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Graham R. Jones,[1] *Ph.D.; Peter P. Singer,*[1] *Ph.D.; and Katherine Rittenbach,*[2] *B.A.*

The Relationship of Methanol and Formate Concentrations in Fatalities Where Methanol Is Detected

ABSTRACT: An automated headspace gas chromatography method was developed for the determination of formate (formic acid) in postmortem specimens, based on the *in situ* sulfuric acid–methanol methylation of formic acid to methyl formate. Diisopropyl ether was used as an internal standard. The method was applied to over 150 postmortem cases where methanol was detected. Of the 153 cases presented, 107 deaths were attributed to acute methanol toxicity. In the vast majority of the remaining 46 deaths, the methanol was determined to be present as a postmortem or perimortem artifact, or was otherwise incidental to the cause of death. Of the 76 victims who were found dead and blood was collected by the medical examiner, all but one had a postmortem blood formate concentration greater than 0.50 g/L (mean 0.85 g/L; $n = 74$). The sole exception involved suicidal ingestion of methanol where the blood methanol concentration was 7.9 g/L (790 mg/100 mL) and blood formate 0.12 g/L. In 97% (72/74) of the cases where blood was available, the blood formate was between 0.60 and 1.40 g/L. In 31 of the 153 cases, the victim was hospitalized and blood obtained on admission or soon after was analyzed for methanol and formate during the subsequent death investigation; the vast majority (27/30) had antemortem blood formate concentrations greater than 0.50 g/L. Cases with samples taken prior to death with blood formate concentrations less than 0.5 g/L can readily be explained by active treatment such as dialysis. The blood formate method has also been useful in confirming probable perimortem or postmortem contamination of one of more fluids or tissues with methanol (e.g., windshield washer fluid or embalming fluid), where methanol ingestion was unlikely.

KEYWORDS: forensic science, methanol, formate, formic acid, postmortem blood, vitreous, poisoning

Methanol is widely available, especially in colder parts of the world where it is commonly used as an antifreeze for both gasoline and windshield washer fluid. Methanol is colorless and has a taste and odor only subtly different from that of ethanol. If mixed with beverages (e.g., orange juice or other fruit drinks), it is difficult to distinguish from ethanol.

Methanol is metabolized to formaldehyde and formic acid (formate), and further to carbon dioxide and water (1–6). Methanol itself is thought to have low direct toxicity, in part because there is a relatively poor correlation between methanol blood or serum concentrations and toxicity or mortality (3,7–10). Formaldehyde is highly toxic but has a short half-life and does not accumulate (11). Formate does accumulate *in vivo* and is thought to be the primary toxic agent produced as a result of methanol metabolism (3,10–15). Because methanol is widely used as a solvent, antifreeze and embalming fluid, it can sometimes be difficult to differentiate ingestion from postmortem contamination on the basis of methanol measurement alone, especially if only one specimen is available. However, measurement of formate can readily differentiate ingestion and metabolism from simple contamination.

The flame ionization detectors (FIDs) normally used to measure methanol and ethanol are almost nonresponsive to formic acid. Enzymatic methods have been used to measure formate (16,17). Headspace gas chromatography (HSGC) with *in situ* methylation has been reported for the detection of formate using sodium propionate as an internal standard (18); HSGC has been used by other investigators employing valeric acid as the internal standard (19,20).

[1]Office of the Chief Medical Examiner, Edmonton, AB.
[2]Neurochemical Research Unit, Department of Psychiatry, University of Alberta, Edmonton, AB.

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We present a HSGC method for the determination of formate in whole blood and other postmortem fluids, which we have applied to over 150 cases where methanol has been detected, since 1986.

Materials and Methods

Selection of Cases

All Medical Examiner-investigated deaths that occurred in Alberta between 1986 and 2005 were reviewed. Cases, where methanol was detected, were included with three exceptions: those where the body was embalmed, those where the postmortem blood or vitreous methanol concentration was <0.2 g/L (20 mg/100 mL), or those cases where death was delayed due to the sequelae of methanol poisoning and a hospital admission blood sample was unavailable. The postmortem cases given in Table 1, were virtually all found dead, or died soon after being found. The cases given in Table 2 are all different from those in Table 1; they are cases where the victim was hospitalized and admission blood was available and analyzed; in most cases there was a significant period of survival (hours to days) before death eventually occurred.

Reagents

Sodium formate, methanol, sulfuric acid, diisopropyl ether, and other reagents were of analytical grade; solvents were commercially distilled-in-glass and used without further purification.

