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TECHNICAL NOTE

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Component Analysis of Illicit Heroin Samples with GC/MS and Its Application in Source Identification

ABSTRACT: A novel method based on GC/MS and GC for component analyses of seized illicit heroin was established by using SKF_{525A} as an internal standard. The main components in illicit heroin products such as heroin, O^3-acetylmorphine, monoacetylcodeine, and O^6-acetylmorphine were determined quantitatively and the organic adulterants such as paracetamol, acetaminophen caffeine and theophylline were detected qualitatively using the developed method. With these obtained data, 500 seized illicit heroin samples were divided into nine groups. The decomposition pattern of heroin was studied. The dependencies of both the decomposition pattern and the content ratios of monoacetylcodeine to heroin and monoacetylcodeine to O^6-acetylmorphine on the source of the seized illicit heroin were observed. This information was used to develop a novel method for its source identification. The examination results were in agreement with the practical cases, thus providing significant information for detection of criminal cases involving illicit heroin.

KEYWORDS: forensic science, heroin, GC/MS, GC, component analysis, source of heroin, decomposition of heroin

The identification of the origin of the seized illicit heroin is effective information for gripping the import way of illicit heroin and fighting the illicit trafficking of this dangerous drug (1). The components of heroin products are closely related to the planting environment of the raw materials and their processing, thus the identification of their geographical origin based on the component analysis has been extensively studied (2–9). O'Neil et al. reported the physical and chemical features of illicitly imported heroin products to distinguish their origin (2–3). Desage et al. used gas chromatography with mass spectrometry (GC/MS) and isotope-ratio MS to study the geographical origin of heroin products (6). The potentials of site-specific isotopic ratio deuterium NMR (7) and multi-element stable isotope analysis (8–10) for regional origin assignment have been studied. This work suggested the component analysis was an effective tool for the identification of the source of seized heroin samples.

The components of heroin products included heroin, monoacetylcodeine, O^3-acetylmorphine, O^6-acetylmorphine and other substances. These substances found in illicit heroin are classified in four groups named as diluents, adulterants, impurities and contaminants (11). These substances and their relative contents may change in heroin samples during the processing and trafficking (1,12). Thus, a wide range of identification on the substances is needed for identification purposes. The component analyses of heroin products can be performed with a series of analytical techniques such as ICP-MS (13), GC/MS (6,14), NMR (7), stable isotope analysis (9) and HPLC (2,3). The levels of some contaminants and thinners such as metals in illicit heroin have been determined with atomic absorption spectrometry (AAS) (14,15), electrothermal AAS and flame atomic emission spectrometry (FAES) (16–18). They have provided the useful information to determine the source and the trafficking routes of the illicit heroin (19–21). The acetylazation by-products of morphine in heroin samples are produced during its processing. The different production routes can form different monoacetylmorphine by-products. Thus these by-products can also be used to distinguish the source of heroin products (22,23). The main method to quantitatively determine the contents of these by-products is GC/MS by using different internal standards (6,13). However, the data based on this technique are often not conclusive enough. The chromatographic profiles obtained are also often dependent on time and instruments (23). This work selects SKF_{525A} as the internal standard of GC analysis to determine the contents of heroin, the acetylazation by-products and some adulterants in 500 seized illicit heroin samples with different sources and to study the natural decomposition patterns of these heroin seizures in one year. A comparative analysis of the contents of the by-products in these samples is described. The dependencies of both the relative contents of heroin to the by-products and the decomposition pattern are observed. These dependencies have been developed to identify the source of these heroin samples with a satisfactory result.

Materials and Methods

Reagents

The pure heroin, O^3-acetylmorphine, O^6-acetylmorphine, monoacetylcodeine were provided by the National Drugs Laboratory (China). SKF_{525A} (2-Diethylaminoethyl-2,2-diphenylvalerate) was obtained from the Forensic Evidence Identification Center, Ministry of Public Security of China. Five hundred samples of illicit heroin seized in China were obtained from different detection institutions.

