Reporting a sudden death due to accidental gasoline inhalation

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ABSTRACT

The investigation of uncertain fatalities requires accurate determination of the cause of death, with assessment of all factors that may have contributed to it. Gasoline is a complex and highly variable mixture of aliphatic and aromatic hydrocarbons that can lead to cardiac arrhythmias due to sensitization of the myocardium to catecholamines or acts as a simple asphyxiant if the vapors displace sufficient oxygen from the breathing atmosphere. This work describes a sudden occupational fatality involving gasoline. The importance of this petroleum distillate detection and its quantitative toxicological significance is discussed using a validated analytical method.

A 51 year-old Caucasian healthy man without significant medical history was supervising the repairs of the telephone lines in a manhole near to a gas station. He died suddenly after inhaling gasoline vapors from an accidental leak. Extensive blistering and peeling of skin were observed on the skin of the face, neck, anterior chest, upper and lower extremities, and back. The internal examination showed a strong odor of gasoline, specially detected in the respiratory tract. The toxicological screening and quantitation of gasoline was performed by means of gas chromatography with flame ionization detector and confirmation was performed using gas chromatography–mass spectrometry. Disposition of gasoline in different tissues was as follows: heart blood, 35.7 mg/L; urine, not detected; vitreous humor, 1.9 mg/L; liver, 194.7 mg/kg; lung, 147.6 mg/kg; and gastric content, 116.6 mg/L (2.7 mg total).

Based upon the toxicological data along with the autopsy findings, the cause of death was determined to be gasoline poisoning and the manner of death was accidental. We would like to alert on the importance of testing for gasoline, and in general for volatile hydrocarbons, in work-related sudden deaths involving inhalation of hydrocarbon vapors and/or exhaust fumes.

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1. Introduction

Gasoline is a complex and highly variable mixture of aliphatic and aromatic hydrocarbons with characteristic odor produced from the light distillates obtained during petroleum fractionation. Gasoline is 100% volatile by volume, giving off invisible vapors with a density five times that of the air. Either the liquid or the vapor may settle in low areas or travel some distance along the ground.

The major route of occupational exposure to gasoline is inhalation [1–7] and occupational fatal reports have included workmen cleaning storage tanks or people entering a petrol tank without a mask respirator [7–9]. Gasoline TLV-TWA (threshold limit value – time weighted average) and TLV-STEL (threshold limit value – short term exposure limit) are 300 ppm and 500 ppm, respectively, with an odor threshold of 0.25 ppm.

Occasionally inhaled gasoline can be a cause of sudden adult death. Death due to gasoline can occur through several mechanisms depending on the modalities of intoxication. In gasoline inhalation, death is generally attributed either to progressive medullar paralysis leading to respiratory failure, or to primary respiratory arrest due to the toxic action on the brain. On the other hand, it has been demonstrated that death can occur from arrhythmia due to cardiac sensitization to endogenous sympathetic amines sometimes precipitated by sudden alarm or exercise [10–12]. Besides, gasoline can act as a simple asphyxiant if the vapors displace sufficient oxygen from the breathing atmosphere [13]. In oral acute hydrocarbon toxicity, death results from aspiration of gasoline and sometimes of the gastric content.
followed by pneumonitis or a hemorrhagic pulmonary edema [9,14].

The investigation of uncertain fatalities requires thorough investigation into the circumstances and complete autopsies with correlation of anatomic findings to these circumstances, all of which are critical in coming to appropriate conclusions. This paper describes a sudden occupational fatality involving gasoline and demonstrates the problems that the forensic pathologists may encounter from the performing medico-legal autopsies in cases of sudden unexpected death. The importance of petroleum distillate detection and its quantitative toxicological significance is discussed using a validated analytical method published by the authors [9].

2. Case history

A 51 year-old Caucasian healthy male died suddenly while working on telephone lines inside a manhole. He worked as a contractor of the telephone company. The street temperature was 23.5°C. The manhole had two connected compartments: a cylindrical one at the entrance, 0.8 m diameter by 1.0 m in height, which was contiguous to a second compartment with right square prism shape 2.6 m long by 1.4 m wide and 2.0 m high. The whole capacity of the manhole was 92.9 kl (Fig. 1). A gasoline line which belonged to a nearby gas station was accidentally punctured and the man was exposed to gasoline vapors while in the manhole without a protection mask. No evaluation was undertaken of the manhole hydrocarbons levels.

Postmortem examination was performed less than 24 h later. The victim was 179 cm in height and weighed 93.0 kg. The rigor mortis was strong at the time of autopsy.

There was a strong smell of hydrocarbons emanating from the body. Extensive blistering and peeling of skin were observed on the skin of the face, neck, anterior chest, back and upper and lower extremities. All this areas lacked vital reaction and had a parchment-like aspect, due to the contact with hydrocarbons in a confined space (Fig. 2). Remainder of the external examination was unremarkable.

