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ETHANOL AND PUTREFACTION: MEDICOLEGAL RELEVANCE OF THE STUDY OF DIFFERENT BIOLOGICAL SAMPLES

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ETHANOL AND PUTREFACTION: MEDICOLEGAL RELEVANCE OF THE STUDY OF DIFFERENT BIOLOGICAL SAMPLES

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INDEX

RESUMO.....7
ABSTRACT.....9
INTRODUCTION.....11
MATERIAL AND METHODS.....14
RESULTS AND DISCUSSION.....15
CONCLUSION.....24
REFERENCES.....25

RESUMO

Na toxicologia forense *postmortem* o etanol é a substância mais frequentemente determinada. A sua interpretação é particularmente difícil quando é colhido sangue de cadáveres em putrefação. Nesta situação é de extrema importância a análise de outras amostras biológicas, como a urina e o humor vítreo, que em circunstâncias normais são positivas para etanol apenas quando ocorre ingestão *antemortem*. A relevância acentua-se quando a concentração de etanol no sangue é menor que 0.5 g/L.

Foi realizado um estudo, por um período de 3 anos, tendo os dados sido retirados da informação presente nas requisições recebidas pelo Serviço de Química e Toxicologia Forense do Instituto Nacional de Medicina Legal e Ciências Forenses de Portugal. Dos casos de putrefação, 66 apresentavam etanol no sangue. Destes, 66.7% tinham uma concentração de etanol entre 0.1 g/L e 0.5 g/L, o que é relevante dado que poderá ter ocorrido produção endógena, tornando a interpretação destes valores desafiante. Esta interpretação pode beneficiar da integração da informação disponível do caso, especialmente da pesquisa de etanol em outras substâncias biológicas. Os dados recolhidos permitiram determinar o número de casos que possuíam uma, duas ou três amostras biológicas e foram escolhidos três casos forenses que representavam cada uma destas categorias, exemplificando situações passíveis de acontecerem no estudo da concentração de etanol em cadáveres putrefactos.

Este estudo realça, através da revisão da literatura e dos casos apresentados, que quando a concentração de etanol é abaixo de 0.5 g/L em cadáveres putrefactos, a análise das três amostras biológicas (sangue, urina, humor vítreo) permite apoiar a presumida origem do etanol.

***Palavras-chave*:** Concentração de etanol no sangue, produção *postmortem*, ingestão *antemortem*, putrefacção, urina, humor vítreo.

ABSTRACT

Alcohol is the most determined substance in postmortem toxicology and its interpretation is especially complicated when the blood alcohol concentration (BAC) is determined from corpses in putrefaction. Therefore, it is of high relevance the analysis of other biological samples, as urine and vitreous humor, that in normal situations are only positive for alcohol when antemortem ingestion has occurred. This is important mostly when the BAC is lower than 0.5 g/L.

This study was made within a period of 3 years, with data material obtained from the Forensic Chemistry and Toxicology Service of the National Institute of Legal Medicine and Forensic Sciences of Portugal. There were 66 cases of putrefaction with a positive result for alcohol. Of these, 66.7% had a BAC between 0.1 g/L and 0.5 g/L, relevant as endogenous production might have happened, making the interpretation of these values challenging. However, this interpretation can benefit from the integration of the available case information, especially the search for alcohol in other biological samples. Using this data, it was determined how many cases presented one, two and three biological samples and three forensic cases were selected, representing each of these categories, exemplifying situations that can easily occur when studying the BAC of putrefied corpses.

This study highlights, through the reviewed literature and the presented cases, that in decomposing bodies, when the BAC is lower than 0.5 g/L, the analysis of all three types of biological samples (blood, urine, vitreous humor) is helpful to reach a conclusion regarding the origin of the alcohol.

Keywords**:** Blood alcohol concentration, postmortem production, antemortem ingestion, putrefaction, urine, vitreous humor.

INTRODUCTION

In postmortem toxicology, alcohol continues to be the most found psychoactive substance (1). The sample of choice for this postmortem analysis is considered to be blood. There is a high relevance in the analysis and interpretation of the BAC because it is commonly used as evidence, in criminal and civil litigation (2,3). It is a rather simple analytical procedure but the interpretation of postmortem BAC and the conclusions that follow, regarding whether there was an endogenous production or ingestion of alcohol prior to death, are fraught with difficulties (1). These are especially aggravated when in situations of putrefaction the BAC is less than 0.5 g/L (4).

When death occurs the supply of oxygen to the body ceases and the process of autolysis begins. The disintegration of cell membranes leads to the softening and liquefaction of the tissue, allowing bacteria from the gastrointestinal track to invade the surrounding tissue and vascular system (1). These enteric bacteria are able to synthetize alcohol, primarily on glucose and, on a smaller scale, on lactate, amino or fatty acids (2).

The degree of putrefaction is proportional to the spread of bacteria throughout the body and vascular system as well as to the production of endogenous alcohol, which is a result of the latter. Therefore, in a putrefied corpse the probability of having contaminated blood by enteric bacteria and subsequent production of ethanol is high (1). In this situation, to establish if the ethanol is of endogenous production and not from antemortem ingestion, it is important to analyze other body fluids, namely urine and vitreous humor.

The concentration of ethanol in blood, body organs and tissues and the speed of distribution depend on the water content of the materials analyzed and the tissue perfusion rate (1).

Urine is a useful sample, as it is mainly composed by water and the risk of being contaminated by microorganisms after death seems to be low (1). Furthermore, the urine of healthy individuals does not have the nutrients required for the postmortem production of ethanol. But in some cases, a limitation might arise, due to some medical conditions, such as diabetes, which can cause elevated levels of glucose in the urine (5,6).
Since alcohol appears in urine shortly after ingestion, if a putrefied body has alcohol present in its blood and absent from its urine, there is a high suspicion that this alcohol is of

endogenous production. However, frequently there is no urine in the bladder to collect a sample (7).

