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CASE REPORT

A case of fatal arsenic poisoning

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Abstract We describe a case of poisoning by arsenic trioxide of a young man found dead at home. There were no obvious external signs of arsenic poisoning; but we observed marked endocardial hemorrhages, hepatomegaly, diffuse gastric mucosal hemorrhages, and slight brain edema at autopsy. The Reinsch test for the stomach contents and liver homogenate was positive for arsenic. Wavelength-dispersive X-ray fluorescence spectrometry combined with the Reinsch test showed that fatal levels of arsenic were present in blood and tissues. The cause of death was diagnosed as circulatory collapse caused by arsenic trioxide.

Keywords Arsenic poisoning · Arsenic trioxide · Tissue distribution · Reinsch test · X-Ray fluorescence spectrometry

Introduction

While fatal poisoning by arsenic is not common in Japan, a few fatalities occur every year for persons who commit suicide by ingesting arsenic [1]. In the summer of 1998, 4 persons died and 63 were hospitalized in Japan after eating curry that had been deliberately mixed with arsenic trioxide.

Arsenic trioxide is cytotoxic, because it inhibits important sulfhydryl-containing enzymes [2]. Acute ingestion of 200–300 mg arsenic trioxide is fatal, and symptoms, such as nausea, vomiting, abdominal pains, and diarrhea, may appear within 30 min. Damage to the capillary endothelium in the gastrointestinal mucous membranes results in exudation of plasma and erosion. Larger amounts of arsenic trioxide can cause death by cardiovascular collapse within several hours. If a victim survives the acute phase, then hepatomegaly (fat degeneration) and anuria due to renal failure may be observed within a few days [3]. After a longer survival period, victims may develop severe encephalopathy and polyneuropathy [4].

We report a case of fatal arsenic poisoning, in which no information suggesting arsenic use was available from police investigations at the time of autopsy; we present the autopsy case of arsenic poisoning, in which arsenic levels was carefully measured in body fluids and tissues using X-ray fluorescence spectrometry.

Case history

A man in his twenties (160 cm tall and weighing 76 kg), who worked for a termite control company, returned home from work around 10 p.m. and complained to his family of severe nausea and vomiting. He took a stomachache remedy and went to bed. He was found dead at home at around 6 a.m. sitting on the floor and leaning on the wall in front of the toilet. He had not complained to his colleagues of any physical problems before leaving work the night before. No information to suggest the use of any poisonous substances was available from the police.

The postmortem interval between death and the autopsy was estimated to be approximately 36 h. No external injuries were present at autopsy. The face was

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markedly congested. No petechial hemorrhage was observed on the conjunctivae of the eyes. The heart weighed 350 g; it contained a large volume of liquid blood and marked endocardial hemorrhage was observed in the left ventricle. The liver weighing 1900 g was markedly degenerated with deposition of fat. The brain weighing 1350 g was slightly edematous. The stomach contained 400 g of slightly brownish liquid. The gastric mucosa was markedly edematous and diffuse hemorrhages were evident. The urinary bladder contained no urine. Other organs were normal except for the presence of congestion.

Toxicological analyses

Sample collection

Small samples (5–10 g) of organs were obtained after the surface was quickly washed with running water and wiped dry with a clean towel. Fluid samples (5–10 ml) were collected using disposable clean syringes. Whole stomach contents were weighed and mixed well in a beaker, and a 50-g portion of the contents was stored for analysis.

Chemicals

Standard solutions of arsenic (100 and 1000 μg/ml) were purchased from Wako (Osaka, Japan), and were diluted with distilled water to prepare 5–500 μg/ml solutions for calibration curves. Other chemicals were of analytical reagent grade.

Screening of drugs and poisons

The stomach contents and liver homogenate (liver: distilled water = 1 : 3 w/w) were tested for heavy metals by the Reinsch test [5]. Blood and stomach contents were tested for alcohols, volatiles, and nonvolatile compounds by gas chromatography, as described previously [6,7].