The internal examination showed a strong odor of gasoline, specially detected in the respiratory tract. The heart weighed 460 g and showed no fibrosis or evidence of myocardial infarction. The right and left anterior descending coronary arteries exhibited mild atherosclerosis. The ventricles, atria, valves, and endocardium appeared normal. The 540 g left lung and 725 g right lung had an appearance of antrachosis and a consistency thicker than normal. The brain weighed 1525 g and showed no anatomical abnormalities. Brain tissue revealed neither evidence of established ischemia or congestion nor was hemorrhages or contusions seen. There was a minimum effusion of light fluid in subarachnoid space and engorgement of veins. The stomach contained 23 ml of a yellowish liquid, and mucosa was eroded. Both the 70 g left kidney and 165 g right kidney had rough surface and dilatation of renal pelvis. Neither bone fractures nor visceral injuries were observed.

Microscopic examination disclosed red cells accumulations in interalveolar walls with haematomas infiltration in the alveoli. The liver showed minimum zone II steatosis. Scarce and disperse lymphocyte accumulations were seen compatible with thyroiditis. Rest of the urogenital, lymphoreticular, endocrine and musculo-skeletal systems were normal.

There was no known drug-abuse history or a present natural disease to account for the death. No injuries or underlying organic diseases were found that could have caused or contributed to death.

Heart blood, urine (30 ml), vitreous humor, liver, lung, and gastric content (23 ml of a yellowish liquid) were sent for analysis to our laboratory.

3. Toxicological examination

A full toxicological analysis was performed on the deceased heart blood and urine. Blood and urine was also examined for ethanol and other common volatiles (methanol, acetone, and isopropanol) using headspace with gas chromatography with flame ionization detector (GC-FID). The percentage of carboxyhemoglobin (%COHb) was also determined in blood using a spectrophotometric procedure previously published by Rodkey et al. [15]. Additionally, urine from the case was screened by immunoassay for propoxyphene, cocaine and benzoylcegonine, methadone, opiates, cannabinoids, benzodiazepines, amphetamine (and related compounds), barbiturates, and tricyclic antidepressants on a Hitachi 902 Automatic Analyzer (Tokyo, Japan) using Cedia® reagents (Microgenics, Fremont, CA). Then, the blood and urine samples were extracted with Bond-Elut Certify columns (Varian Sample Preparation Products, Harbor City, CA) collecting together the acidic–neutral and basic eluates. The sample extracts were analyzed by gas chromatography with nitrogen–phosphorus detector (GC-NPD) and by gas chromatography mass-selective detector (GC–MS) for screening/confirmation analysis following an analytical method described in our previous work [16]. Finally, all the autopsy samples were analyzed for petroleum distillates/hydrocarbons by means of gas chromatography with flame ionization detector (GC-FID) and confirmation was performed using gas chromatography–mass spectrometry (GC–MS) following our previously described method [9].

3.1. Quantitation of gasoline

3.1.1. Chemicals and reagents

Sodium sulphate, diethyl ether, and methanol of analytical grade were obtained from Scharlau (Barcelona, Spain). Unleaded gasoline neat standard was purchased from NSI Solutions, Inc. (Raleigh, NC), and n-octylbenzene (internal standard, IS) was purchased from Fluka–Sigma–Aldrich (Buch, Switzerland). The density of the gasoline standard, 0.75 g/mL, was used as a multiplication factor in order to convert microliters to milligrams. Stock solutions of gasoline (1 and 10 mg/mL) and IS (1 mg/mL) were prepared by dissolving the appropriate amount in methanol. These stock solutions were stored in glass tubes and maintained at −25°C until used to prepare blood calibration standards by spiking the appropriate amounts of gasoline to negative blood samples obtained from non-intoxicated individuals.

3.1.2. Sample preparation

Specimens collected at autopsy were preserved and frozen until analysis. Heart blood, urine, vitreous humor, liver, lung, and gastric content, blank blood, control, and calibrators, all of them refrigerated at 4°C, were processed according to the one step liquid–liquid extraction procedure described below. Liver and lung were homogenized using a mixer blended. Due to the strong smell to petroleum distillates/hydrocarbons that clung to these solid tissues and also to the abdominal contents, they were diluted with deionized water by a factor of 10 prior to the extraction in accordance with our experience with cases due to massive exposure of these toxic substances. Then diluted tissues were sonicated for 3 min and cooled at 4°C previously to their extraction.