Ethanol and Methanol Assay

Ethanol and methanol concentrations were determined by HSGC with FID, using a minor modification of a published method (21). The analytical column was 2×3 mm OD Nickel 200 column

TABLE 1—*Concentrations of methanol, formate, and ethanol in postmortem blood, vitreous humor, urine, and bile (g/L) in cases where death was attributed to methanol toxicity.*

	Methanol				Formate				Ethanol	
Case No.	Blood	Vitreous	Urine	Bile	Blood	Vitreous	Urine	Bile	Blood	Circumstances
1	4.9	5.3	5.6	–	0.83	0.65	–	–	0.0	Died on the route to hospital
2	3.1	3.2	4.4	–	1.06	0.53	4.00	–	0.0	Found dead in hotel room
3	1.7	1.8	2.0	–	0.74	–	5.60	–	0.0	Found dead in residence
4	5.2	5.3	6.5	–	1.00	0.98	4.80	–	0.0	Found dead in field
5	1.3	1.6	1.9	1.4	0.72	0.98	14.60	1.60	0.0	Died in hospital emergency department
6	3.0	3.7	4.1	3.4	1.07	0.94	10.40	1.43	0.0	Suicide; found in ex-wife's house
7	3.4	4.6	–	–	0.79	0.77	–	–	0.0	Found dead at home; drinking methanol earlier
8	6.5	7.9	8.0	–	0.94	0.86	5.90	–	0.0	Found dead in bed
9	1.3	1.6	–	1.5	0.85	–	–	1.50	0.0	(History not available)
10	4.0	5.1	5.5	–	1.03	0.85	5.90	–	0.0	Found dead in half-way house
11	2.7	3.3	3.3	3.0	0.64	0.56	4.70	2.10	0.0	Found dead in bed
12	1.2	–	1.0	1.2	0.65	–	2.07	0.63	0.0	Found dead in hotel room
13	0.9	1.0	1.0	1.0	0.57	0.30	4.60	1.70	0.0	Found dead in bedroom
14	0.7	0.8	0.9	–	0.69	0.88	1.38	–	0.0	Drank methanol & slashed wrists
15	6.3	9.3	7.4	–	0.66	1.23	5.20	–	0.0	Drank windshield washer fluid; dead at home
16	4.7	5.6	–	–	1.10	0.80	–	–	0.0	Found dead at home
17	2.6	3.1	3.6	–	1.20	1.20	4.70	–	0.0	Found dead in bed
18	3.7	4.2	4.5	–	1.14	1.40	2.81	–	0.0	Found dead at home
19	2.4	2.9	–	–	0.83	0.79	–	–	1.3	Drinking methanol from vodka bottle
20	–	3.2	–	–	–	0.71	–	–	0.0	Found dead on couch at home
21	4.3	5.1	–	5.0	0.92	0.72	–	2.02	0.0	Found dead in bed at home
22	4.4	5.2	–	–	0.88	0.87	–	–	0.0	Found dead at home
23	1.4	–	–	–	0.74	–	–	–	0.0	Difficulty breathing; died 3 h later
24	1.5	1.7	2.0	–	1.40	–	14.70	–	0.0	Found dead: suicide note
25	1.8	2.0	–	1.9	0.82	0.84	–	0.90	0.0	Developed seizures; died later in hospital
26	7.9	9.3	4.1	–	0.12	0.04	0.18	–	0.0	Found dead at home; methanol suicide
27	5.7	6.5	6.6	6.2	1.30	0.96	5.50	1.20	0.0	Found dead in bed
28	1.2	1.5	1.6	1.3	0.74	0.75	8.70	0.58	0.0	Found dead on kitchen floor
29	0.7	0.8	–	0.7	0.80	1.04	–	0.47	0.0	Headache, vomiting; died after 4 h in hospital
30	4.0	5.0	5.0	–	1.03	0.98	3.00	–	0.0	Found dead in his room
31	2.0	2.3	2.3	–	0.87	0.86	3.60	–	0.0	Found dead in his room
