND B. ZENKER, *Zucker-Beih.*, 4 (1960) Heft 1.

<sup>10</sup> H. M. BREGOFF, E. ROBERTS UND C. C. DELWICHE, *J. Biol. Chem.*, 205 (1953) 565.

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## Wick systems in circular paper chromatography

Many techniques for the controlled transport of solvent from the reservoir to the paper have been studied in order to improve the reliability, simplicity, convenience, and speed of circular paper chromatography. These techniques have ranged from the introduction of solvent by means of a self-regulating pipette<sup>1</sup> to the use of various wick systems.

In one group of wick systems, direct contact between solvent and paper is effected by a small, cut-out portion of the chromatogram dipping into a solvent reservoir. RUTTER<sup>2</sup> used a single cut-out strip for the entire chromatogram, whereas PHILIPPU<sup>3</sup> used an individual wedge for each segment of the chromatogram. In a second group of wick systems, indirect contact between solvent and paper is effected by means of

some connecting agency. Among the devices that have been used are capillary tubes<sup>4</sup>, single or multiple cotton threads<sup>5</sup>, and detachable filter paper wicks of various shapes<sup>6</sup>, such as a cylinder of rolled paper, a small paper cone, or simply a paper strip.

A new indirect contacting technique, based on the use of an inert, porous cylinder as the wick, has been found in practice to be somewhat simpler and more convenient than the above techniques. The desired properties of this type of wick are satisfactorily evinced by an unglazed ceramic filtering crucible (style No. 528-30, Laboratory Equipment Corp., St. Joseph, Michigan). The crucible, as a rigid unit, is also ideally suited as a center support for the paper in large-diameter (11 inch), circular chromatographic chambers (Pyrex pie plates, Corning Glass Works, Corning, New York, edges ground flat). The same unit may be used repeatedly, facilitating reproducibility among runs, although its low cost permits it to be disposed of after use. A typical system is shown in Fig. 1. Similar application of other devices made from porous or sintered glass or metal, foamed plastic, etc., in hollow or full shapes is readily apparent.

The porous cylinder wick technique appears equivalent in performance to several widely used wick systems when judged by the ability to separate a mixture of amino acids. The data listed in Table I indicate that the wick system does not affect the individual amino acid  $R_F$  values within experimental error. A similar lack of dependence was shown by the zone widths.

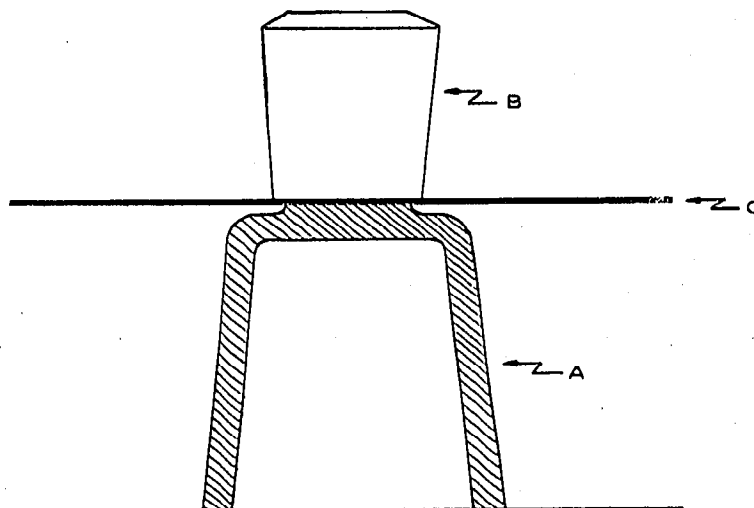


Fig. 1. Porous cylinder wick system. A = unglazed ceramic crucible; B = glass weight; C = chromatogram.

All four wick systems gave symmetrical, mildly eccentric, elliptical development patterns, with an average ratio of major to minor axis of  $1.11 \pm 0.01$ .

The rate of solvent flow for each wick technique can be influenced by the geometry and characteristics of the wick and by the distance between paper and solvent. It has been shown<sup>5</sup> for the cotton thread technique that the square root of the solvent flow is proportional to the length of the wick and that the rate depends on the number and the thickness of the strands, and on their position with respect to the center of the paper disc. The rate of flow in the capillary technique similarly depends on the

TABLE I

OBSERVED  $R_F$  VALUES<sup>a</sup> OF INDIVIDUAL AMINO ACIDSSolvent system: organic phase of the mixture *n*-butanol-glacial acetic acid-water in the ratio 40:10:50

	Wick system			
	Capillary tube	Cotton plug	Porous cylinder	Philippu
DL-Aspartic acid	0.37 ± 0.01	0.34 ± 0.02	0.36 ± 0.02	0.34 ± 0.01
β-Alanine	0.44 ± 0.02	0.42 ± 0.01	0.44 ± 0.01	0.41 ± 0.01
DL-Valine	0.58 ± 0.01	0.56 ± 0.01	0.57 ± 0.01	0.56 ± 0.01
DL-Isoleucine	0.68 ± 0.01	0.67 ± 0.01	0.69 ± 0.01	0.67 ± 0.01

<sup>a</sup> Values represent the average of four runs and the average deviation.

diameter and height of the capillary<sup>1</sup>. We have found that the rate may also be varied by the use of either a bundle of thin-walled capillaries (*e.g.*, melting point tubes) or a series of concentric tubes with capillary spacing. The solvent flow rate may be modified in the porous cylinder technique by use of a device of different porosity, size or shape, and in the Philippu technique, by use of a different effective wick length and width. The average rate of chromatographic development observed for each wick system, in units of time (h) for the solvent front to move 12.3 cm, was as follows: capillary 5.1, cotton plug 4.4, porous cylinder 4.5, and Philippu, 2.5.

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