Glass Strip Collector for Column Chromatography

THE glass strip collector for column chromatography to be described presents a visible result of an analysis in a manner similar to one-dimensional paper chromatography, but without the need for specific development reagents. Fractions can be isolated and estimated quantitatively.

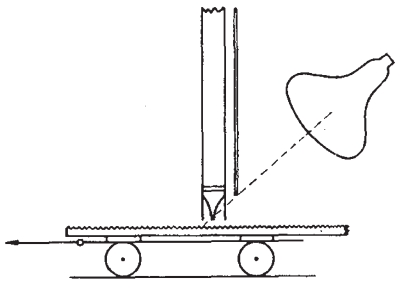

Fig. 1

A serrated glass strip (Fig. 1), 2–4 in. wide, rests on a carriage pulled along at a suitable constant speed beneath the drip point. The solvent is evaporated by heat from an infra-red lamp. A shield protects the column and the dried fractions. Excessive heat may cause longitudinal cracks in the strip. 'Vitrolite' panelling glass is used, serrated on the back with twelve serrations per inch. For colourless crystalline substances black glass is used; in other cases white or coloured glass may be preferable. Oily fractions are 'developed' by dusting with talc and brushing off the excess: this also in some cases promotes crystallization, which may take up to an hour for completion. The drip-point is shrouded to surround it with saturated vapour. This prevents creeping. For 2-in. glass a drop rate of 7 drops/min. of petroleum ether is suitable.

Fig. 2 shows the result before and after talc development from a crude benzene hexachloride extract, using a 1-cm. column with 30 gm. silica gel[1] and a speed of 0·7 in./hr.

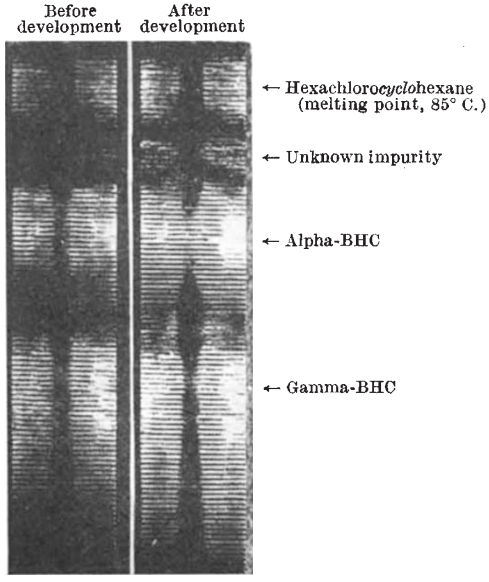

Fig. 2. The same strip before and after talc development

For isolation and quantitative recovery of fractions, the strip is hung on edge in a loop of braided nylon radio panel cord, with a smaller knotted loop at the lowest point to direct drips of solvent into a receiver. The selected fraction is washed off with any suitable solvent applied with an eye-dropper. With pure gamma-B.H.C., 20-mgm. amounts gave 98 per cent recovery when the minimum requisite heat was applied.

Flat glass was used in the early development of the method, with a T-shaped distributor hooked on to the drip-point of the column. Levelling and flow-rate were highly critical and creep was also troublesome. With the serrated glass the management is much less critical, surface tension providing the effect of closed-end troughs.

The carriage has been used also for mounting a straight rack of test-tubes as described by Grubhofer[2].

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[1] Aepli, O. T., Munter, P. A., and Gall, J. F., *Anal. Chem.*, **20**, 610 (1948).
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Biosynthesis of Acetylcholine Precursor with Choline containing Carbon-14 showing the Presence of Labile Phosphorus

SOME years ago, we found that scarcely any preformed acetylcholine exists in the organs of rabbit or frog[1–3]. Working on organs frozen in liquid air or ethyl ether mixed with solid carbonic acid until the solutions to be prepared were free from proteins and then at the lowest possible temperature, we showed that the acetylcholine found with ordinary methods was in reality present in a bound state which we called the acetylcholine precursor. We were able to show its presence in two essentially different ways[1–4]. Nevertheless, some workers were unable to show the precursor, probably due to difficulties in technique. We have been working on a method of demonstrating the precursor which is essentially different from the earlier methods. Now we are able to report the demonstration of acetylcholine precursor, which is biosynthesized from choline containing carbon-14 in its methyl groups.

Earlier, it was shown that young rats on a choline-free diet lose about two-thirds of their precursor contents. It increases swiftly to normal values when choline is given[1–3]. Therefore, young rats weighing 23–25 gm. were placed on a choline-free diet. After 7 days on this diet we injected 2 millicuries of the radioactive choline intraperitoneally. After 2 hr. the animals were killed. The hearts, livers and muscles from their hind limbs were taken out with great rapidity and frozen in a mixture of ethyl ether and solid carbonic acid. The precursor was extracted with 2 vol. of 96 per cent alcohol. The alcohol was evaporated to a final volume of 1 ml. The residue