The vitreous humor is a watery fluid (99%) and studies have demonstrated that the mean vitreous humor/blood ratio of ethanol is nearly the values estimated from the distribution of water in these two biological specimens (1). The other advantage is its anatomical location. The vitreous humor is in the posterior eye cavity, shielded by the bone structure of the eye. It is not close to any large vessels or the gastrointestinal tract (1). Therefore, if the corpse has undergone putrefaction, the vitreous humor is in a more protected site, unlikely to become contaminated by bacteria. As only a small volume is needed for analysis and due to its protection, vitreous humor is a more available sample (7). It is important to acknowledge that the vitreous humor can contain glucose, one of the substrates for ethanol production (1) but because the infiltration of bacteria into the vitreous humor does not occur until later in the putrefactive process, any alcohol present is considered to be of exogenous origin (7).

It is claimed in a study that with positive results in these three mentioned biological samples – blood, urine and vitreous humor – ingestion of alcohol is the most likely event (7).

The interpretation of low alcohol concentrations can be harder in the presence of putrefaction, because of the possibility of postmortem alcohol production (4). This is because lower BAC (less than 0.3 g/L) are more likely to be formed postmortem rather than higher concentrations (1). So it is important to additionally analyze other biological samples, as urine or/and vitreous humor (4). A BAC lower than 0.1 g/L is normally reported as negative (1). Other studies showed that values less than or equal to 0.2 g/L have an equal probability of being from antemortem ingestion and postmortem production (4,8). It has been discussed that in the presence of a BAC lower than 0.3 g/L the determination of alcohol in urine and vitreous humor must be conducted in order to support evidence of the BAC, otherwise its significance is debatable (1). Another study concluded that a BAC lower than 0.3 g/L is frequently associated with a negative alcohol concentration of vitreous humor, implying that postmortem production might have happened (8). On the other hand, when the BAC is higher than 0.3 g/L, more than 50% of the cases are associated to a positive alcohol concentration of urine and/or vitreous humor. This suggests that a BAC equal or greater than 0.3 g/L is most likely to be due to alcohol ingestion rather than endogenous production (4,8).
Another study reported that with a BAC of 0.5 g/L there is an 87% probability of a positive result for alcohol in humor vitreous and with the increase of this concentration the probability goes up to 99% (9).

In the present study, the authors made a statistical survey, within the period of three years, of the cases that indicated putrefaction in which the medical expert required the analyses of the BAC. This information was obtained through the requisitions, from the Forensic Chemistry and Toxicology Service (FCTS) of the National Institute of Legal Medicine and Forensic Sciences (NILMFS), Centre branch. The aim of this study was to assess the different types of biological samples (blood, urine or/and vitreous humor) available in these cases and understand their relevance when interpretating the BAC.

From all the cases evaluated, three forensic cases were selected, which represent situations that might occur when studying the BAC of a putrefied corpse. These cases were chosen to highlight the possible benefits of analyzing multiple biological samples, when present, for confirmation of the BAC.

MATERIAL AND METHODS

The data material of the current study was collected from a statistical survey, conducted through the Starlims program from the FCTS of the NILMFS, within the period of three years, regarding the autopsies with information of putrefaction, in which the medical expert requested the analysis of the blood alcohol concentration. There were 130 cases that fulfilled these criteria. The alcohol analysis was executed in the FCTS of the NILMFS, Centre branch, using the different biological samples (blood, urine and vitreous humor).
Considering the purpose of the study, the positive results were the ones evaluated.

Biological Samples

The *post mortem* samples (blood, urine and vitreous humor) were collected from autopsies performed in the Forensic Pathology Service of that institution or in the Medico-Legal Offices from central regions of Portugal.
The samples of blood collected were preserved with 1% of fluoride sodium.

Analytical procedure

The preparation of the samples, controls and calibrators were performed using the procedure described by several authors for the determination of volatile analyses (1,10,11). Prior to analysis by gas chromatography, all samples, including the calibrators, were diluted: 100 µL of urine, vitreous humor or whole blood were diluted with 1 mL aqueous solution of *n*-propanol (100 mg/L), used as internal standard (IS). No inorganic salt was used.

Instrumentation: HS-GC/FID

An Agilent 6890N GC equipped with a flame ionization detector (FID) and coupled to an Agilent G1888 headspace injector capable of injecting 1 ml (fixed volume) of sample was used for determination of alcohol in biological samples. To enhance specificity, a dual-column confirmation was performed. The first column used was a DB-ALC2 obtained from Agilent (Santa Clara, CA), with dimensions of 30 m x 0.320 mm x 1.2 µm; the second column was an Agilent DB-ALC1, with dimensions of 30 m x 0.320 mm x 1.8 µm. The HS-GC-FID conditions are described in table 1. Analytical data was processed using the Agilent ChemStation Rev B.03.01 [317] software. It was obtained a total run time of 5 minutes.

RESULTS AND DISCUSSION

In the period of 3 years, there were 4264 medical-legal autopsies executed, in the NILMFS, Centre branch, in which the medical expert required the determination of the blood alcohol concentration. Of these cases, a total of 130 had information stating the presence of putrefaction. They were related to different causes of death (intoxication, drowning, trauma, natural deaths and undetermined cause) and had a postmortem interval between one and six days. The blood alcohol concentration measured determined 64 cases with a negative result and 66 with a positive result. Of these 66 cases, there were 17 (25.8%) with only one biological sample available (peripheral blood or cardiac blood), 32 (48.5%) with two biological samples available (peripheral blood and urine; peripheral blood and cardiac blood; cardiac blood and vitreous humor; cardiac blood and urine) and 17 (25.8%) with three or more biological samples available (eg: cardiac blood, vitreous humor and urine) (Figure 1).