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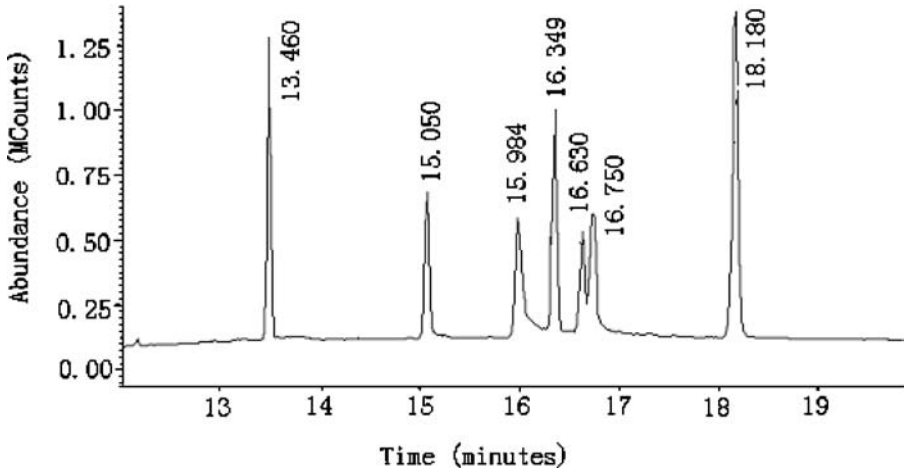


FIG. 1—*GC/MS total ion chromatogram of a typical heroin sample. The peaks occurred at 13.46, 15.05, 15.98, 16.35, 16.63, 16.75 and 18.18 min are attributed to internal standard, codeine, morphine, monoacetylcodeine, O^3-acetylmorphine, O^6-acetylmorphine and heroin, respectively.*

Other reagents were of analytical grade or higher. The sample solutions were prepared by dissolving 5–10 mg heroin products in 8.0 mL anhydrous ethanol followed by adding 2.0 mL 1.0 mg/mL SKF_{525A} as the internal standard. The ethanol solutions for studying the natural decomposition of heroin, monoacetylcodeine and O^6-acetylmorphine contained 0.2 mg/mL internal standard, respectively. These solutions were maintained for different periods of 1, 2, 3, 6, 9 and 12 months.

Apparatus

An HP 5890 II model gas chromatographic analyzer with a FID Detector, an autosampler and a BP-1 quartz capillary column (25 m × 0.22 mm id × 0.25 μm) was used for quantitative analysis of the components in seized heroin samples. The injection port temperature was kept at 280°C. The column temperature increased from 160 to 280°C at 10°C/min and was held at 280°C for 6 min. The detector temperature was 280°C. Nitrogen was used as the carrier gas. The GC column pressure was held constant at 13.0 psig.

The GC/MS experiments were carried out on a Varian Saturn 3 Model GC/MS analyzer with a BPX-5 capillary column (30 m × 0.22 mm id × 0.25 μm). Helium was used as the carrier gas at a flow rate of 0.6 mL/min. The GC injection port temperature was maintained at 280°C, column temperature program was, isothermal at 160°C for 1 min, increased at 10°C/min to 280°C, and held at 280°C for 23 min. The split ratio was 10:1. The MS was operated in an EI model with 70 eV EI voltage and the emission current was 10 μA. The ionic trap temperature was 200°C. The MS scanning rate was 1 scan/sec with a scan range of 40–450 amu.

Results and Discussion

Total Ion Current Scan of Heroin Samples

GC/MS is a well-known technique commonly used for component analysis of seized heroin samples. It can detect the presence of active components and organic adulterants in heroin seizures, even if they are not completely resolved on a GC column. The total ion current scan of a heroin sample containing the internal standard, SKF_{525A}, and six components showed the retention times of 13.46, 15.05, 15.98, 16.35, 16.63, 16.75 and 18.18 min (Fig. 1). These peaks were attributed to SKF_{525A}, codeine, morphine, monoacetylcodeine, O^3-acetylmorphine, O^6-acetylmorphine and heroin, respectively. Adapted GC/MS parameters resolved the main components in a typical heroin sample well. They were used in this study for component analysis of seized heroin samples, with SKF_{525A} as the internal standard. Figure 2 shows the mass spectra of the four main components in the heroin sample and the internal standard.