32	4.4	5.1	5.4	4.8	0.91	0.67	5.50	2.10	0.0	Found dead in bathroom
33	6.7	7.9	8.3	–	1.30	6.80	7.80	2.20	0.0	Drinking windshield washer fluid
34	5.6	6.0	7.3	–	0.62	0.99	4.80	–	0.2	Suicide note; dead in apartment
35	0.9	0.9	1.2	–	0.79	0.97	5.26	–	0.0	Found dead at home
36	3.7	4.2	4.4	3.8	0.89	0.74	3.50	2.60	0.0	Found dead in bed at home
37	3.8	4.5	4.7	4.4	0.97	1.10	6.40	2.50	0.0	Death in police cell
38	4.7	6.1	5.9	–	0.86	0.84	8.30	–	0.0	Found dead in bed; drinking 5 days
39	5.6	6.2	–	–	0.98	0.70	–	–	0.0	Found dead in her home
40	0.8	1.0	1.1	–	0.60	0.70	6.70	–	0.0	Diabetic found dead at home
41	4.1	5.3	5.6	–	0.70	0.91	11.00	–	0.0	Found dead in home
42	4.8	–	–	–	0.65	–	–	0.81	0.0	Acute methanol toxicity
43	3.5	4.4	4.8	–	1.25	0.82	4.11	–	0.0	Drinking windshield washer fluid
44	3.4	4.3	4.6	–	0.74	0.87	4.19	–	0.0	Dead in hotel bed; drinking windshield washer fluid
45	4.5	5.2	4.9	–	0.74	0.60	3.52	–	0.0	Found dead at home
46	1.1	–	1.1	1.2	0.80	–	0.51	0.95	0.0	Found dead at home on couch
47	1.9	2.2	–	2.2	0.80	0.81	–	2.10	0.0	Found dead in his apartment
48	2.5	3.2	3.3	–	0.77	0.84	10.00	–	0.0	Found dead in his home
49	2.6	3.2	3.1	–	0.83	0.83	2.70	–	0.2	Found dead in friend's garage
50	2.1	2.0		2.3	1.05	0.82	–	0.92	0.0	Ethanol abuse, depression, found dead by parents
51	4.3	5.2	5.5	–	0.95	0.81	7.30	–	0.0	Alcoholism, depressed; found dead by wife
52	1.7	2.2	–	–	0.90	0.92	–	–	0.0	Suicidal methanol poisoning
53	4.0	5.1	–	–	0.64	0.71	–	–	0.0	History of alcohol abuse
54	1.2	1.5	–	–	0.81	0.85	–	–	0.0	Suicidal methanol poisoning
55	1.8	2.2	–	–	0.80	1.20	–	–	0.0	History of alcohol abuse
56	1.1	–	1.6	1.2	0.78	–	1.90	0.56	0.3	Found dead in his residence; decomposed
57	2.2	2.6	3.0	–	0.73	1.59	11.70	–	0.0	Alcoholic found dead by roommate
58	5.9	6.7	7.2	–	0.92	0.96	–	–	0.0	Alcoholism
59	4.4	–	5.7	4.4	0.84	–	4.70	1.54	0.0	Alcoholic, depressed
60	3.8	4.8	4.3	–	0.72	0.30	–	–	0.0	Dead outside with windshield fluid; suicide
61	5.0	6.1	–	–	1.00	0.91	–	–	0.0	Chronic alcoholic
62	2.1	2.5	2.8	–	0.82	0.74	–	–	0.0	Found dead with windshield washer fluid
63	4.8	5.6	6.5	–	0.84	0.84	5.40	–	0.0	Arrested, later found dead in cells
64	3.6	4.0	–	3.9	0.87	0.73	–	0.89	0.0	Drug and ethanol abuse; pregnant
65	2.5	3.2	–	2.9	0.75	0.72	–	–	0.0	Alcoholic, depression
66	–	2.6	–	3.0	–	0.86	–	1.26	0 (Vi)	Alcoholism and depression, heart disease
67	1.1	1.4	1.5	–	0.79	0.90	1.87	–	0.0	Laying by camper, suicide note
68	1.8	2.3	–	–	0.67	0.70	–	–	0.0	Alcohol abuse, found dead in mobile home
69	5.4	–	6.9	–	0.94	–	7.30	–	0.0	Alcoholism, hepatitis C

