TECHNICAL NOTE

Dion T. Manhoff,[1] *M.D.; Ian Hood,*[1] *M.B., Ch.B.; Frank Caputo,*[1] *B.S.; Jeffrey Perry,*[1] *B.A.; Samuel Rosen,*[1] *Ph.D.; and Haresh G. Mirchandani,*[1] *M.D.*

Cocaine in Decomposed Human Remains

REFERENCE: Manhoff, D. T., Hood, I., Caputo, F., Perry, J., Rosen, S., Mirchandani, H. G., **"Cocaine in Decomposed Human Remains,"** *Journal of Forensic Sciences*, JFSCA, Vol. 36, No. 6, Nov. 1991, pp. 1732–1735.

ABSTRACT: From March 1988 through March 1990, at the Philadelphia Medical Examiner's Office toxicology laboratory, samples from 77 decomposed human bodies were tested for the presence of cocaine, employing gas chromatography/mass spectrometry (GC-MS). The material analyzed included decomposed soft tissue, bloody decomposition fluid, mummified tissue, maggots, and beetle feces. Twenty-two cases (28.6%) were positive for cocaine, many of these cases in states of advanced decomposition. These findings indicate the usefulness of testing decomposed tissue for cocaine in all cases where its presence is suspected. This is contrary to what might be expected, since cocaine is generally labile and rapidly broken down by both enzymatic and nonenzymatic mechanisms.

KEYWORDS: toxicology, cocaine, tissues (biology), decomposition

Cocaine is generally quite labile in human tissues and has a plasma half-life of less than 1 h [*1,2*]. It is rapidly hydrolyzed by both enzymatic [*3,4*] and nonenzymatic [*5*] reactions and has been reported to be undetectable in animal brain tissue 6 to 8 h after exposure [*6*]. Despite this, it has been our observation at the Philadelphia Medical Examiner's Office (and undoubtedly at other toxicology laboratories, as well) that cocaine and its metabolites are frequently detectable in decomposed human remains. Other authors have also noted that decomposition did not prevent detection of benzoylecgonine by radioimmunoassay (RIA) [*7*]. In this study, which used gas chromatography/mass spectrometry (GC-MS) in combination with solid-phase extraction methods for decreasing background interference, cocaine was detected even in advanced cases of decomposition and mummification. In some cases in which only relatively small amounts of dessicated decomposed tissue remained on essentially skeletal material, positive results were still obtained.

Received for publication 17 Jan. 1991; revised manuscript received 15 April 1991; accepted for publication 16 April 1991.

[1]Pathology resident, deputy medical examiner, chemists, toxicologist, and medical examiner, respectively, Medical Examiner's Office, Philadelphia, PA.

Methods

There were 77 cases in which decomposed human tissue samples were subjected to toxicologic analysis at the Philadelphia Medical Examiner's Office between March 1988 and March 1990. These cases were selected for toxicologic analysis using the same criteria as for undecomposed cases and did not represent a selected group in which circumstantial evidence had suggested that high concentrations of cocaine might exist. The analyzed samples consisted of decomposition fluid from within the circulatory system and body cavities, decomposed and/or mummified soft tissue, which often included maggots, and, in one case, a dust-like tilth of decomposed tissue and beetle feces.

For the solid samples, supernatants were obtained from 3 to 4 g of accurately weighed material, which was mixed and ground in a homogenizer with 15 mL of deionized water spiked with 10 μL of 40 mg/L (0.17-mmol) ketamine in methanol as the internal standard. The resultant homogenate was adjusted to pH 4.5 to 5.0 with 0.1*M* sulfuric acid and centrifuged, and the supernatant was retained. For fluid samples, supernatants were obtained from 2 mL of sample mixed with 7 mL of deionized water and 1 mL of 0.1*M* sulfuric acid, spiked with ketamine internal standard, and pH adjusted, if necessary, as was done with the solid samples. A cocaine standard solution of 0.1-mg/L (0.3-μmol) concentration was prepared from 2 mL of known drug-negative blood in the same way, but with the addition of 5 μL of 40-mg/L (0.13-mmol) cocaine in methanol.