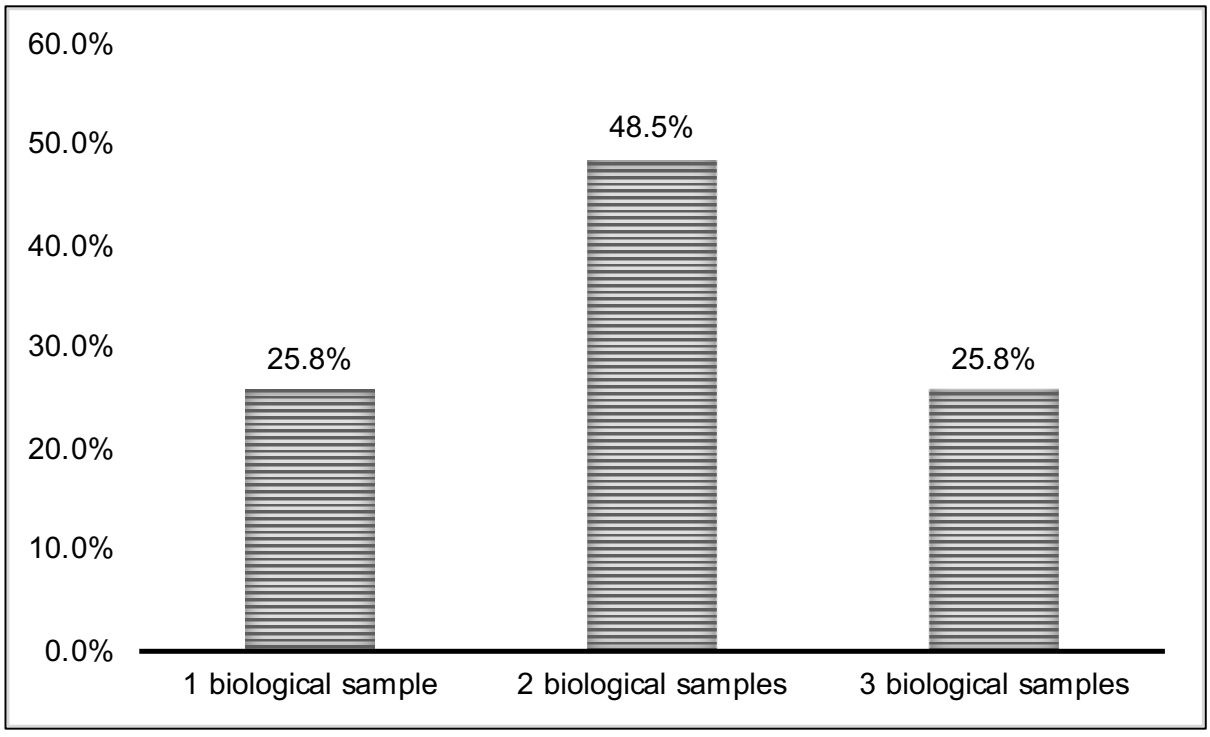

Figure 1. Distribution of the percentage of cases according to number of biological samples available.

Analyzing these 66 positive cases, 39.4% were greater or equal to 0.1 g/L and lower than 0.3 g/L, 27.3% greater or equal to 0.3 g/L and lower than 0.5 g/L, 16.7% greater or equal to 0.5 g/L and lower than 1.2 g/L and 16.7% greater or equal to 1.2 g/L (Figure 2).

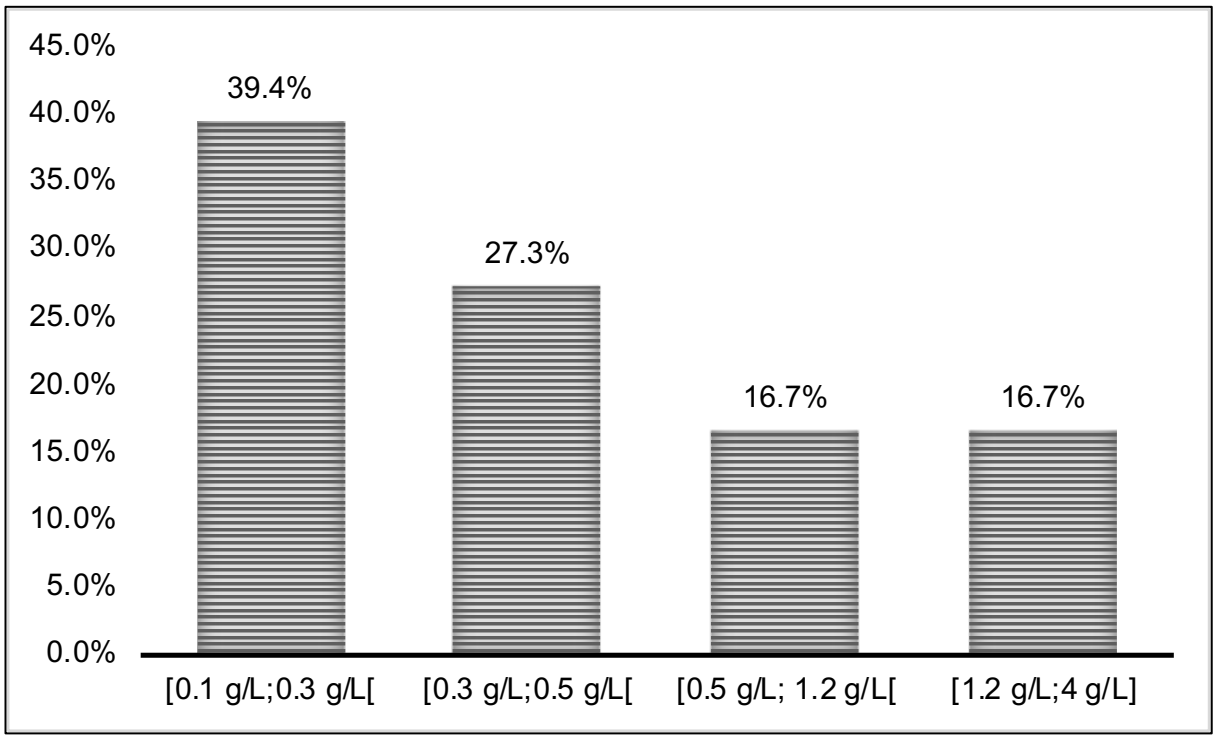

Figure 2. Distribution of the percentage of cases according to the blood alcohol concentration.

