

Accuracy Profile Validation of a New Analytical Method for Propane Measurement Using Headspace-Gas Chromatography–Mass Spectrometry

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Propane can be responsible for several types of lethal intoxication and explosions. Quantifying it would be very helpful to determine in some cases the cause of death. Some gas chromatography–mass spectrometry (GC–MS) methods of propane measurements do already exist. The main drawback of these GC–MS methods described in the literature is the absence of a specific propane internal standard necessary for accurate quantitative analysis. The main outcome of the following study was to provide an innovative Headspace-GC–MS method (HS-GC–MS) applicable to the routine determination of propane concentration in forensic toxicology laboratories. To date, no stable isotope of propane is commercially available. The development of an *in situ* generation of standards is thus presented. An internal-labeled standard gas (C_3D_7) is generated *in situ* by the stoichiometric formation of propane by the reaction of deuterated water (D_2O) with Grignard reagent propylmagnesium chloride (C_3H_7MgCl). The method aims to use this internal standard to quantify propane concentrations and, therefore, to obtain precise measurements. Consequently, a complete validation with an accuracy profile according to two different guidelines, the French Society of Pharmaceutical Sciences and Techniques (SFSTP) and the Gesellschaft für toxikologische und Forensische Chemie (GTFCh), is presented.

Introduction

Propane is an odorless, colorless and flammable gas. Present in natural gas (3–18 vol%), it is an important fuel source and aerosol propellant (1). Up to levels of 1,000 ppm in the inhaled air, propane is not considered as a dangerous substance, even though at higher levels it may have some narcotic and asphyxiating properties (2–5). Above this threshold, propane becomes toxic and has been the cause of death in several types of domestic and industrial accidents.

Death usually occurs because of the asphyxiation of the tissues by oxygen depletion. Effectively at high concentrations, propane can substitute for air which is no longer available to oxygenate the organism (5). In general, it is difficult to assess the exact cause of death related to poor knowledge of the precise inhalation of air without O_2 . Two types of death due to propane are usually reported: accidents and suicides (6–11). Drug inhalation (11–22), domestic incidents (23–25) and industrial disasters (26–29) are the usual types of reported accidents. Propane is issued from natural gas purification, and can easily be found commercially, as it is present in aerosols, gas lighters, and is a well-known energy source for boilers, barbecues and similar appliances. Inhaling propane will first cause hallucinations, a loss of inhibition and an impaired judgement. These are the main reasons why propane is commonly used as a volatile drug; this practice is called huffing: it consists in inhaling fumes from common household products to get ‘high’ (17, 30–33). It is

especially known for drug abuse by teenagers as it is cheap and easy of access. Moreover, if a significant quantity is inhaled, drowsiness, narcosis, asphyxia, frostbite, brain damage and even cardiac arrhythmia will occur. As said earlier, propane is also frequently used in suicides. Victims are usually found with a bag over their head, which will induce a rapid suffocation and asphyxia leading to a cardiac arrest. Usually a few seconds before death, a rapid cardiac acceleration is observed, as oxygen of the body is excreted into the atmosphere with very strong movements when respiration is forced to perform in oxygen-depleted circumstances (5).

When propane is suspected as being a cause of death, blood samples, as well as lung, liver, brain, fat tissue and heart samples, should be analyzed.

At the present time, analytical measurements of propane have already been performed using gas chromatography (GC) (34). It was either coupled to a flame ionization detector (13, 24, 27, 28), or to a mass spectrometer (MS) (9, 14, 22, 35). Gas liquid chromatography was also used (36). These procedures are satisfying to prove the presence of propane, and to have an approximate concentration, but with an internal standard even more precise results could be obtained. Difficulties due to gas losses during sampling would be avoided. One of the reported techniques used a liquid solution of 1,1,2 trichlorotrifluoroethane in *t*-butyl methylether as an internal standard (35), and another one used pentane as an internal standard (24). These internal standards are not sufficiently specific to propane. Therefore, the most suitable internal standard would be deuterated propane. Previous studies on methane, deuterated methane, butane and deuterated butane generated *in situ* have already been carried out (37, 38). To produce methane, a possible reaction would be the reaction between the Grignard reagent methylmagnesium chloride and water. Subsequently, deuterated methane was produced from methylmagnesium chloride and deuterated water. Similarly, deuterated propane may be produced by the reaction of propylmagnesium chloride and deuterated water.