TABLE 1—*(Continued)*

	Methanol				Formate				Ethanol	
Case No.	Blood	Vitreous	Urine	Bile	Blood	Vitreous	Urine	Bile	Blood	Circumstances
70	6.9	8.5	9.3	–	1.00	1.00	–	–	0.0	Ethanol abuse and depression, suicidal, lost job
71	3.0	3.9	4.4	–	0.90	0.85	6.40	–	0.0	Depression and alcoholism
72	3.7	3.8	4.5	–	0.63	0.73	3.00	–	0.0	Alcoholism and heart disease, ran out of money
73	3.7	4.5	–	–	0.78	0.77	–	–	0.0	Drinking antifreeze
74	4.0	5.0	5.5	–	0.83	0.88	6.10	–	0.0	Drinking antifreeze
75	1.5	1.9	2.0	–	0.73	0.79	4.70	–	0.0	Alcoholism; found dead
76	2.1	2.6	2.8	–	0.66	0.64	6.45	–	0.0	Found dead after drinking unknown liquid

Vi, vitreous humor; –, specimen not available or analysis not performed.

TABLE 2—*Concentrations of methanol, formate, and ethanol in antemortem blood and urine (g/L) in cases where death was eventually attributed to methanol toxicity.*

	Methanol		Formate		Ethanol	
Case No.	Blood	Urine	Blood	Urine	Blood	Circumstances
77	3.0	–	0.77	–	1.6	Unknown
78	2.3	–	0.98	–	0.0	Alcohol abuse
79	–	0.9	–	4.60	0.0	Alcohol abuse
80	5.3	–	0.53	–	0.0	Alcohol abuse
81	1.6	–	0.55	–	0.9	Suicide by methanol and meprobamate
82	3.7	–	0.89	–	0.0	Accidental methanol poisoning
83	4.5	–	0.96	–	0.0	Depression; suicidal methanol poisoning
84	4.9	–	0.83	–	0.0	Chronic alcohol abuse
85	1.3	–	0.56	–	1.9	Chronic alcohol abuse
86	4.0	–	0.78	–	0.4	Alcohol abuse; methanol and solvent containers
87	3.5	–	1.70	–	0.0	Found unconscious in hotel
88	2.4	–	0.68	–	0.0	Alcohol abuse; drank windshield washer fluid
89	3.7	–	0.78	–	0.0	History of substance abuse
90	4.0	–	0.65	–	0.0	Died on weekend pass from rehab center
91	4.7	–	0.75	–	0.0	Chronic alcohol abuse
92	0.9	–	0.04	–	0.0	Acute methanol toxicity; delayed death
93	1.4	–	0.75	–	1.2	History of alcoholism
94	0.3	–	0.58	–	0.6	Alcoholic; drank windshield washer fluid
95	0.5	–	0.23	–	0.8	Accidental—thought was drinking ethanol
96	3.2	–	1.10	–	0.0	Drank windshield washer fluid
97	4.5	–	1.10	–	0.0	History of ethanol abuse
98	1.2	–	0.40	–	0.0	Chest pain; deteriorating vision
99	2.0	–	0.52	–	0.8	Alcoholic admitted to hospital with back pain
100	1.2	–	0.77	–	0.0	History of alcohol abuse
101	1.0	–	0.80	–	0.0	Admitted to hospital for vision loss
102	3.2	–	1.10	–	0.3	Alcoholism, recently lost job; methanol toxicity
103	3.2	–	0.72	–	0.0	Alcoholism and depression, admitted nausea, diarrhea
104	2.9	–	0.84	–	0.0	Previous suicide attempts, drank 2 cups windshield fluid
105	4.7	–	0.93	–	0.0	Drank antifreeze, died in hospital
106	4.2	–	0.69	–	0.0	Alcoholism, found confused, seizures, died in hospital
107	1.4	–	0.84	–	0.0	Drinking from methanol bottle; died in hospital 9 days later

–, specimen not available or analysis not performed.

packed with 5% Carbowax 20 M on Carbopack B 60/80 mesh (Supelco #1-1766, Bellefonte, PA). In mid-1995, the column was changed to a Rtx-BAC1 column, 30 m × 0.53 mm ID, 3.0 μM film thickness (Restek #18001, Bellefonte, PA).

Formate Assay

Whole blood (0.1 mL) or other specimen was pipetted into a 20-mL capacity headspace vial. Methanolic internal standard (20 μL of 0.125% v/v diisopropyl ether in methanol) and concentrated sulfuric acid (36 M, 20 μL) were then added, and the vial was immediately capped and vortexed for 5 sec. Calibrators were prepared by adding known concentrations of sodium formate to previously tested whole blood (fresh human or sheep blood) at concentrations of 0, 0.1, 0.2, 0.5, 1.0, and 2.0 g/L. Outdated blood bank blood was found not to be suitable due to unidentified interference. An in-house whole blood control was independently prepared at a concentration of 0.6 g/L. The blank, calibrators, and in-house control were run singly; specimens were run in duplicate. Example chromatograms for a blank, calibrator, and a case are shown in Fig. 1.

Headspace Gas Chromatography

A Perkin Elmer F-45 HSGC, equipped with FID and a 2 × 3 mm Nickel 200 column packed with 10% SP-1000 on 80/100 Supelcoport (Supelco #1-872), was used until May 1995. The carrier gas was nitrogen (flow 30 mL/min). Conditions: vial oven 60°C, needle temperature 150°C, injector 120°C, oven 85°C, detector 130°C, sample time 4 sec, sample equilibrium 20 min.

