A modified applicator for the uniform coating of thin-layer plates

The application of uniform layers of adsorbent to thin-layer chromatography (TLC) plates is the most difficult and important step in TLC. The need for a uniform layer becomes even more critical when photometric methods[1] are used for quantitative analysis on the chromatoplate. Uniform adsorbent layers are difficult to prepare because of the variation in the thickness of the glass chromatoplates, and because most commercial applicators use a smooth surface template upon which the glass plates are placed. The spreader is passed over the uneven top surfaces of the plates resulting in a non-uniform adsorbent layer.

A commercially available applicator has been modified to enable the preparation of uniform adsorbent layers on TLC plates. Advantage was taken of the inflatable bag arrangement available in an applicator distributed by Colabs* that permits the top surface of the glass plate to be even and perfectly flat during the application of the adsorbent layers. Non-uniformity of adsorbent layers resulting from variation in chromatoplate thickness is eliminated by the leveled top surface. In order to accommodate the 2 × 20 cm plates used for zonal scan analysis[2] and the selection of desired solvent systems, the bed rollers originally supplied with the plate holder were removed and an 8 in. × 45 in. × $^1/_8$ in. aluminum bed plate (A) was used instead as shown in

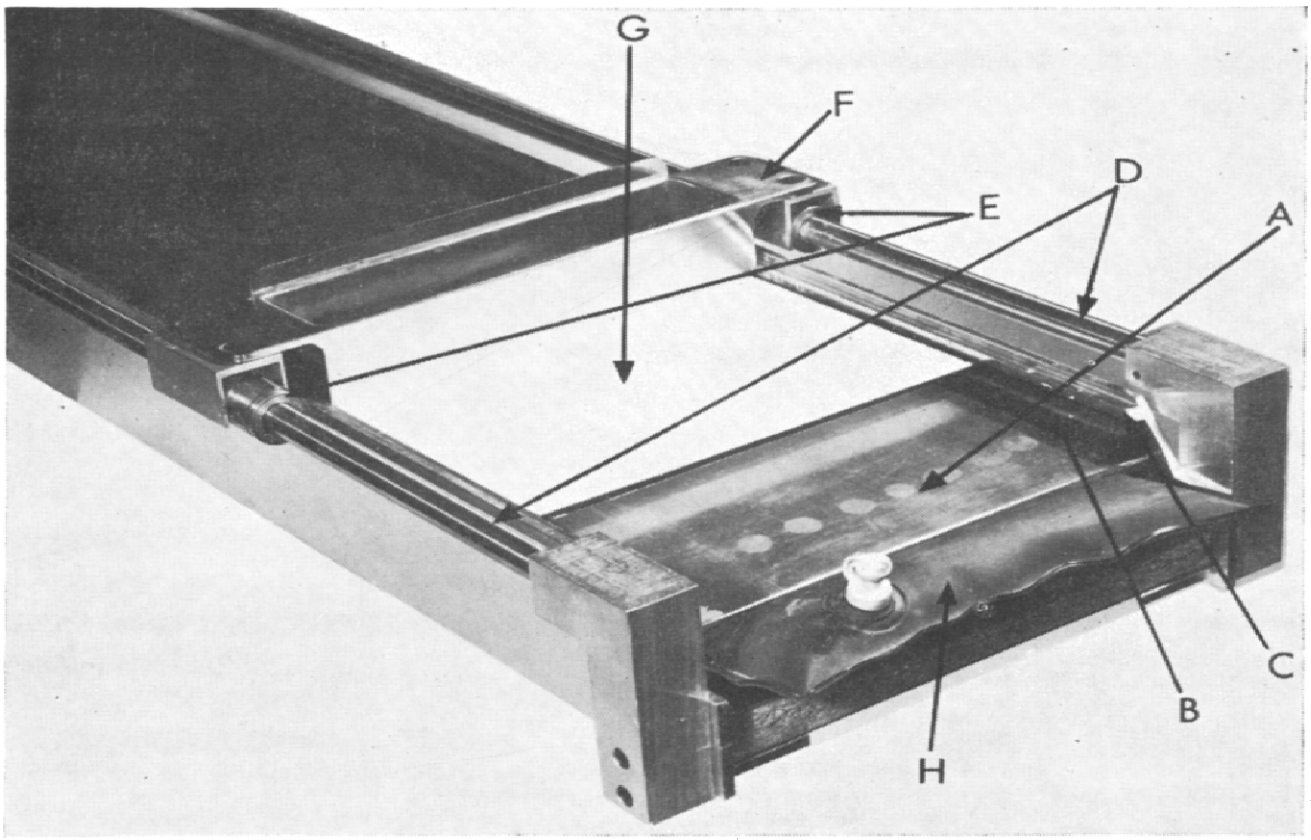

Fig. 1. A modified TLC applicator for the preparation of uniform layers. The labeled parts are: (A) aluminium bed plate; (B) foam rubber strip; (C) securing edge; (D) stainless steel guide bars; (E) ball bushings; (F) spreader; (G) chromatoplate; and (H) the inflatable bag.

* Available from Colabs Laboratories, Inc., Chicago Heights, Ill.

Fig. 1. A $^1/_2$ in. $\times$ $^3/_8$ in. piece of foam rubber (B) affixed to each side of the bed plate holds the chromatoplates (G) firmly against the securing edges (C) when the leveling bag (H) is inflated. The addition of guide bars* (D) equipped with ball bushings (E), also shown in Fig. 1, allows the spreader (F) to be moved evenly and smoothly across the plate, producing a very uniform layer of adsorbent on all plate sizes ranging from 2 $\times$ 20 cm up to 20 $\times$ 20 cm. Micrometer heads** can also be added on the applicator's adjustable plate to allow quick, accurate, and reproducible selection of layer thicknesses.

This applicator is routinely used in this laboratory for the preparation of all TLC plates. Many of the chromatoplates are used for the quantitative analysis of lipids by the densitometric technique of BLANK *et al.*[3], where uniformity of the adsorbent layer improves the quantitativeness of the measurement.

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* One-half-in. stainless steel ball bushings and $^1/_2$-in., 60-core hardened and ground stainless steel rods obtained from Thompson Industries, Inc., Manhasset, N.Y.

** Calibration (0.0–0.500-in. graduations of 0.001 in.) obtained from Brown and Sharp, Precision Park, Northkingston, R.I.

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Thin-layer chromatography of phosphate esters of biologic interest*

The methods available for the separation and quantitation of phosphate esters from tissue extracts are based on either paper or ion-exchange chromatography or electrophoresis. In general, the procedures are either time consuming, not easily adapted to microquantities, or not capable of affording adequate separation. Thin-layer chromatography (TLC) offers the possibility of combining these features. WARING AND ZIPORIN[1] have described a two-dimensional procedure for the separation of a limited number of sugar phosphates with acidic solvent systems, but the ability of the method to separate these compounds from cell extracts was not noted. In our experience the use of acid solvent systems in the first phase produced excessive admixture and uneven solvent fronts, and did not provide adequate resolution. The following two-dimensional procedure has been used satisfactorily for the separation and quantitation of phosphate esters of human red cells. The method has proven

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