The following study aims to present an innovative HS-GC–MS method of quantification that could be applied to routine determination of propane present in biological matrices. First, the analytical protocol is fully described and validated according to two guidelines (SFSTP and GTFCh), and then secondly, the method was applied to the measurements of propane concentration present in autopsied cadavers, following death from intoxication and explosions.

Experimental

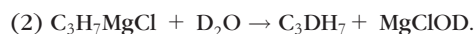
Materials and reagents

Propylmagnesium chloride (C_3H_7MgCl) 2.0 M diethyl ether was purchased from Sigma-Aldrich (Saint-Louis, USA). Deuterated

water was obtained from Cambridge Isotope Laboratories (CIL), Inc. (Andover, USA). All headspace (HS) extractions were carried out in 20 mL of HS vials. A certified butane C106 cylinder from Camping gaz (Givisiez, Switzerland) was used to perform an external control. The technical data sheet of C106 butane cylinder indicates a certified propane concentration of ~20%.

Extraction method

Propane (C_3H_8) and deuterated propane (C_3DH_7) were generated separately in two different 20 mL HS vials. The reactions of the Grignard reagent with water and deuterated water are given below:



Due to a really high reactivity of these reactions, it is important to proceed quickly (propylmagnesium chloride reacts with water coming from the ambient air) and safely (under a fume hood). Grignard reagent and water are added without any contact in an aluminum cap, which has no septa and no hole previously introduced in a HS vial. Then, the vial must be rapidly and hermetically closed, before being vortexed to allow the reaction of propane generation at room temperature. Precise volumes of gas (C_3H_8 and C_3DH_7) were sampled (automatically or manually) by a HS gas syringe through the vial septum and directly introduced in the GC injector.

GC-MS analysis

To perform the GC separation of the gaseous samples, an Agilent 6890N Gas Chromatograph (Agilent Technologies, Palo Alto, CA, USA) combined with a HS gas autosampler and equipped with an Agilent Select Permanent Gases column was used. This column is specially designed for gas analysis. It is made of two capillary columns set in parallel: a molecular sieve 5 Å PLOT capillary column (10 m × 0.32 mm) and a Porabond Q (50 m × 0.53 mm). The temperature program was set as follows: 100°C, held for 2 min and raised at 10°C/min to 250°C; the injector (splitless mode) set to 100°C and the interface MS temperature to 230°C. Helium was used as the carrier gas. The detection was performed with an Agilent 5973 mass spectrometer (Agilent Technologies), operating in the electron ionization mode at 70 eV. The selected ion monitoring mode was used to acquire the C_3H_8 and the C_3DH_7 signals at m/z 44 and 45, respectively (Figure 1).

Calibration standards and controls

Six working calibration standards with concentrations corresponding to 6.30, 12.5, 18.8, 25.0, 37.5 and 50.0 nmol of propane/mL of HS vial were prepared daily by reacting propylmagnesium chloride with water.

Intermediate quality control samples were also prepared daily from the same reaction at the same concentrations. The internal standard was prepared by the reaction of propylmagnesium chloride with deuterated water. Concerning its sampling, 250 µL of the 0.1 µmol/mL of HS vial working internal standard was sampled in a HS gas syringe, which means that 25.0 nmol deuterated propane were introduced each time and used to proceed to the calibration.