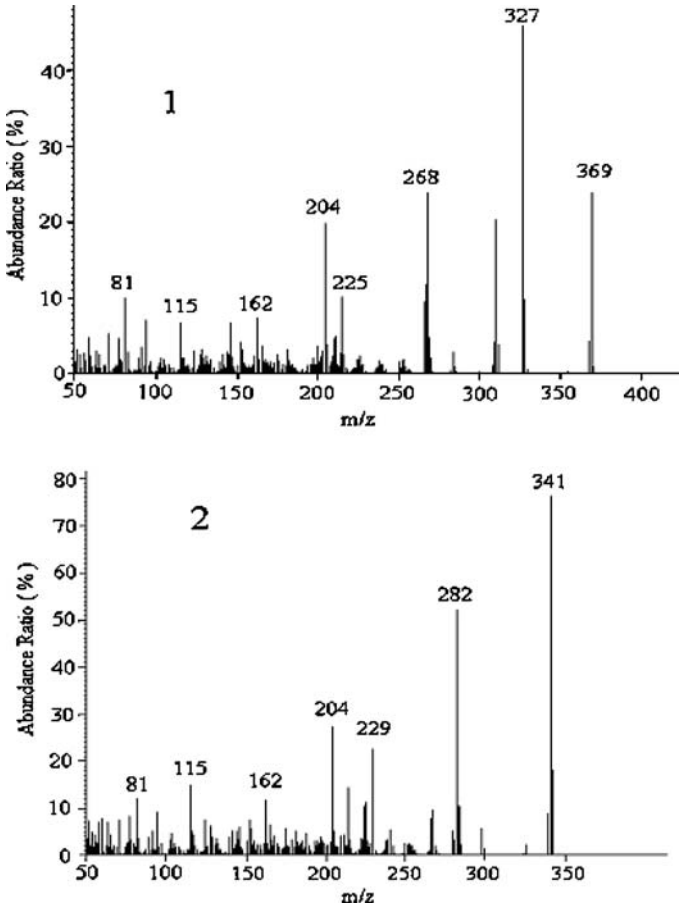
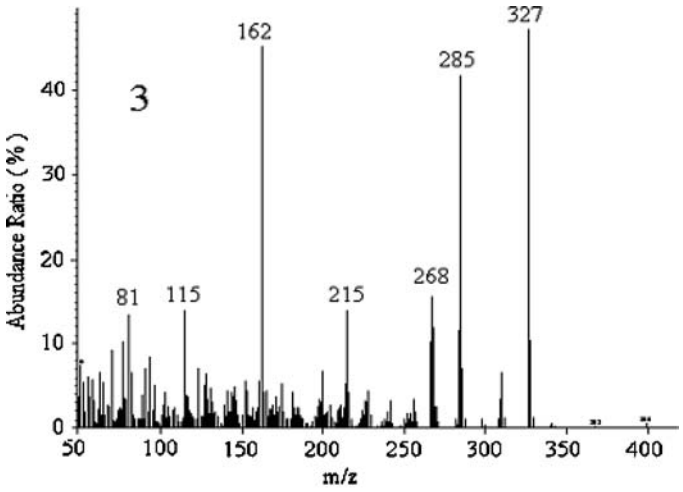


FIG. 2—*Mass spectra of (1) heroin, (2) monoacetylcodeine, (3) O^3-acetylmorphine, (4) O^6-acetylmorphine and (5) SKF_{525A}.*