A 3 mL aliquot of each liquid sample (or 3 g of each solid tissue) was transferred to a 10 mL screw-capped glass tube and added with 100 μL IS solution (n-octylbenzene methanolic solution of 100 mg/L), 1 mL of diethyl ether (cooled at 4°C) and 15 mg of anhydrous sodium sulphate, vortex mixed for 3 min, centrifuged at 4400 rpm for 15 min (at 4°C). Then the organic layer was collected and transferred to a gas vial and 3 μL were injected first for GC-FID
screening analysis and quantitation, and then for GC–MS for confirmation of the obtained results. Room temperature for all analytical procedures, including chromatographic analysis, must be maintained below 18 °C.

3.1.3. Instrumental analysis

GC-FID analysis was performed with an HP 6890 Series equipment provided with a 25-m (0.20-mm i.d., 0.11-µm film thickness) Ultra-1 HP cross-linked methylsilicone column (all from Agilent, Avondale, PA). Helium (Air Liquid, Madrid, Spain) carrier gas was delivered at a column head pressure of 22 psi, split ratio was 1:24, injector temperature was 280 °C, oven temperature began at 40 °C for 3 min, increased at 10 °C/min to 280 °C, and detector temperature was 300 °C. The detector gases were hydrogen and air (Air Liquid), delivered at a flow rates of 40 and 400 mL/min, respectively. Under these conditions, gasoline components including n-octylbenzene (IS) eluted between 1 and 15 min. They can be easily identified by comparing chromatography of the gasoline standard to that of the samples using this GD-FID screening method. Although no doubt was maintained about gasoline as source of poisoning, GC–MS analysis was performed with an Agilent 5975 C mass-selective detector from Agilent Technologies (Avondale, PA) for specific peak confirmation. The GC conditions were as mentioned above and the column was a 30-m (0.25 mm i.d., 0.25 µm film thickness) UI-HP-5 cross linked 5% phenyl–methyl silicone capillary column from Agilent Technologies (Avondale, PA). The mass spectrometer was operated in the total ion chromatogram (TIC) mode (m/z 35–650), electron impact (EI) ionization energy was 70 eV, and transfer line and ion-source temperatures were both maintained at 280 °C. Under these conditions extract ion chromatograms were obtained for ions: 91.00 (toluene), 106.00 (ethylbenzene and xylenes isomers), 120.00 (trimethylbenzene isomers), and 190.00 (n-octylbenzene, IS). An identification table of gasoline components by their GC retention time and mass spectra under the described chromatographic conditions was provided in our previously published work [9].

3.1.4. Calibration curve for gasoline

Quantitative analysis was undertaken by GD-FID using a four point blood calibration curve. The gasoline concentrations in the blood calibrators were 1, 25, 50, and 100 mg/L. The calibrators were prepared by adding gasoline to aliquots of human whole

![Fig. 1](image1.png)  
**Fig. 1.** (A) Plan view of the manhole with dimensions given in centimeters. (B) Manhole view from the street. (C) Manhole view with the corpse.

![Fig. 2](image2.png)  
**Fig. 2.** Extensive blistering and peeling of skin, with parchment-like aspect, due to the contact with hydrocarbons.
blood and n-octylbenzene as IS. The m,p-xylene peak, present in the gasoline blood calibrators, was used as the reference peak (marker) for all gasoline calculations. Calibration curves for gasoline were obtained by plotting the peak area ratios ($y$) of m,p xylene to n-octylbenzene as IS versus gasoline concentrations ($x$). Additionally, in the batch an extracted gasoline spiked blood sample (25 mg/L) and a gasoline standard in diethyl ether (75 mg/L), both prepared from a different stock solution of gasoline, were assayed as controls. $R^2$ value in the linear range was $>0.997$. The limits of detection and quantitation were 0.3 and 1.0 mg/L. Accuracy was 77.6–98.3%, and intraday ($n = 6$) and interday ($n = 10$) precisions had a CV $\leq 5.4\%$ between 1 and 100 mg/L.

The toxicological screening and quantitation of gasoline was performed by means of gas chromatography with flame ionization detector (GC-FID) and confirmation was performed using gas chromatography–mass spectrometry (GC–MS).

4. Results and discussion

We present a case of acute gasoline intoxication at the scene of working inside a manhole. The victim died immediately after the accidental inhalation of gasoline vapors.