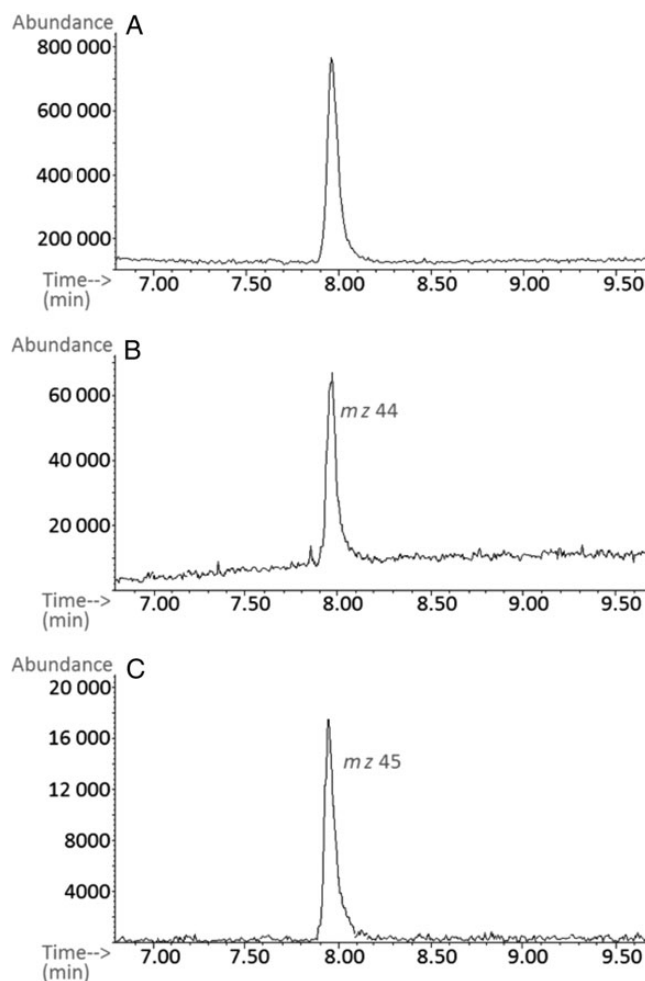


Figure 1. (A) Total ion current (TIC) chromatogram of 25.0 nmol propane with 25 nmol deuterated propane, and the gray peak corresponds to propane. Extracted ion chromatograms of propane ($m/z = 44$) (B) and deuterated propane ($m/z = 45$) (C) obtained from the TIC chromatogram.

Regarding the gas sampling protocol, once the sampling of the internal standard in a HS gas syringe was completed, and a sampling of calibrators (or the real sample) was performed using the same syringe. Hence, all different gases were mixed together in the HS gas syringe and the total gas volume was then injected in the GC injector. While not in use, propylmagnesium chloride was stored at +4°C and deuterated water was stored at room temperature.

Validation procedure

The validation procedure was performed using two different protocols. The first one followed the guidelines of the 'French Society of Pharmaceutical Sciences and Techniques' (SFSTP) (39). The second protocol used the guidelines of the 'Gesellschaft für Toxikologische und Forensische Chemie' (GTfCh) (40). Because of this double validation, obtained results of both calibrations may be compared confirming the accuracy of the method. Both are based on the following criteria: selectivity, response function (calibration curve), linearity, trueness, precision (repeatability and

intermediate precision), accuracy, limit of detection (LOD) and limit of quantification (LOQ).

In each validation procedure, the points of the calibration curve were defined as being the area ratio of propane to

deuterated propane, but frequency and number of repetitions of measurements differ.

By following the SFSTP procedure, three calibration curves of propane concentrations were completed in triplicate ($n = 3$) over 3 nonconsecutive days ($P = 3$), during a period of 2 weeks. The calibration curves were done with calibration standards at six different concentration levels ($k = 6$): 6.30, 12.5, 18.8, 25.0, 37.5 and 50 nmol/mL of HS vial. The lowest coefficient of determination of the three curves was equal to 0.9912 (Table I). Control samples were used to validate the method, and they were measured at the same six concentration levels ($k = 6$) in triplicate ($n = 3$) for each curve.

According to the guidelines of the GTFCh, one calibration curve of propane concentrations, repeated nine times over 3 nonconsecutive days (3×3), was done with calibration standards at the same six concentration levels. The determination coefficient of the curve was equal to 0.996 (Table II). In addition, controls samples were measured at the same six concentration levels ($k = 6$). Control samples were realized on nonconsecutive days eight times ($P = 8$) in duplicate ($n = 2$) for each concentration level.

The trueness of the method was therefore assessed by these control repetitions and by an external control (certified gas cylinder containing ca. 20% of propane) for both approaches.

