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Note

A simple, disposable, streaking sample applicator for thin-layer chromatography

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Thin-layer chromatography (TLC) on a preparative scale constitutes a rapid, easily monitored and versatile means of purification of substances. The technique requires that the sample be applied as a continuous, fairly uniform streak along the starting line of the chromatogram which is then developed in the usual way. This application can be achieved either manually by multiple transfers on adjacent spots of small volumes of solution with a capillary tube or by streaking with a capillary pipette. These procedures are time consuming and require considerable skill in order to avoid scratching the adsorbant layer with loss of material, and, in the case of radioactive substances, contamination of developing tanks etc. In order to avoid these inconveniences, more sophisticated applicators have been devised[1–4]; some of which are commercially available. In most of these applicators the sample is delivered from a microsyringe mounted in a more or less mechanized frame to assure uniform delivery of the solution along the application line. These instruments are costly and present the disadvantage that the microsyringes are extremely difficult to decontaminate after being used with radioactive substances. A number of simpler applicators have been proposed which make use of wedges[5], wicks[6], tiny brushes[7], capillaries[8], or threads[9].

EXPERIMENTAL

The apparatus described here is constructed from inexpensive disposable laboratory supplies. It presents the advantages of simplicity of design, ease of manipulation and quantitative transfer of the samples. As illustrated in Fig. 1, it is made of a length of thin-walled glass capillary tubing (*e.g.* a 100-μl volumetric micropipette) drawn at both ends to a smaller diameter in order to fit at one end into the silicone rubber insert of a Drummond Microcaps® holder and at the other end into a 20-mm length of polyethylene capillary tubing (I.D. about 0.6 mm) drawn to a fine curved tip by heating gently above a microburner flame and then pulling. After filling the pipette by aspiration with the rubber bulb, the assembly, held horizontally, is seated in a cradle carved of styrofoam in such a way that when the base of the rubber bulb rests against the shoulder, the tip overhangs the edge by about 25 mm (Fig. 2).

Fig. 1. Components of the streaking applicator. From left to right: polyethylene capillary tip, modified micropipette, silicon rubber insert, glass tube and rubber bulb of Microcaps® holder.

