I. I. Ziderman,[1] *B.Sc., M.Sc., Ph.D.*

A Simple Technique for Comparing Paper Samples by Their Performance as a Chromatographic Sorbent (Inverse Paper Chromatography)

REFERENCE: Ziderman, I. I., **"A Simple Technique for Comparing Paper Samples by Their Performance as a Chromatographic Sorbent (Inverse Paper Chromatography),"** *Journal of Forensic Sciences,* JFSCA, Vol. 26, No. 2, April 1981, pp. 387-392.

ABSTRACT: Evidence in the form of paper documents can be highly valuable in crimes of fraud. Available methods of examination do not provide absolute certainty that two samples have the same origin. A new technique for testing paper, known as inverse paper chromatography, is based on using the paper as a chromatographic sorbent medium on which a known mixture of dyestuffs is separated under standard conditions. The resulting chromatogram will uniquely designate the paper grade and composition. This principle has been validated in a standard test with more than 30 types of paper. The experimental technique is simple, inexpensive, and largely nondestructive. Only a small specimen is required, and it provides a permanent exhibit for evidence. While printing on the paper or wetting it does not alter its chromatographic behavior, artificial aging by heat or exposure to sunlight does. The individualization of paper specimens for unambiguous identification requires further research.

KEYWORDS: criminalistics, questioned documents, papers, chromatographic analysis, inverse paper chromatography

Evidence in the form of paper can be highly valuable in crimes of fraud. Currency, certificates, wills, or identification documents may be shown to be counterfeit. Furthermore, an exhaustive analysis of the paper may lead to the conclusion that the alleged date of preparation of a document is not possible. While documents are the more frequently encountered and generally more important, other types of paper evidence, such as pieces of cardboard at a bomb scene or blotting paper at a clandestine drug operation, are also available. Bearing in mind these considerations, Lyter and Brunelle [*1*] described a systematic approach for the comparison of paper evidence comprising five sequential stages of examination:

(1) measurement of physical characteristics: color, dimensions, weight, opacity, and fluorescence;

(2) watermark examination, including embossings, aided with ultraviolet illumination;

(3) microscopic fiber analysis after staining;

(4) chemical analysis of ingredients such as sizing and loading materials, fillers, whiteners, plasticizers, and waxes; and

Received for publication 18 Sept. 1980; accepted for publication 9 Oct. 1980.

[1] Visiting scientist, Research & Development Division and Analytical Laboratory of Criminal Identification Branch, Israel Police Headquarters, Jerusalem; permanent position, senior research scientist, Israel Fiber Institute, Jerusalem.

(5) trace elemental analysis by neutron activation analysis and X-ray fluorescence, scanning electron microscopy, or emission spectrography.

While these procedures provide numerous points of comparison that may reveal conclusive evidence that paper samples are different, Lyter and Brunelle [*1*] conclude that it is still not possible to determine with absolute certainty whether or not the specimens have the same origin. It would be useful to develop additional methods for paper examination. Furthermore, simpler and inexpensive techniques are required since few laboratories are capable of maintaining the whole array of experts and equipment needed for implementing the entire systematic approach on a current basis. A research program has been initiated in Jerusalem to evaluate novel approaches to paper identification; the program includes such techniques as inverse paper chromatography and gel permeation chromatography.

In paper chromatography [*2*], a sheet of paper is used as a stationary medium in which mixtures are separated into their component compounds by virtue of their selective interactions with the stationary phase and the solvent (mobile phase) moving along the paper by capillarity. This principle has been used to identify unknown compounds by their migration on a specific filter paper having fixed and reproducible properties. It is now proposed to invert the objective: to identify an unknown specimen of paper by the separation achieved when a known mixture of compounds is chromatographed on it (inverse paper chromatography). The migratory behavior of both the test compounds and the solvent should be influenced by all components of the paper—cellulose, fillers, and sizings. The effects of aging and of wetting the paper on its chromatographic performance are also reported in this communication. The simplest methods and equipment were specially chosen to provide a facile and inexpensive technique that may be implemented without the resources of a fully equipped chemical laboratory and that requires only a small specimen of paper.

Methods

The direction in which the paper moved through the paper-making machine (DM), and thus the direction in which the wood fibers are oriented, was found by cutting a strip from each of two perpendicular edges of the sheet and then holding them together horizontally (Fig. 1). The stiffer strip is designated DM. The direction perpendicular to DM is denoted by CM (cross-machine).

Preparation of the Paper

A strip of paper (19 by 1.5 cm) was folded 1.5 cm from one end and stapled across 1 cm from the fold, thus forming a slot in which a glass rod (1.7 by 0.3 cm) was inserted so as to keep the paper extended on suspension later. The solution of dyes was applied to the paper 3.5 cm from the fold as a spot (2 to 3 mm wide) by means of a tapered capillary glass tube that had been prepared by heating the middle of the tube in a gas flame until it was soft, gently pulling the two ends apart by about 2 cm, and then breaking the tube in half. The spotted paper strip was dried with a hair drier. It was folded 1 cm from the other end, and then the flap obtained was fixed with a thumbtack to a cork wrapped with aluminum foil so that the new fold was in the center of the cork face (Fig. 2).