Toxicological investigations and examination of the scene of death are critical to identify hydrocarbons intoxication as the cause of death, because anatomic autopsy findings will be typically

Fig. 3. Representative GC-FID chromatograms of the forensic case: (A) gasoline standard (75 mg/L), (B) blank blood, (C) gasoline spiked blood (25 mg/L), (D) heart blood obtained from the case, and (E) lung obtained from the case dilution (1:10). Peak identification: 1 = isooctane, 2 = toluene, 3 = ethylbenzene, 4 = m,p-xylene, 5 = o-xylene, 6 = 3-ethyltoluene, 7 = 1,2,4-trimethylbenzene, 8 = 1,2,3-trimethylbenzene, 9 = 1,2,4,5-tetramethylbenzene, 10 = naphtalene, 11 = 1-methylnaphtalene, 12 = n-octylbenzene (IS), 13 = ionol (diethyl ether stabilizer), and 14 = fatty acids.
nonspecific. Investigations should involve the search of gasoline components using highly sensitive and specific analytical methods, being the evaporation of the substance from postmortem samples one of the main difficulties. Besides, as illegitimate exposure to solvent vapor is a possibility, it is necessary to take a broad approach to the problems posed by volatile substance abuse and other situations where poisoning by volatiles may be encountered. The proposed analytical method allows a comprehensive toxicological screening for solvents and other petroleum distillates. The screening method is based in the pattern recognition method widely applied in many areas of forensic science [9,17].

Fig. 3 shows representative GC-FID chromatograms of gasoline standard, blank blood, gasoline spiked blood, and heart blood and lung obtained from the forensic case. Although no doubt was maintained regarding the identity of the source of poisoning after analyzing the samples on the GC-FID, additionally we re-analyze the heart blood sample on the GC–MS for specific peak confirmation. Fig. 4 shows a zoom of the TIC and mass ions obtained from the heart blood sample under the described chromatographic conditions. Selected ions for target compounds and internal standard were detected by constructing chromatograms of molecular ions/principal fragment ions as described previously [9].

Toxicological findings are showed in Table 1. Regarding alcohol and other volatiles, abuse and therapeutic drugs screens the results were negative for all of them. The detected %COHb was normal in healthy people.

We found 35.7 mg/L of gasoline in heart blood. Quantitative analysis was undertaken by GD-FID using blood calibration curves for gasoline in the range of 1–100 mg/L and using the m,p-xylene peak, present in the gasoline calibrators, as the reference peak for all gasoline calculations. The gasoline concentrations obtained in the present case are comparable with other report of fatal inhalation described previously by us, in which the same analytical method was applied [9]. A drawback for comparison of gasoline

Fig. 4. Zoom of the total ion chromatogram (TIC) (A) and mass ions (B) obtained from the case heart blood sample by GC–MS. Peak identification: 1 = toluene (ion 91.00), 2 = ethylbenzene and xylene isomers (ion 106.00), 3 = trimethylbenzene isomers (ion 120.00), and 4 = n-octylbenzene (IS) (ion 190.00).
evaluation of manhole gasoline concentrations in our case was not undertaken.

Sudden death due to the inhalation of halogenated hydrocarbons is a well-documented phenomenon in the scientific and medical literature. There is also information suggesting that nonhalogenated hydrocarbons may cause potentially fatal cardiac arrhythmias [26,27] and cardiac arrest is described as a consequence of an armoured vehicle rollover accident where petroleum fuel spilt into the ditch in which a vehicle came to lie [28].

The proposed mechanism of death in our case is myocardial sensitization to endogenous catecholamines and this is supported by the absence of any other significantly life threatening injury and the absence of any evidence of other mechanisms such as complications of vomiting aspiration, coma or convulsions. The death can be partly due to oxygen deprivation caused by poor ventilation in the confined space. Resuscitation from sudden death due to cardiac arrhythmia associated may not be successful, as the death is often unnoticed. In this case, lack of an adequate ventilation system tragedized the situation.

Inhaled compounds of gasoline may rapidly attain high concentrations in vital, well-perfused organs such as the brain and heart. Should death occur, this situation is “frozen”, but if exposure continues, the compound will slowly accumulate in less accessible (poorly perfused) tissues. This would explain why high gasoline concentrations were demonstrated in the lungs and liver in our case. In accordance with Carnevale, urine was negative because metabolites presented in this fluid need another extraction method [14]. The low concentration obtained for gasoline in vitreous humor also agrees with our previous experience [9] indicating that lipophytic substances do not concentrate in this biological fluid.

5. Conclusions

We have reported a case of sudden death due to gasoline inhalation in a confined space. Although the job was not directly related to chemicals management, alert must be made about occupational conditions in maintenance works especially when they are taken near gas stations or big tanks of volatile hydrocarbons.

Autopsy was not specific as death occurred too sudden. The finding of burns, in light of the history of sudden death in the manhole and the odor at the scene were highly suggestive of gasoline exposure. Based upon the toxicological data along with the autopsy, the cause of death was determined to be gasoline poisoning and the manner of death was accidental. We would like to alert on the importance of testing for gasoline, and in general for volatile hydrocarbons, in work-related sudden deaths involving inhalation of hydrocarbon vapors and/or exhaust fumes.

Conclusions regarding sudden unexpected death must take into account multiple factors that may have played a role, as well as anticipate the forensic issues that will arise. In addition, the present work add a collection of important comparable data to the scarce literature regarding toxic gasoline concentrations.

References