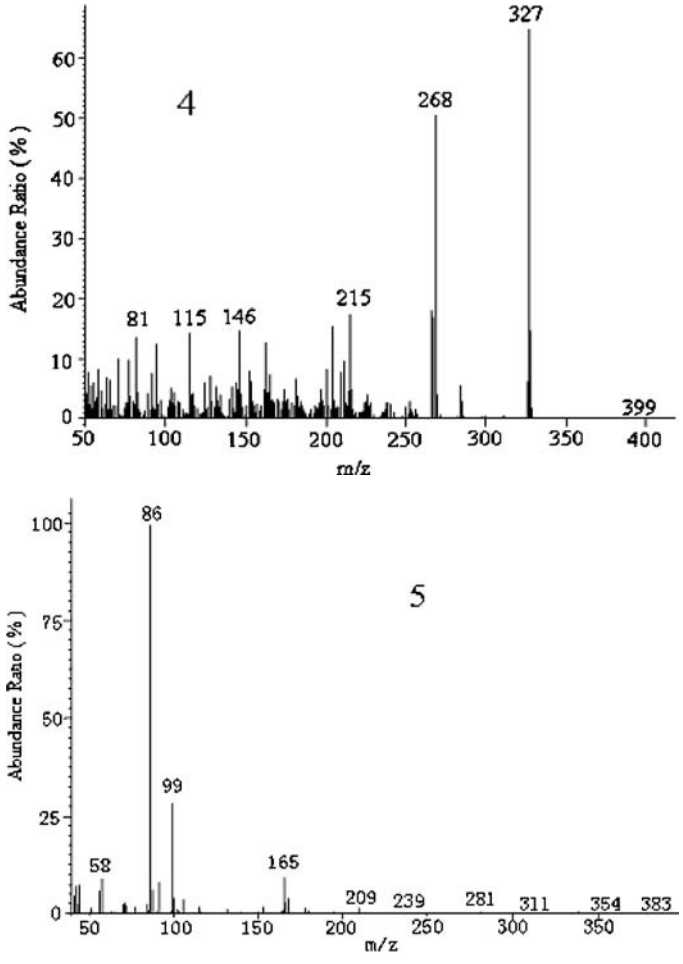


FIG. 2—*Continued*

Gas Chromatogram Analysis of Components of Heroin Samples

An important factor for the component analysis of heroin samples by GC or GC/MS is the separation between heroin and other main components with similar molecule structure such as monoacetylcodeine, O^3-acetylmorphine, O^6-acetylmorphine. As shown above, under optimized conditions, this work achieves their complete separation. Due to the similar structure and retention time, nalorphine is commonly used as the internal standard for GC analyses of morphine and its derivatives. Since it is unstable for the natural decomposition study of heroin, SKF_{525A} was adapted in this study. It has a good stability and a retention time compatible to the main components and adulterants commonly found in heroin samples. Figure 3 shows the gas chromatogram of a seized heroin sample, in which the peaks occurred at the retention times of 5.56, 7.15, 8.29, 8.49, 12.28, 14.07, 14.21 and 15.53 min for paracetamol, caffeine, theophylline, phenobarbital, SKF_{525A}, monoacetylcodeine, O^6-acetylmorphine and heroin, respectively. Their peak areas are related to their concentrations in heroin sample.

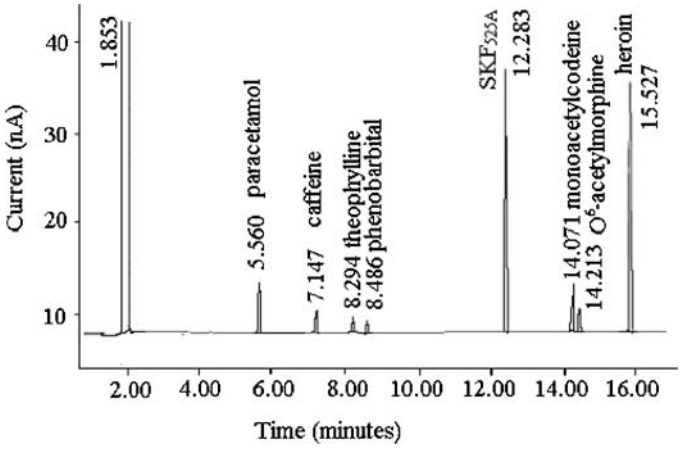


FIG. 3—*Gas chromatogram of a seized heroin sample and internal standard for component analysis.*

