

R. E. Zumwalt,¹ M.D., R. O. Bost,² Ph.D., and
Irving Sunshine,³ Ph.D.

Evaluation of Ethanol Concentrations in Decomposed Bodies

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ABSTRACT: Blood samples obtained from 130 putrefied bodies were studied for endogenous ethanol (postmortem neoformation). An objective method of establishing the degree of putrefaction was developed and correlated with the determined ethanol concentration. The ethanol concentration of the vitreous humor was found to be helpful in establishing whether any ethanol detected in blood or putrefied fluid samples was endogenous or exogenous.

KEYWORDS: pathology and biology, alcohol, toxicology, vitreous humor, bacteriology, ethanol, postmortem decomposition

Determining whether ethanol identified in the blood or putrefied fluid of a decomposing body was ingested before death (exogenous) or was formed in the body after death (endogenous) is a frequent forensic science problem. In making this determination, forensic scientists have relied on circumstantial evidence and chemical evaluations of other body fluids and tissues. If a person had no documented access to alcohol before death, ethanol found in his putrefied blood was assumed to have been formed after death [1]. The lack of ethanol in a deceased's urine also suggested that ethanol found in the blood was endogenous [2,3]. In this study, the amount of ethanol in vitreous humor was ascertained to determine if this result could contribute to the assessment of postmortem alcohol formation.

Endogenous ethanol is a result of microbial activity, primarily on glucose [4], and to a lesser extent on lactate, glycerol, and amino acids [5]. Some microorganisms will form ethanol and others will not, and those that form ethanol may use different biochemical pathways to accomplish this change [5]. Attempts have been made to establish the maximum amount of postmortem ethanol production [1,6-8]. Different investigators have come to different conclusions [9]. No two people are biologically alike, and no two bodies have the same potential for postmortem ethanol production. Our studies of specimens from a large number of putrefied bodies assessed whether or not any ethanol detected was endogenous or

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¹Assistant professor of pathology, University of Cincinnati College of Medicine, and deputy coroner, Hamilton County, OH; formerly, deputy coroner, Cuyahoga County, OH, and assistant professor of forensic pathology, Case Western Reserve University, Cleveland.

²Associate toxicologist, Cuyahoga County Coroner's Office, Cleveland, OH, and instructor in toxicology, Case Western Reserve University, Cleveland.

³Chief toxicologist, Cuyahoga County Coroner's Office, Cleveland, OH, and professor of toxicology, Case Western Reserve University, Cleveland.

exogenous, reevaluated the range of postmortem ethanol production, and attempted to correlate the degree of putrefaction with the amount of endogenous ethanol. The specimens analyzed were blood, vitreous humor, and urine. The use of the ethanol analyses of these specimens was evaluated as an indicator of postmortem ethanol formation.

Material and Methods

One hundred thirty decomposing bodies received at the Cuyahoga County Coroner's Office (Cleveland, OH) were evaluated for degree of putrefaction and for the concentrations of ethanol in their "blood," urine, and vitreous humor. Designation of degree of putrefaction as mild, moderate, or severe was inadequate and was abandoned because such assessments were subjective and were not reproducible. An objective system of establishing the degree of putrefaction was developed. Eight physical changes that may occur in putrefaction were defined (Table 1): skin slippage, mummification, changes in the eyes, marbling, presence or absence of rigor mortis, bloating, purging of fluids from mouth and nostrils, and discoloration. The system became known by the acronym SMELLBAD. A point value was developed for each change based on the extent that each of these changes was present. The sum of the assigned points was used to define four categories of putrefaction: mild (less than five points); mild to moderate (six to twelve points); moderate (13 to 20 points); and severe (21 to 29 points). In autopsied cases, "blood" was removed directly from the heart. If no "blood" was present in the heart or vessels, putrefied fluid from the pleural cavities was used. In nonautopsied cases, heart blood or putrefied pleural fluid was recovered by syringe and large bore needle. In tabulating our results, we did not distinguish between "blood" and putrefied pleural fluid. Urine samples were taken directly in autopsied cases or removed by syringe in nonautopsied cases. Vitreous humor was removed by syringe and 18-gauge needle.

"Blood" samples were preserved with calcium oxalate and sodium fluoride. No preservative was added to urine or vitreous humor samples. An aliquot of each specimen was diluted with an aqueous solution of *n*-propanol as the internal standard in the subsequent analysis. The samples were then tested for ethanol using a Perkin-Elmer F-42 headspace gas chromatograph containing a column packed with Carbowax B coated with 5% Carbowax 20M. Because our procedure often reported background concentrations of ethanol of 0.5 to 2.0 mg/dL and because concentrations less than 10 mg/dL were not considered to be significant for our purposes, a cutoff level of 10 mg/dL was adopted.

