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Interpretation of Low Postmortem Concentrations of Ethanol

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ABSTRACT: The interpretation of postmortem blood ethanol concentrations (BAC), especially those less than 0.05 g/dL can be complicated by postmortem ethanol formation. One method used by the toxicologist to respond to this possibility is to analyze multiple specimens for ethanol. Two useful specimens to analyze are vitreous humor and urine, because they are less susceptible to the putrefaction process. A negative vitreous humor and/or urine ethanol would suggest that the measured ethanol resulted from postmortem formation. Data were collected from the Office of the Chief Medical Examiner, State of Maryland and the Armed Forces Institute of Pathology on blood specimens with ethanol concentrations less than 0.05 g/dL to develop a reasonable threshold for interpretation in the absence of other specimens. A total of 381 cases with a BAC between 0.01 and 0.04 g/dL were studied over a 2 year period. Urine and vitreous humor specimens were tested where available. At a BAC of 0.01 g/dL, 54% of the cases were associated with a positive vitreous humor and/or urine ethanol concentration. This percentage increased to 63% when BAC equals 0.02 g/dL. Seventy-three percent and 92% of the cases had a positive alternate specimen if the BAC was 0.03 g/dL and 0.04 g/dL, respectively. In addition, 90% of the cases where both vitreous humor and urine were analyzed showed consistent results, that is both specimens were positive or negative. This suggests that in the absence of additional information, a BAC of 0.04 g/dL or higher probably resulted from ethanol consumption.

KEYWORDS: toxicology, blood, ethanol, postmortem ethanol concentration

One of the most vexing problems faced by the postmortem forensic toxicologist is the question of whether a measured BAC indicates antemortem ethanol consumption or postmortem ethanol formation. Studies have shown that ethanol is produced in postmortem blood by the activity of a variety of microorganisms on glucose, amino or fatty acids [*1–6*]. This formation may occur in the intact body between death and autopsy or in vitro in specimens collected at autopsy. One way of sorting out the differences between ethanol consumption and postmortem formation is by analyzing for ethanol in different body fluids [*7*]. We had previously studied vitreous humor to blood ratios [*8*]. Urine can

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also be useful. Both vitreous humor and urine are easily obtainable at autopsy and vitreous humor can be collected without an autopsy. Each specimen is easy to work with analytically. More important, each specimen is more resistant to postmortem ethanol formation than is blood [7]. This means that a positive blood ethanol, plus a positive urine or vitreous fluid ethanol would likely indicate ethanol consumption rather than postmortem formation.

Occasionally, only a postmortem blood specimen is collected for ethanol analysis. Nonetheless, the interpretation of BAC in terms of whether it arises from consumption or postmortem formation is often required. As expected, the greatest number of difficulties arise when the BAC is low, less than 0.04 g/dL. The data presented are intended to facilitate the interpretation of blood specimens with low BAC.

Experimental

Specimen Collection

Blood, urine, and vitreous humor were collected from autopsies performed by pathologists of the Office of the Chief Medical Examiner, State of Maryland or of the Armed Forces Medical Examiner. Specimens were collected in 120 mL plastic containers; no preservative was added. The site of blood collection was unspecified, but believed to be heart blood in the majority of the cases. Specimens were refrigerated between autopsy and analysis. All blood specimens with a BAC greater than or equal to 0.01 g/dL had the associated vitreous humor and/or urine specimens analyzed for ethanol. Blood specimens with a BAC less than or equal to 0.04 g/dL were tabulated for this study.

Specimen Preparation

An aliquot of biological fluid was diluted with water containing an internal standard (n-propanol or 2-butanone, 1 part fluids: 9 parts water) and placed in a 23 mL headspace vial. Specimens were analyzed in duplicate.

Instrumentation

A Perkin-Elmer 8500 gas chromatograph with a HS-101 head space analyzer was used for ethanol analysis. Specimens were equilibrated for 35 min prior to gas chromatographic analysis. The column packing was a 0.2% Carbowax 1500 on 80/100 Carbopak C. The temperatures were as follows: needle, 90°C; transfer line, 90°C, oven, 120°C, detector, 250°C. The times were the following: pressurization, 0.5 min; injection, 0.08 min; withdrawal, 0.2 min. The carrier gas was nitrogen flowing at 30 mL/min.

Quantitation

The area ratio of ethanol to internal standard of the specimen were compared to aqueous ethanol standards. Values less than 0.01 g/dL were reported as negative; values above 0.01 g/dL were rounded to two decimal places.

Results and Discussion

A total of 381 cases were included in this study. Fifty-four percent of the cases had a positive vitreous humor ethanol concentration (VAC) and/or urine ethanol concentration (UAC) when the BAC was 0.01 g/dL. This percentage increased to 63% when BAC was

TABLE 1—*Distribution of blood ethanol concentration (BAC), vitreous humor ethanol concentration (VAC) and urine ethanol concentration (UAC).*

	BAC (g/dL)											
	0.01			0.02			0.03			0.04		
	VAC−	VAC n. a.	VAC+	VAC−	VAC n. a.	VAC+	VAC−	VAC n. a.	VAC+	VAC−	VAC n. a.	VAC+
UAC−	13	5	2	21	4	3	11	5	1	1	1	0
UAC n. a.	22		18	23		38	9		25	4		23
UAC+	0	4	21	3	11	27	4	8	31	1	16	26

+ = >0.01 g/dL.
n. a. = not analyzed.
− = <0.01 g/dL.

