TECHNICAL NOTE

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Xylazine Toxicity—Literature Review and Report of Two Cases

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ABSTRACT: Xylazine is a veterinary sedative, analgesic or general anesthetic. Its pharmaceutical action results in sympathetic discharge via stimulation of alpha-2-adrenoceptors. In humans, toxicity consists of central nervous system depression, bradycardia and hypotension. The dosages known to produce toxicity in humans vary from 40 mg up to 2400 mg.

Because of decomposition, xylazine blood concentrations in two homicide victims were unknown; however, the concentrations in the brain, liver, and kidneys were much higher in the 23-year-old female versus the 33-year-old male victim. A bottle of xylazine found on the crime scene had a concentration of 100 mg/mL. This 50 mL bottle had 32 mL remaining. Therefore at some point in time 18 mL had been utilized. The amount of available milligrams of xylazine (1800 mg) were enough to cause toxicity in both the woman and the man. Of interest was the fact that the partially skeletonized heads were found remote from the torsos, however, the concentration of xylazine in the body tissues provided a toxicological match of which head belonged to which body. Xylazine toxicity in humans and its relationship to these homicides will be the focus of this report.

KEYWORDS: forensic science, forensic pathology, forensic toxicology, xylazine, toxicity, homicide

Xylazine (Rompun, Bay 1470, Bayvet) is a veterinary sedative, analgesic or general anesthetic for large animals such as deer, ruminants, and horses. Its pharmaceutical action (similar to clonidine) results in sympathetic discharge via stimulation of alpha-2-adrenoceptors. The drug is available in solutions of 20, 50, and 100 mg/mL and as dry product consisting of a vial containing 500 mg (1). In humans, xylazine toxicity consists of central nervous system depression (tiredness, faintness, coma, respiratory suppression), bradycardia, hypotension and hyperglycemia (1–7).

Toxicological analysis of organs from the dismembered bodies of two homicide victims revealed xylazine in the torsos which were found remote from the heads. The concentrations in the brains and viscera of these victims provided a toxicological match of which head belonged to which body. The purpose of this report will be to review the literature of xylazine toxicity and its relationship to these homicides.

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TABLE 1—*Toxicological results* for 23-year-old woman.*

Substance	Specimen	Result
Ethanol	Brain	0.01 g/dL
Ethanol	Liver	Detected < .01 g/dL
Drugs	Gastric	Undetected
Xylazine	Liver	42 mg/kg
Xylazine	Kidney	28 mg/kg
Xylazine	Brain	19 mg/kg

*See text for testing methods.

Case Reports

The decomposed torsos of a 33-year-old man and a 23-year-old woman were found each within their respective 55 gallon drum dumped in a Dade County waterway. Approximately one month later, the severely decomposed heads were found in $3^1/_2$ gallon buckets in a remote location. The man had evidence of antemortem blunt cranial trauma, however, the woman did not have any detectable antemortem physical injuries. The heads and torsos were identified by various conventional methods including dental, medical, radiological, anthropological and circumstantial evidence. DNA testing from the bodies was also conclusive for positive identifications. The manners of death were both classified as homicide.

At the crime scene, there was a 50 mL vial of xylazine (100 mg/mL) with 32 mL of solution remaining. In addition, police recovered syringes and a hypodermic-dart gun of the type used to subdue dangerous animals.

Toxicological testing revealed the presence of xylazine in the brain and viscera of both victims (Tables 1 and 2). The xylazine

TABLE 2—*Toxicological results* for 33-year-old man.*

Substance	Specimen	Result
Ethanol	Brain	0.04 g/dL
Ethanol	Liver	0.08 g/dL
Drugs	Gastric	Undetected
Xylazine	Brain	0.16 mg/kg
Xylazine	Liver	0.26 mg/kg
Xylazine	Kidney	0.15 mg/kg

*See text for testing methods.

was assayed because of the scene investigation, however, the drug would have been detected by standard toxicological testing procedures utilized in our laboratory.

Materials and Methods

Materials

All chemicals used in the extraction were reagent grade. The organic solvents n-butyl chloride, hexane and methanol were Distilled-in-Glass quality from Burdick-Jackson (VWR Scientific, Atlanta, GA.). Sodium borate and hydrochloric acid were obtained from Baker Chemicals. The internal standard, cyclizine, was obtained from Burroughs-Wellcome. Xylazine standards were prepared from a pharmaceutical grade solution of Rompun 100 (100 mg/mL as the hydrochloride; Miles Inc. Agricultural Division, Shawnee Mission, KS).