In cases where no ethanol was detected in any sample, neither antemortem consumption (exogenous) or postmortem formation (endogenous) had occurred. When blood or putrefied fluid contained ethanol and other samples did not, the ethanol present was considered to be endogenous. If all specimens contained ethanol, some or all of the ethanol was considered to be exogenous. When only one sample was obtained and it contained ethanol, no determination of endogenous or exogenous source was made.

Results

Of the 130 cases in the study, 23% were mildly decomposed, 25% were in the mild to moderate stage of decomposition, 32% were moderately decomposed, and 18% were severely decomposed (Table 2). The three desired specimens, "blood," urine, and vitreous humor, were obtained in 24% (31 cases). Blood and urine were obtained in 12% (15 cases). Urine was available in 35% of the putrefied bodies (46 cases), while vitreous humor was present in 70% of bodies (91 cases) (Table 3). In order to determine the extent of bacterial infiltration into the vitreous humor, samples from several decomposed bodies (and one nondecomposed body as a control) were submitted for bacteriological examination. Even for cases of moderate decomposition, very few bacteria were detected (Table 4). Thus, the probability of bacterial action resulting in neof ormation of ethanol in vitreous humor was judged to be negligible.

TABLE 1—SMELLBAD system of grading eight physical changes that may occur in putrefaction of human bodies.

Characteristics	Rating					Total Possible Points ^a
	1 Point	2 Points	3 Points	4 Points	5 Points	
S—skin slippage	slippage over areas of pressure	skin slips easily with handling	sloughing of skin without pressure	generalized slippage (except where mummified)	...	4
M—mummification	drying of lips, finger tips, and exposed mucous membranes	drying and shrinking of fingers and toes	drying of hands and feet and around eyes	extensive drying with distortion of facial features	all skin leather-like	5
E—eyes	cloudy cornea	eyes soft, slightly sunken	eyes collapse with pressure, some vitreous left	no vitreous	...	4
L—livid streaks, marbling	few streaks along vessels of torso	marbling of torso and extremities	generalized marbling	3
L—limpness (rigor)	receding rigor	no remaining rigor	2
B—bloating	mild abdominal distension	abdominal distention and subcutaneous emphysema	possible protrusions of viscera through other orifices	breakdown of skin	...	4
A—appalling effusions (purging)	slightly malodorous; bloody fluid from mouth and nose	gray or red effusions from mouth and nose	malodorous fluids from all orifices	3
D—discoloration	green discoloration of right lower quadrant	patchy green and purple discoloration of abdomen and torso	purple and green discoloration of torso and extremities	generalized discoloration	...	4

^aOverall total = 29.

TABLE 2—Number and percentage of cases studied by stage of decomposition.

Stage of Decomposition	Number of Cases	Percentage
Mild to moderate	30	23
Mild to moderate	33	25
Moderate	42	32
Severe	23	18
Unclassified	2	2
Total	130	100

TABLE 3—Samples obtained in 130 decomposing bodies.^a

Specimen	Number of Cases	Percentage
Blood only	24	18
Vitreous humor only	3	2
Blood and urine	15	12
Blood and vitreous humor	57	44
Blood, urine, and vitreous humor	31	24
Totals	130	100

^aVitreous humor obtained in 70% of all cases; urine obtained in 35% of all cases.

TABLE 4—Vitreous humor bacteriology.

Case	Degree of Decomposition	Results
180301	moderate	few <i>Proteus morganii</i> (<i>Morganella morganii</i>)
181402	mild/moderate	one half dozen mixed gram + bacteria, none predominating
181446	moderate	no growth
181448	mild/moderate	few colonies <i>Clostridium</i> species
180491	mild/moderate	few slow growing anaerobes, including "bacter" species and anaerobic streptococcus
181670	mild/moderate	no growth
180305	control	no growth

In mildly decomposed bodies, 80% contained no ethanol. This compares with our experience that 80% of all coroner's cases are negative for ethanol. There was only one case in the mildly decomposed group in which postmortem production of ethanol (endogenous) could be presumed. Among the 33 bodies in the mild to moderate stage of decomposition, 18 (55%) had no ethanol. Endogenous ethanol was presumed in nine cases (27%). No determination about the source of ethanol present could be reached in two cases because only one specimen was available for analysis.

