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Case Report / Приказ случаја

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Fatal consequences caused by prolonged chloroform inhalation in a child

Фатални исход изазван пролонгираним тровањем детета хлороформом

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SUMMARY

Introduction Chloroform intoxication as a result of inhalation is usually associated with occupational exposure. Fatal cases of accidental and intentional chloroform poisoning are extremely rare.

The aim of this work was to report a case with fatal consequences caused by prolonged chloroform inhalation in a child.

Case outline A three-month-old child found dead by its mother in their home. Investigation provided the data about two refrigerators in the house, which were not entirely functional, and were filled with a refrigerant based on chloroform, and about a recent house spraying with a pesticide also based on chloroform. Forensic autopsy was performed next day. Autopsy established the existence of malnutrition, dehydration, brain and lungs edema, the presence of very scarce contents in the digestive tract and the enlargement of the abdominal lymph nodes. An initial stage of liver degeneration was established on histopathological examination. Chemical-toxicological analysis of organ tissue samples (brain, lungs, spleen, liver with gall bladder, kidney and bladder, stomach, small and large intestine) performed using the techniques of headspace gas chromatography with mass detector, confirmed chloroform poisoning.

Conclusion The emaciation underlying the dysfunction of organ systems provided a grim frame for a child to succumb to chloroform toxicity. Low levels of chloroform inhaled from the room air may have fatal consequences in a susceptible individual.

Keywords: chloroform, intoxication, toxicology, autopsy, forensic pathology

САЖЕТАК

Увод Тровање услед удисања хлороформа је обично повезано са професионалним излагањем хлороформу. Смртни случајеви задесног и намерног тровања хлороформом су екстремно ретки.

Циљ овог рада је био да прикаже случај тровања детета хрлороформом са фаталним исходом.

Приказ случаја Тромесечно дете је мајка пронашла мртво у кући. Истрагом је утврђено да су у кући постојала два непотпуно исправна фрижидера са расхладним гасом на бази хлороформа и да је у кући у недавно коришћен пестицидни спреј на бази хлороформа. Судско-медицинска обдукција је извршена наредног дана и утврђено је постојање потхрањености, дехидратације, отока мозга и плућа, присуства веома оскудног садржаја у дигестивном тракту и увећања трбушних лимфних чворова. Хистопатолошким прегледом нађен је почетни степен дегенерација јетре. Хемијско-токсиколошка анализа узорака органа (мозак, плућа, слезина, јетра са жучном кесом, бубрег са бешиком, желудац, танко и дебело црево) применом технике гасне хроматографије са масеним детектором, потврдила је тровање хлороформом.

Закључак Исцрпљеност уз дисфункционалност система органа утицали су на околности под којима је дете подлегло токсичном ефекту хлороформа. Ниске концентрације хлороформа удахнутог из ваздуха у просторији могу бити фаталне код осетљивих појединаца.

Кључне речи: хлороформ, тровање, токсикологија, обдукција, форнезичка патологија

INTRODUCTION

Chloroform (trichloromethane – CHCl3) is a colorless, volatile liquid, whose potent anesthetic properties were recognized early after its synthesis during the 30thies of the 19th century [1]. However, soon after the first described applications of chloroform as anesthetic during surgical procedures in a clinical context, its acute toxicity was observed, and chloroform was established as a cause of death in a number of physically fit patients [2]. In spite of the fatal complications, chloroform continued to be used as a potent anesthetic agent over the next 50 years. Only in 1912, the Committee on Anesthesia of the American Medical Association proclaimed that the use of chloroform as the anesthetic for major surgery was no longer justifiable [3]. The use of chloroform as an anesthetic is abandoned because exposing the organism to high concentrations may cause hypotension and fatal cardiac arrhythmias [4, 5]. Fatal cardiac arrhythmias, as cause of death, may be also induced by inhalation abuse of toxic substances such as chloroform. From the sixties of last century, deaths caused by inhalation of toxic substances are termed “sudden sniffers death” [6, 7].

Nowadays, chloroform has a wide application in industry, and is a subject to strict regulations as a hazardous substance. The main use of chloroform is the production of fluorocarbons used in the synthesis of tetrafluoroethylene and polytetrafluoroethylene, as a refrigerant and propellant. It is also used as an organic solvent in industry and in analytical laboratories, as an ingredient of pharmaceuticals, drugs, cosmetics, grain fumigants, dyes and pesticides [4]. Before 1989 and The Montreal Protocol on substances that deplete the ozone layer, chloroform was also a popular refrigerant [8].

Acute chloroform poisoning, resulting in respiratory and central nervous system depression, has been described in accidental, suicidal and homicidal cases [9-11]. Intentional fatal chloroform intoxications usually concern self-poisoning acts, while homicides by chloroform are very rare. Accidental poisoning more frequently occurs in the setting of occupational exposure, but may also be encountered in domestic environment [11, 12]. Chronic exposure to inhalation of chloroform leads to liver damage, and causes central nervous system depression as well [3, 8].