Materials

Only colored dyestuffs were used as markers in the chromatography experiments so as to obtain immediate visualization of the chromatogram, avoiding the need to spray the paper. The first three compounds in Table 1, which were colored yellow as used, are acidic

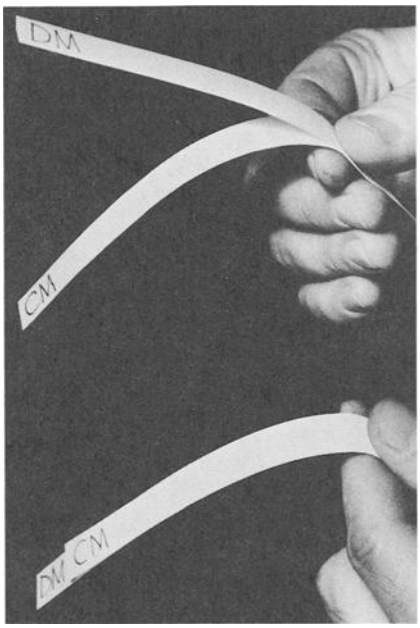

FIG. 1—*Test to reveal machine direction* (DM) *in paper sheet. Strips cut from each of two perpendicular directions are held together horizontally. The stiffer strip is designated* DM *and the other,* CM.

materials commonly used as pH indicators, and chemically they are all γ-sultones of the sulfonephthalein series. The other compounds in Table 1 are all basic dyes with various chemical structures. Two ionic types of dyestuffs were chosen to study the ion-exchange effects resulting from the inorganic fillers in the papers and from the presence of carboxylic acid functional groups in the cellulosic fibers. Solutions in methanol (0.5%) were used to apply the dyes to the papers. Unless otherwise stated, the experiments were performed with a 60-g wood-free writing paper.

A fixed chromatography time (3 h) was adopted to reveal differences in paper capillarity as an additional identifying characteristic and to simplify the experimental procedure.

Chromatographic Technique

A graduated cylinder (50 mL), charged with *n*-butanol (about 10 mL), was closed with a suitably sized cork wrapped in foil, to which the paper strip had been previously attached as described above. The spot of dyes was initially above the solvent surface. Care was taken to insert the paper strip so that it did not touch the inner wall of the cylinder (Fig. 3). The loaded cylinder stood in a draft-free place while the solvent ascended the paper strip, forming the chromatogram. After exactly 3 h, the strip was removed from the cylinder. The height attained by the solvent was immediately marked with a pencil and the chromatogram was suspended to dry near an open window or, preferably, in a ventilated hood.

An additional dimension of discrimination may often be achieved by exposing the chromatogram to acid vapor (such as formic acid) or alkaline vapor (such as ammonia) in which the pH indicators used as markers may change color.

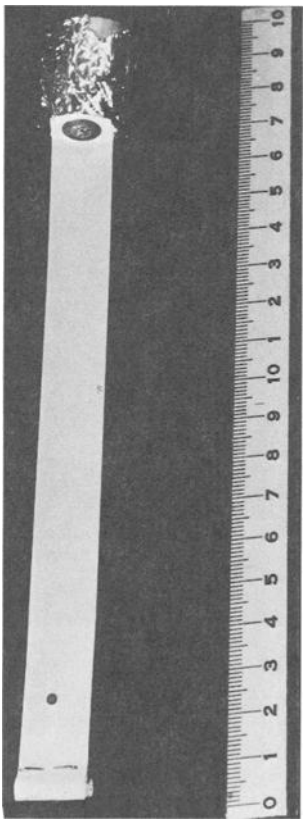

FIG. 2—*Preparation of the paper for chromatography.*

Results

The eight colored compounds were chromatographed separately and together on a 60-g wood-free writing paper in the DM and CM directions. The result was different in each case: CM gave less tailing but the solvent front advanced less. Various mixtures of the stains were assayed. The combination of cresol red, fuchsine, methylene blue, and malachite green seemed to give the best chromatogram (R_f = 0.40, 0.76, 0.11, and 0.63, respectively, in the DM direction).

Fourteen different commercial papers were tested in both the CM and DM directions for their behavior as chromatographic media. The paper samples were clearly distinguishable from each other in either direction with respect to both the distance advanced by the solvent front and the chromatographic separation of the dyestuff mixture. The contrast between papers taken from print-out rolls supplied with two different calculating machines was especially dramatic: front mobility and methylene blue R_f were, respectively, 3 cm and 0.83 for one and 12.7 cm and 0.08 for the other.