Endogenous ethanol was presumed in 19% (eight cases) of the 42 moderately decomposed bodies, while no ethanol was found in 12 (29%). Exogenous ethanol was found in 14 (33%) and no determination could be reached in seven cases (17%). For the 23 severely decomposed bodies, 13% (three cases) exhibited endogenous ethanol, while no ethanol was detected in three (13%). Exogenous ethanol was found in seven (30%) and no determination was reached in ten (43%).

Twenty-three of the 130 cases had presumed endogenous ethanol. Nineteen of the 23 cases had levels of 70 mg/dL or less. The other four had levels of 110, 120, 130, and 220 mg/dL. Cases with presumed endogenous ethanol are listed in Table 5.

Discussion

Since ethanol may be a casual or contributory factor in deaths, it is essential that forensic scientists determine whether the ethanol found in the "blood" and other fluids from putrefying bodies is exogenous or endogenous. Although circumstantial evidence may give presumptive evidence that no alcohol was ingested in a particular case, this is hearsay and has minimal value. Furthermore, in most cases no such evidence is available. Finding other putrefactive products, particularly other alcohols, may suggest that the ethanol identified in putrefied blood was also endogenous [3]. However, most investigators do not attempt to detect those products because usual methods are insensitive to the low concentrations of these products.

Much importance has been placed on the determination of ethanol concentration in the urine obtained from putrefied bodies [2]. Ethanol will appear in the urine shortly after ingestion, but generally will not be formed in the urine during putrefaction [7,8]. Thus, the determination of the presence of ethanol in "blood" but not in the urine of a putrefied body has suggested that the ethanol in the "blood" is endogenous. However, frequently there is no urine in the bladder, precluding any such interpretation.

Some vitreous humor usually remains until advanced putrefaction, and only a small volume is needed for analysis. The use of vitreous humor for postmortem ethanol analysis is well established and recent studies have compared the vitreous humor and blood ethanol levels [10,11]. Although vitreous humor contains glucose, infiltration of bacteria into the vitreous humor does not occur appreciably until late in the putrefactive process. Thus, any ethanol found in the vitreous humor would be presumed to be exogenous. Vitreous humor is

TABLE 5—Twenty-three cases with presumed endogenous ethanol.^a

Number	Case	Age	Sex	Race	Ethanol Concentration, mg/dL		
					Blood	Vitreous	Urine
1	175823	84	f	w	10	neg	...
2	176433	76	f	w	10	neg	...
3	176575	73	f	w	10	neg	...
4	176596	64	m	w	10	neg	...
5	173466	57	m	w	20	...	neg
6	175843	60	m	w	20	...	neg
7	176284	67	f	w	20	neg	...
8	181620	71	m	w	20	neg	neg
9	172682	50	m	b	30	neg	...
10	175563	62	m	w	30	neg	...
11	172949	54	f	b	40	neg	...
12	173404	87	f	w	40	neg	...
13	175882	19	m	w	40	neg	...
14	173723	64	m	w	50	neg	...
15	173922	64	m	w	50	neg	...
16	175474	29	m	w	50	neg	...
17	177521	75	m	w	50	neg	...
18	174630	87	m	w	60	neg	neg
19	176965	65	m	w	70	neg	...
20	174887	28	m	b	110	neg	neg
21	174886	40	f	b	120	...	neg
22	176698	69	f	w	130	neg	...
23	174670	47	m	b	220	...	neg

^aNeg = negative; ... = no sample obtained.

readily accessible in most bodies, and in the present series was obtained in twice as many cases (70% versus 35%) as was urine. Therefore, vitreous humor may be preferred over urine as a corroborating sample in the determination of exogenous or endogenous ethanol.

Using our criteria for endogenous ethanol production, there were four instances in our series where greater than 100 mg/dL ethanol was found in the blood.

Conclusion

An objective method for assessing the degree of putrefaction has been developed. This method has been applied in a study of postmortem ethanol formation in 130 bodies. Analysis for ethanol in "blood" in a decomposing body is important and can give reliable and useful information. In a significant proportion of cases, the ethanol found can be classified as exogenous or endogenous. In order to properly evaluate the antimortem ethanol status of the deceased, more than one specimen should be analyzed. Vitreous humor and urine are both suitable additional specimens, but because of its greater availability, vitreous humor may be more frequently used in these investigations.

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Address requests for reprints or additional information to
 Ross E. Zumwalt, M.D.
 University of Cincinnati
 College of Medicine
 231 Bethesda Ave.
 Cincinnati, OH 45267