Standard ethanol solutions containing 0.2 mg/mL SKF_{525A} and 0.1, 0.2, 0.3, 0.5, 0.8 or 1.0 mg/mL of heroin, O^3-acetylmorphine, O^6-acetylmorphine or monoacetylcodeine were prepared independently to establish respective calibration curves for these corresponds. Linear regression equations derived from the peak area ratio of heroin, O^3-acetylmorphine, O^6-acetylmorphine or monoacetylcodeine to SKF_{525A} and the concentrations are shown in Table 1. The linear ranges of all four components were adequate for quantitative analysis.

Classification of 500 Heroin Samples Based on Heroin Content and Their Components

The analytical results of heroin content and organic adulterants in 500 seized heroin samples indicated that O^6-acetylmorphine and monoacetylcodeine existed in all heroin samples, only 11% of heroin samples did not contain the organic adulterants. Of these seized heroin samples, 79% contained paracetamol. Fifty-four percent of these samples contained both paracetamol and theophylline. Caffeine existed in 54% of these samples. Fifty-six percent of these samples contained three or more kinds of organic adulterants. Seven percent of samples contained procaine, niacinamide and rimifon. Only 1% of these samples contained phenobarbital. These organic adulterants have the chemical character similar to heroin and can improve the activity of heroin, so that they are often used as the adulterants of heroin products.

According to the components of these organic adulterants, the 500 seized heroin samples could be divided into nine groups as shown in Table 2. The nine groups of the 500 heroin seizures

TABLE 1—*The Linear regression equations for determination of heroin, O^3-acetylmorphine, O^6-acetylmorphine and monoacetylcodeine.*

Component	Parameters in y = a + bc (n = 3, c: mg/mL) a	b	Correlation Coefficient	Linear Range (mg/mL)	Detection Limit at 3S/N (μg/mL)
Heroin	0.065	2.317	0.999	0.1–1.0	1.0
O^3-Acetylmorphine	0.027	3.986	0.998	0.1–1.0	1.0
O^6-Acetylmorphine	0.032	4.031	0.999	0.1–1.0	1.0
Monoacetylcodeine	0.051	4.126	0.999	0.1–1.0	1.0

TABLE 2—*Classify of 500 heroin samples based on their components.*

Classify	Content of Heroin	Organic Adulterations	Percent in Total Samples	Sample Source (in China)
1	>70%	not detected	11%	Yunnan, Guangdong and Jilin (Yanbian)
2	>50%	caffeine	8%	Xinjiang, Gansu, Nei Mongol and Eastern-north region
3	~30% (15–60%)	paracetamol and caffeine	18%	Xinjiang, Gansu, Nei Mongol and Eastern-north region
4	~20% (7–40%)	paracetamol, caffeine and theophylline	19%	Xinjiang, Gansu, Nei Mongol and Eastern-north region
5	>40%	paracetamol	2%	Xinjiang, Gansu, Nei Mongol and Eastern-north region
6	20% ±	paracetamol and theophylline	5%	Xinjiang, Gansu, Nei Mongol and Eastern-north region
7	20–40%	paracetamol, theophylline and phenacetin	29%	Xinjiang, Gansu, Nei Mongol and Eastern-north region
8	20–45%	caffeine, procaine paracetamol, niacinamide and rimifon	7%	Xinjiang
9	~20%	caffeine, paracetamol theophylline and phenobarbital	1%	Xinjiang