Table I

Parameters for validating propane measurements according to the SFSTP procedure

| Calibration curve (6.30–50 nmol/mL of HS vial) ($k = 6, n = 3, P = 3$) | | | |
|--|---------------|------------------------|---------|
| | Day 1 | Day 2 | Day 3 |
| Slope | 49.01 | 47.44 | 48.38 |
| Intercept | 1.41 | 1.50 | 1.52 |
| r^2 | 0.99759 | 0.99752 | 0.99124 |
| Linearity (6.30–50 nmol/mL of HS vial) ($k = 6, n = 3, P = 3$) | | | |
| Slope | 0.9846 | | |
| Intercept | 0.0005 | | |
| r^2 | 0.9958 | | |
| Trueness (relative bias %) ($k = 6, n = 3, P = 3$) | | | |
| Levels (nmol/mL HS) | Trueness (%) | | |
| 6.30 | -8.6 | | |
| 12.5 | -0.1 | | |
| 18.8 | 6.2 | | |
| 25.0 | 3.1 | | |
| 37.5 | 2.8 | | |
| 50.0 | -2.9 | | |
| Precision (RSD %) ($k = 6, n = 3, P = 3$) | | | |
| Levels (nmol/mL HS) | Repeatability | Intermediate precision | |
| 6.30 | 5.50 | 12.7 | |
| 12.5 | 4.50 | 4.50 | |
| 18.8 | 1.80 | 4.40 | |
| 25.0 | 4.30 | 5.80 | |
| 37.5 | 1.60 | 2.20 | |
| 50.0 | 1.60 | 2.20 | |

Table II

Parameters for validating propane measurements according to the GTFCh procedure

| Calibration curve (6.30–50 nmol/mL of HS vial) ($k = 6, n = 2, P = 8$) | | |
|--|---------------|------------------------|
| All 5 days together | | |
| Slope | 48.27 | |
| Intercept | 1.48 | |
| r^2 | 0.99272 | |
| Linearity (6.30–50 nmol/mL of HS vial) ($k = 6, n = 2, P = 8$) | | |
| Slope | 0.9753 | |
| Intercept | 0.0007 | |
| r^2 | 0.9960 | |
| Trueness (relative bias %) ($k = 6, n = 2, P = 8$) | | |
| Levels (nmol/mL HS) | Trueness (%) | |
| 6.30 | -6.5 | |
| 12.5 | 0.7 | |
| 18.8 | 5.2 | |
| 25.0 | 2.4 | |
| 37.5 | 2.3 | |
| 50.0 | -3.5 | |
| Precision (RSD %) ($k = 6, n = 2, P = 8$) | | |
| Levels (nmol/mL HS) | Repeatability | Intermediate precision |
| 6.30 | 8.30 | 9.00 |
| 12.5 | 3.90 | 7.50 |
| 18.8 | 3.20 | 8.10 |
| 25.0 | 3.50 | 7.50 |
| 37.5 | 1.70 | 3.00 |
| 50.0 | 1.70 | 1.90 |

Results and discussion

Selectivity of the method

More than thirty biological samples such as blood, kidney, lung, liver, heart, urine and fat tissue from autopsied cadavers were analyzed to check that no co-eluting chromatographic peaks would interfere with the detection of propane and deuterated propane. As no interference peak was observed at propane retention time and m/z 44, it indicates that the method is selective enough to quantitatively determine the amount of propane. An assessment of propane quantity generated during deuterated propane production was assessed and found very weak, repeatable, reproducible and taken into consideration in the validations. Deuterated propane contribution during propane generation was found negligible.

Calibration curve for the method

To check the validity of the method with both protocols, a linear relationship was established between propane concentrations and the measured response in the calibration range. The calibration range was deliberately selected between 6.30 and 50.0 nmol/mL of HS vial, as it is a suitable range from a forensic point of view (Table III).

Then, calculated concentrations of each calibrator were compared with target values and were found to be within $\pm 21\%$ when following the SFSTP protocol, and within $\pm 22\%$ when following the GTFCh protocol. All results of the calibration curves for this validation procedure are compiled in Tables I and II.

Linearity of the method

The linearity was assessed by fitting back-calculated concentrations of the control samples versus their theoretical concentrations. First, in order to respect the SFSTP guidelines, control

Table III

Concentrations of propane from various samples of lethal cases in which propane was part of a gaseous mixture or alone ($\mu\text{g/g}$)