0.02 g/dL. At blood concentrations of 0.03 and 0.04 g/dL, 73 and 92% of the cases, respectively, had an alternate specimen positive for ethanol.

The individual distribution of VAC and UAC at BAC of 0.01 to 0.04 g/dL respectively are given in Table 1. At a BAC of 0.01 g/dL, 54% (41 of 76) of the cases had a positive VAC and 56% (25 of 45) of the cases had a positive UAC. At a BAC of 0.02 g/dL, 59% of the cases had a positive VAC (68 of 115) and/or UAC (41 of 70). At a BAC of 0.03 g/dL, 70% (57 of 81) of the cases had a positive VAC and 72% (43 of 60) of the cases had a positive UAC. At a BAC of 0.04 g/dL, 89% (49 of 55) of the cases had a positive VAC and 96% (43 of 48) of the cases had a positive UAC.

A total of 165 cases were obtained where both vitreous humor and urine were collected. Over 90% of these cases demonstrated consistent results between VAC and UAC. Forty-six cases had negative VAC and UAC and 105 cases had positive VAC and UAC. Fourteen of the 165 cases showed some inconsistency, that is where one specimen was positive and the other specimen was negative. The data from these 14 cases are presented in Table 2. One-half of these cases had a positive VAC and a negative UAC while, the other half of the cases had a positive UAC and a negative VAC. These cases warrant further study. The circumstances surrounding these deaths and the apparent alcohol concentration of the specimen less than 0.01 g/dL are also given in Table 2. It was hoped that a review of the circumstances might provide information to facilitate interpretation of the analytical findings. For example, a hospital blood with a positive antemortem blood ethanol or witnessed drinking would indicate drinking prior to death. On the other hand, a decomposed body would suggest that measured ethanol resulted from postmortem formation. Unfortunately, no cases had hospital specimens submitted. Three cases were decomposed; one of these cases was a diabetic. Since the combination of high glucose and microorganism can cause the production of large amounts of ethanol in blood or urine, this would suggest that the measured ethanol was produced by postmortem for-

TABLE 2—*Specimens with inconsistent results.*

No.	BAC	VAC	UAC	Circumstances
1.	0.01	0.02	0(0.0071)	Acute MI
2.	0.01	0.01	0(0.0088)	Chest injuries/accident
3.	0.02	0(0.0097)	0.03	Suicidal gunshot wound (GSW) chest, mild to moderate decomposition
4.	0.02	0.05	0(0.0063)	GSW homicide
5.	0.02	0.02	0(0.0047)	Multiple injuries, driver hit by tractor-trailer
6.	0.02	0(0.0013)	0.01	Cirrhosis, chronic alcoholism
7.	0.02	0.01	0(0.0063)	GSW chest, homicide
8.	0.02	0(0.0009)	0.03	Intracerebral bleed
9.	0.03	0(0.0047)	0.02	Diabetic, decomposition
10.	0.03	0.01	0(0.0089)	GSW head, homicide
11.	0.03	0(0.0018)	0.06	Drug overdose
12.	0.03	0(0.0067)	0.02	Drug overdose
13.	0.03	0(0.0000)	0.01	Suicidal GSW chest
14.	0.04	0(0.0000)	0.01	Blunt force injuries, accident; mild decomposition

BAC = blood ethanol concentration.
VAC = vitreous humor ethanol concentration.
UAC = urine ethanol concentration.
MI = myocardial infarction.
GSW = gun shot wound.
All concentrations are g/dL. The numbers in parentheses represents the apparent ethanol concentrations.

mation. Cases with BAC ≥ VAC ≥ UAC can be explained pharmacokinetically; if the individual were in the absorptive phase, then this pattern would be expected [9]. Four of the 13 cases demonstrated this pattern.

One assumption in the interpretation of this data is that a positive VAC and UAC indicate antemortem consumption and not postmortem ethanol formation. Although glucose is present in the vitreous humor, microorganism contamination of the fluid is limited during the early stages of the decomposition process. In healthy individuals, no glucose should appear in the urine. The urine should also be free of microorganism contamination. One report of in vitro formation of ethanol in urine was from a diabetic patient infected with *Candida albicans* [3].

Data from Table 1 suggests that a BAC of 0.01 g/dL is associated with a positive VAC and/or UAC approximately half of the time. This implies that a BAC of 0.01 g/dL can result from antemortem ethanol consumption or postmortem ethanol formation with about equal probability. At a BAC of 0.03 g/dL, the probability is favoring antemortem ethanol consumption; greater than two-thirds of the cases with a BAC of 0.03 g/dL were associated with positive VAC and/or UAC. At a BAC of 0.04 g/dL greater than 90% of cases were associated with a positive VAC and/or UAC. This suggests that in the absence of vitreous humor or urine, one can reasonably predict that a BAC of 0.04 g/dL resulted from ethanol consumption.

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