TABLE 3—*The ratio changes of AC/H, AC/AM and AC/(AM + H) upon adulteration.*

Samples Number	Before Adulteration AC/H	AC/AM	AC/(AM + H)	After Adulteration AC/H	AC/AM	AC/(AM + H)
1	0.0941	0.8500	0.0847	0.0949	0.8288	0.0852
2	0.1309	2.7674	0.1250	0.1326	2.8952	0.1268
3	0.1223	1.7724	0.1144	0.1217	1.7117	0.1136
4	0.1392	4.4051	0.1349	0.1408	4.3457	0.1364
5	0.1120	2.1832	0.1065	0.1114	2.2191	0.1061

were trafficked from Yunnan, Gansu, Xinjiang, Nei Mongol, Jilin (Yanbian), Guangdong and Eastern-north region of China.

Effect of Adulteration on Content Ratio of Different By-products in Heroin Sample with Different Sources

Generally, heroin samples contain some by-products due to the requirements of syntheses and technological processes. These by-products include mainly monoacetylcodeine, O^3-acetylmorphine and O^6-acetylmorphine. Their relative contents may change by adding the organic adulterants during the trafficking of the illicit drug. However, as shown in Table 3, after the heroin samples are adulterated by adding different amounts of paracetamol and caffeine, the content ratios of monoacetylcodeine to heroin (AC/H) and monoacetylcodeine to O^6-acetylmorphine (AC/AM) are found to have a very little change with a variation coefficient of 4.15% and 3.53% ($n = 6$), respectively, during a three month period. Furthermore, the ratio of AC/(AM + H) remains basically at a constant value upon the addition of adulteration. Thus, these ratios provide a base to identify the source of heroin seizures. The values of AM/H, AC/AM and AC/(AM + H) of different heroin samples were determined by GC. When these ratios of different heroin samples were respectively similar, they were considered to be from the same source.

Natural Decomposition Pattern of Heroin and Dependence on Heroin Source

In order to study the natural decomposition pattern of heroin and the by-products in heroin samples upon storage, the solutions and mixtures with four different concentrations of heroin, monoacetylcodeine and O^6-acetylmorphine were prepared by dissolving pure substances in absolute ethanol or by adding solid caffeine and inorganic diluents into the above pure substances. After the solutions and mixtures were stored in air at room temperature for 1, 2, 3, 6, 9 and 12 months, they were analyzed quantitatively by GC under the optimal conditions mentioned above. The changes of the heroin, monoacetylcodeine and O^6-acetylmorphine concentrations were reported in Fig. 4. At different concentrations, all of the solid samples showed greater concentration changes of heroin and O^6-acetylmorphine than the solution samples upon storage under the same conditions. In both solid and liquid phases, with the increasing storage period the heroin contents decreased, the O^6-acetylmorphine contents increased and monoacetylcodeine contents almost retained its original values. The ratio of AC/(AM + H) showed a small change upon the storage. O^6-acetylmorphine is one of the by-products during preparation of heroin product and formed from the hydrolysis and decomposition of heroin. Thus, its content increased in the storage of heroin product. Codeine is a component

