A Fatal Intoxication Following the Ingestion of 5-Methoxy-N,N-Dimethyltryptamine in an Ayahuasca Preparation*

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Abstract

A case of a 25-year-old white male who was found dead the morning after consuming herbal extracts containing β-carbolines and hallucinogenic tryptamines is presented. No anatomic cause of death was found at autopsy. Toxicologic analysis of the heart blood identified N,N-dimethyltryptamine (0.02 mg/L), 5-methoxy-N,N-dimethyltryptamine (1.88 mg/L), tetrahydroharmine (0.38 mg/L), harmaline (0.07 mg/L), and harmine (0.17 mg/L). All substances were extracted by a single-step n-butyl chloride extraction following alkalinization with borate buffer. Detection and quantitation was performed using liquid chromatography-electrospray mass spectrometry. The medical examiner ruled that the cause of death was hallucinogenic amine intoxication, and the manner of death was undetermined.

Introduction

Ayahuasca, also commonly known as yagé, hoasca, and daimê, refers to a decoction of the woody vines of Banisteriopsis caapi. The extract is prepared as a tea formulation, usually with additional plant material containing psychoactive drugs. Ayahuasca has a long history of use in various South American medical and sacramental practices, and its use has expanded into North America and Europe in recent years (1). The foundation of the ayahuasca beverage is the β-carboline alkaloids: harmine, harmaline, and tetrahydroharmine (THH) found in B. caapi. Harmine and harmaline are competitive, reversible inhibitors of monoamine oxidase type-A (MAO), whereas THH is believed to inhibit presynaptic serotonin uptake (2). Harmine, harmaline, and B. caapi extract have also demonstrated increased dopamine release in vitro (3). The harmala alkaloids themselves have mild sedative effects, possibly through interaction with the benzodiazepine receptor (4).

The ayahuasca brew is commonly prepared with the leaves of Psychotria viridis, which contains the U.S. Drug Enforcement Administration Schedule I hallucinogen N,N-dimethyltryptamine (DMT), although nicotine, cocaine, atropine, and other alkaloid combinations have been described (5). Hallucinogenic drugs are generally composed of two groups: phenethylamines and indoleamines. DMT is the prototypical drug of the tryptamine subgroup of the indoleamine hallucinogenic drug group. Other tryptamine derivatives that have been identified as hallucinogenic drugs include 5-methoxy-α-methyltryptamine, α-methyltryptamine, 5-methoxy-N,N-DMT (5-MeO-DMT), N,N-diethyltryptamine, N,N-dipropyltryptamine, and 5-methoxy-N,N-diisopropyltryptamine. All of these compounds have high affinity for the serotonin 5-HT₂ receptors; hallucinogenic potency is well correlated with the relative affinity of these compounds for the 5-HT₂ receptor (6).

DMT and its analogues are metabolized by MAO in the liver. When it is administered orally, first pass metabolism prevents significant absorption of DMT. However, when taken as part of an ayahuasca beverage, the presence of the harmala alkaloids significantly reduces the first-pass metabolism of DMT. As a result, more DMT is free to circulate to the central nervous system where it binds to serotonergic receptors and exerts its psychedelic effects (7). The onset of action of an ayahuasca preparation is between 35 and 40 min with effects felt for up to 3 or 4 h (8–10). Subjective effects have been reported to include nausea, dose-dependent visual and auditory perception changes, personal psychological introspection, increased alertness and stimulation, and changes in gauging the passing of time (7,9,11). Cardiovascular effects have included moderate increases in systolic/diastolic blood pressure and heart rate (7,11) with only diastolic increases significant over placebo (7).

We report a fatal case involving the recreational administration of an ayahuasca-like preparation containing 5-MeO-DMT. Quantitative measurements of 5-MeO-DMT, DMT, harmine, harmaline, and THH are reported for postmortem samples. 5-MeO-DMT has been reported to have greater potency than DMT when smoked (12). Presumably this translates...
to an equivalent potency difference when both are administered orally in combination with a monoamine oxidase inhibitor. This premise was advanced by the self-experimentation reported by Ott (8), which showed oral activity of 5-MeO-DMT at lower doses than DMT when in the presence of identical harmaline doses.

Case History

A 25-year-old white male went camping with family and friends in a national park. According to these individuals, he drank some type of “herbal tonics” and went to sleep. He was found dead the next morning. Subsequent investigation indicated that the decedent ingested a preparation from a South American tree bark, “oasca” (sic), and approximately 4 h later, he ingested tryptamines.

