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CASE REPORT

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A case of drowning linked to ingested sulfides – a report with animal experiments

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Abstract An adult male was found dead beneath a pool of sewage in the pump room of a fish market. Autopsy revealed the cause of death to be suffocation after aspirating sewage into the respiratory tract. Since hydrogen sulfide gas was detected in the atmosphere at the scene of the accident, gas poisoning was suspected and toxicological analysis of sulfides in body tissues was performed. The concentrations of sulfides in the blood, lung and kidney were 0.95 μmol/ml, 0.22 and 0.38 μmol/g, respectively. These values were remarkably higher than those in previously reported cases involving exposure to hydrogen sulfide gas. Therefore, oral intake of sulfides was assumed and the distribution of sulfides in tissues following oral administration of sodium sulfide solution was examined by means of animal experiments using rats. The concentration of sulfides in the blood from rats following oral intake was much higher than that seen following gas exposure. Based on these results, we concluded that the victim had been exposed to hydrogen sulfide gas and had then collapse into a pool of sewage containing sulfides. The sulfides which were distributed throughout the body tissues had mainly issued from the alimentary tract prior to death by drowning.

Key words Toxicology · Drowning · Sulfides · Human tissue · Distribution

Introduction

Although several poisoning cases due to hydrogen sulfide have been reported from the aspect of clinical medicine or industrial hygiene [1–3], as far as we know, only a few papers have mentioned the concentration of sulfides in the body tissues in cadavers from the aspect of forensic medicine [4–8]. Moreover, no investigation has been reported concerning the distribution of sulfides following oral administration.

In this paper, we report a fatal case involving ingested sulfides, and present an evaluation of sulfide distribution in tissues based on animal experiments.

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Case report and autopsy findings

A 37-year-old man and his colleague entered an underground pump room to repair a drainage pump in a fish market (Fig. 1). When the man loosened a check valve on the pump, both gas and sewage gushed out. Because his colleague smelled hydrogen sulfide gas, he told the victim to leave the room and left to fetch a ventilator. When he returned, the victim was lying in a prone position with his face beneath the surface of a pool of sewage which covered the floor to a depth of about 50 cm. After the room had been ventilated, the victim was taken to hospital in an ambulance, in which he vomited about 800 ml of gastric contents. Intensive cardiopulmonary resuscitation including gastric lavage was performed in the hospital, but this proved to be in vain.

At the time of the inspection of the scene, it was noted that the concentration of hydrogen sulfide gas was 123 ppm in the atmosphere and that a stop valve between the pump and the sewage tank was rusted and had been left open.

A forensic autopsy was carried out 24 hours after the accident in order to diagnose the cause of death. The cadaver was kept at 0° C until autopsy to prevent the postmortem production of sulfides.

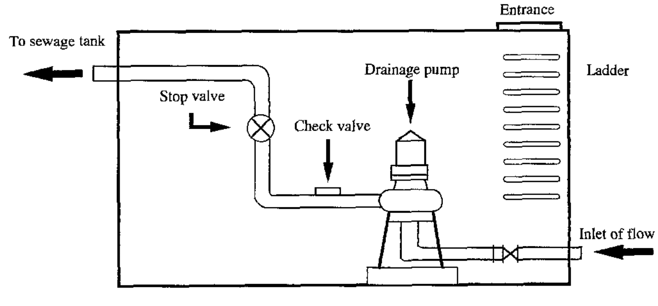

Fig. 1 Schematic diagram of the underground pump room

There were no injuries or wounds to the body. In addition, no findings of histopathological significance were observed, which could have brought about the victim's sudden death. However, ca. 200 ml of fluid was found in the stomach with the obnoxious odor of bad eggs, with minute bubbles in the bronchi and tracheae. Findings associated with acute death, such as petechiae beneath the mucosa, congestion of organs and dark red non-coagulated blood in the heart were also found. Microscopically, many plankton were observed in the samples obtained from the lung, liver and kidney. Blood and solid tissues were collected and stored at –20° C until toxiological analyses. Urine could not be obtained from the cadaver.

Materials and methods

Animal experiments

Four adult male Wistar rats weighing about 300 g were anesthetized with ether and administered 1 ml of 10% Na_2S solution orally, which was the minimum lethal dose known from preliminary experiments. All the rats died by poisoning within 5 min after the administration. The tissue samples including whole blood were collected immediately after death and stored at –20° C until analysis. All the animal experiments, including preliminary ones, were carried out under the control of the Guideline for Animal Experiment in the Faculty of Medicine, Kyushu University and the Law (No. 105) and Notification (No. 6) of the Government.

Analytical procedure for sulfides

Since sulfides are easily oxydized or volatilized by the means of homogenization [9, 10], we had developed a new method to analyse sulfides in the slices of frozen samples, using extractive alkylation technique [11]. The procedures are described in brief.

A 0.5 ml volume of 20 m*M* pentafluorobenzyl bromide solution in toluene and 2.0 ml of ethyl acetate containing 10 μ*M* 1,3,5-tribromobenzene (TBB) as internal standard were placed in a 10-ml glass-stoppered test tube with 0.8 ml of 5 m*M* tetradecyldimethylbenzylammonium chloride solution in oxygen-free water saturated with sodium tetraborate. To this mixture was added 0.2 ml (or g) of test sample, and the preparation was stirred vigorously for 1 min at room temperature. About 0.1 g of potassium dihydrogenphosphate was added as a buffer to stop excessive alkylation by tissue protein. The preparation was stirred vigorously for 10 s, and then centrifuged at 2500 rpm for 10 min. Using the extractive alkylation technique, bis(pentafluorobenzyl) sulfide (BPFBS) was derived from sulfide.