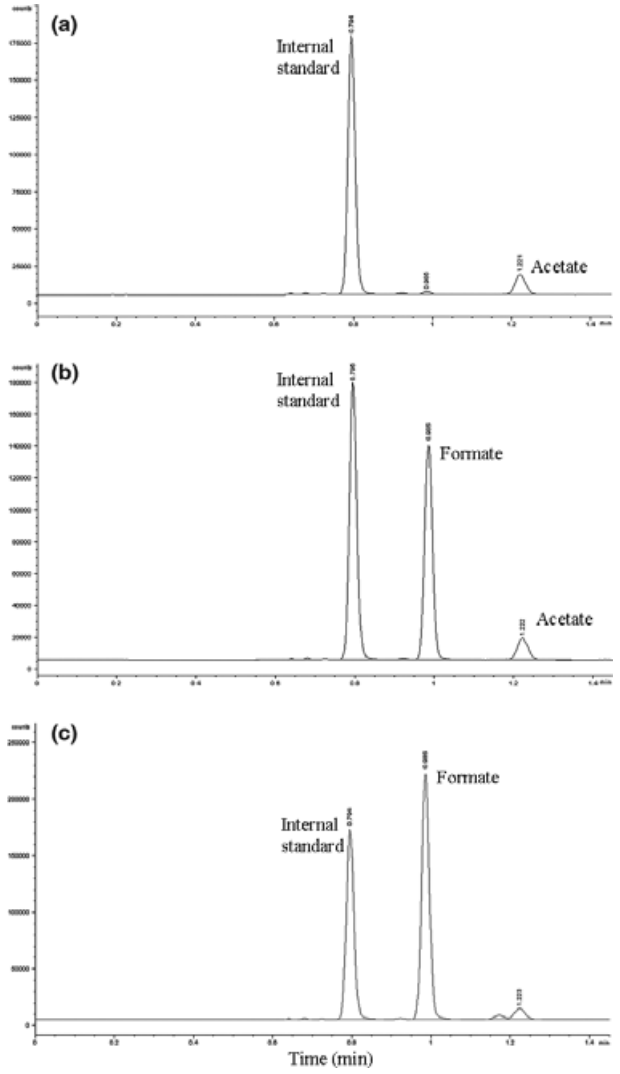

FIG. 1—*Headspace GC/ flame ionization detectors (FID) chromatograms of blood formate determinations: (a) blank blood, (b) 0.5 g/L blood-based calibrator, (c) case blood with 0.89 g/L formate.*

Chromatography and integration of peak areas were measured on a Hewlett Packard 3390 integrator. Typical retention times were as follows: diisopropyl ether 0.90 min, methyl formate 1.20 min, ethyl formate 1.53 min, methyl acetate 1.55 min, and methanol 1.90 min.

From May 1995 to October 1998, a Tekmar 7000 series HSGC linked to a Hewlett Packard 5890 Series II GC with FID was used, equipped with a Stabilwax fused silica column, 30 × 0.53 mm ID, 1.0-μm film thickness (Restek #10655). The column was connected to the carrier gas line of the injection port and run in the split mode (*c.*1:10). The transfer line back pressure (about 7 psi) was adjusted to give an injection port pressure of 5 psi. Conditions: vial oven 60°C, transfer line 70°C, GC injector 120°C, oven 50°C, detector 200°C. The sample equilibration time was 20 min. Chromatography and integration of peak areas were measured on an HP 3390 ChemStation. Typical retention times were as follows: diisopropyl ether 1.33 min, methyl formate 1.66 min, methyl acetate 2.06 min, and methanol 2.66 min. After October 1998 the Tekmar headspace unit was replaced by a HP 7694 headspace unit, run with the same GC and column under similar conditions.

The method has an LOD of 0.025 g/L and an administrative LOQ of 0.1 g/L formate in postmortem blood. Calibrations were run with five points covering 0.1–2.0 g/L and were typically linear with a correlation coefficient (r^2) of better than 0.999. The precision is estimated as 7% CV based on analysis of 76 control samples run over the last 10 years (spiked value averaging 0.57 g/L; experimental mean 0.559 ± 0.039 g/L).

Formate Assay Variables

The following variables were examined during development of the assay: volumes of specimen, methanol, and concentrated sulfuric acid; reaction temperature (HSGC-heated sample block); length of time in the heated block; and the effect of elevated ethanol concentrations.