Methods

Xylazine was initially detected in a basic drug screen. The screen was performed on a Hewlett Packard Model 5890 gas chromatograph equipped with dual nitrogen-phosphorus detectors and a single split/splitless injector. The chromatographic columns, DB-1 and DB-17, were 30 m × 0.25 mm ID capillary columns with 0.25 micron film thickness (J&W Scientific). The column oven was programmed from 50°C to 190°C at 20°C per minute with an initial hold time of 1 min. The column was then programmed to 290°C at 5°C per minute and held for 30 min. The injector and detectors were set to 250°C and 300°C respectively. The carrier gas was helium at 1.5 cc/min. The detector gases were hydrogen at 3.1 cc/min, air at 100 cc/min, and nitrogen make-up at 30 cc/min. Extracts were analyzed simultaneously on the two columns with a single splitless injection. The nitrogen phosphorus detectors were optimized to a bead current of 20 picoamps.

Xylazine was confirmed in tissue samples using a Finnigan Model ITS-40 Ion Trap gas chromatograph/mass spectrometer. Identification was made by comparing the full spectra to the data system library and to published reference spectra (8). The chromatographic column was a 15 m × 0.25 mm ID. DB-5MS capillary column with a 0.25 micron film thickness (J&W Scientific). The column oven was programmed from 50°C to 300°C at 20°C per minute. The initial and final hold times were 1 min. The injection port and ionizer temperatures were 250°C and 220°C respectively. The injection was made in the splitless mode with a 0.5 min hold time. The carrier gas was helium at 2 cc/min. The instrument was scanned from 50–450 amμ at 1 sec/scan (3 microscans).

Quantification of xylazine in tissue samples was performed on a Hewlett Packard Model 5890 gas chromatograph equipped with a nitrogen-phosphorus detector and a Model 7673A autosampler. The chromatographic column was a 15 m × 0.53 mm ID. DB-1 capillary column with a 1.5 micron film thickness (J&W Scientific). The column was programmed from 150°C to 270°C at 10°C per minute. The initial and final hold times were 1 min. The carrier gas was helium at 15 cc/min. The detector gases included hydrogen at 3.1 cc/min, air at 102 cc/min and nitrogen make-up at 16 cc/min. The detector and injector temperatures were 250°C and 300°C respectively. The injector was a packed column inlet with a 1 mm ID straight liner packed with a small plug of glass wool. The detector was optimized at a voltage setting sufficient to generate a bead current of 20 picoamps. A personal computer based Perkin Elmer Turbochrom version 4.0 data system was used to evaluate chromatograms and calculate concentrations.

Extraction

The analyses were performed on kidney, liver, and brain. The extraction procedure for the initial basic drug screen and the quantification were the same. Ten grams of tissue were weighed and combined with 10 mL of water and homogenized. A 2 mL aliquot was combined in a 15 mL screw top culture tube with 1 mL of saturated sodium borate buffer and 1 mL of 0.5 mg/L cyclizine internal standard solution. The tube was vortexed and extracted with 8 mL of n-butyl chloride on a rotator for 15 min. After centrifugation the solvent layer was transferred to a second tube and back-extracted with 1 mL of 0.1N hydrochloric acid on a rotator for 15 min. After centrifugation the solvent layer was aspirated to waste and 5 mL of hexane added to each tube. The tubes were vortexed and centrifuged. The hexane layer was aspirated to waste. The acid layer was alkalinized with 1 mL 0.1 N sodium hydroxide and 1 mL of saturated sodium borate buffer. The alkaline aqueous layer was extracted with 8 mL of n-butyl chloride on a rotator for 15 min and then centrifuged to separate the layers. The solvent layer was transferred to a third tube and evaporated to dryness under nitrogen at 50°C. The residue was dissolved in 50 μL of methanol and transferred to an autosampler vial for analysis.

Calibration

A 1 mg/mL working stock solution of xylazine was prepared using an aqueous 100 mg/mL xylazine hydrochloride solution received with the case. A calibration curve for xylazine was prepared by spiking 1 mL of water with the working stock to yield 0.05, 0.1, 0.5, and 1.0 mg/L standards. Data analysis and calculations were performed using the Perkin Elmer Turbochrom 4.0 software. A least squares linear regression analysis was performed using peak area ratios of analyte to internal standard versus concentration. The r^2 correlation coefficient for the xylazine standard curve was 0.9959. When necessary, tissue samples were diluted to yield concentrations within the limits of the calibration curve.

Discussion

Toxicological data regarding human dosage of xylazine to achieve signs and symptoms is scant; however, veterinary sedation of a red deer requires 0.3 to 0.5 mg/kg (75–125 mg dose to sedate a 250 kg animal) and a fallow deer requires 0.5–1.0 mg/kg (e.g., 50–100 mg dose to sedate a 100 kg animal) (1). Other dosages include 0.4 mg/kg for caesarean section and 0.1 to 0.2 mg/kg for superficial surgery on cattle, 2 to 5 mg/kg for superficial surgery on a horse, 1.1–2.0 mg/kg for anesthesia/sedation for cats and dogs, and 0.22 mg/kg for respiratory suppression of goats (2,3).