An autopsy was performed the day after the body was discovered. External examination identified the presence of lividity that was fixed on the posterior surface of the body, except in areas exposed to pressure. The body was in full rigor. No remarkable external findings were noted. Internal examinations, both gross and microscopic, were unremarkable except for some tissue congestion and edema. Specimens were sent to the laboratory for toxicologic analysis.

Experimental

Materials

DMT was obtained from the Drug Enforcement Administration central laboratory (Atlanta, GA). 5-MeO-DMT was purchased over the internet from JMAR Chemical Corporation. Harmine hydrochloride, ammonium formate, 5-fluorotryptamine, and sodium borate decahydrate were obtained from Sigma-Aldrich (St. Louis, MO). Harmaline hydrochloride was purchased from Acros Organics (Morris Plains, NJ), and sodium borohydride was purchased from Fisher Scientific (Pittsburgh, PA). All solvents were high-performance liquid chromatography (HPLC) grade and were purchased from Fisher Scientific.

Preparation of tetrahydroharmine

The synthesis of THH was based on a previously reported method (13). Harmaline HCl (500 mg) was added to 50 mL of methanol in a 0°C ice bath. The methanol was acidified by the addition of 4M HCl, and sodium borohydride was slowly added until the solution turned pale yellow and gas was no longer formed. After 30 min, the solution was alkalinized (pH > 10) by the addition of 1N NaOH. The solution was extracted twice with 10 mL of n-butyl chloride, and the solvent was transferred to a 100-mL beaker to evaporate at room temperature. The resulting crude tetrahydroharmine crystals were dissolved in methanol, and 100-μL portions were injected into an Agilent 1100 HPLC (Palo Alto, CA). The instrument consisted of a vacuum degasser, quaternary pump, well-plate autosampler, column compartment, diode-array detector (DAD), and fraction collector. An isocratic mobile phase of 25% acetonitrile, 75% 0.02M ammonium formate was used at a flow rate of 0.8 mL/min. The separation column was a Phenomenex Prodigy-ODS (150 × 4.6 mm, dp = 5 μm) maintained at 35°C. Time-based collections of the tetrahydroharmine peak were made, and fractions were pooled after each injection. Pooled fractions were alkalinized with ammonium hydroxide and extracted into n-butyl chloride. The solvent was evaporated, and the crystals were assayed for purity using the aforementioned HPLC–DAD apparatus. The chromatographic purity of the tetrahydroharmine was 87.4%, and its identity was verified using the full scan mass spectrometry parameters listed herein.

Sample preparation

Blood (central and peripheral), urine, gastric contents, bile, kidney, brain, and liver were collected at autopsy and stored, unpreserved, at –15°C. Standard curves were prepared in blood and urine at 0.010, 0.025, 0.050, 0.10, 0.50, 1.0, and 2.5 mg/L for DMT, 5-MeO-DMT, and harmine. Separate standard curves were prepared for THH and harmaline due to the lower purity of these standard reference materials.

A 5-fluorotryptamine internal standard was prepared at a concentration of 0.01 mg/mL in methanol. One-milliliter volumes of blood, urine, bile, and gastric were initially assayed, but final quantitation was based on respective dilutions of each. Tissue samples (0.5 g) were homogenized in 3 mL of saturated sodium borate buffer using a Brinkmann PT3000 tissue homogenizer (Westbury, NY). One-milliliter aliquots of homogenate were extracted.

Samples and/or their dilutions were added to clean, labeled 16 × 100-mm tubes, and 2 mL saturated sodium borate was added. Fifty microliters of the 5-fluorotryptamine internal standard solution was added to each tube along with 2 mL of n-butyl chloride. The tubes were capped and mixed for 10 min on an orbital mixer. After centrifuging the tubes for 10 min at 3500 rpm, the solvent was transferred to 10-mL conical tubes, a drop of 1% HCl in methanol was added, and the extracts were evaporated to dryness under nitrogen at 40°C. The sample residue was reconstituted with 100 μL of HPLC mobile phase (75% 0.02M ammonium formate/25% acetonitrile) and transferred to autosampler vials. Two microliters was injected for analysis.

Instrumentation

Biological extracts were analyzed using an Agilent 1100 LC–mass selective detector (SL) equipped with an orthogonal electrospray ionization interface. Separation was performed using an XTerra® MS-C18 column (100 × 3.0 mm, dp = 3.5 μm, Waters, Milford, MA) held at 35°C. The mobile phase consisted of 0.02M ammonium formate (75%) and acetonitrile (25%) at a flow rate of 0.4 mL/min.

