

Thin Layer Chromatography Apparatus

A simple and inexpensive apparatus for thin layer chromatography (TLC) has been developed which can be assembled from commonly available laboratory glassware and chemicals, and which obviates the applicators and supporting templates usually used. The technique should be of value in the student laboratory as well as being satisfactory for routine use. It consists of glass plates cut into two convenient sizes: 20×20 cm and 5×20 cm (which are grooved in order to contain the adsorbent layer), a glass developing chamber² large enough to accommodate the larger plates, and a 1-liter graduated cylinder cut off at the 600-ml mark which serves as a developing chamber for the smaller plates. A desiccator is unnecessary if the adsorbent layer is to be of cellulose or its derivatives, but for layers which require storage in a dry atmosphere (e.g., activated silica gel or alumina), a large desiccator can be constructed from a 12- \times 12-in. Pyrex cylindrical jar with cover and fitted with a No. 5 porcelain desiccator plate and small Petri dishes for the desiccant.

Preparation of the Grooved Plate

The grooved plates are formed by masking the areas which will later contain the adsorbent with $\frac{1}{2}$ -in. wide plastic electrical tape spaced about $\frac{1}{8}$ in. apart and then coating the entire plate, front and back, with Glyptal varnish.³ After the resin coat has dried, the tape is stripped off and the plate is immersed in an etching bath containing 114 g of ammonium bifluoride per liter of water. A polyethylene dishpan serves as an inert tray, and the solution is stored in a polyethylene container which can be kept tightly closed. After approximately $7\frac{1}{2}$ hr at room temperature, during which the solution

is gently agitated by a polyethylene-coated mechanical stirrer, the plate is removed, rinsed with hot water and then with acetone to remove the Glyptal, and finally cleaned in dichromate cleaning solution followed by thorough rinsing with distilled water. The depth of the grooves (which is quite uniform) is about 0.3 mm, depending on the type of glass used, and can be varied according to the duration of immersion; this should be determined on small trial pieces of the glass to be used.

Preparation of Adsorbent Layer

Slurries of the various adsorbents are prepared according to usual TLC techniques.⁴ A cellulose layer for a 20- \times 20-cm plate (which can contain about 12 channels) is prepared by vigorously stirring 3.5 g of cellulose powder⁵ with 18 ml of acetone in a 50-ml Erlenmeyer flask. A magnetic bar works very well for this purpose. The slurry is poured evenly across one end of the grooved plate, then spread with a straight edge (glass plate) across the remainder of the plate in the direction of the grooves. After drying, the ridges between the grooves are scraped free of adsorbent and the plate is ready for use. If only two or three compounds are to be chromatographed, a 5- \times 20-cm plate is prepared using 1 g of cellulose and 5 ml of acetone.

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² Developing tanks large enough for the 20 \times 20-cm plates are commercially available from Brinkmann Instruments, Inc., 115 Cutter Mill Road, Great Neck, N. Y., and No. 6944 from Corning Glass Works, Corning, N. Y.

³ General Electric Glyptal varnish, No. 1202.

⁴ RANDEATH, K., *Biochem. Biophys. Res. Comm.*, **6**, 452 (1962); RANDEATH, K., AND STRUCK, H., *J. Chromatog.*, **6**, 365 (1961); WOLLISH, E. G., SCHMALL, M., AND HAWRYLYSHYN, M., *Anal. Chem.*, **9**, 1139 (1961); HOFMANN, A. F., *Anal. Biochem.*, **3**, 145 (1962).

⁵ Adsorbents for TLC, average particle size $<10 \mu$, are available from Macherey, Nagel and Co., Düren, Germany; distributed by Brinkmann Instruments, Inc.