Fifteen new grades of paper in the catalogue of Israel's main paper mill were assayed. The results again demonstrated that the chromatographic performance of each grade is unique. Papers of the same grade (wood-free, coated both sides, polished) but differing slightly in weight (90 and 118 g) gave similar chromatograms. A difference in color among papers of

TABLE 1—*Dyestuffs used for chromatography.*

Name	*Colour Index*[a] Number	Source of Sample	R_f	
			DM	CM
Cresol red	(pH indicator)	Merck & Co.	0.30	0.43
Chlorophenol red	(pH indicator)	Merck & Co.	0.28	0.42
Bromothymol blue	(pH indicator)	Riedel-de Haen	0.67	0.75
Fuchsine (rosaniline)	CI 42510	Riedel-de Haen	0.48	0.68
Methylene blue	CI 52015	Merck & Co.	0.12	0.16
Malachite green	CI 42000	BDH	0.21	0.44
Rhodamine B	CI 45170	Merck & Co.	0.34	0.61
Gentian violet	CI 42555	Allied Chemical	0.51	0.71

[a] Compiled and published jointly by the Society of Dyers and Colourists of Great Britain and the American Association of Textile Chemists and Colorists.

the same grade (70-g wood-free offset) evidently affects their chromatographic behavior. The same grade of paper from different sources may not be distinguishable by this method.

Applications

Once it was established that behavior as a chromatographic sorbent may serve as a criterion for comparing different grades of paper, several forensic science aspects of the technique were studied. Printing a counterfeit bank note does not alter the chromatogram obtained from the plain paper. Artificial aging of paper in a hot oven for 30 min at 200°C increases the mobility of both the solvent and the fast-moving fuchsine but retards the slow-moving methylene blue. Paper that has been wetted to remove soluble impressions retains its chromatographic characteristics.

Exposure to an atmosphere of high humidity overnight or to indirect sunlight (through a pane of glass) for 56 h does not affect the paper's chromatographic behavior. However, 14 h of direct sunlight causes a significant increase in mobility of the yellow markers, cresol red and chlorophenol red, from $R_f = 0.59$ to $R_f = 0.78$.

Discussion

The inverse paper chromatography method for characterizing papers is simple, inexpensive, and largely nondestructive. Only a small specimen is required and provides a permanent exhibit for presentation as evidence. This technique may also be applicable to other fields of investigation involving paper, such as quality control. When necessary, the original sample may be regenerated by a suitable elution of the markers from the chromatogram.

To promote the use of this new method, the development of a complete kit would be helpful and would encourage routine use of paper testing outside the confines of well-equipped chemistry laboratories. Furthermore, the chromatographic data may be quantified in terms of front mobility and R_f values to permit computer-aided comparison of an unknown specimen with a reference collection of chromatograms.

Even when this new method for comparing paper samples is used with other methods, a unique individualization of paper, the only scientific basis for determining with absolute certainty that two specimens have the same origin, is still not possible. We intend to pursue this investigation of paper identification and to attempt to determine the polymeric molecular weight distribution of paper's major constituent, cellulose, in a manner that will provide a unique assay of paper quality.

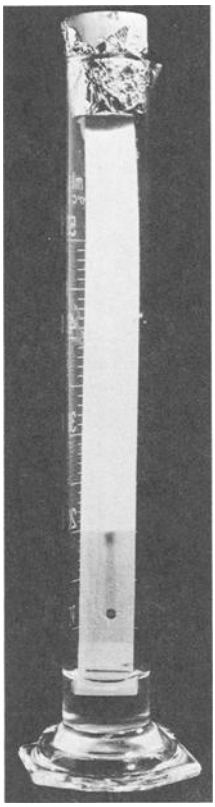

FIG. 3—*Chromatographic technique.*

Acknowledgments

Dr. J. Almog, deputy head of the Research and Development Division, and Dr. S. Kraus, director of the Analytical Laboratory, Criminal Identification Branch, and their staffs are sincerely thanked for their kind hospitality and cooperation in providing the facilities for carrying out this project at the Israel Police Headquarters. Capt. A. Betsaleli of the Documents Laboratory is also thanked for helpful discussions of the forensic science aspects. The excellent work of the Photography Laboratory is greatly appreciated.

References

[*1*] Lyter, A. H. and Brunelle, R. L., *Identification News*, Vol. 27, No. 7, July 1977, pp. 3-6.
[*2*] Sherma, J., in *Handbook of Chromatography*, Vol. 2, Z. Gunter and J. Sherma, Eds., Chemical Rubber Co. Press, Cleveland, 1972, pp. 73-87.

Address requests for reprints or additional information to
I. Ziderman, Ph.D.
Israel Fiber Institute
P.O. Box 8001
91 080 Jerusalem, Israel