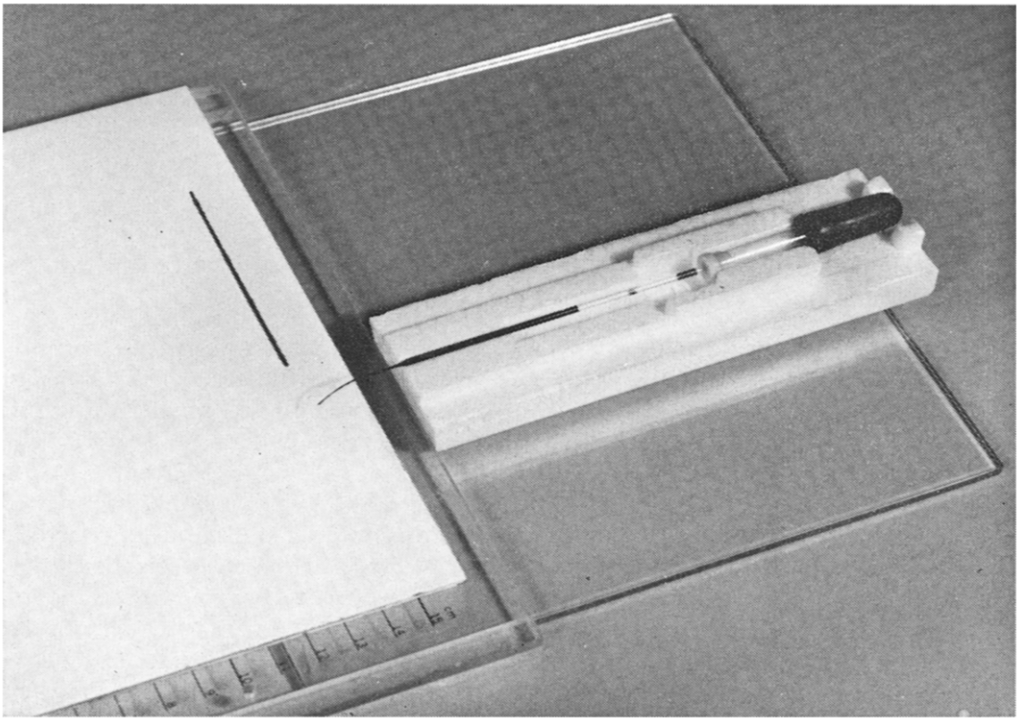

Fig. 2. Assembled applicator seated in styrofoam cradle. Note that the capillary tip does not touch the layer of absorbant and that the base of the rubber bulb fits against the shoulder of the styrofoam cradle.

A fresh TLC plate is positioned on top of a plastic template under which a 20 × 20 cm glass plate is partly inserted. This glass plate and the edge of the template form a smooth guide against which the end of the styrofoam cradle can slide. The tip of the polyethylene tip is trimmed with scissors in such a way that when the cradle rests on the glass plate, the tip does not touch the TLC plate: it should be about 2 mm above. By tilting the cradle, the tip is brought in contact with the absorbent layer and the sample solution flows (Fig. 3). Uniform application along the starting line is made by sliding the cradle along the template edge. Bringing the cradle back into a horizontal position lifts the tip and stops the flow of the solution. Large volumes of solution can be applied in multiple overlapping streaks along the same line when the refilled pipette is re-positioned against the cradle shoulder. Refilling the pipette with pure solvent allows for quantitative application of the sample.

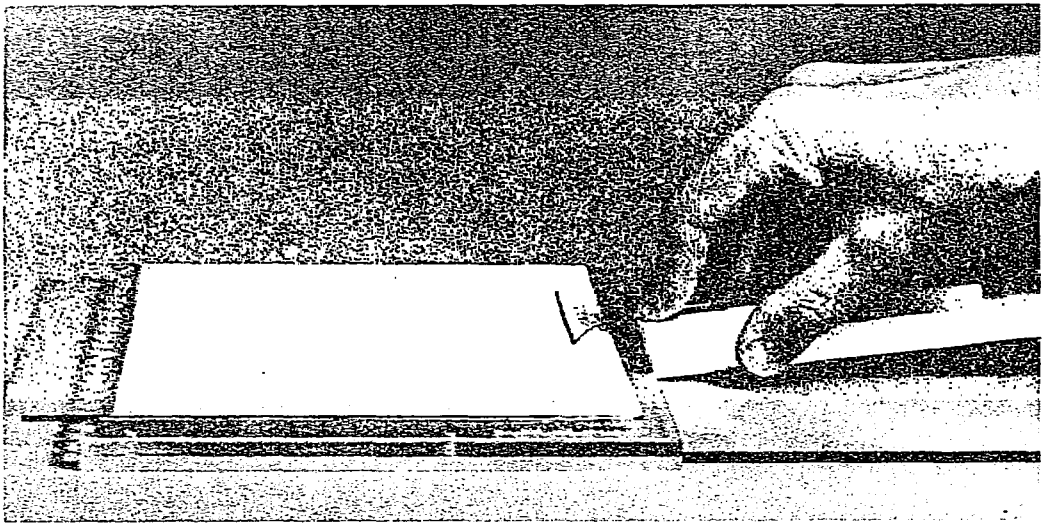

Fig. 3. The assembly, tilted in order to bring the polyethylene tip in contact with the absorbant layer, is slid along the template edge.

RESULTS

This simple instrument is very easily maneuvered to deliver the sample solution uniformly. The polyethylene tip is soft and does not scratch the absorbant layer even after multiple applications. Tips made of PTFE were found to be too hard. No contamination originating from the polyethylene was found when plates onto which chloroform or chloroform–methanol mixtures had been applied, were sprayed with sulfuric acid and heated at 130°. Aqueous solutions do not start to flow easily when the tip touches the absorbant layer. Addition of 10–20% alcohol or acetone to these solutions overcomes this difficulty.

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