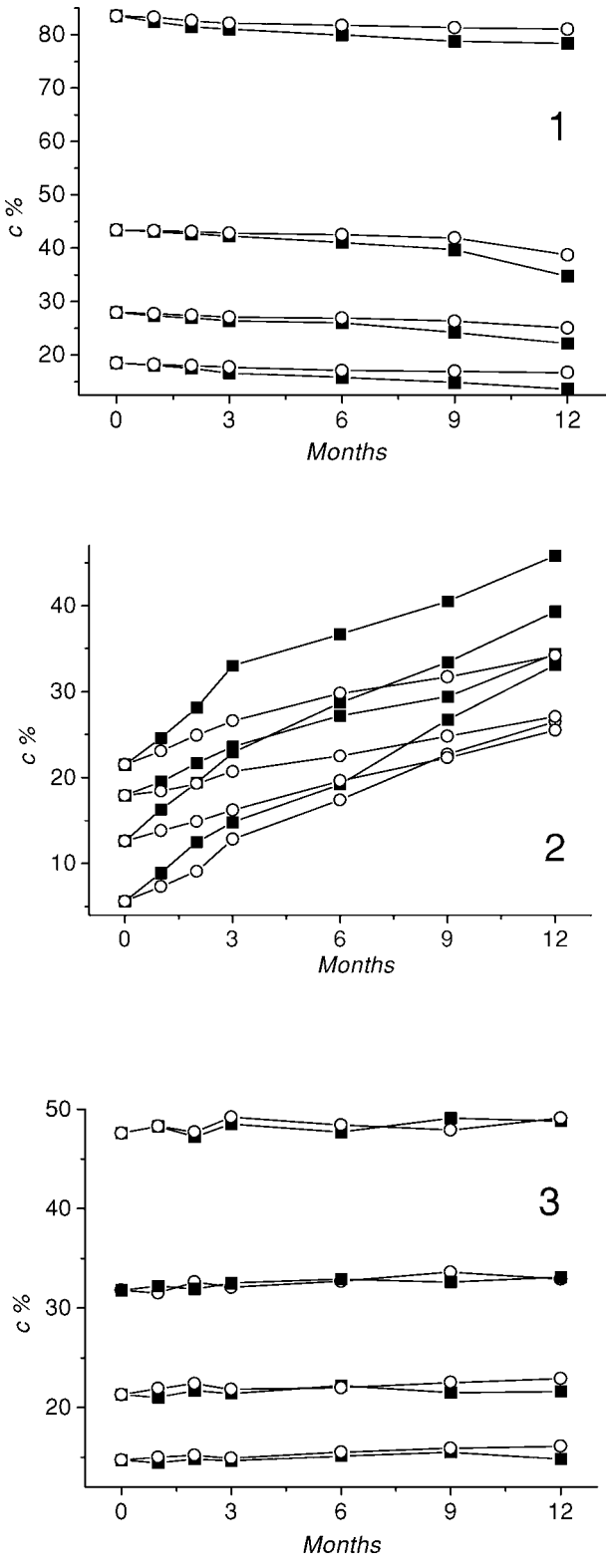


FIG. 4—*Content changes of pure heroin (1), O⁶-acetylmorphine (2) and monoacetylcodeine (3) at four concentrations in solid (■) and liquid (○) phases upon storage at room temperature in air.*

of morphine raw material for preparation of heroin product and has only an acetyl group. The acidylation reaction of codeine in the preparation process of heroin product produces monoacetylcodeine that is very stable, thus its content did not change during 12-month storage. The changes in heroin and O^6-acetylmorphine content resulted in different AC/H and AC/AM values, which would lead to the deviation of heroin source identification. In this case, the source identification based on the ratio of AC/(AM + H) was a more suitable method.

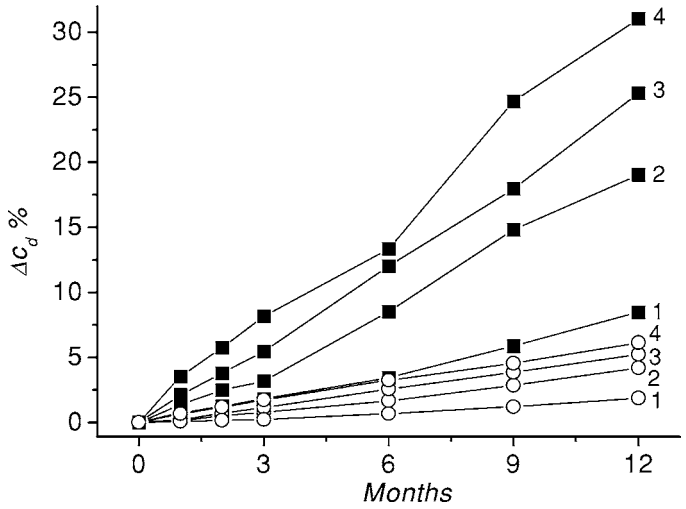


FIG. 5—*Average decrease percentage (ΔC_d%) of heroin contents in four groups of heroin products with different heroin concentration ranges of less than 20% (1), 20–50% (2), 50–70% (3) and more than 70% (4) in solid (■) and liquid (○) phases upon storage at room temperature in air.*