Based on the information from published articles, the dosages of xylazine known to produce toxicity in humans vary from 40 mg up to 2400 mg (1–7) (Table 3). In most of the cases, the concentrations of administered xylazine were 100 mg/mL. Dosage information based on body weight is available through Spoerke et al. (6), who describe usage of 0.73 mg/kg and 22 mg/kg by a 29-year-old woman and a 37-year-old woman, respectively. The symptoms of the woman with the lesser dose included disorientation, hypotension and bradycardia whereas the woman with the higher dose was somnolent, apneic, bradycardic and hypotensive. Both women recovered, however, the woman with the larger dose required four days of hospitalization.

The scant toxicological data are not of great assistance in comparing xylazine blood or tissue concentration with the two cases under consideration (Tables 1–3). In one tissue distribution study, Poklis et al. (5) determined a blood concentration of 0.2 mg/L as well as

TABLE 3—*Cases of xylazine toxicity.*

References	Age/Sex Race[1]	Manner of Usage[2]	Route[3]	Vial mg/mL	Amount Used (mL)	Dose (mg)	Toxicologic[4] Results	Comments
Carruthers et al. (1979)(2)	34 M	A	IM	100	10	1000	—	Tx of insomnia access to horses
Gallanosa et al. (1981)(3)	20 WF	S	Oral	100	4	400	Plasma: Neg Urine: Pos	Fight with boyfriend, horse trainer, prior suicide attempts
Lewis et al. (1983)(4)	39 F	A	IM/SQ	—	—	—	Serum: .03 mg/L Urine: 1.7 mg/L	Tx of horse bite, veterinary surgeon's wife
Poklis et al. (1985)(5)	36 WM	A	IV	—	—	—	Blood*: 0.2 mg/L Urine: 7 mg/L Viscera: 0.05–0.9 mg/kg Ethanol (Serum): 0.38 g/dl Nordiazepam (Serum): 2.5 mg/L	Recreational abuse, fatality[2]
Spoerke	29 F	—	IM	40	1	40(0.73 mg/kg)	—	—
et al.	37 F	S	IM	100	24	2400(22 mg/kg)	—	Depression
(1986)(6)	29 F	—	IV	—	—	—	—	—
Samanta et al. (1990)(7)	19 M	A	SQ	100	2	200	—	Veterinary nurse

[1]M = Male, F = Female, W = White.
[2]A = Accidental intoxication, S = Suicide attempt/Only one fatality is reported (Poklis et al.).
[3]IM = Intramuscular, SQ = Subcutaneous, IV = Intravenous.
[4]All results are for xylazine unless otherwise specified.

concentrations of 0.05 to 0.9 mg/kg in various body tissues. The latter data, however, are not useful in that the decedent had a large blood ethanol concentration (0.38 g/dL) combined with blood concentrations of nordiazepam (2.5 mg/L). Lewis et al. (4) found a 0.03 mg/L serum concentration after an intramuscular or subcutaneous injection in a 39-year-old woman who was treating herself for horse bite. She suffered drowsiness, blurred vision and bradycardia. The blood concentration of xylazine is hard to interpret because it is not certain when the medication had been taken. Xylazine has also been detected in the urine (3–5) suggesting that urine may well be a satisfactory screening specimen for xylazine toxicity.

Because of decomposition, the xylazine blood concentrations in the cases of the two homicide victims are unknown. The concentrations in the brain, liver, and kidney were much higher in the female versus the male victim (Tables 1, 2). The bottle of xylazine found on the crime scene had a concentration of 100 mg/mL. This 50-mL bottle had 32 mL remaining. Therefore, at some point in time, 18-mL had been utilized. The amount of available xylazine (1800 mg) is enough to produce toxicity in both the woman and the man. In the article by Spoerke et al. (6) doses of 0.73 mg/kg (40 mg total dose) and 22 mg/kg (2400 mg total dose) resulted in severe symptomatology in the female victims weighing 54.5 kg (119.9 pounds) and 109 kg (239.8 pounds). Such data are worth comparing with the two studied cases. In addition, it is noteworthy that only 50–100 mg and 500 mg will sedate a 100 kg fallow deer and a 454 kg horse, respectively. The ethanol concentrations in the man's brain and liver are consistent with putrefication and/or ethanol ingestion. In either event, the ethanol concentrations are considered negligible.

The final consideration of this report is the linking of dismembered remains by toxicological matching. Analysis of Tables 1 and 2 reveal much lower concentrations in brain and viscera for the man compared with the woman. The differences are such that it is reasonable to conclude which head belonged to which torso based on these comparisons. Xylazine use in man is so unusual that the odds against a coincidence (i.e., head/viscera from other persons in the community) under these circumstances are astronomical.

In conclusion, xylazine is a potentially hazardous drug in humans. Small dosages may produce toxicity and larger dosages may be survived with medical help. The concentrations in the female victim are consistent with fatal overdose. The male victim had other injuries which could explain his death, however, xylazine could have been utilized for sedative purposes. Finally, the medical examiner should consider use of toxicological matching of dismembered remains as a method of verifying identification.

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