Extraction of the supernatants was performed on 300-mg Clean Screen® DAU columns (Worldwide Monitoring, Horsham, Pennsylvania), which had been preconditioned by being washed twice with 3 mL of methanol and twice with 3 mL of deionized water, using a vacuum manifold. The supernatants were loaded onto the wet columns in reservoirs and suctioned through at a flow rate of 1 to 2 mL/min, followed by washing with 3 mL of deionized water and 3 mL 0.1*M* hydrochloric acid. After being dried under 10-mmHg vacuum for 1 to 2 min, the columns were washed with two 3-mL aliquots of methanol and dried again, and the compounds of interest were eluted with 3 mL of 0.3*M* (1%) ammonium hydroxide (NH_4OH) in methylene chloride/isopropanol (4:1).

For GC-MS analysis, the eluate was dried under nitrogen and reconstituted in 50 μL of ethyl acetate. Eight microlitres of this solution was injected into a Shimadzu GC-9A gas chromatograph (using a 1-m-long, 3.4-mm inside diameter (ID) glass column packed with 3% OV 17 and a helium flow of 30 mL/min) at an initial temperature of 200°C, raised 20°C/min to 280°C. The retention time for cocaine was 3.6 min, and ions of 82, 182, 272, and 303 atomic mass units (amu) were monitored in the selected ion monitoring mode on a Shimadzu QP1000 quadrupole mass spectrometer. For quantitation, the ratio of the 303 amu ion from cocaine to either the 180 or 209 amu ion from ketamine (internal standard) was compared with the same ratios in standards of known concentration. For qualitative screening, the same procedure was followed but the ketamine internal standard was omitted. This permitted determination that the specimen was negative for ketamine and had no other ions that might interfere with quantitation studies.

In some cases, bloody effluent was analyzed using a solid-phase iodine-125 (^{125}I) radioimmunoassay (Coat-a-Count®, Diagnostic Products Corp., Los Angeles, California). This assay was designed primarily for urine, and, although negative results were always confirmed as such with GC-MS, false positives occurred in more than 70% of decomposed samples. This assay therefore proved of little practical use as a screening procedure for these samples.

The GC-MS procedure was found to detect cocaine reliably at concentrations as low as 0.003 mg/L (10 nmol), and such a concentration or greater was considered a positive result for this study. Although standard curves were not established for each assay, they were performed regularly and produced slopes of 0.66, zero intercepts, and correlation

TABLE 1—*Detection of cocaine in decomposed human remains.*

Case	Tissue/ Decomposition Fluid	Cocaine, mg/L or kg
1	bloody effluent	1.800
2	bloody effluent	0.008
3	muscle	0.900
4	bloody effluent	0.910
5	bloody effluent	16.000
6	muscle	0.480
7	muscle	0.420
8	muscle	0.100
9	muscle	1.500
10	bloody effluent	1.400
11	bloody effluent	0.500
12	bloody effluent	0.580
13	bloody effluent	0.770
14	bloody effluent	0.050
15	bloody effluent	0.720
16	bloody effluent	1.000
17	bloody effluent	1.000
18	mummified tissue and beetle feces	0.090
19	bloody effluent	0.200
20	bloody effluent	0.300
21	muscle	0.010
22	muscle	0.003

coefficients of 0.99. The procedure was reliably linear for concentrations between 0.005 and 1.0 mg/L.

Results

Twenty-two of the 77 cases (28.6%) were positive for cocaine. All the types of material submitted for analysis (bloody decomposition fluid, muscle, and mummified tissue) produced positive results at all degrees of decomposition (mild decomposition to complete mummification or skeletonized remains). In one case, even a homogenate of blowfly larvae obtained from the decomposed remains was positive for cocaine. The distribution and quantitation of positive results is shown in Table 1.