Measurement of arsenic

An analytical method described by Ozo et al. [8] was modified. Briefly, 4 ml of body fluid homogenate (body fluid: distilled water = 1 : 3 v/v), 4 g of stomach content homogenate (stomach contents: distilled water = 1 : 3 w/w), or 4 g of tissue homogenate (tissue: distilled water = 1 : 3 w/w) was mixed with 1 ml concentrated HCl. The mixture was heated with three pieces of copper plate (10 × 5 mm), which had been cleaned by dipping into 1 N nitric acid, at 100°C for 30 min in a heating block. The copper plate was then washed with distilled water and ethanol and dried. The copper arsenide sticking to the plate was eluted into 0.5 ml of methanol/concentrated ammonia (8 : 2) at 60°C for 15 min in the heating block. The resulting solution was dropped onto a filter paper with a plastic ring (external diameter: 48 mm, center filter diameter: 20 mm) (Micro Carry, Rigaku, Tokyo, Japan) and dried. The copper plate was again rinsed in 0.25 ml of methanol/concentrated ammonia (8 : 2), and the solution was also dropped onto the same filter paper and dried. The filter paper was set on a wavelength-dispersive X-ray fluorescence spectrometer with a rhodium anode tube (RIX 3001, Rigaku). Tube voltage and current were 50 kV and 80 mA, respectively.

Results

The Reinsch test was positive for the stomach contents and liver homogenate. The sublimates obtained by strongly heating the precipitates on the copper plate were recognized microscopically as crystals of arsenic trioxide (Fig. 1). Routine screening by gas chromatography was negative for common organic compounds.

Arsenic completely reacted with copper, regardless of the sample matrix and the limit of quantification was 5 μg/ml (or 5 μg/g) using X-ray fluorescence spectrometry combined with the Reinsch test procedure. A calibration curve for arsenic concentrations versus secondary X-ray (Kα ray) intensity was linear in the range of 5–500 μg/ml (r = 0.995). Determinations of within-day precisions for 5 and 500 μg/ml were fairly good with coefficients of variation (n = 3) of 17.5% and 11.3%, respectively.

High levels of arsenic were detected in body fluids and tissues (Table 1). The highest concentrations were obtained for the liver and bile. A fatal amount of arsenic

Table 1 Arsenic concentrations in various body fluids and tissues

Sample	Arsenic concentration (μg/ml or μg/g)
Blood	
Left cardiac chamber	6.3
Aorta	5.1
Bile	22.9
Lung	
Left	6.3
Right	8.4
Liver	23.0
Right kidney	16.4
Stomach contents	489 (196)[a]

[a] Total amount given in parentheses in milligrams, which is equivalent to 255 mg arsenic trioxide

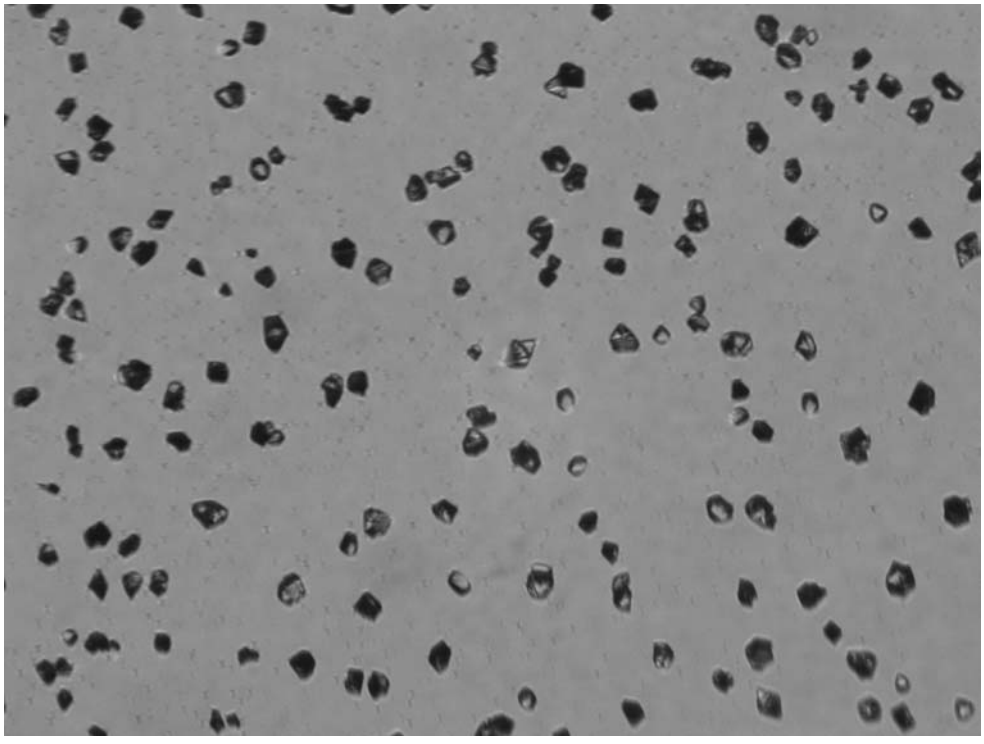

Fig. 1 Microscopic appearance of sublimates obtained by strongly heating copper plates used for the Reinsch test for the stomach contents (×100)