Assay Results and Discussion

Formate Assay Variables

Diisopropyl ether was chosen as an internal standard rather than a short-chain carboxylic acid to avoid the potential for interference from naturally occurring substances—especially in postmortem blood. The volume of methanol reagent was set at 20 μL and was not changed. Sulfuric acid volumes of 5, 10, 20, and 50 μL resulted in 0%, 80%, 95%, and 90%–95% maximum reaction, respectively and so 20 μL was optimal. The reaction time was evaluated at 60°C for 10, 20, 30, and 45 min. The reaction was about 80%–90% of maximum at 10 min; all other reaction times gave close to the maximum response. With instruments where the vials are heated in a single fixed platen (combined tray and oven; e.g., Perkin Elmer F-45), it was determined that reproducible results could be obtained up to heating time of about 90 min; absolute recoveries declined thereafter. The reaction was examined at vial oven temperatures of 60°C and 80°C. The 80°C temperature gave slightly better response, but this was probably attributable to greater partition into the headspace rather than a more complete reaction. A vial oven temperature of 60°C was chosen for convenience (the same temperature as our ethanol assay), and to enhance stability of the reaction mixture in analyzers without constant heating (e.g., Perkin Elmer F-45). The specimen volume (whole blood) was examined at 0.1 and 1.0 mL, with the volumes of reagents scaled accordingly. The 0.1 mL volume gave better sensitivity and precision, possibly due to the smaller clot and larger headspace volume compared with 1.0 mL of sample.

In postmortem case samples, ethanol is often present in varying amounts. Ethanol, at concentrations of 240 and 400 mg/100 mL, spiked into 0.5 g/L of formate standards caused no measurable reduction in the concentration of formate. It should be noted that at very high concentrations, ethanol slowly reacts with formic acid with sulfuric acid catalysis to produce ethyl formate. In addition, the presence of small amounts of endogenous acetate in the body, or the higher concentrations formed by the metabolism of ethanol, result in the formation of methyl acetate. Under both the chromatographic conditions described above, ethyl formate and methyl acetate co-elute and are separated from the target analyte, methyl formate, and therefore do not cause interference.

Case Results and Discussion

Methanol-Caused Fatalities

The deaths attributed to methanol poisoning where the victim was found dead are summarized in Table 1 (cases 1–76). Table 2 lists additional cases where death was attributed to methanol poisoning, but where the victim was found alive and hospitalized,

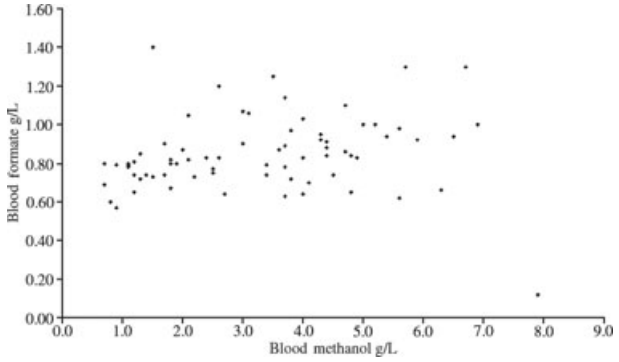

FIG. 2—*Correlation of postmortem blood methanol versus blood formate concentrations in 73 methanol fatalities (r = 0.1463).*

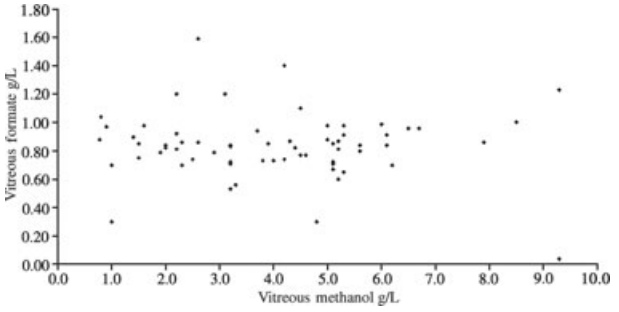

FIG. 3—*Correlation of vitreous methanol versus vitreous formate concentrations in 64 methanol fatalities (r = 0.0632; excluding case 33).*

death was delayed, and where at least one blood or urine sample close to the time of admission to hospital was available (cases 77–107). The correlation between methanol and formate concentrations in blood is poor ($r = 0.1463$), as shown in Fig. 2. Postmortem blood methanol concentrations in the methanol-caused fatalities varied widely, ranging from 0.7 to 7.9 g/L (mean 3.3 g/L; cases 1–76, $n = 74$). The vitreous methanol concentrations also varied considerably, ranging from 0.8–9.3 g/L (mean 3.95 g/L; $n = 69$); see Fig. 3.