The apparatus used to analyse BPFBS and TBB was a Shimadzu GC-14AE gas chromatograph equipped with a Ni^{63} electron capture detector, controlled with a Shimadzu C-R5A model Chromatopac computer. The column was a glass tube of 2.1 m × 3 mm I.D. packed with 5% Apiezon grease L on Uniport HP, 60–80 mesh. The column temperature was maintained at 220° C and the injection port and detector at 270° C. Nitrogen was used as the carrier gas at a flow rate of 30 ml/min. The lower detection limit for sulfide was 0.3 nmol/ml (or g).

Results and discussion

Based on the autopsy findings, the cause of death of the victim was diagnosed as suffocation after aspirating sewage into the respiratory tract. Since hydrogen sulfide gas was detected in the atmosphere at the scene, toxicological analysis focusing on sulfides was carried out. We did not determine the sulfide concentration in the gastric contents collected at autopsy, because from the case history it appeared that the sulfide concentration in gastric contents at the time of autopsy was not equatable to the concentration at the time of drowning.

Table 1 Sulfide concentrations in tissues in autopsied individuals (μmol/ml or μmol/g)

	Our case	Case reported by Kimura et al. [8]		
		A	B	C
Blood	0.95	0.02	0.01	N.D.
Lung	0.22	0.02	0.01	0.01
Brain	0.08	0.01	0.03	0.01
Liver	0.08	0.05	0.04	0.04
Kidney	0.38	0.03	0.05	0.01
Muscle				
abdominal	0.11	N.A.	N.A.	N.A.
femoral	0.14	N.A.	N.A.	N.A.

N.A.: Not analysed. N.D.: Below lower detection limit (< 0.3 nmol/ml or nmol/g)

As shown in Table 1, the concentrations of sulfides in the blood, lung and kidney in this case were extremely high when compared to the concentrations in cases where drowning has occurred following exposure to hydrogen sulfide gas. In the report by Kimura et al. [8], our extraction procedure was used for analysis, so we selected the report for the comparison of sulfide concentrations. We previously reported the postmortem generation of hydrogen sulfide in cadavers [12], but the possibility of increasing sulfide concentrations after death in this case was refuted on the basis of having handled the cadaver and samples according to the correct procedure.

We, therefore, considered the feasibility of oral intake of sewage containing sulfides, followed by distribution of sulfides from the alimentary tract throughout the whole body. To verify this hypothesis, sulfides were administered orally to rats and sulfide concentrations in tissues were determined.

The results are shown in Table 2, together with our previous data obtained from animal experiments relating to the exposure to hydrogen sulfide gas [12]. The sulfide concentration in the blood following oral intake was 0.32 μmol/ml, which was about 32 times higher than that following gas exposure. Sulfide concentrations in the other tissues were about the same, whatever the method of administration.

According to our experimental results, the remarkably high sulfide concentration in the victim's blood supposedly stemmed from the oral intake of sewage containing sulfides. Since the sulfide concentrations in the rat lungs following oral administration were similar to those following gas exposure, we consider the rather high sulfide concentration in the victim's lungs to have been due to aspiration of the sewage.

In this case, it seems that the high sulfide concentration in the blood derived from the alimentary tract rather than the respiratory tract, because the sulfide concentration in the blood was higher than that in the lungs. The reason

Table 2 Sulfide concentrations in rats following oral intake of sulfides and exposure to hydrogen sulfide gas. (µmol/ml or µmol/g, $n = 4$)

	Oral intake					Gas exposure
	No. 1	No. 2	No. 3	No. 4	Mean ± S.D.	Mean ± S.D.
Blood	0.23	0.68	0.13	0.22	0.32 ± 0.25	0.01 ± 0.00
Lung	0.01	0.01	0.01	0.02	0.01 ± 0.01	0.02 ± 0.01
Brain	0.02	0.04	0.05	0.01	0.03 ± 0.02	0.01 ± 0.00
Liver	0.04	0.04	0.09	0.17	0.08 ± 0.06	0.05 ± 0.01
Kidney	0.02	0.02	0.04	0.07	0.04 ± 0.04	0.04 ± 0.04
Muscle						
abdominal	0.00	0.03	0.04	0.06	0.03 ± 0.03	0.01 ± 0.01
femoral	0.00	0.00	0.00	0.01	0.01 ± 0.00	0.01 ± 0.00

Lower detection limit: 0.3 nmol/ml (or nmol/g)

why a high concentration of sulfides was detected in the victim's kidney needs to be further investigated, although it may have resulted from the fact that the kidneys serve as an excreting organ of sulfides.

From the findings of the autopsy and experimental results, it was considered that the victim had been exposed to hydrogen sulfide gas, and had then suffocated to death by aspiration of sewage containing sulfides and that the sulfides had become distributed mainly from the alimentary tract into the body followed by the cessation of circulation.

In conclusion, a case linked to ingested sulfides is reported. The results obtained from autopsy findings, toxicological analysis and animal experiments led to the following conclusion: the victim swallowed sewage containing sulfides into the alimentary tract and the sulfides were then distributed throughout the whole body until death occurred by drowning.

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