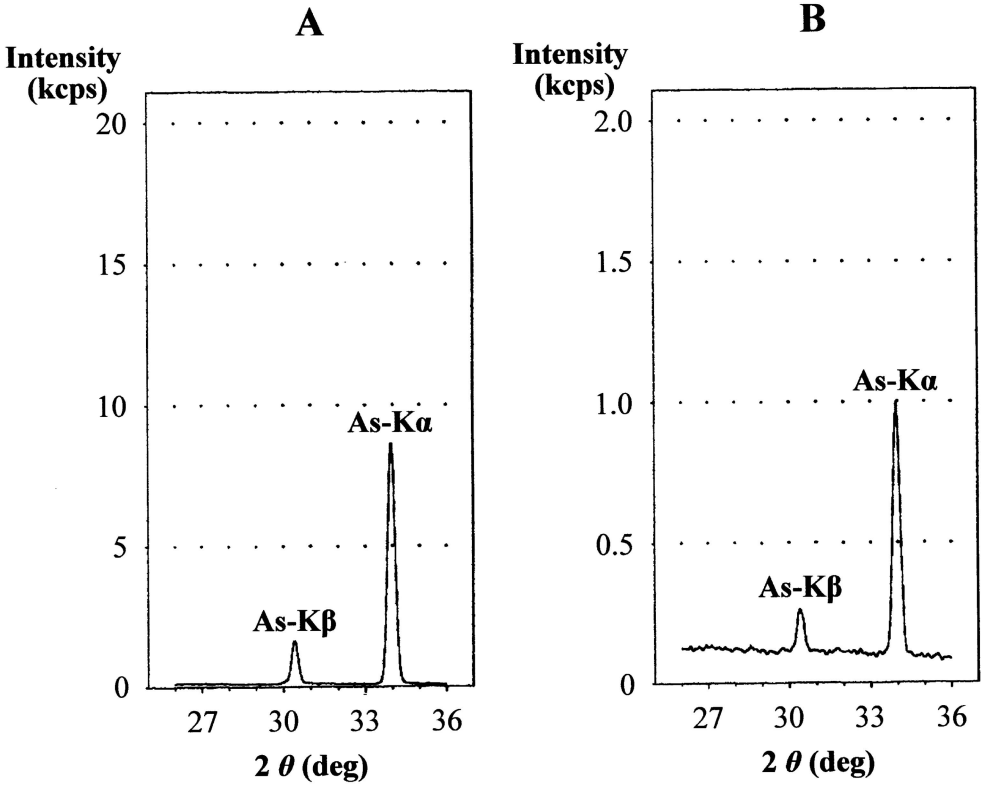

Fig. 2 X-Ray fluorescence spectra of a reactant with copper plate in **A** 4 g stomach content homogenate (stomach contents: distilled water = 1 : 3) and in **B** 4 ml of distilled water containing 100 µg arsenic

(196 mg: 255 mg as arsenic trioxide) remained in the stomach. An X-ray fluorescence spectrum obtained from the stomach contents is shown in Fig. 2.

The cause of death, therefore, was diagnosed as circulatory collapse caused by arsenic trioxide.

Discussion

Police scientific methods and investigations failed to find any evidence of criminal acts in the present case. However, our autopsy findings of marked edema and diffuse

hemorrhages of the gastric mucosa and fat degeneration of the liver were strongly suggestive of chemical poisoning by arsenic [3]. Therefore, we performed the conventional Reinsch test for gastric contents and liver homogenate in addition to the routine gas chromatographic screening for general organic compounds. As expected, positive results were obtained in the Reinsch test and the sublimates were microscopically recognized as crystals of arsenic trioxide. Ozo et al. [8] failed to obtain positive results for pentavalent organic and inorganic arsenic by the Reinsch test. Only arsenic trioxide can react with copper to form copper arsenide (Cu_5As_2). This is important for forensic chemists, because arsenic trioxide is the most usual form of arsenic to be used in criminal cases.