In contrast, most postmortem blood formate concentrations fell into a much narrower range of 0.53–1.40 g/L (mean 0.86 g/L; $n = 73$, case 26 excluded—this case had an extremely high blood methanol concentration of 7.9 g/L and an unusually low formate of 0.12 g/l, indicating that direct methanol toxicity was likely the mechanism of death, rather than formate toxicity). Although many other cases contained methanol concentrations that were well above 4.0 g/L (400 mg/100 mL), it must be borne in mind that methanol has as much as twofold less *direct* toxicity than ethanol (22). The toxicity of alcohols increases as the carbon number increases (23).

However 97% of the methanol-caused fatalities had blood formate concentrations in the range 0.60–1.10 g/L (Fig. 2). The mean vitreous formate concentration is 0.84 g/L ($n = 64$, range 0.30–1.59 g/L, excluding cases 26 and 33) giving a vitreous:blood formate ratio of 0.98. Case 33 was reported to have an extremely high vitreous formate concentration of 6.8 g/L, just over eight times the mean for all postmortem cases, and suspected to reflect a reporting error. Unfortunately, the original analytical data for this case is no longer available for review.

In contrast, the formate concentrations in urine are almost invariably much higher than in the blood or vitreous humor suggesting some degree of active transport. Other studies have also shown unequal distribution of formate with high levels in the kidneys (18). The ratio of concentrations of methanol in blood versus vitreous is consistent with that reported for ethanol, as might be expected because methanol has similar physical properties and volume of distribution ($r = 0.9859$; $y = 1.1959x - 0.0044$; Fig. 4). Cursory examination of the data for postmortem blood and vitreous formate concentrations indicates that all are high and of the same general order of magnitude. Also, all the blood formate concentrations and the vast majority of vitreous formate concentrations in postmortem methanol caused fatalities are around 0.5 g/L or greater. However, the quantitative correlation between vitreous and postmortem blood formate concentrations is relatively poor ($r = 0.2646$; Fig. 5).

It is noteworthy that 88% of the deaths attributed to methanol poisonings were male victims and that most had a well-established history of chronic ethanol abuse or strong suspicion thereof.

As expected, in those cases where the individual was found critically ill, but ultimately died despite extensive medical treatment, the formate concentrations in the admission antemortem blood samples are comparable to those in the postmortem blood of the victims found dead. In those cases where dialysis and other medical treatment prolonged survival, the methanol and formate concentrations at the time of death were considerably lower or zero (data not presented). In many of these cases, ethanol was administered as active treatment to inhibit methanol metabolism to formate (10). In those cases where medical treatment with ethanol had been instituted, but where ethanol was not present at the time of death, the medical records indicated it had been discontinued when the blood

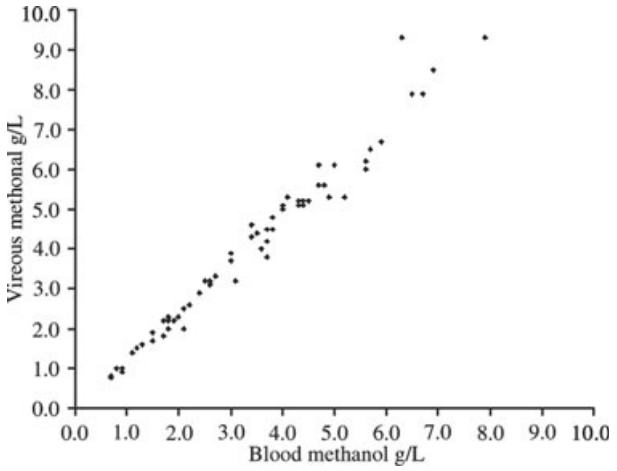

FIG. 4—*Correlation of blood methanol versus vitreous methanol concentrations in 66 postmortem cases (r = 0.9859).*

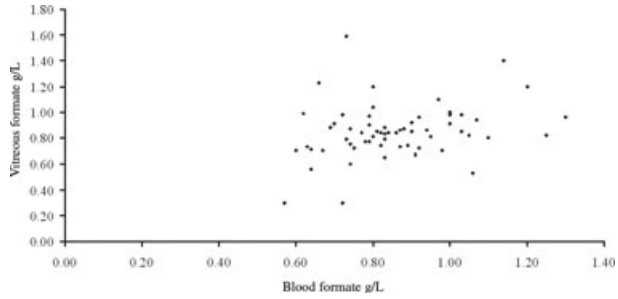

FIG. 5—*Correlation of postmortem blood formate versus vitreous formate concentrations in 61 methanol fatality cases (r = 0.2646; excluding cases 26 and 33).*

methanol was close to zero, but where hypoxic brain damage was recognized as irreversible and life support discontinued. In all of the deaths in this category, the medical cause of death was attributed to methanol poisoning or the consequences thereof (e.g., severe acidosis or hypoxic brain damage). The mean postmortem blood and antemortem blood formate concentrations were similar (mean 0.85 g/L, $n = 73$ and mean 0.76 g/L, $n = 30$, respectively).

