CHROM. 3611

Recording of thin layer chromatograms

In the course of an investigation of differences in amino acids and phenols in diseased and healthy cotton plants, we found it necessary to record the results of large numbers of thin layer chromatograms (TLC). This note describes a method that we have found convenient for analyzing the data of several TLC plates over a period of time.

Various methods have been described for recording the results of thin layer chromatograms. These include utilization of tracing paper[1,2] spraying with plastic emulsions[2-4] and production of photocopies[5-7]. Currently, the preferred method for preserving the data is Polaroid[8,10,11] or standard[3,9] color photography. These have been used primarily with photography in visible light; however, color[8,9] and black and white film[10,11] have also been used for photography in ultraviolet light (U.V.).

The normal procedure when spotting a plate is either to code each spot with a number, and refer to a separate sheet for identification, or attempt to write the identification of the spot directly on the layer. The method described below represents another alternative.

Thin Layer Chromatogram

TLC No._____ Technician_____

Literature: Date _____

Solv. Syst. _____ Adsorption Layer_____

Solv. Dist. _____ cm. Time out:_____
Time in:_____

Quant. Appl._____

Detection:

Remarks: Yes____ No____ (over) Type Recording

Date _____ Photo_____

TLC No._____ Neatan_____

Other_____

	Substance	Notes
1		
2		
14		

Fig. 1. Legend sheet (large) for use adjacent to 8 × 8 in. plate. Portion to left of dashed line is included in photograph of plate.

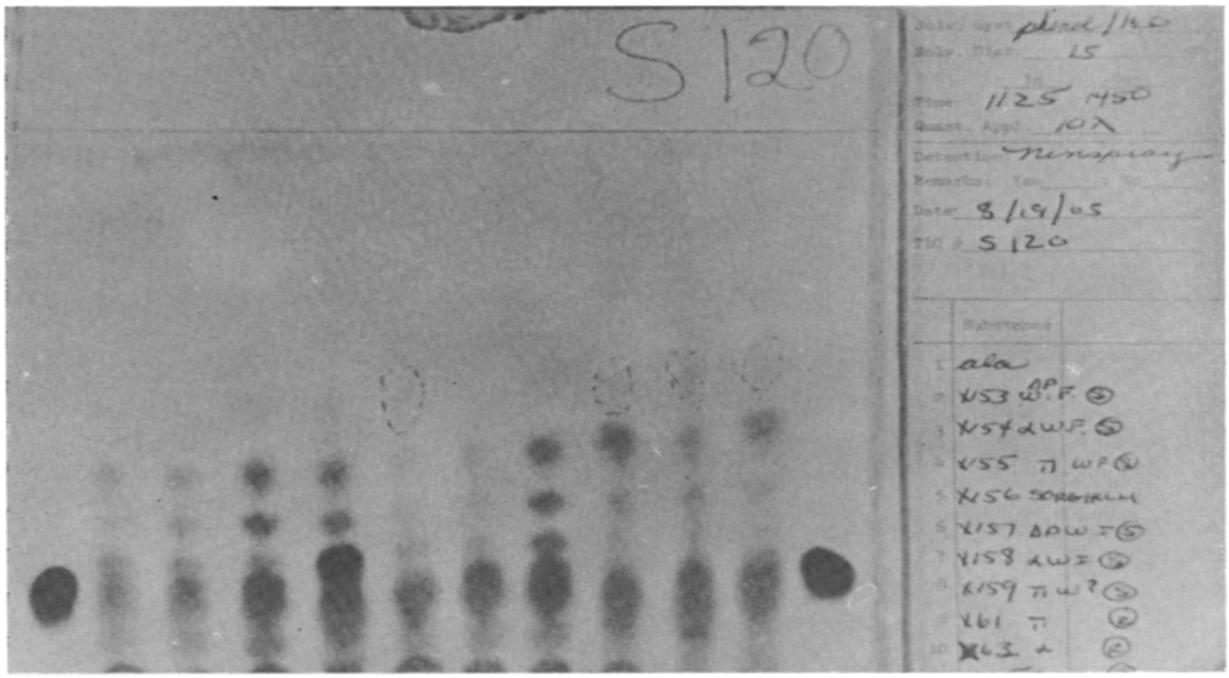

Fig. 2. Photograph using sheet shown in Fig. 1. Amino acids from cotton. Detection: Ninhydrin Photography: Visible light as described in text.