The literature contemplating this subject of study highlights how difficult it is to reach a conclusion on the origin of the alcohol when in a blood sample from a putrefied corpse the BAC is between 0.1 g/L and 0.5 g/L. As in a decomposing body endogenous alcohol production might occur, it is important to distinguish this phenomenon from antemortem ingestion of alcohol. In this study, of the positive cases determined there are 66.7% with a BAC between 0.1 g/L and 0.5 g/L, which implies that the interpretation of this results can be questioned. Therefore, in order to support and help unravel the alcohol provenience, a wide range of information is available which can be integrated. It is, for example, the detection of volatile compounds known to be present when postmortem alcohol production occurs or the search of alcohol in other biological samples, which were available when the autopsy was conducted.

Notwithstanding, results greater than 0.5 g/L must have this same careful examination because despite being frequently associated with antemortem ingestion, it is possible that, adding to this, postmortem alcohol production might have happened. The case 1 exposed is representative of this situation.

In order to highlight the advantages and situations where it can be of relevance the multiple sample evaluation mentioned above, a more detailed statistical analysis was made, entailing the blood samples with alcohol concentrations greater or equal to 0.1 g/L and lower than 0.5 g/L, from the period of time of three years.

There were 44 cases with concentrations within this value range. Of these, 13 had one biological sample available, which was peripheral blood in 5 of the cases and cardiac blood in

the remaining 8. The cardiac blood is a specimen that has special characteristics that have to be taken into account when interpretating the concentration of alcohol. Studies have showed that concentrations of alcohol in cardiac blood are normally higher than the ones in peripheral blood (1). It is also vital to acknowledge that the tubes used to transport and collect the peripheral blood samples to the laboratory contain sodium fluoride as a preservative, essential to inhibit the additional production of alcohol between the time of the autopsy and the analysis in the laboratory (12). Within the situations with only one sample, case 1 is reported with a postmortem cardiac blood measured in a context of submersion, where 1-butanol was determined, a volatile compound normally produced during decomposition of the body (1).

There were 20 cases with two biological samples available and of these, 16 had the second sample supporting the alcohol positivity in the blood sample. The described case 2 had a postmortem cardiac blood determination of 0.38 g/L and a negative concentration of alcohol in vitreous humor. This is one of the 4 cases that do not have a second sample supporting the result obtained from the blood sample.

At last, there were 11 cases with three available biological samples and of these, 9 supported the alcohol concentration determined in the blood sample**.** One of these is case 3, where a decomposed body of a male was found in his home and the result obtained from the blood sample was confirmed by the two alternative samples collected, urine and vitreous humor.

Case 1

A 44-year-old homeless male, who was reported missing for 5 days, was found at high sea by a ship. The autopsy was performed on the day after being found. In the external examination there were obvious signs of putrefaction, the marbling of the skin throughout the body, the slippage of skin, the destruction of both eye cavities and the subcutaneous emphysema, especially marked in the scrotum. The autopsy, in the internal examination, revealed that both lungs had the pulmonary parenchyma and bronchi filled with a diluted hematic fluid, that had a characteristic sea odor. The stomach contained 750 mL of a fluid compatible with sea water. Both of these findings were consistent with the documented mechanism of death, drowning. The pathologist collected a sample of blood from the only available site, the pulmonary artery, consequence of the osmotic phenomena, typical in salt water drownings. This sample was subjected to a toxicological analysis of drugs and alcohol. The BAC then determined was of 0.77 g/L (Table 1). According to literature, this BAC, above 0.5 g/L, and the detection of acetaldehyde in the chromatogram (Figure 3), which is the first metabolite of alcohol, are findings compatible with antemortem ingestion (1,13). In this case, what can be questioned is the determined concentration as it is a submerse corpse in an advanced state of putrefaction, where it was possible to detect and quantify a volatile organic compound in the blood, 1-

butanol, with a concentration of 0.43 g/L (Figure 4). In bodies recovered from water, the presence of 1-butanol is considered a good indicator that postmortem synthesis of ethanol might have occurred during immersion (1). Therefore, the BAC is probably a result of both antemortem ingestion and endogenous production. An alternative biological sample, if available, would have been of use as it could support the result obtained from the blood.

Table 1 - Toxicological results of the presented case 1

	Analytes			
	Acetaldehyde	Ethanol	n-propanol (IS)	1-butanol
Retention Time (min) FID 1A	1.932	2.625	4.650	8.003
Concentration (g/L)	**0.10**	**0.77**	**D**	**0.43**

