## Investigations of greenheart alkaloids by paper electrophoresis

Some applications of electrophoresis to the separations of alkaloids have been reviewed by LEDERER[1]. The limited use of this technique in alkaloid chemistry can be partly attributed to lack of suitable apparatus and partly to the lack of suitable current sources. It is the purpose of this paper to describe electrophoresis experiments on alkaloids from the British Guiana greenheart (*Ocotea rodiaei*), using an ordinary glass chromatography tank. The construction of a suitable D.C. supply is also described.

*Apparatus.* The apparatus consisted of a glass tank 30 × 35 × 45 cm fitted with a glass cover having a central hole (Fig. 1). The paper was held in the vertical position

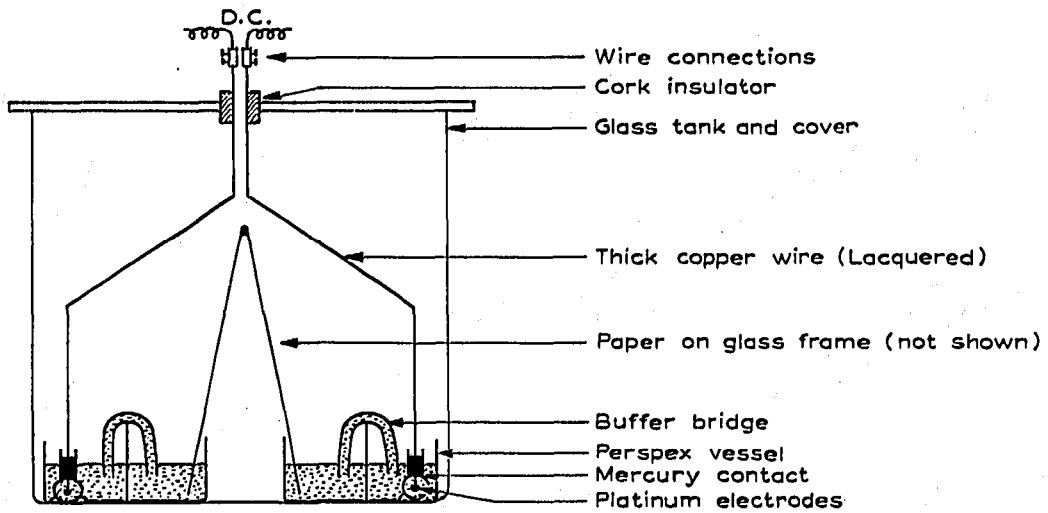


Fig. 1. Electrophoresis tank.

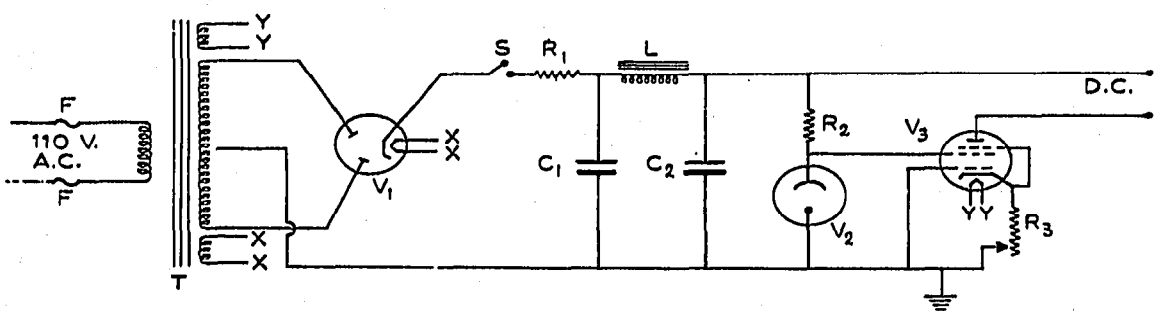


Fig. 2. Constant current D.C. circuit. $C_1$: 16 mfd. electrolytic 500 V; $C_2$: 32 mfd. electrolytic 500 V $R_1$: 100 Ω, 1 W; $R_2$: 133 kΩ, 1 W; $R_3$: 5 kΩ potentiometer 1 W; S: Switch; T: Transformer 250-0-250–60 mA; $V_1$: 5U4G; $V_2$: VR 150; $V_3$: 6B.W.6; L: 10 Henry choke, 300 Ω, 40 mA F: 1 A fuses.

by means of a glass frame. The electrodes consisted of horizontally placed platinum wires sealed into glass tubing with holes. The platinum wires were connected to the external copper leads through mercury contacts. The copper leads were lacquered to prevent corrosion. A simple D.C. source is shown in Fig. 2. This circuit using the stabilising valves $V_2$ and $V_3$ supplies a constant current with slightly varying resistive load of the paper, which may be due to evaporation taking place.