Method

All TLC plates are photographed with an accompanying legend, using either visible light or U.V. light. Prints are enlarged to 3.5 × 5 in. ("3-R" size), which are convenient for filing after analysis. Two general methods can be used for this documentation:

*Use of 8.5 × 11 in. legend sheets** (*Fig. 1*). Data are placed on the sheet prior to chromatography, with any additional data or remarks added at detection. The sheet is then placed adjacent to the TLC plate and photographed (Fig. 2). After photography, the sheet is filed in a loose leaf folder for cross referencing to the subsequently filed photographs. The layer can be preserved further with "Neatan"[3] if desired, as an additional documentation step.

Use of 2.75 × 8 in. legend sheet (*Fig. 3*). Data are placed on the sheet prior to chromatography and the sheet is placed on the upper half of the TLC plate for photography (when the solvent front is 10 cm this fits immediately above the solvent front). After photography, the layer may be preserved with "Neatan" and the legend sheet taped back on the layer in the same position as when photographed.

Photography of plates

Visible light: Kodacolor X film, f 5.6 for 1/50 sec, two 15 Watt fluorescent bulbs placed 18 in. above the plate. U.V. light: High Speed Ektachrome film, f 8 for 12 sec, two 15 Watt U.V. bulbs (General Electric F15T8 BLB, 3200–4000 Å) placed 18 in. above the plate. The film is developed commercially.

Discussion

With both of these methods, the only item to be written on the plate is the TLC

* This sheet is an adaptation of that originally suggested by STAHL[2].

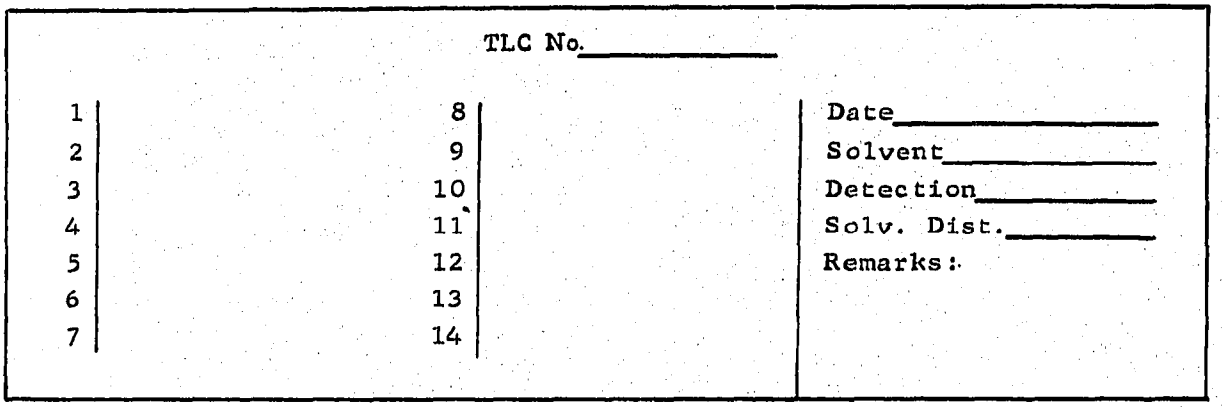

Fig. 3. Legend sheet (small) for use on 8 × 8 in. plate.

number (for reference to the appropriate legend sheet during processing). Further, either sheet can be used with U.V. light by first photographing the TLC plate in U.V. light followed by a covering of the plate with an opaque material and rephotographing the legend in visible light (the camera must permit a double exposure).

This photographic technique has facilitated the comparison of data derived over time. If necessary, the photographs can be mounted on punched cards (*e.g.* Unisort[12]) for further indexing.

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