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Note

Instant copy technique for documentation of thin-layer chromatograms

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Documentation of TLC using for example the Polaroid system is not quite satisfactory, especially when weak spots are to be visualized or when a natural-size chromatogram is required. An alternative, sensitive and cheap instant copy technique, which does not suffer from the shortcomings of the Polaroid system or other photographic methods, is reported here.

The developed thin-layer plate (Fig. 1) is transferred into a dark room. An ordinary photographic paper (plastic type; Ilfospeed 2.1M, glossy 2; Ilford, Essex, Great Britain) is placed on a table with the emulsion side upwards. Then, the chromatogram plate is carefully placed on the photographic paper with the layer downwards, and exposed for ca. 1 sec with a 25 W opaque bulb from a distance of 150 cm. The photographic paper is developed in a standard developer for ca. 1.5 min at room temperature, treated with a common acid fixative for ca. 3 min and washed in running water for 1 min. The excess of moisture is removed with a filter-paper and the sheet is dried in a horizontal position.

The method allows tracing of weak spots otherwise impossible to detect visually. The best results were achieved when the exposure was adjusted to give a medium-grey tint of the background. Using black and white papers, all spots, irrespective of colour, are reproduced in shades of white-grey. The colour sensitivity of the photographic paper has its peak between 400 and 500 Å (the blue region of the spectrum). Consequently all coloured spots outside this area will have ideal detection conditions. Of course, even blue spots will be reproduced but with a decreased detection level.

As the plastic papers are practically non-shrinking the natural-size copy can be superimposed on the actual TLC plate. Thus, holding a lamp behind the combined copy-plate, the exact zones can be easily scraped off. This makes the

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Fig. 1. Instant contact copy (\times 1/2) of a thin-layer chromatogram of a reference PGE₂ sample (10-200 ng/mm line) visualized in iodine after developing according to Srivastava¹, except for the TLC plates used (Merck silica gel 60 F₂₅₄, layer thickness 0.25 mm). The right hand band (10 ng sample applied per mm) is invisible to the naked eye without the copy (representing the visibility limit for the actual TLC plate).

procedure convenient for preparative work, especially if the zones have faded (*i.e.*, by vaporization of iodine).

Moreover, the modern plastic type glossy papers possess an extremely evenly distributed emulsion layer. Therefore densitometric scanning may be performed directly on these instant copies.

In conclusion, the technique described represents a valuable aid for a rational documentation of thin-layer chromatograms because of its sensitivity, reproducibility and simplicity.

REFERENCE

1 K. C. Srivasatava and V. K. S. Shukla, Z. Anal. Chem., 293 (1978) 45-48.