*Electrophoresis run.* Experiments were done in a temperature-controlled room ($T = 23 \pm 2°$). Whatman No. 1 paper sheets 53 × 23 cm were spotted near the apex with approximately 500 $\mu$g of the alkaloid mixture or 50 $\mu$g of single components. The buffer used consisted of a mixture of sodium acetate–acetic acid, pH 4.4, ionic strength 0.133. The paper was first wet with the buffer solution and after a short period for draining, the current was switched on and adjusted to 7 mA (*ca.* 200 V), with the variable resistance $R_3$. After 12–16 h the paper was removed and dried. The alkaloid spots were detected by dipping the paper in a saturated solution of iodine dissolved in petroleum ether. This was found to be a more satisfactory method of application than the iodine vapour detection method used by MUNIER AND MACHEBOEUF[2]. The alkaloids showed up as yellow or brown spots on a faint yellow or white back ground.

*Results.* The results of a typical run are shown in Fig. 3. The components of the

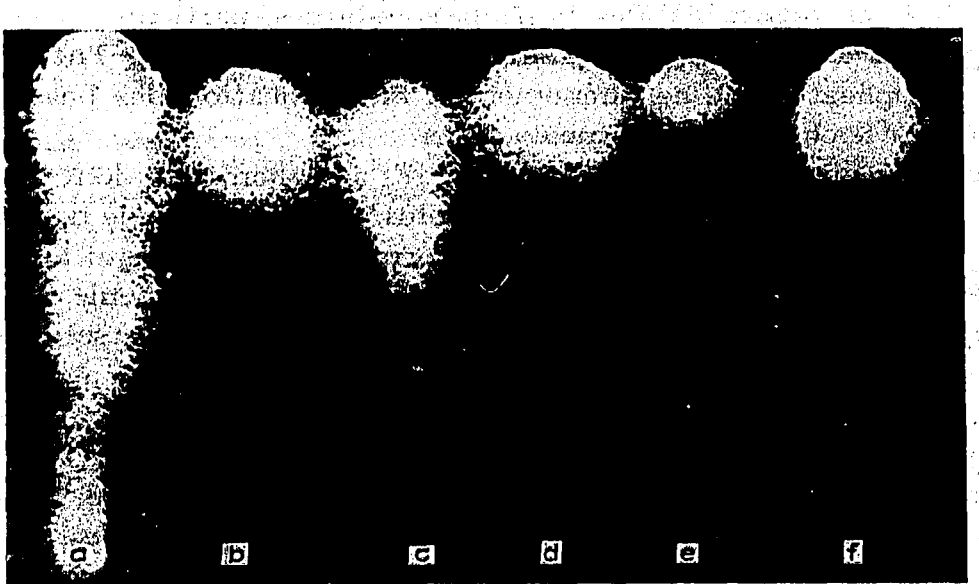


Fig. 3. a = Mixture of Greenheart alkaloids; b = Base III; c = Base III and IV; d = Base II. e = *d*-Tubocurarine chloride; f = *l*-Curine hydrochloride.

mixture were only partially resolved. The similar electrophoretic behaviour of Base II and Base III isolated by counter-current distribution techniques was not surprising. Other chemical evidence indicated them to be very similar bases. Detailed structural investigations will be published elsewhere.

Two very closely related bisbenzyl isoquinoline alkaloids *l*-curine and *d*-tubocurarine chloride which were used as reference compounds also show similar electrophoretic behaviour.

*Chemistry Department, University College of the West Indies, Jamaica (West Indies)* A. W. SANGSTER

[1] M. LEDERER, *Introduction to Paper Electrophoresis and Related Methods*, Elsevier, Amsterdam, 1955, p. 92.
[2] R. MUNIER AND M. MACHEBOEUF, *Compt. rend.*, 230 (1950) 1177.

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