The positive, pseudomolecular ions of DMT (m/z 189), 5-MeO-DMT (m/z 219), 5-FT (ISTD, m/z 179), THH (m/z 217), harmaline (m/z 215), and harmine (m/z 213) were formed by electrospray ionization and selectively monitored for quantitation. Pneumatic-assisted nebulization utilized nitrogen at
Results

The heart blood and urine specimens from this case were tested for volatile substances and therapeutic and abused drugs. This included volatile testing for methanol, ethanol, acetone, and isopropanol by headspace gas chromatography (GC); acid/neutral drug testing by GC–nitrogen-phosphorus detection (NPD); alkaline drug testing by GC–NPD; morphine by radioimmunoassay; and acetaminophen and salicylate by color test. No ethanol or other volatile substances were detected in the case. Diphenhydramine was detected in the urine; no diphenhydramine was detected in the blood at a limit of quantitation of 0.05 mg/L. In addition, an unidentified peak was detected in both the blood and urine on the alkaline drug screen that eluted around chlorpheniramine. Subsequent mass spectral analysis tentatively identified the substance as 5-MeO-DMT (14). This was verified when an authentic drug standard was subjected to the same extraction, chromatographic, and mass spectral conditions.

Table I lists the distribution of the tryptamines and β-carbolines in the specimens, and Figure 1 shows representative chromatograms of the heart blood sample and a negative blood control. Quantitative values were calculated by multi-point, linear regression of the peak-area ratio of individual compounds to that of the 5-FT internal standard. Regression lines for the five analytes' standard curves were between 0.989 and 0.999. The limits of detection, quantitation, and linearity for all drugs were 0.005, 0.01, and 2.5 mg/L, respectively. All of the analytes were identified in the specimens except for DMT, which was not present in the brain, kidney, or liver. In samples where DMT and 5-MeO-DMT were both measured, 5-MeO-DMT was present at higher concentrations. THH was present in higher amounts than both harmaline and harmine in all samples with the exception of the gastric contents, where the amount of harmine was 10-fold greater than that of THH.

Discussion

To the authors' knowledge, this is the only reported case of death following
ingestion of hallucinogenic tryptamines contained in an ayahuasca preparation. Morano et al. (15) reported a fatal case of ethyltryptamine intoxication in a 19-year-old female who consumed a beer allegedly containing Ecstasy. The heart blood concentration in this case was 5.6 mg/L. In addition, methamphetamine (0.12 mg/L) and amphetamine (0.05 mg/L) were detected. Warren (16) presented an acute nicotine intoxication resulting in death following ayahuasca ingestion during a healing ritual. Harmane and harmaline presence were reported in the ayahuasca brew.

In addition to fatalities from these preparations, there have also been several non-fatal intoxications from hallucinogenic tryptamine use. Meatherall and Sharma (17) published a case report of a 21-year-old male who ingested 5-methoxy-N,N-di-isopropyltryptamine, also known as “Foxy.” A urine concentration of 1.7 mg/L was measured. A metabolite, 5-methoxy-indoleacetic acid (1.3 mg/L) was also detected. Within 4 h of oral ingestion, the hallucinations stopped and the patient was discharged from the hospital. Another nonfatal case of 5-methoxy-N,N-diisopropyltryptamine was reported by Vorce and Sklerov (18); the urine concentration was 0.229 mg/L. In addition, a monoisopropyl metabolite was tentatively identified. A single case of intoxication in a 17-year-old college student was reported after the ingestion of the extract of three Peganum harmala seeds and a combined smoked and snorted dose of 25–30 mg of 5-MeO-DMT (19). This was accompanied by emesis, hyperthermia, increased heart rate, and rhabdomyolysis. However, in this case, only harmine and harmaline were detected in urine.

Oral dosing of 15 volunteers with a traditional ayahuasca preparation was reported by Callaway et al. (13). Average doses, based on a 59-kg individual, were 28.8, 204, 24, and 128.4 mg for DMT, harmine, harmaline, and THH, respectively. The DMT plasma range reported for this trial was 0.011–0.025 mg/L, which is similar to both central and peripheral values for this case. Plasma carboline concentrations were given in the following ranges: THH, 0.049–0.134 mg/L; harmaline, <0.001–0.009 mg/L; and harmine, 0.036–0.222 mg/L. The present case had all three alkaloids near or above these upper ranges with the largest difference seen for THH. In this report, the unknown interval between dosing and death and the large concentrations found in both urine and gastric contents make it impossible to put the values in any context of peak concentrations.

The medical examiner ruled that the cause of death in this case was hallucinogenic amine intoxication, and the manner of death was undetermined.

Acknowledgments

The authors would like to thank Dr. Jerry Zweigenbaum of Agilent Technologies for the loan of the HPLC fraction collector.

This work was funded in part by the American Registry of Pathology, Washington, D.C. 20306-6000.

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Manuscript received November 29, 2004; revision received March 18, 2005.