According to the analytical results of heroin content in 500 seized heroin samples, heroin products could be divided in four groups containing, respectively, less than 20%, 20–50%, 50–70% and more than 70% heroin. With the increasing storage period, all of the average decrease percentages of heroin content in four groups increased (Fig. 5). With an increasing heroin content in the heroin products, the average values of varied percentage increased. These values in solid phase were much larger than those in solutions after more than 9-month of storage. When the heroin content was more than 70%, the average values of varied percentage in solid phase reached 31% with a storage period of 12 months, while it was 6.1% in solution. All of the average values of varied percentage of heroin content in four groups in both solid phase and solutions within three months were less than 8.5%. Thus, three-month storage in air at room temperature did not affect the identification of heroin source based on the ratios of AM/H, AC/AM and AC/(AM + H). After a three-month storage, the sample solution of heroin product must be corrected.

Application of Component Analysis in Heroin Source Identification

The proposed method for identification of heroin source has been applied to many illicit heroin trafficking cases. Table 4 gives some examples of heroin source identification in eight illicit heroin trafficking cases. The component ratios of AC/H and AC/AM indicated that the illicit heroin products numbered 98–1112 and 98–1116, 98–0544 and 98–0545 attributed the same source, respectively, and those numbered 98–1067A and 98–1067B, 98–5164 and 98–5260 came from different regions. These results were in good agreement with the information derived from the investigation process. Thus the proposed method could satisfy the requirement for source identification of heroin samples.

Conclusions

GC/MS technique can be used for the component identification of illicit heroin samples. The qualitative and quantitative analytical

TABLE 4—*Practical applications of component analyses in heroin source identification.*

Case	Heroin Content and Orangic Adulterations	Component Ratios		Source Identification the Same Source?	Real Source
98–1112	30.81%	AC/H	0.155	Yes	Xinjiang
	paracetamol, caffeine	AC/AM	2.737		
98–1116	32.43%	AC/H	0.153		Xinjiang
	paracetamol, caffeine	AC/AM	2.584		
98–0544	36.74%	AC/H	0.119	Yes	Xinjiang
	caffeine, procaine, rimifon, niacinamide	AC/AM	0.531		
98–0545	37.71%	AC/H	0.117		Xinjiang
	caffeine, procaine, rimifon, niacinamide	AC/AM	0.532		
98–1067A	37.14%	AC/H	0.185	No	Gansu
	caffeine, procaine, theophylline	AC/AM	4.713		
98–1067B	24.51%	AC/H	0.091		Nei Mongol
	caffeine, procaine, theophylline	AC/AM	2.143		
98–5164	84.19%	AC/H	0.136	No	Yunnan
	not detected	AC/AM	6.674		
98–5260	82.83%	AC/H	0.121		Guangdong
	not detected	AC/AM	9.059		

methods of illicit heroin components have been established by GC and GC/MS techniques. According to the statistical results of the organic adulterants, the 500 seized heroin samples can be divided into nine groups. Both the content ratios of monoacetylcodeine to heroin and monoacetylcodeine to O^6-acetylmorphine are related to the heroin source. Thus, a method to identify the heroin source is proposed. The natural decomposition pattern of heroin products indicated a three-month storage in air at room temperature does not affect the identification of heroin source based on these ratios. After storage of more than three months, the source identification based on the content ratio of monoacetylcodeine to the sum of O^6-acetylmorphine and heroin is a more suitable method. The proposed method has been satisfactorily used for the source identification of 500 seized heroin samples, thus providing useful information for detecting the illicit heroin cases.

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