The aim of this paper was to present a case of a child death by accidental prolonged chloroform poisoning at home, under unusual circumstances.

CASE REPORT

Circumstances of chronic exposure to chloroform inhalation

In the early morning, mother found her three-month-old child dead. She called the police, and a forensic autopsy was ordered. During the investigation, data about underprivileged, multi-member family (four adult persons and six children) who lived in a house with two rooms were obtained. There were two refrigerators in the house, which were not entirely functional, and were filled with a refrigerant based on chloroform. A short period before the relevant event, the house was sprayed with disinsection agent, a pesticide also based on chloroform. This information was obtained from investigative authorities but did not specify the name of the preparation used on that occasion. Child was found on the mattress on the floor, where he slept, in the room where these refrigerators were. All that was happening during the winter and because of the poorly ventilated rooms in this period, chloroform poisoning of the infant was suspected.

Autopsy findings

The autopsy was performed next day. The three months old male child was malnourished, with signs of dehidratation. The weight of the body was 3 kilograms, and the length was 49 centimetres, which according to the table of development of infants corresponds to the age of about a month. Cadaver had reduced muscle and bone mass, with almost complete absence of subcutaneous adipose tissue (Figure 1 and 2). On the skin of the lower part of the back, decubitus wounds were noted. No signs of injury were found on the body of the child. The examination revealed signs of brain and lungs edema. Inside the digestive tract only slight amounts of content were found. Abdominal lymph nodes were enlarged. The examination of bones revealed no fractures or any other remarkable changes. In

Figure 1. Cadaver aged three months with reduced mass of bones and muscles and almost complete absence of subcutaneous adipose tissue-external findings.

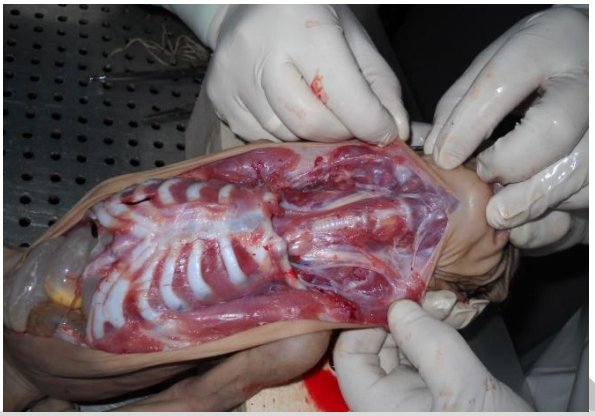

Figure 2. Cadaver aged three months with reduced mass of bones and muscles and almost complete absence of subcutaneous adipose tissue-internal findings.

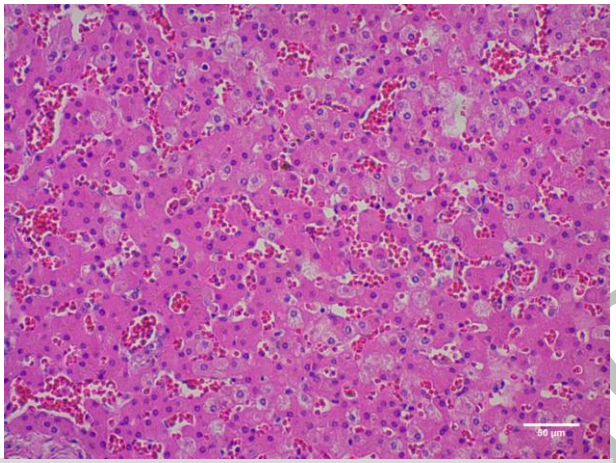

Figure 3. Cytoplasmic vacuolation and swelling of the hepatocytes. (H&E, ×20).

addition to pulmonary edema, the histopathological analysis demonstrated the hydropic liver degeneration. Cytoplasmic vacuolation, and swelling of the hepatocytes, with a mild architectural disarrangement of the plates was observed in the liver tissue (Figure 3). The rest of the morphological findings were unremarkable and classified as normal.

Chemical-toxicological analysis of internal organs samples (brain, lungs, spleen, liver with gall bladder, kidney and bladder, stomach, small and large intestine), using the techniques of headspace gas chromatography with mass detector, showed the presence of chloroform. We performed chemical-toxicological analysis using the techniques head space gas chromatography with flame ionization detection (HS-GC/FID) and head space gas chromatography mass spectrometry (HS-GC/MS).

The following chemicals were used in the process: chloroform and isopropanol, GC purity (J.T. Baker, Mallincrodt, Netherlands).