| Propane concentrations ($\mu\text{g/g}$) | | | | | | Administration | Reference |
|---|-------|-------|------|--------|------------|----------------------------|-----------|
| Blood | Brain | Liver | Lung | Kidney | Fat tissue | | |
| Cases where propane was identified as one of the compounds present in the gaseous mixture | | | | | | | |
| 0.27 | 11.0 | | | | 13.0 | LPG inhalation + explosion | (9) |
| 1.90 | 7.10 | | | | 1.00 | LPG inhalation + explosion | (9) |
| 1.20 | 1.10 | 3.90 | 1.50 | | | Inhalation | (10) |
| 31.2 | 5.18 | 33.2 | 35.5 | | | Inhalation | (10) |
| 30.0 | 89.0 | | | | | Inhalation | (11) |
| 0.07 | 0.14 | 0.28 | 0.09 | 0.07 | | Inhalation | (12) |
| 20.9 | 18.5 | 105 | 14.9 | 25.9 | | Inhalation | (21) |
| 3.90 | | 1.36 | 1.05 | 0.45 | | Propane/butane inhalation | (22) |
| 1.17 | | 0.87 | 0.45 | 0.93 | | Propane/butane inhalation | (22) |
| 36.7 | 1.60 | 9.20 | 0.20 | 1.10 | | Inhalation + explosion | (26) |
| 0.20 | 1.00 | 0.30 | 0.50 | 0.20 | | Inhalation + explosion | (26) |
| 0.07 | 0.15 | 0.16 | | | 1.10 | LPG inhalation | (33) |
| Cases where propane was identified as the unique compound in the gaseous mixture | | | | | | | |
| 69.4 | 130 | 94.8 | | | 75.0 | Inhalation | (10) |
| 10.2 | 43.5 | 70.7 | 4.15 | 5.85 | 68.3 | Inhalation | (12) |

samples were measured at six concentration levels ($k = 6$) in triplicate ($n = 3$). Control sample concentrations were calculated using the calibration curve made for each measurement day. In the range of 6.30–50 nmol/mL of HS vial, a good linearity was obtained (Table I), with a slope of 0.9846, and the coefficient of determination was equal to 0.9958. Following the GTFCh guidelines, control samples were measured at six concentration levels ($k = 6$) eight times in duplicate ($n = 2$) over 8 days. Similarly, control sample concentrations were calculated using the calibration curve made for each measurement day. In the range of 6.30–50 nmol/mL of HS vial, good linearity was obtained (Table II), the slope equaled 0.9753 and the coefficient of determination was equal to 0.996.

Trueness of the method

Also called the bias, the trueness test expresses how close experimental average values and accepted reference values are. This test detects systematic errors and is expressed as a percent deviation from the accepted reference value. Daily repetitions of control samples were analyzed over several weeks at their respective concentrations. The obtained results were used to establish a true value at each concentration. An additional trueness evaluation was performed using an external quality control made with the same procedure as for control samples. As exposed in Table I, according to the SFSTP guidelines, the trueness was found to be within the acceptance criteria [$\pm 15\%$ of the accepted reference value and within 20% at lower limit of quantification (LLOQ), 8.0 nmol of propane/mL of HS vial]. Similarly for the GTFCh protocol, the trueness was within the acceptance criteria. Thus, the method is reliable to quantify propane. The evaluation of trueness involving the external quality control was performed with a certified gas cylinder. Propane is present in the gaseous mixture at $20 \pm 5\%$, whose density is comprised between 0.5 and 0.595 g/cm³. After several dilutions, and by taking into account the allowance, the expected values should be between 13.3 and 26.6 nmol/mL of HS vial. Four repetitions were made over 2 different days. The obtained average values for both days were found in the expected range.

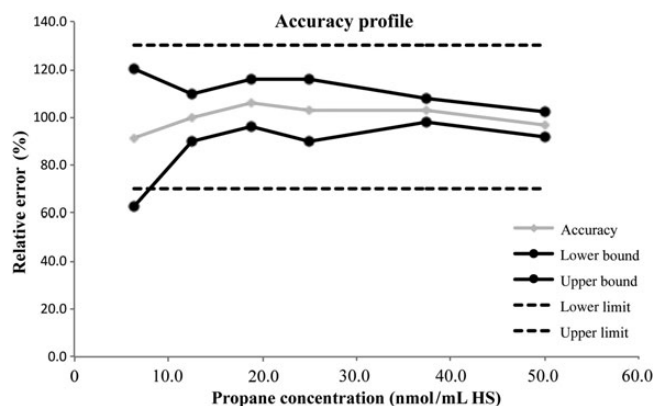


Figure 2. Accuracy profile of propane gas according to the SFSTP protocol using a simple linear regression model within a range of 6.30–50 nmol/mL of HS vial (continuous light gray line: trueness, black dotted lines: acceptance limits set at $\pm 30\%$, black lines: lower and upper accuracy limits in relative values).