X-ray fluorescence spectrometry cannot discriminate between trivalent and pentavalent arsenic. However, it can exclusively detect toxic trivalent arsenic trioxide, when it is combined with the Reinsch test procedure [8]. Ozo et al. [8] successfully detected as little as 0.8 μg of arsenic trioxide in 2 ml of serum using X-ray fluorescence spectrometry combined with the Reinsch test procedure. We applied this method to body fluids and tissues of the present case, because it was much simpler than other methods such as inductively coupled plasma mass spectrometry with ion chromatography [9]. X-Ray fluorescence spectrometry allowed us to quantify arsenic regardless of the kind of sample matrix, although it was not as sensitive as that reported by Ozo et al. [8]. The low sensitivity of our X-ray fluorescence spectrometry may be due to the fact that we dropped arsenic-containing eluate on the entire area of the filter of Micro Carry. Ozo et al. [8] dropped their solution onto filter paper that was held under a hair dryer to prevent the drop from diffusing.

We confirmed high levels of arsenic in the liver and kidney (Table 1) [10]. Although blood levels of arsenic were relatively low even in severely intoxicated patients [4,11,12], the values we observed in blood were much higher than the reported levels. Bile may be a suitable fluid for detecting arsenic, because its level was as high as those found in the liver and kidney.

When a fatal amount of arsenic trioxide is ingested, gastrointestinal symptoms usually appear within 30 min. Because his colleagues alleged that the victim appeared in normal physical condition before leaving work, he might have died within several hours after being intoxicated with arsenic trioxide. It is thought that hepatomegaly is usually induced a few days after acute arsenic intoxication [3]. Our findings, however, indicate that fat degeneration of the liver may really occur within a matter of several hours. A marked endocardial hemorrhage was noteworthy in our case and its mechanism should be investigated.

References

1. National Research Institute of Police Science (2001–2005) Annual case reports of drug and toxic poisoning in Japan (in Japanese). Nos. 43–47
2. Goyer RA (1996) Toxic effects of metals. In: Klaassen CD (ed) Casarett and Doull's toxicology: the basic science of poisons, 5th edn. McGraw-Hill, New York, pp 691–736
3. DiMaio DJ, DiMaio VJM (1993) Forensic pathology. CRC, Boca Raton, pp 439–487
4. Dueñas-Laita A, Pérez-Miranda M, González-López MA, Martín-Escudero JC, Ruiz-Mambrilla M, Blanco-Varela J (2005) Acute arsenic poisoning. Lancet 365:1982
5. Curry AS (1988) Poison detection in human organs, 4th edn. Thomas, Springfield, pp 83–113
6. Moriya F, Ishizu H (1994) Can microorganisms produce alcohol in body cavities of a living person? A case report. J Forensic Sci 39:883–888
7. Moriya F, Hashimoto Y (1998) Medicolegal implications of drugs and chemicals detected in intracranial hematomas. J Forensic Sci 43:980–984
8. Ozo Y, Yoshizawa M, Murata A, Shimazaki S, Kajiwara M, Takagi T, Sato Y (2004) Simple quantitation of arsenic by energy dispersive fluorescence X-ray spectrometer using Reinsch's test (in Japanese with English abstract). Jpn J Toxicol 17:359–364
9. Suzuki S, Suzuki Y (2005) Arsenic compounds and other organic poisons. In: Suzuki O, Watanabe K (eds) Drugs and poisons in humans: a handbook of practical analysis. Springer, Berlin Heidelberg New York, pp 637–647
10. Baselt RC (2002) Disposition of toxic drugs and chemicals in man, 6th edn. Biomedical, Foster City, pp 79–82
11. Sanz P, Corbella J, Nogué S, Munné P, Rodriguez-Pazos M (1989) Rhabdomyolysis in fatal arsenic trioxide poisoning. J Am Med Assoc 262:3271
12. Benramdane L, Accominotti M, Fanton L, Malicier D, Vallon J-J (1999) Arsenic speciation in human organs following fatal arsenic trioxide poisoning: a case report. Clin Chem 45:301–306