Gas Chromatographic Determination of Ethanol in Pharmaceuticals. Comparison of the Flame Ionization and Thermal Conductivity Detectors

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The use of a flame ionization detector has been investigated as a means of reducing the time necessary for the analysis of ethanol. Results obtained are in complete agreement with those found using a thermal conductivity detector.

GAS CHROMATOGRAPHY has been reported as an analytical tool for the determination of ethanol in wine (1), cosmetics (2), mixed solvents (3), and pharmaceuticals (4, 5). In a collaborative study sponsored by the Pharmaceutical Manufacturers Association (6), gas chromatography was found comparable in accuracy and precision to the official U.S.P. and N.F. distillation method for the determination of ethanol. Subsequently, the gas chromatographic procedure was adopted in the N.F. XII (7).

All of these methods specified a thermal conductivity detector which necessitated the elution of water from the column before injection of the next sample. Even with efficient columns which reduced or eliminated the tailing of water, most chromatograms required at least 12 min. It is interesting to note that in N.F. XII First Supplement (8) no detector is specified.

The use of a flame ionization detector would considerably reduce the analytical time, but the presence of water diminishes the flame detector's over-all sensitivity (9). The need to keep the flame ionization detector at constant sensitivity is the basis for obtaining results that would compare favorably with those using a thermal conductivity detector. By designing a column which gives efficient separation of acetone (internal standard) and ethanol while allowing the water to bleed off, it was found possible to use the flame ionization detector with constant sensitivity. This water bleeding phenomenon was probably responsible for the excellent results obtained by Parker *et al.* (10), in the determination of ethanol in blood using the flame ionization detector.

EXPERIMENTAL

Flame Ionization Detector

Apparatus.—Perkin-Elmer model 801 flame ionization gas chromatograph.

Column Packing.—A 6 ft., 1/4-in. o.d., copper column packed with 20% polyethylene glycol 4000[1] on Gas Pack RP 60/80 mesh (Applied Science Laboratory, State College, Pa.).

Operating Conditions.—Column temperature, 115°; injector temperature, 150°; detector temperature, 150°; He flow rate, 40 ml./min. Figure 1 illustrates a typical chromatogram.

Received June 9, 1966, from the Pharmaceutical Research and Development Laboratories, Warner-Lambert Research Institute, Morris Plains, N. J. 07950.
Accepted for publication August 24, 1966.

[1] Marketed as Carbowax by the Union Carbide Corp., New York, N. Y.

Thermal Conductivity Detector

Apparatus.—Perkin-Elmer model 154C gas chromatograph equipped with thermistor detector.

Column Packing.—A 6-ft., 1/4-in. o.d., copper column packed with 20% polyethylene glycol 4000 on 60/80 mesh Gas Pack F (a perfluorocarbon impregnated diatomaceous earth, Chemical Research Services, Inc., Addison, Ill.).

Operating Conditions.—Column temperature, 110°; injector temperature, 150°; He flow rate, 80 ml./min. Figure 2 illustrates a typical chromatogram.

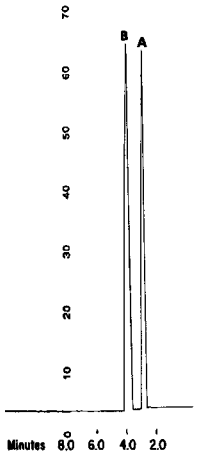

Fig. 1.—A typical chromatogram. (Flame ionization detector.) Key: A, acetone; B, ethanol.

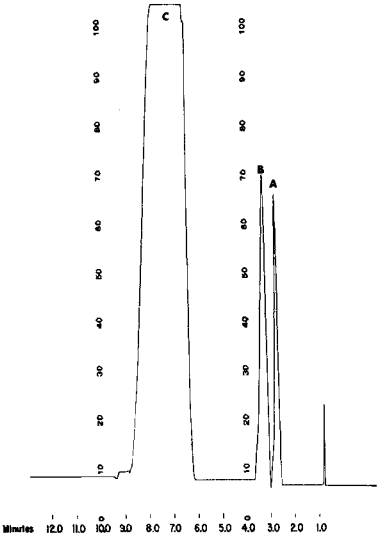

Fig. 2.—A typical chromatogram. (Thermal conductivity detector.) Key: A, acetone; B, ethanol; C, water.

Quantitation was performed, following the procedure outlined in the collaborative study of the Pharmaceutical Manufacturers Association (6).

RESULTS AND DISCUSSION

In order to demonstrate that the flame ionization detector technique was comparable to thermal conductivity detector technique, six repetitive analyses were done by each method on an experimental mouthwash product. The results obtained are summarized in Table I.

It is obvious from Table I that both methods give comparable results.