Precision (repeatability and intermediate precision) of the method

Precision was assessed by calculating the repeatability (intraday precision) and intermediate precision (interday precision) for each control sample concentration. The repeatability variance was estimated by calculating the intraday variance (S_r^2), and the intermediate precision variance was estimated by adding the intra- and interday variances (S_{IP}^2). As summarized in Table I, the relative standard deviation values for repeatability and intermediate precision according to the SFSTP were between 0.10 and 8.60%, and 1.60 and 12.7%, respectively. As reported in Table II, the relative standard deviation values for repeatability and intermediate precision according to the GTFCh were between 0.70 and 6.50%, and 1.70 and 9.00%, respectively.

Accuracy and LOQ of the method

The accuracy expresses the total error defined by the sum of trueness (systematic error) and precision (random error). Both accuracy profiles are given in Figures 2 and 3, and they both prove the ability of the method to provide analytical results

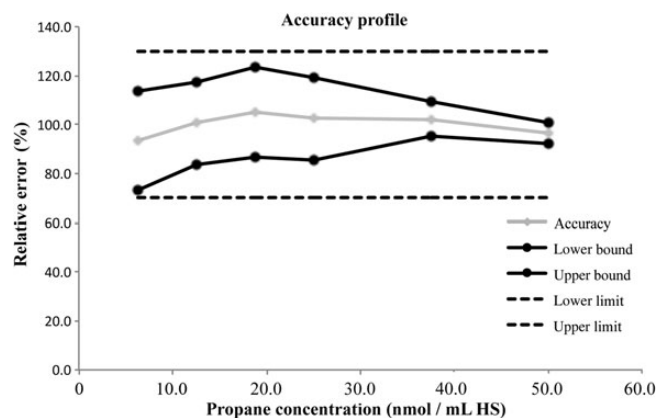


Figure 3. Accuracy profile of propane gas according to the GTFCh protocol using a simple linear regression model within a range of 6.30–50 nmol/mL of HS vial (continuous light gray line: trueness, black dotted lines: acceptance limits set at $\pm 30\%$, black gray lines: lower and upper accuracy limits in relative values).

using systematic and random errors with a risk of $\alpha = 5\%$ at each concentration level. The mean bias (%) confidence interval limits for the control samples were within the $\pm 30\%$ acceptability limits typically allowed by Swiss forensic laboratories.

With a threshold of 30% as the acceptability limit, an LLOQ of propane was assessed to be at 8.0 nmol/mL of HS vial according to the SFSTP guidelines. With the GTFCh protocol, the LLOQ was not reached, but seemed really close to 8.0 nmol/mL of HS vial. This difference is due to the variable parameters of validation of both procedures.

LOD of the method

To determine the LOD of propane by this new approach, HS extractions of blank samples containing water and propylmagnesium were done. After several consecutive dilutions of a propane HS vial having a concentration of 2.50 $\mu\text{mol/mL}$ of HS vial, the LOD was assessed using a signal-to-noise ratio (S/N) of >3 . Estimating the noise by measuring >10 blank samples, the LOD of propane quantification was evaluated at 4.0 nmol/mL of HS vial.

Application to real cases

The boiling point of propane is approximately -42°C , which means that propane will be easily extracted above this temperature, whatever the sample is (blood or tissue) and the state of the sample (solid, liquid, putrefied). What remains critical during the handling part is the potential loss of propane during sampling. Even though the matrix is not a crucial parameter, a minimum amount is necessary to obtain a propane signal above the LOD. Propane concentrations are initially expressed in $\mu\text{mol/mL}$ of HS, but it could easily be expressed in $\mu\text{g/g}$ by applying the following formula:

$$\text{Concentration } (\mu\text{mol/mL}) \times M(\text{g/mol}) \times V(\text{mL})/m(\text{g}) = \text{Concentration } (\mu\text{g/g}),$$

where M is the molar mass; V , the headspace volume and m , the mass sample.

Table IV

Obtained concentrations of propane from two autopsied cases of our legal medicine center ($\mu\text{g/g}$)

| Propane concentrations ($\mu\text{g/g}$) | | | | | | | Administration |
|--|-------|-------|------|--------|------------|-------|--------------------------------|
| Blood | Brain | Liver | Lung | Kidney | Fat tissue | Heart | |
| 40 | | | | | | | Man (car explosion + gas tank) |
| 130 | Nd | 10 | 100 | 150 | Nd | 80 | Worker (gas leak + explosion) |

Nd: not detected.