D – Detected

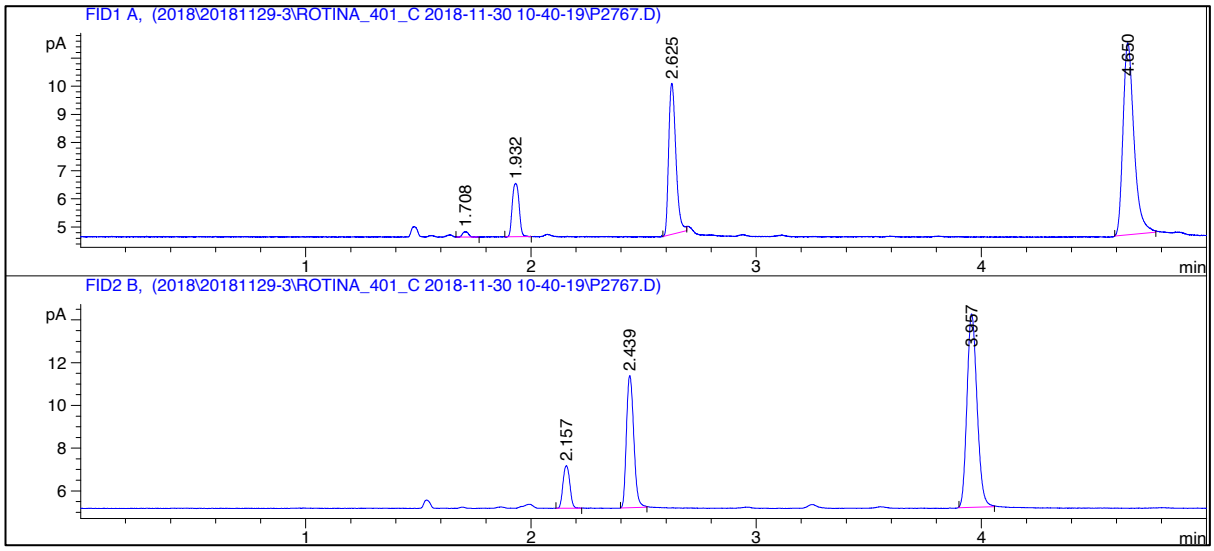

Figure 3. Chromatogram analysis of ethanol in cardiac blood by HS-GC/FID.

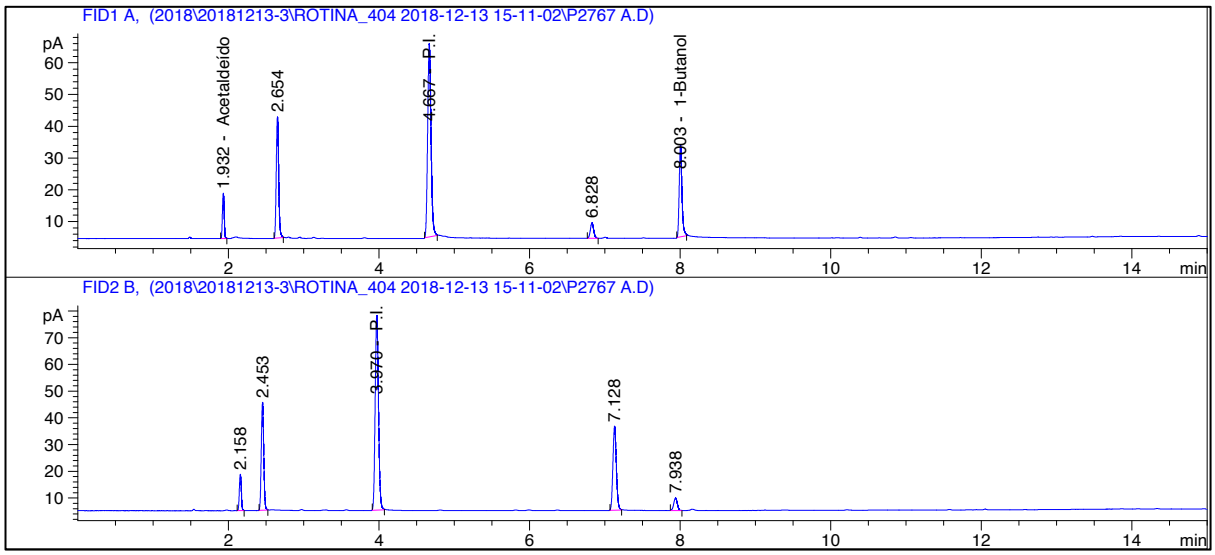

Figure 4. Chromatogram analysis of volatile substances in cardiac blood by HS-GC/FID.

Case 2

A 10-year-old female child entered the hospital in cardiopulmonary arrest. The medical information available referred that the child suffered from Larsen syndrome and cerebral palsy. There was not any record of possible medication. The autopsy was performed two days after death. Signs of putrefaction were discovered as the external examination revealed an extensive green abdominal stain. The internal examination highlighted that both lungs contained a purulent pleural effusion, which is consistent with the medical information that referred a “lung infection”. The pathologist collected biological samples, blood from the heart cavity and vitreous humor, throughout the autopsy. These were to be subjected to a toxicological analysis of drugs and alcohol. The quantification of the BAC was of 0.38 g/L (Figure 5 and Table 2) and the analysis executed on vitreous humor was negative for alcohol (Figure 6). The presence of a second biological sample was of value as it gave additional information. The negative result in the vitreous humor suggests that the alcohol found in the blood was probably a result of postmortem production. This is supported by the visible putrefaction on the corpse and the BAC obtained being lower than 0.5 g/L (4). The tubes used to collect the cardiac blood sample did not contain sodium fluoride and the time between the collection of the blood sample and the analysis was significant, consequently the value of the BAC might have been caused by in vitro production (12). Additional information that can help distinguish these two phenomena might be provided by a microbiological analysis of the blood sample, containing routine bacterial and fungal cultures.

Table 2 - Toxicological results of the presented case 2.