The basic standard chloroform solution was prepared at a concentration of 1 mg/mL in methanol. A series of dilutions for calibration curve, in the range of 1-50 mg/L, was prepared out of the basic solution. Isopropanol was used as the internal standard (IS) at a concentration of 0,50 mg/mL in water. The identification was done by comparing mass sample spectra with mass spectra databases Willy 7 and Pfleger Maurer Weber, and the quantification was done by internal standard method.

Conditions for gas chromatographic determination:

HS-GC/FID, Shimadzu QP 2010, Ultra, Autosampler AOC -5000.

GC column: InterCap 624 (30 m × 0.53 mm i.d., film thickness 3 μm). The temperature program starts at 40^0C, and lasts for 1min, and then the temperature was raised to 120^0C, at a rate of 50^0C/min. Injector 250 ^{0}C, split ratio 20. Detector FID 260 ^{0}C. Carrier gas He, 30 mL/min flow, Hydrogen, 40 mL/min flow, Air 400 mL/min flow.

HS-GC/MS, Shimadzu QP 2010, Ultra, Auto sampler AOC -5000.

GC column: DB-5 ms (30 m × 0.25 mm i. d., film thickness 0.25 μm), isocratic at 40^0C for 3 min, at a flow rate of 0.55 mL/min. Injector 200^0C, split ratio 30. Ion source 200^0C. Interface 200^0C. Scan range: m/z 28-100, scan rate 0.5 scan/sec. Incubation temperature 55^0C. Incubation time 900 s. Syringe temperature 90^0C. Agitator speed 300 rpm. Fill speed 500 μL/s. Injection speed 500 μL/s. Pre inject delay 500 ms. Post inject delay 500 ms. Flush time 10 s. GC runtime 180 s.

Chloroform concentrations detected in organ samples were: in the liver with gall bladder 17.35 mg/kg (Figure 4), in the stomach 10.29 mg/kg (Figure 4), in the small and large intestine 10.58 mg/kg (Figure 4), in the brain 27.64 mg/kg (Figure 5), in the lungs 27.64 mg/kg (Figure 5), and in the kidney and bladder 25 mg/kg (Figure 5). The results of this analysis led to the conclusion that chloroform poisoning was the cause of death.

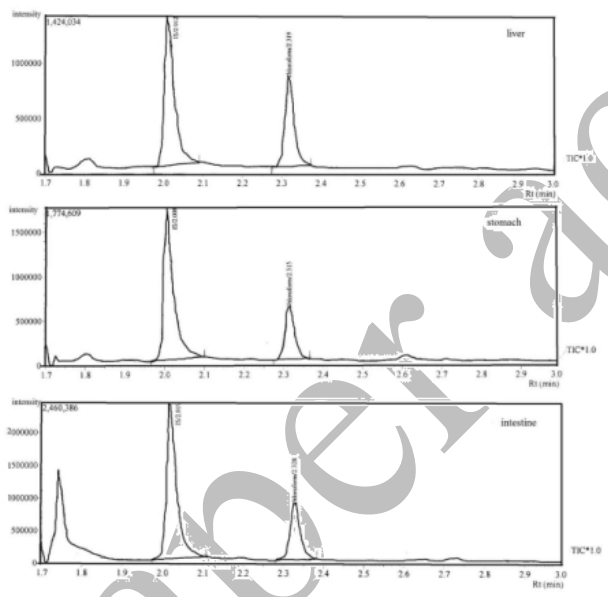

Figure 4. Chromatogram of chloroform concentration detected in liver with gall bladder, stomach, small and large intestine.

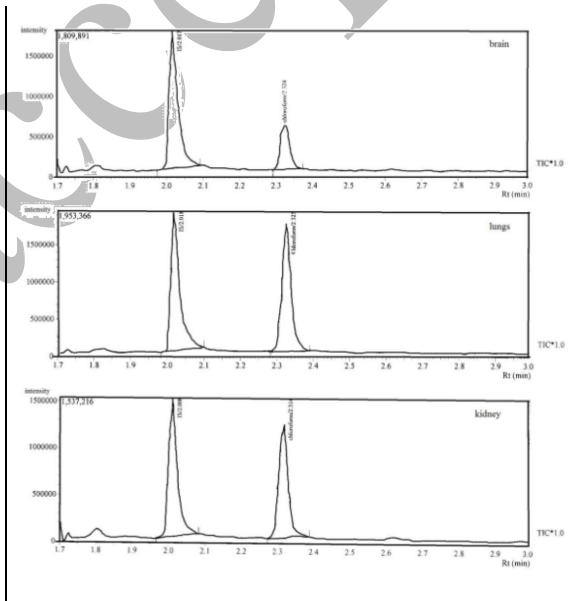

Figure 5. Chromatogram of chloroform concentration detected in brain, lungs, and kidney and bladder.