To assess the propane exposure that a victim has been subjected to, obtained concentration results from the different samples must be combined. Propane is a lipophilic gas, which means that, in the case of a long exposure to propane and a death by anoxia, a high concentration in fat tissues, brain and kidney should be detected. On the contrary, after a short exposure to propane subsequent to a propane outburst, it is more likely to find higher concentrations in lungs than in fat tissue or brain. Furthermore, a postmortem distribution due to the volatility of propane may exist, enhancing some variations.

The evaluation of the role played by propane in lethal intoxication depends on several parameters: state of health of the victim, the exposure time-period, the circumstances of the exposure (gas outburst, anoxia, sniffing bags. . .), if reanimation on the deceased was attempted or not. The propane concentration will therefore differ from one organ to another, so the matrix has its importance in results interpretation. Moreover, propane is usually present in a gas mixture (such as LPG), so it will not necessarily be the only cause of death, but will contribute to it. It seems difficult to assess norms above which propane becomes fatal.

The main lethal cases concerned by the contribution of propane to death available in the literature were listed in Table III. All propane measurements were expressed in $\mu\text{g/g}$ of the sample. Consequently, propane concentrations (alone as well as in gas mixtures) range from 0.07 to 69.4 $\mu\text{g/g}$ in blood ($n = 14$), from 0.14 to 130 $\mu\text{g/g}$ in the brain ($n = 12$), from 0.16 to 105 $\mu\text{g/g}$ in the liver ($n = 11$), from 0.09 to 35.5 $\mu\text{g/g}$ in the lungs ($n = 9$), from 0.07 to 25.9 $\mu\text{g/g}$ in the kidney ($n = 7$) and from 1.00 to 75.0 $\mu\text{g/g}$ in the fat tissue ($n = 5$). From these results, it seems clear that, in cases of solely propane inhalation, the minimum observed concentrations were higher whatever the matrix than in gas mixtures. Our method of quantification allows very precise measurements of propane at concentration starting at 8.0 nmol/mL, which equals 7.04 $\mu\text{g/g}$. As shown by two examples in Table III, when propane is considered as responsible of the death, concentrations are much greater than the limit of quantitative determination.

As shown by results compiled in Table IV, the method was applied to real cases and has given relevant results. The first victim has suffered from a short-time exposure before the explosion, not many propane has been metabolized, whereas the second victim has suffer from a long-time exposure before the explosion leading to a high metabolization of propane. In this case, the obtained concentrations in the blood, lung, heart and kidney of the second victim were high enough to constitute a potential cause of death, even though in that case the victim had suffered intoxication from a mixture of several gases: propane, carbon monoxide and cyanide.

Conclusion

The method described here is a new approach to the quantification of propane that appears to be very selective and sensitive. Thus, it allows propane concentration measurements in post-mortem samples even though these samples may be of poor quality, which is common in postmortem cases. The idea of generating a stable labeled isotope from a Grignard reagent has already been used in other alkane quantification such as methane and butane, but was never applied to propane. The procedure was first validated according to the guidelines of the SFSTP, before being validated again according to the guidelines of the GTFCh. Accurate and reliable measurements ($\pm 30\%$) of propane concentrations have been made with this method in a range of 8.00–50.0 nmol/mL of HS. The method remains safe as the propane is generated in a hermetically closed HS vial. Moreover, the quantification is very precise as deuterated propane is used as the internal standard, and is particularly useful in cases where only small amounts of tissue are available. Therefore, the described method provides reliable, accurate and repeatable propane concentrations of various samples (blood, tissue . . .) whatever their state. Both validation procedures can be used to assess this new method of quantification, even though due to their differing guidelines, small differences could be observed in their accuracy profile. The main difference was the lower limit of quantification that was reached with the first procedure (SFSTP), while the second procedure (GTFCh) only got close to it. Anyway, the range of quantification is fully satisfying to provide forensic results to complete an autopsy, or even to determine the exact cause of death in cases where the origin is in suspicious circumstances. Moreover, this new analytical protocol of quantification can be applied to a range of applications other than forensic sciences, such as measuring propane concentrations in aerosol propellant, liquefied petroleum gas lighters, propane grills and in environmental analysis. It could help assess safety requirements for a wide variety of equipment.

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