Analytes	Cardiac blood concentration (g/L)	Vitreous humor concentration (g/L)
Acetaldehyde	ND	ND
Ethanol	**0.38**	**Negative**
n-propanol (IS)	D	D

D – Detected; ND – Not Detected

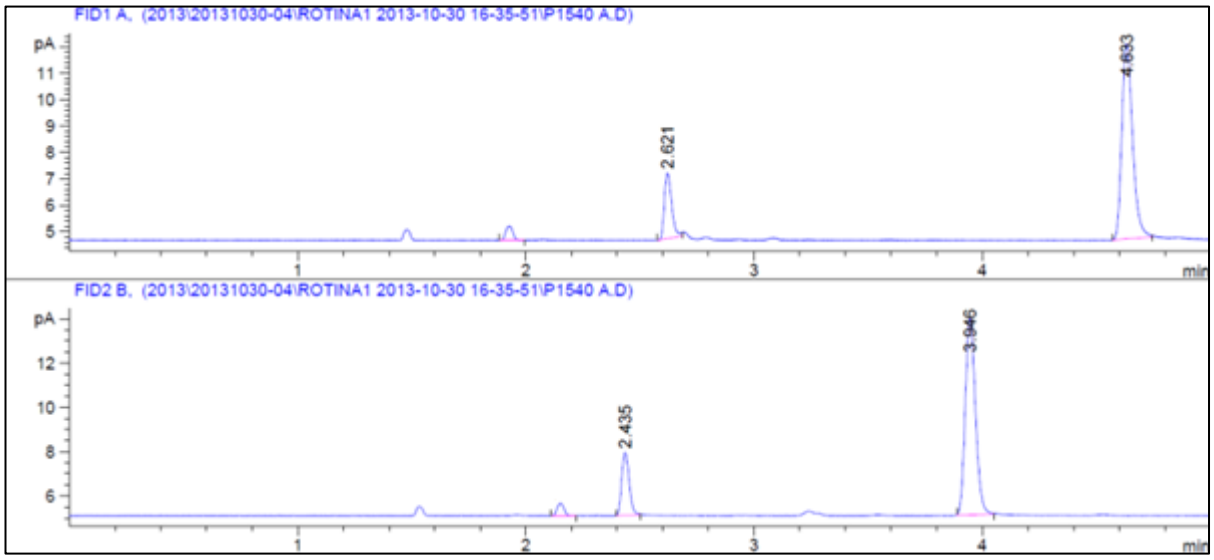

Figure 5. Chromatogram analysis of ethanol in cardiac blood by HS-GC/FID.

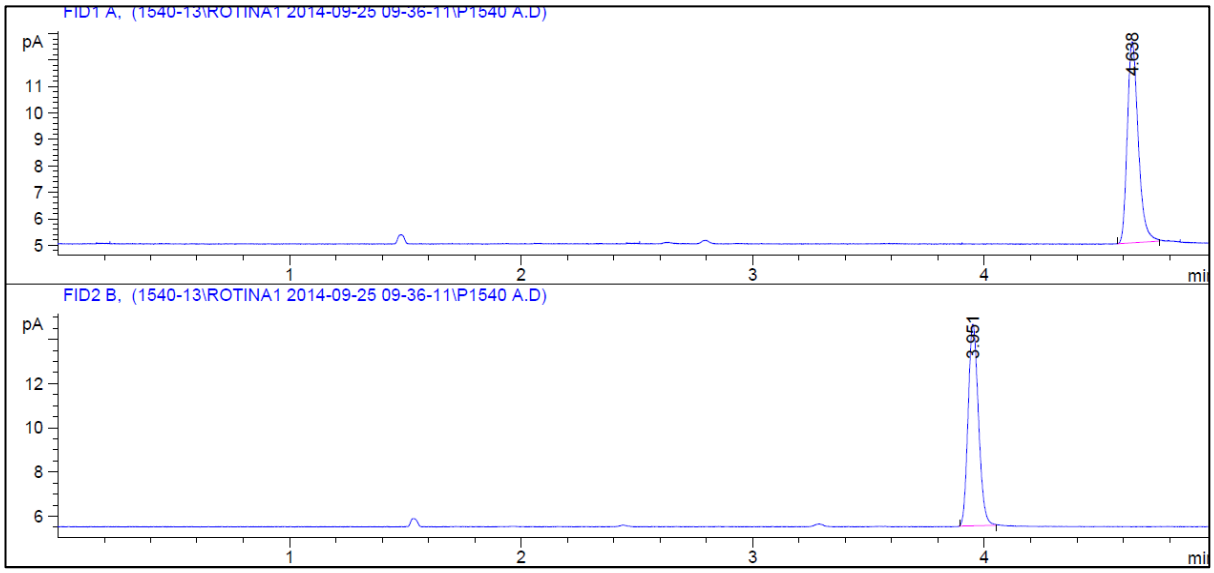

Figure 6. Chromatogram analysis of ethanol in vitreous humor by HS-GC/FID.

Case 3

A 51-year-old male was found and declared dead in his home after being unreachable for two days. The autopsy was performed two days after being found. The deceased had a medical leave because, in the past month, had a considerable weight loss and had been feeling unwell with multiple complains, as headaches, dyspnea and weakness. In the external examination, a green putrefactive stain in the neck, thorax and abdomen and marbling of the skin in the sacral region were visible. The findings in the internal examination were compatible with the degree of putrefaction primarily seen and there was clear evidence of atherosclerosis in multiple arteries of the body, namely the aorta and the coronaries. The heart had findings of an extensive fibrosis with hemorrhagic areas. The cause of death was assumed to be related to the chronic ischemic cardiomyopathy that the deceased had, probably a result of the atherosclerotic cardiopathy.

During the autopsy several biological samples were collected: blood from the heart cavity, urine and vitreous humor. The conducted toxicological analysis determined a cardiac blood alcohol concentration of 0.36 g/L (Figure 7), an alcohol concentration of 0.13 g/L in the urine (Figure 8) and of 0.14 g/L in the vitreous humor (Figure 9). In this case, the presence of these two additional biological samples and their positive results for alcohol were important to presume that the alcohol found in the blood sample was due to antemortem ingestion**.** Levine, et al. reported that in a BAC higher than 0.3 g/L more of 50% of the cases are associated to a positive alcohol concentration of urine and/or vitreous humor, suggesting that these values of BAC are most likely associated with alcohol ingestion rather than an endogenous production (4).

