Gel electrophoresis of alkaloids*

Many papers have appeared in the literature describing the electrophoretic separation and identification of alkaloids on paper[1–5]. This paper describes briefly a method for separating alkaloidal mixtures via gel electrophoresis. The use of a solid gel media in electrophoresis has been described for proteins, amino acids, and other substances[6], but not for alkaloids. Although the method has no definite advantage over column chromatography as a preparative method, it may have utility for mixtures that do not lend themselves readily to chromatographic separation.

Experimental

The gel used in this laboratory was prepared via the method described by SMITHIES[7]. Eighteen grams of acetone washed Merck starch[6] were boiled under vacuum in an Erlenmeyer flask with the electrolyte, 1 *N* acetic acid (100 ml). The gel was poured into plastic trays (17 × 7 × 1.5 cm) and when the gel was nearly congealed (usually about 1 h) filter paper wicks were inserted on each end of the tray. The trays were then placed in the electrophoresis unit** and the wicks placed in contact with the electrolyte tanks (Fig. 1).

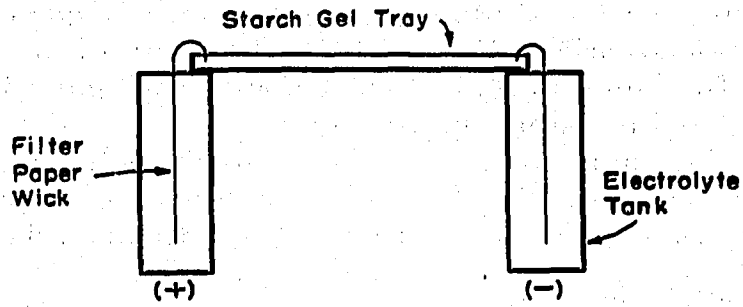

Fig. 1. Starch gel tray with filter paper wicks in contact with the electrolyte tanks.

A small cavity was cut into the congealed starch gel about 1 cm from the end with a razor blade or spatula insuring that it did not reach to either edge nor to the bottom of the starch gel tray (Fig. 2).

The "window" method described for paper chromatography and paper electrophoresis by POPOWICZ[8] works satisfactorily for applying the sample to the prepared gel tray. A piece of 17MM Whatman paper was cut to the size 1 cm × 5 cm and the alka-

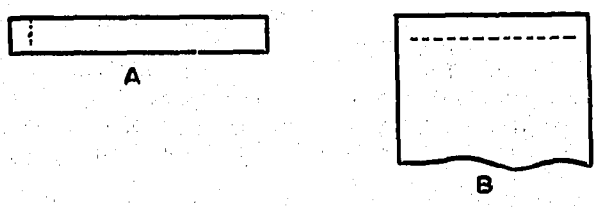

Fig. 2. (A) Side view of starch gel tray showing depth of cavity for insertion of "window" containing the alkaloid sample. (B) Partial top view of starch gel tray showing width of cavity for insertion of filter paper "window".

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** Spectrolator, supplied by Microchemical Specialties Co., Berkley, Calif.

loid mixture was adsorbed onto this strip and allowed to dry. This strip was then placed into the cavity.

To demonstrate the gel electrophoresis procedure two alkaloid mixtures were prepared that would contain alkaloids that fluoresced under ultraviolet light. Mixture No. 1 contained berberine HCl and quinine while mixture No. 2 contained berberine HCl and hydrastine.

Each mixture contained 10 mg of each alkaloid. The alkaloids were dissolved in methanol and adsorbed onto a Whatman strip as described and placed into the prepared starch gel tray.

The electrophoretic run was carried out at 50 mA and 400 V for 2 h. The migration distances for mixture No. 1 were 20 mm for berberine and 70 mm for quinine while mixture No. 2 had migration distances of 20 mm and 40 mm for berberine and hydrastine respectively. These distances were measured from the center of the fluorescent zones. The respective zones were cut out of the trays and placed in beakers. The gel was then mixed with water and solution effected on a steam bath. The final step of the procedure depends on the chemical nature of the alkaloid and this step actually represents a disadvantage over column chromatography and continuous curtain electrophoresis. The gel solutions of quinine and hydrastine were made alkaline with ammonium hydroxide and the alkaloidal bases extracted with chloroform. Berberine can be extracted from an acid solution with chloroform and so extraction of the gel solutions of berberine were effected in this way. The percent recovery of the alkaloids was not determined for this procedure.

For preparing larger quantities a pyrex dish (34 cm × 30 cm × 7 cm) was employed following the same procedure described and using 1000 ml of starch gel. Instead of the "window" method for applying the sample a "trough" is made in the gel by inserting a plate glass in the gel while still mobile and removing the glass plate when the gel is congealed. The sample is dissolved in the electrolyte (1 *N* acetic acid), starch added, and a gel prepared. When the gel is cool, but still mobile it is poured into the "trough" and allowed to congeal before applying the current.

This is a preliminary report and work with crude alkaloidal extracts is in progress.

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