
THE USE OF MULTIPLE STRIPS IN ONE-DIMENSIONAL PAPER CHROMATOGRAPHY

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In the separation of the flavonoid constituents of complex plant extracts by paper partition chromatography, one often finds that there are too many colored substances in the extract to achieve complete separation on one strip of filter paper unless extremely dilute solutions are used. With such dilute solutions, the individual zones are often so small alone as to render identification difficult. This handicap has been recently overcome in this laboratory by use of multiple strips of filter paper.

EXPERIMENTAL. An extract of a lemon peel infusion was paper chromatographed, using the method of Wender and Gage (1). The butanol layer of the 40% butanol — 50% water — 10% acetic acid equilibrated system was used as the flowing solvent. Since most of the flavonoid and related pigments of the lemon peel extract are fluorescent in ultraviolet light, the strip was examined under a "black light" after development. Fourteen fluorescent bands were observed on the strip. Fifteen other similar strips had been treated simultaneously in a like manner.

Each of the fourteen bands was cut from each of the sixteen strips. Band No. 1 from all the strips was shredded into the same container and extracted in a small Soxhlet apparatus with ethanol. Bands No. 2 through No. 14 were individually treated in the same way. Thus, each extract from the combined sixteen strips contained a sufficient concentration (of one pigment) for further purification.

The ethanol extract of each band was then chromatographed again using a flowing solvent different from butanol. Ethyl acetate, saturated with water, was usually used.

As a result of this treatment, each of the bands was separated from traces of its original neighboring bands due to different rates of movement of the various substances in the different solvents, and also sufficient quantities were obtained for identification studies. For example, the twice chromatographed bands were suitable for use in the determination of the absorption spectrum.

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LITERATURE CITED

1. WENDER, S. H. and GAGE, T. B. 1949. Paper chromatography of flavonoid pigments. *Science* 109: 287-289.
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