Table 3 - Toxicological results of the presented case 3.

Analytes	Cardiac Blood concentration (g/L)	Urine concentration (g/L)	Vitreous humor concentration (g/L)
Acetaldehyde	D	ND	ND
Ethanol	**0.36**	**0.13**	**0.14**
n-propanol (IS)	D	D	D

D – Detected; ND – Not Detected

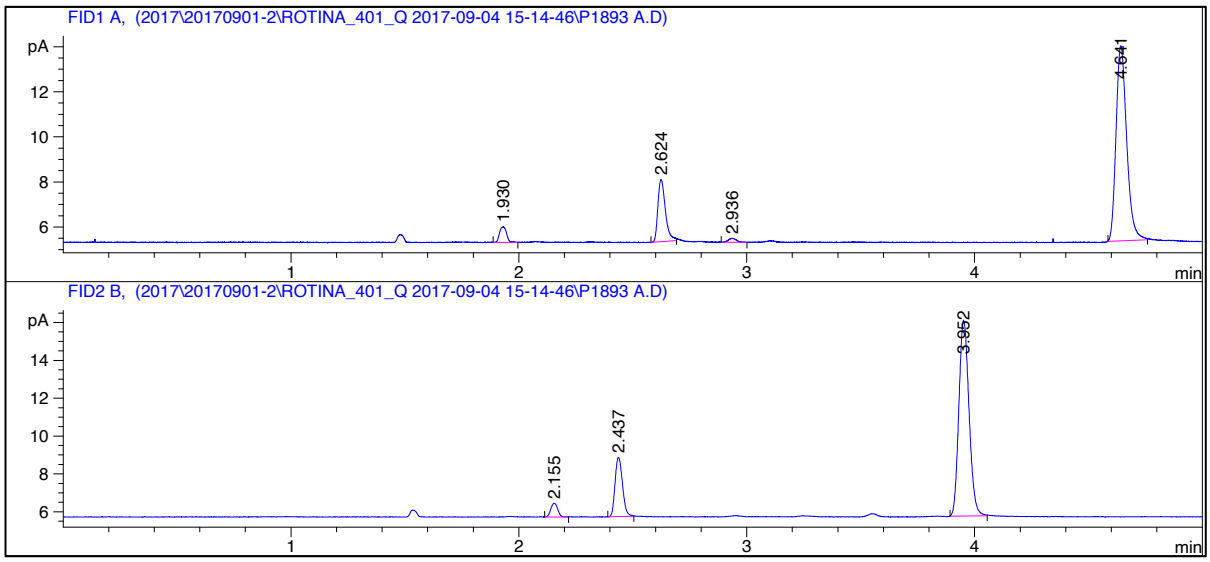

Figure 7. Chromatogram analysis of ethanol in blood by HS-GC/FID.

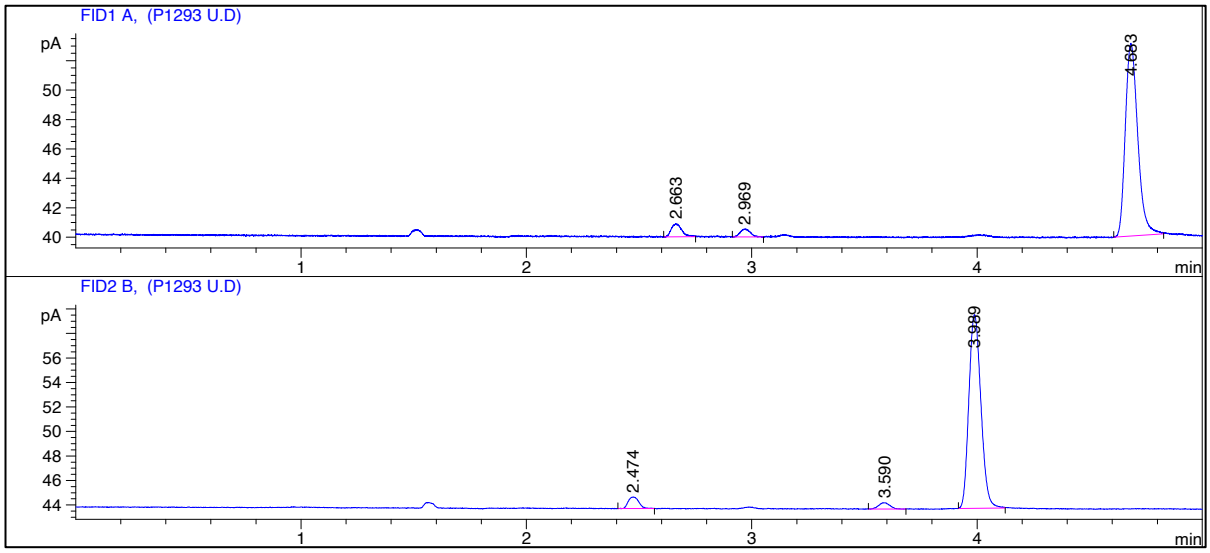

Figure 8. Chromatogram analysis of ethanol in urine by HS-GC/FID.

