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Determination of heroin and some common adulterants by capillary gas chromatography

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ABSTRACT

A method has been developed for the simultaneous determination of heroin along with some of the commonly occurring adulterants that have been found in samples in recent years. The method utilizes simple sample dissolution along with an internal standard followed by capillary gas chromatography on a non-polar DB-1 column attached to a flame ionization detector. Column temperature programming was used along with a programmed temperature vaporizer in the injection port. Linearity studies were conducted on heroin base, heroin hydrochloride and several of the other drug adulterants. Excellent results were obtained and the method is applicable to a broad range and type of heroin sample.

INTRODUCTION

Forensic drug analysis always seems to present the analyst with new and unusual types of samples with which existing methods have had difficulty coping. Recently a new type of heroin sample has been appearing in our laboratory in which the sample is cut with a multitude of diluents and adulterants thereby rendering a once relatively simple analysis much more complex. In addition to the normally occurring components such as acetylcodeine, O^6- and O^3-monoacetylmorphines, codeine, morphine, papaverine and noscapine, a large number of other components have been occurring with some regularity. These include such drugs, in varying combinations, as caffeine, nicotinamide, phenobarbital, procaine, methaqualone, phenacetin, cocaine, acetylprocaine, lidocaine, diazepam, antipyrine, chloroquine, acetaminophen, quinine, N-phenyl-2-naphthylamine, phenolphthalein and occasional antihistamines along with sugars and inorganic salts such as calcium carbonate and sodium chloride.

While high-performance liquid chromatography (HPLC) methods [1–15] exist for the determination of heroin, they do not address the problem of quantitation of a large number of adulterants. Similarly HPLC screening techniques [16–22] do not address quantitation, although some authors [18,19] do list a comparison of UV response factors *vs.* electrochemical response factors. The same situation occurs in gas chromatography. Packed columns do not readily lend themselves to the analysis of these complex multicomponent mixtures. On the other hand, capillary columns are

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ideally suited to provide a simultaneous separation and quantitation of these multicomponent samples. There are capillary gas chromatography (GC) methods for the determination of heroin [23–27] as well as methods giving qualitative retention time data for a large number of drugs [28–37]. In addition, there is an extensive listing of retention time indices in the second edition of Clarke [38]. Again however, none of them deal with the quantitative aspects of samples that contain a large number of components.

Capillary GC in combination with a programmable temperature vaporizer (PTV) operated in the split mode was the technique used. The PTV allows a sample to be injected at low, or cold, temperatures which is then followed by a rapid rise in temperature to ensure immediate vaporization of all components. This eliminates mass discrimination effects as well as greatly reducing changes due to thermal breakdown of heat labile substances. Increased accuracy, precision and high resolution result from using this type of injection system. It can also allow pre-column separation of the solvent when used in the solvent purge mode. In standard injection systems, the syringe needle is exposed to high temperatures before the transfer of the sample into the injector which can result in selective volatilization as well as possible degradation and or adsorption. Poy and co-workers [39,40] showed excellent relative standard deviations with a PTV as compared to poorer results when using a standard hot split injection system. Schomburg and co-workers [41,42] showed excellent standard deviations of absolute and relative peak areas indicating high precision and accuracy. Loyola *et al.* [43] showed that the PTV gave the best accuracy and precision in his study and indicated that it provided better performance than the classical split injector technique.

This paper gives quantitative data on a selected number of drugs, in particular, those drugs which seem to occur most frequently in samples submitted to this laboratory. Excellent results were obtained.

EXPERIMENTAL

Instrumentation

A Perkin–Elmer Sigma 2000 gas chromatograph, equipped with a hydrogen flame ionization detector and interfaced with a Perkin–Elmer 7500 data system was used. The capillary column used was a fused silica, cross-linked and bonded DB-1 30 m × 0.25 mm I.D. with a 0.25 micron film thickness (J&W Scientific, Folsom, CA, U.S.A.). The carrier gas was hydrogen (zero grade) with an average gas velocity of 41 cm/s. The oven temperature program used started with an initial temperature of 200°C with a 1-min hold, then ramped at 12°C/min to 280°C followed by a final hold of 8 min. A PTV from Perkin–Elmer was used with an initial temperature of 75°C, held for 0.1 min then immediately ramped to 285°C. The detector temperature was held at 285°C. A split ratio of 25:1 was used for all injections.