Previously during a 5-hr. period, 20 ethanol analyses could be done. Now, using a flame ionization detector, 60 analyses can be run in a 5-hr. period.

Table I.—Per Cent Ethanol Content in Experimental Mouthwash

Sample	Flame Ionization	Thermal Conductivity
1	13.0	13.1
2	13.2	13.2
3	13.1	13.1
4	13.0	13.0
5	12.9	13.0
6	13.0	13.0

REFERENCES

(1) Bouthelet, R. J., Caputi, A., Jr., and Ueda, M., *J. Assoc. Offic. Agr. Chemists*, **44**, 410(1961).
(2) *Ibid.*, **49**, 242(1966).
(3) Casazza, W. T., and Stellenkamp, R. J., *J. Gas Chromatog.*, **3**, 253(1965).
(4) Kawai, S., *Eisei Shikensho Hokoku*, **77**, 2733(1959).
(5) Wesselman, H. J., *J. Am. Pharm. Assoc., Sci. Ed.*, **49**, 320(1960).
(6) "Pharmaceutical Manufacturers Association Yearbook," Pharmaceutical Manufacturers Association, Washington, D.C., 1964–1965, vol. 6, p. 571.
(7) "National Formulary," 12th ed., Mack Publishing Co., Easton, Pa., 1965, p. 432.
(8) *Ibid.*, 1st Suppl., p. 20.
(9) Hill, D. W., and Newell, H. A., *Nature*, **206**, 708 (1965).
(10) Parker, K. D., Fontan, C. R., Yee, J. L., and Kirk, P. L., *Anal. Chem.*, **34**, 1234(1965).

Anticonvulsants III. Alkyl Esters of 4-Bromo-2-sulfamoylbenzoic Acid and 4-Chloro-2-sulfamoylbenzoic Acid

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In addition to steric factors, electronic effects may also be important in the anticonvulsant activity of alkyl *o*-sulfamoylbenzoates and related compounds. To further explore the relationship between electronic properties and antielectroshock activity, three esters of 4-bromo-2-sulfamoylbenzoic acid and four esters of 4-chloro-2-sulfamoylbenzoic acid were prepared by the alcoholysis reaction of passing hydrogen chloride into a refluxing solution of 6-bromo- or 6-chlorosaccharin in the appropriate alcohol. The following alkyl 4-bromo- and 4-chloro-2-sulfamoylbenzoates were thus obtained: methyl; ethyl; *i*-propyl; and *sec*-butyl. In these compounds the bromine or chlorine atom is in the 4-position *para* to the alkoxycarbonyl group; thus, they do not possess the steric interactions between the large halogen atom and the ester moiety, which are believed to be necessary in the anticonvulsant activity of the related, potent *ortho*-substituted alkyl 6-chloro-2-sulfamoylbenzoates. Preliminary pharmacological results indicate that isopropyl 4-bromo-2-sulfamoylbenzoate lacks antielectroshock effects in mice.

Recent work has shown that alkyl esters of 2-sulfamoylbenzoic acid, 4-amino-2-sulfamoylbenzoic acid, and 6-chloro-2-sulfamoylbenzoic acid (I) possess marked anticonvulsant activity as indicated by their prevention of the effect of strychnine or maximal electroshock in mice (1–5).

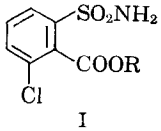

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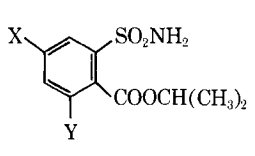
Table I.—Anticonvulsant Activities of *ortho*- and *para*-Substituted Isopropyl Sulfamoylbenzoates[a]



Compd.	X	Y	Antielectroshock ED_{50}, Mice, mg./Kg.
1	NH_2	H	13
2	H	H	39
3	NO_2	H	240
4	Cl	H	Less active than compd. *2* above[b]
5	H	Cl	100[c]

[a] The pharmacological testing was performed by Smith Kline & French Laboratories, Philadelphia, Pa. [b] *Reference 1.* [c] Pharmacological testing was performed by Riker Laboratories, Northridge, Calif.

Received April 25, 1966, from the School of Pharmacy, University of Southern California, Los Angeles 90007.
Accepted for publication September 9, 1966.
This investigation was supported in part by research grant NB 03328 from the National Institute of Neurological Diseases and Blindness, U. S. Public Health Service, Bethesda, Md.
Presented to the Medicinal Chemistry Section, A.Ph.A. Academy of Pharmaceutical Sciences, Dallas meeting, April 1966.
Previous paper: Hamor, G. H., and Farraj, N., *J. Pharm. Sci.*, **54**, 1265(1965).