DISCUSSION

In the case of acute chloroform poisoning, sings as edematous swelling of the lips, focal mummifications of the facial skin and the chloroform-soaked soft covering of the airways could be very useful in determination of the cause of the death [12]. In chronic poisoning, there are no previous nor indicative signs.

In the case of exposure to poison in the gaseous state, blood levels peak already within a few minutes after exposure because of the extensive capillary surface area of the lungs. Because of their

lipid solubility, volatiles, including chloroform, quickly cross lipid membranes, and distribute to well-perfused organs such as brain, liver, heart, and kidneys. This condition will be retained if sudden death occurs, but if exposure is prolonged, toxic substance will slowly accumulate in poorly perfused parts also, such as muscle and fat tissue [13].

In fatal chloroform poisoning cases, its concentration ranges were 10.0–48.0 μg/ml in the blood, 50.4–156.0 μg/g in the brain, 16.0–27.0 μg/g in the kidney, 6.0–86.2 μg/g in the liver and 0–60.0 μg/ml in urine [14]. In the case reported herein, only the concentration of chloroform in the brain (27.64 mg/kg) was under the fatal level.

Non-fatal chronic inhalation of chloroform is usually associated with features of hepatic damage that develop 2–5 days after exposure [15-17]. These pathological changes are similar to those noted in the case of the prolonged anesthesia. Signs of hepatic injury have been reported after occupational exposure in the industry, up to workplace air chloroform concentrations up 205 ppm (1005 mg/m^3) [18]. Pathologic changes that may be observed in liver tissue probably reflect the major role of hepatocytes and cytochrome P-450 enzymes in chloroform metabolism. Depending on the concentration and quantity of oxygen, chloroform can be metabolized in the body by oxidative or reductive processes. The oxidative metabolism of chloroform, which is more commonly encountered, creates phosgene by CYP2E1 (high affinity–low capacity enzyme), while the reductive metabolism produces a dichloromethyl free radical. Both metabolic products of chloroform are highly cytotoxic and, due to their liposolubility, they rapidly penetrate into cells causing necrosis primarily of liver and kidney cells and tissue [5, 19-21]. A priori, it might be expected that the oxidative pathway of chloroform metabolism would predominate in vivo, because tissues of healthy individuals are oxygenated. Moreover, the toxic effect of chloroform includes manifestations as ventricular fibrillation, respiratory paralysis, and even multi-organ failure caused by chloroform-induced systemic inflammatory response syndrome [11, 22].

Fat liver degeneration that begins on the periphery and progresses to the centre of lobe, infiltration of lymphocytic lymphocytes and plasmocytes, and compression of liver sinus with initially expressed fibrosis represent the basic microscopic characteristics of liver changes in malnutrition [23]. In the present case, unlike the foregoing, as a result of prolonged chloroform intoxication the microscopic examination showed the hydropic liver degeneration with a mild architectural disarrangement of the plates. This suggests that liver changes were more likely to be caused by prologed chloroform poisoning. (See figure 3)

The circumstances recorded at the death scene suggest that the deceased infant might have been constantly exposed to chloroform inhalation in its home. The age of the infant, the cold in the winter months when death occurred, chloroform concentrations' in tissue samples, and the findings of skin decubitus on the lower part of back point to the conclusion that the child was staying for longer in the place where it was found dead. However, other family members, including elder children in presented case, showed no signs of intoxication and did not complain of any health problems. The fact is that

they went out of the house every day, which was the way of detoxification for them, thus their exposure was not constant. During the investigating procedure, the sample of the air from the family house had not been taken therefore we do not have the data about gas analysis and chloroform level in the air in the room where the child was found dead.

The hepatic damage as an effect of chloroform toxicity occurs more frequently in patients with predisposing factors, such as hypoxia, hypercapnia, dehydration, acidosis, and alcoholism [1, 3]. During the external and internal examination of the body of the child, signs of severe dehydration and malnutrition were registered, which were certainly a predisposing factor for faster appearance of toxic effects of chloroform. The emaciation underlying the dysfunction of organ systems provided a grim frame for a child to succumb to chloroform toxicity.

In the context of mental functions disorders that occur during chronic chloroform poisoning, a wide range and different degrees of disturbances have been described, from a lack of concentration to the most severe ones, such as stupor and coma [1, 15]. It is possible that the child showed some signs of passivity, but the parents failed to correctly recognize them.

In recent times, chloroform intoxication has been mostly limited to professional exposure, and it has been mainly related to the period prior to the adoption of regulations on ventilation of the working area. This accidental chronic chloroform poisoning implies that the use of products containing a certain even small amount of chloroform in a household can have the fatal consequences. The proper use and maintenance of domestic appliances, and careful application of chemical agents indoors is an imperative.

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