Materials

Chloroform and methanol (Mallinckrodt) were reagent grade. All drugs were of pharmaceutical grade or better, with the exception of N-phenyl-2-naphthylamine. Caffeine was obtained from K&K Labs., procaine and tetracosane from Pfaltz and Bauer, nicotinamide from Eastman-Kodak, phenacetin from Dow Chemical, metha-

qualone from William B. Rorer, phenobarbital, cocaine hydrochoride and cocaine base from Merck & Co., heroin hydrochloride from Research Triangle, quinine hydrochloride from Mallinckrodt and heroin base was synthesized in our laboratory and is the DEA "house" standard. N-Phenyl-2-naphthylamine was obtained from Aldrich and was used as received.

Internal standard solutions

The internal standard solution for the linearity study was prepared by making a 0.1 mg/ml solution of tetracosane in chloroform. The internal standard solution for the quantitative analysis of the samples was prepared by making a 1.0 mg/ml solution of tetracosane in chloroform.

Preparation of drug stock solutions for linearity study

Three stock solutions were prepared for each drug, the second and third of which were dilutions of the first. The primary stock solutions of the drug standards were prepared by weighing 100 mg of each drug into separate 100 ml volumetric flasks, dissolving in a minimum amount of methanol and bringing to volume with chloroform. This solution was then diluted (1:10) into a second volumetric flask to produce the second stock solution and this in turn was further diluted (1:10) into a third volumetric flask to provide the third stock solution. Aliquots were then transferred from each of the three stock solutions for each drug, into separate glass stoppered tubes and evaporated to dryness. Aliquots of 1, 3, 5, 8 and 15 ml were transferred from the primary stock solution and 1-, 3-, 5- and 8-ml aliquots were transferred from the second and third stock solutions. The thirteen tubes for each drug were reconstituted with 1.0 ml of internal standard solution (0.1 mg/ml tetracosane in chloroform). Equal volumes of all solution were injected on the GC.

Quantitative analysis of the samples

A standard solution was prepared by weighing an amount of the appropriate standards into a 25-ml flask, dissolving in a minimum amount of methanol, adding 2.0 ml of internal standard solution (1 mg/ml tetracosane in chloroform) and diluting with chloroform. Generally 4–5 mg of standard is a suitable amount. In the case of nicotinamide approximately 10 mg is required, and for phenobarbital 8–10 mg is required. Quinine, which usually occurs as the hydrochloride, requires about 15–20 mg.

Preparation of the sample

The weight of sample to be used for the analysis depends on the concentrations of the particular components which are present. This may quickly be determined by performing a preliminary qualitative GC screening analysis on the material and estimating the concentrations based on their relative peak areas. An appropriate amount of sample, at least 100 mg, was weighed into a volumetric flask, mixed with a small amount of methanol and then brought to volume with chloroform. An aliquot was transferred to a 25-ml flask followed by the addition of 2.0 ml of internal standard solution (1.0 mg/ml) and diluted to a suitable volume. The final solution may be filtered if necessary.

RESULTS AND DISCUSSION

Good separation and quantitative results were achieved. Fig. 1 shows a chromatogram of a complex sample which was submitted to the laboratory. Linearity studies of the standards were conducted over a wide range of concentrations and correlation coefficients were calculated. These values along with the linearity ranges determined are listed in Table I.

Relative standard deviations were calculated from a sample that was injected repetitively eight times and the values are listed in Table II. Excellent results were obtained with values that ranged from 0.7 to 3.4%. This largest value was represented by nicotinamide indicating slightly less reproducibility for this component. Low levels of nicotinamide and quinine exhibited very poor reproducibility so care should be taken not to attempt an analysis below the stated limits. Results for heroin, caffeine and procaine compare favorably to quantitative results obtained by HPLC. These results are listed in Table III. Other components could not be quantitated by HPLC because of interferences with the peaks of interest.

It should be noted that while cocaine and N-phenyl-2-naphthylamine have virtually the same GC retention time, they have not been found to occur in combination with each other in any sample that has been analyzed to date. Noscapine and phenolphthalein are also a pair of compounds that co-elute under the existing temperature programming conditions. Although lowering the starting temperature as

