THE CALIBRATION OF PIPETTES FOR PAPER PARTITION CHROMATOGRAPHY

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In the field of quantitative paper partition chromatography the use of micro pipettes is essential. One of the requirements for such a pipette is that it deliver small aliquots of solution ranging from 0.0001 ml to 0.01 ml. When volumes larger than 0.01 ml are transferred to the filter paper strips in one operation, the liquid tends to spread unduly and forms a larger spot than desirable. There are several types of commercial pipettes which will deliver small aliquots of liquid, but their cost is high. This paper will describe a satisfactory and inexpensive micro pipette which may be prepared from an ordinary medicine dropper.

The glass tip of a medicine dropper was heated in the flame of a micro burner until soft and then drawn out to a capillary. The bulk of the capillary was discarded by breaking the filament about one inch from the shoulder of the dropper. The tip of the capillary was then ground with carborundum powder and, finally, on a carborundum stone (No. 000) until a smooth shoulder was obtained. The progress of the grinding operation was easily followed by occasionally checking the condition of the capillary tip under the low power field of a microscope. The tip of the capillary must be smooth, since delivery of the pipette is measured in drops.

Each pipette thus prepared was calibrated by determining the number of drops of alcohol required to fill a 1 ml volumetric flask. The pipettes selected for use delivered from 100 to 200 drops per ml.

A check on the uniformity of delivery of each pipette was made by determining the optical transmission of solutions prepared from one or more drops of stock material appropriately diluted. The optical transmission of solutions containing 1, 2, 3, and 4 drops of quercetin (3, 5, 7, 3', 4'-pentahydroxy flavone) stock solution was determined in the Beckman model DU spectrophotometer. The measurements were made at 375 m_{μ}, corresponding to one of the absorption maxima of quercetin.

Small deviations in delivery were noted in 1-drop samples. These deviations were much less noticeable in the 3 and 4-drop samples. Consequently, in using this type of pipette, it is advisable to adjust solutions to a concentration requiring the measurement of at least 3 or 4 drops per sample.

It is important that the pipette be held in a vertical position when drops are being measured. The glass must be clean and the drop rate uniform. After filling the pipette, a few drops should be discarded immediately prior to measurement. The pipette must be recalibrated for each solvent with which it is to be used since an appreciable change in surface tension will alter the volume delivered per drop.

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