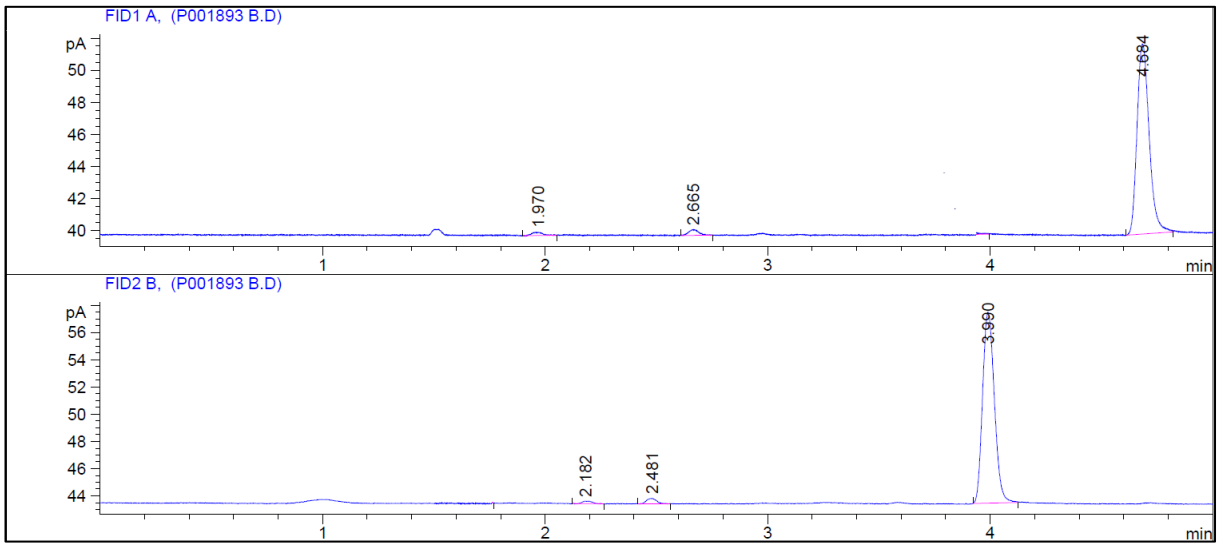

Figure 9. Chromatogram analysis of ethanol in vitreous humor by HS-GC/FID.

CONCLUSION

An analysis and interpretation of the BAC is often required in death investigations, as it can be a casual or contributory factor. This information is commonly vital in civil and criminal litigations. It is considered to be one of the most demanding tasks in forensic toxicology, especially when performed on a putrefied corpse.

In this specific situation, several origins for the alcohol detected postmortem are probable. Possible causes include antemortem ingestion, postmortem endogenous production or in-vitro production, these last two being a consequence of fermentation processes by enteric bacteria. In order to distinguish these situations, it is essential to evaluate and integrate all available information. The history of the case must be taken into account, as well as the type and quality of the collected samples, which rely highly on the storage method and on the conservation state of the body, which in turn is influenced by the environmental conditions (temperature and humidity) and time elapsed between death and the autopsy. Other tests can be executed in order to identify microorganisms present in the sample, the presence of volatile compounds known to be produced postmortem and to determine the alcohol concentration in other available biological samples.

In this study we discussed how the determination of the alcohol concentration in urine and vitreous humor can be a valuable tool, especially in the presence of putrefied corpses and low concentrations of alcohol in the blood, when trying to determine whether the blood alcohol concentration was of exogenous origin, endogenous production or both. The 3 cases were presented to illustrate real situations, in which the additional examination of other available biological samples demonstrated to be of invaluable value to the final conclusions.
This suggests that the analysis of these three types of biological samples (blood, urine, vitreous humor) can be of use in reaching a conclusion regarding the origin of the alcohol and making it less susceptible to dispute.

REFERENCES

1. Kugelberg FC, Jones AW. Interpreting results of ethanol analysis in postmortem specimens: A review of the literature. Forensic Sci Int. 2007;165(1):10–29.
2. De Lima IV, Midio AF. Origin of blood ethanol in decomposed bodies. Forensic Sci Int. 1999;106(3):157–62.
3. Costa N, Silva R, Mendonça CM, Real FC, Vieira DN, Teixeira HM. Prevalence of ethanol and illicit drugs in road traffic accidents in the centre of Portugal: An eighteen-year update. Forensic Sci Int. 2012;216(1–3):37–43.
4. Levine B, Smith ML, Smialek JE, Caplan YH. Interpretation of Low Postmortem Concentrations of Ethanol. J Forensic Sci. 1993;38(3):13453J.
5. Canfield D V., Brink CJD, Johnson RD, Lewis RJ, Dubowski KM. Postmortem Ethanol Testing Procedures Available to Accident Investigators. 2007;(August):1–4.
6. Antonides H, Marinetti L. Ethanol production in a postmortem urine sample. J Anal Toxicol. 2011;35(7):516–8.
7. Zumwalt RE, Bost RO, Sunshine I. Evaluation of Ethanol Concentrations in Decomposed Bodies. J Forensic Sci. 1982;27(3):12167J.
8. Caplan, Yale H; Levine B. Vitreous Humor in the Evaluation of Postmortem Blood Ethanol Concentrations.pdf.
9. Pinto M, Eusébio E, Monteiro C. Development and Validation of an Analytical Method for Volatiles with Endogenous Production in Putrefaction and Submersion Situations. J Anal Toxicol. 2020;(October):1–8.
10. Monteiro C, Franco J M, Proença P, Castañera A, Claro A, Vieira DN C-RF. Qualitative and quantitative analysis of a group of volatile organic compounds in biological samples by HS-GC/FID: application in pratical cases. Forensic Sci Int. 2014;Vol. 243:137–43.
11. Monteiro C, Proença P, Tavares C, Castañera A C-RF. Interference of anesthetics in blood alcohol analysis by HS-GC/FID: a case report. Forensic Sci Int. 2016;Vol. 265:65–9.
12. Quintas MJ, Costa P, Melo P, Castro A, Franco JM, Teixeira HM. Postmortem in vitro ethanol production—It could be more common than we think! Forensic Sci Int [Internet]. 2017;274:113–6. Available from: http://dx.doi.org/10.1016/j.forsciint.2016.12.040
13. AW J. Alcohol, its absorption, distribution, metabolism, and excretion in the body and pharmacokinetic calculations. Wires Forensic Sci. 2019;Vol. 1:1–26.