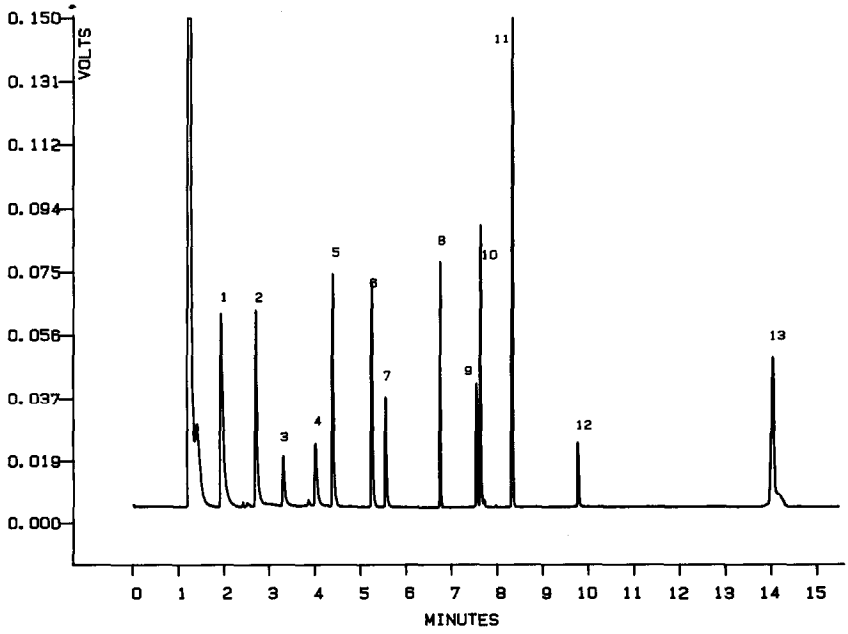

Fig. 1. Chromatogram of a complex heroin sample. Peaks: 1 = nicotinamide; 2 = phenacetin; 3 = caffeine; 4 = phenobarbital; 5 = procaine; 6 = methaqualone; 7 = N-phenyl-2-naphthylamine; 8 = tetracosane; 9 = acetylcodeine; 10 = O^6-monoacetylmorphine; 11 = heroin; 12 = papaverine; 13 = noscapine.

TABLE I

RESULTS FOR LINEARITY STUDY

Drug	Linearity range (μg on column)	Correlation coeff.
Heroin base	0.0008–0.6	0.9998
Heroin HCl	0.0008–0.6	0.9998
Cocaine base	0.0004–0.6	0.9998
Cocaine HCl	0.0008–0.3	0.9999
Caffeine	0.0004–0.6	0.9999
Nicotinamide	0.02–0.6	0.9999
N-Phenyl-2-naphthylamine	0.0012–0.2	0.9999
Methaqualone	0.0004–0.6	0.9999
Phenobarbital	0.002–0.6	0.9999
Procaine HCl	0.02–0.6	0.9993
Phenacetin	0.002–0.6	0.9994
Quinine HCl	0.04–0.6	0.9987

TABLE II

RESULTS OF ANALYSIS (8 REPETITIVE INJECTIONS)

R.S.D. = Relative standard deviation.

Drug	(%)	R.S.D. (%)
Nicotinamide	19.3	3.4
Phenacetin	11.7	1.8
Caffeine	4.5	1.0
Phenobarbital	5.0	2.2
Procaine	12.1	0.8
Methaqualone	7.0	0.7
N-Phenyl-2-naphthylamine	3.0	0.9
Heroin	16.2	0.8

TABLE III

COMPARISON OF GC AND HPLC RESULTS OF HEROIN, CAFFEINE AND PROCAINE

	Sample 1		Sample 2		Sample 3		Sample 4		Sample 5	
	GC	HPLC	GC	HPLC	GC	HPLC	GC	HPLC	GC	HPLC
Heroin	35.1	36.4	24.9	25.6	3.1	3.7	19.5	18.9	18.4	18.4
Caffeine	4.3	4.6	9.8	10.2			29.8	29.6	16.8	17.2
Procaine	9.8	9.5							2.6	2.7

well as the oven program rate will allow for some separation, for quantitative work it is necessary to treat the sample with a derivatizing agent. The preparation of a trimethylsilyl (TMS) derivative of phenolphthalein resolves the two peaks and allows for quantitation [27,44]. Similarly, in the quantitative analysis of O^6-monoacetylmorphine and acetylcodeine, the preparation of a TMS derivative of O^6-monoacetylmorphine is required for a satisfactory quatitation of both components [27,44].

The rationale for the presence of a noxious chemical like N-phenyl-2-naphthylamine in a heroin sample cannot be explained. The Aldrich catalog [45] describes the chemical as an irritant. One can speculate that it probably has some, if not a considerable degree of toxicity as well.

CONCLUSIONS

The nature of samples currently being examined are too complex for analysis by packed column gas chromatography, however capillary GC has been shown to be suitable for both qualitative and quantitative analysis of these samples. Excellent results were obtained with small relative standard deviations and a wide range of linearity for the compounds studied. Other drugs will be studied in future work.

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