Discussion

This study demonstrates the usefulness of testing decomposed human remains for cocaine in all cases where its presence is suspected. Approximately 30% of all cases tested for cocaine at the Philadelphia Medical Examiner's Office were positive over the period of this study, which compares well with the figure of 28.6% found in these unselected decomposed cases. Such results were the primary determinant of the cause of death in several instances and were corroborated by investigation and circumstantial evidence. With current GC-MS techniques, cocaine can be reliably detected at low concentrations, especially when preparatory solid-phase extraction techniques are used to reduce background interference. Why cocaine remains detectable in cases of advanced decomposition despite its normally rapid enzymatic and nonenzymatic degradation is conjectural. It is possible that the enzymatic pathway may itself be lost in the process of decomposition while some substrate cocaine remains, but nonenzymatic degradation would be expected to continue. However, this process is known to be temperature and pH dependent [*8*], and the generally low temperature and acid conditions that pertain

in decomposing tissue could be expected to result in reduced rates of cocaine degradation in such cases. It may be that protection from degradation is afforded by binding of cocaine to specific receptors or other compounds and that such complexes are more resistant to degradation, but this theory requires further investigation. In light of the findings of this study, we would recommend that, in those cases warranting it, analysis for cocaine be performed on decomposed remains without reservation.

References

[1] Wilkinson, P., Van Dyke, C., Jatlow, P., Barash, P., and Byck, R., "Intranasal and Oral Cocaine Kinetics," *Clinical Pharmacology and Therapeutics*, Vol. 27, No. 3, March 1980, pp. 386–394.

[2] Chow, M. J., Ambre, J. J., Ruo, T. I., Atkinson, A. J., Bowsher, D. J., and Fischman, M. W., "Kinetics of Cocaine Distribution, Elimination and Chronotropic Effects," *Clinical Pharmacology and Therapeutics*, Vol. 38, No. 3, Sept. 1985, pp. 318–324.

[3] Stewart, D. J., Inaba, T., Tang, B. K., and Kalow, W., "Hydrolysis of Cocaine in Human Plasma by Cholinesterases," *Life Sciences*, Vol. 20, No. 9, 1 May 1977, pp. 1557–1564.

[4] Inaba, T., Stewart, D. J., and Kalow, W., "Metabolism of Cocaine in Man," *Clinical Pharmacology and Therapeutics*, Vol. 23, No. 5, May 1978, pp. 547–552.

[5] Stewart, D. J., Inaba, T., Lucassen, M., and Kalow, W., "Cocaine Metabolism: Cocaine and Norcocaine Hydrolysis by Liver and Serum Esterases," *Clinical Pharmacology and Therapeutics*, Vol. 25, No. 4, April 1979, pp. 464–468.

[6] Misra, A. L., Nayak, P. K., Bloch, R., et al., "Estimation and Disposition of (^{3}H)-1-Benzoylecgonine and Pharmacological Activity of Some Metabolites," *Journal of Pharmacy and Pharmacology*, Vol. 27, No. 10, Oct. 1975, pp. 784–786.

[7] Robinson, K. and Smith, R. N., "Radioimmuno-assay of Benzoylecgonine in Samples of Forensic Interest," *Journal of Pharmacy and Pharmacology*, Vol. 36, No. 3, March 1984, pp. 157–162.

[8] Isenschmid, D. S., Levine, B. S., and Caplan, Y. H., "A Comprehensive Study of the Stability of Cocaine and Its Metabolites," *Journal of Analytical Toxicology*, Vol. 13, No. 5, Sept./Oct. 1989, pp. 250–256.

Address requests for reprints or additional information to
Haresh G. Mirchandani, M.D.
Medical Examiner's Office
321 University Ave.
Philadelphia, PA 19104