Not surprisingly, ethanol was absent in virtually all (72/76) postmortem cases. Ethanol is preferentially metabolized and blocks the metabolism of methanol to formaldehyde and formate. Therefore any of the victims who were consuming ethanol prior to or during the consumption of methanol would have cleared the ethanol first, and only then started to metabolize the methanol, initiating the accumulation of formate and subsequent toxicity. The ethanol detected in case 19 was the result of the therapeutic administration of ethanol during medical treatment—death occurring shortly after due to the already advanced state of methanol/formate toxicity. The other cases with low concentrations of ethanol can easily be attributed to postmortem formation.

Nonmethanol-Caused Fatalities

The cases in Table 3 are fatalities attributed to causes other than methanol poisoning and warrant further explanation. The distribution of methanol in all but three of the cases 108–138 is not consistent with the ingestion of methanol during life, because either methanol is present in only one specimen and is very low or absent in others, or formate was absent. In cases 109, 110, 128, and 135, the deaths were classified as suicidal due to the causes other than methanol toxicity (gunshot wound or carbon monoxide poisoning)

TABLE 3—*Concentrations of methanol and formate in postmortem blood, vitreous humor, urine, and bile (g/L) in cases where death was attributed to causes other than methanol toxicity.*

	Methanol				Formate				Ethanol	
Case No.	Blood	Vitreous	Urine	Bile	Blood	Vitreous	Urine	Bile	Blood	Circumstances
108			5.3				0.00		0 (Ur)	MVA driver; went through stop sign
109	2.9		4.1		0.53		7.90		–	Methanol intoxication and GSW suicide
110	0.2	0.2	0.2		0.23	0.40	3.90		0.0	GSW suicide
111	0.6								–	MVA
112	0.1	0.2							–	Asphyxia/hanging
113	0.5				0.07				0.3	Gastrointestinal hemorrhage
114	0.0	1.6	0.0						3.8	GSW head
115	0.3	0.0	1.6		0.00	0.00	0.00		3.0	MVA driver: rolled vehicle
116	1.2	5.4	0.0		0.00	0.00	0.00		3.6	MVA driver: rolled vehicle
117		0.2	0.2			0.00	0.00		0 (Vi)	Presumed carbon monoxide poisoning
118	0.0	1.7	0.0		0.00	0.00	0.00		0.2	GSW suicide
119	3.3				0.10				1.3	GSW head
120			2.3	1.9					–	GSW head
121	0.8				0.00				1.8	MVA with drowning
122	0.0	2.7			0.00	0.00			2.0	Suicidal hanging
123	0.6	0.5			0.00	0.00			0.0	MVA; driver of semi-trailer
124	0.2	0.0							1.6	Massive closed head injury
125	0.2	0.1							–	Carbon monoxide poisoning
126	0.0	1.0	0.0		0.00	0.00			0.4	MVA/train—car driver
127	1.1	7.7			0.00	0.00			0.4	GSW Suicide; samples collected funeral home
128		0.5				0.12			0 (Vi)	GSW to head
129	10.7				0.00				0.2	GSW suicide
130	0.0	0.3	0.0						0.3	Pedestrian hit by car on highway
131	0.2								0.0	MVA driver with massive head and chest injury
132	0.3								0.0	Cold exposure
133	0.5	0.1							0.2	GSW suicide
134	0.2	0.3	0.3		0.10	0.00			2.2	Blunt head trauma
135		0.9				0.22			0 (Vi)	Presumed CO toxicity in garage (no blood)
136		0.9				0.00			0 (Vi)	MVA passenger with massive injuries
137	0.0	5.1	0.0						1.4	Cardiovascular accident
138	0.6	0.0			0.00				0.0	Cardiac arrhythmia
Solvent abuse cases										
139	2.9	6.3	0.3		0.00	0.00	0.00		2.6	Lacquer thinner abuse: toluene, MEBK, methanol
140	0.3	0.4							0.6	Lacquer thinner absue
141	0.2		0.2	0.2					0.0	Solvent abuse
142	0.4								0.5	Laqcuer thinner toxicity; toluene, MEK
143	0.3		0.3		0.00		0.06		0.0	Toluene and methylethylketone toxicity
145	0.2	0.2		0.2					0.0	Acute solvent toxicity
144	0.2	0.2		0.2					0.0	Acute solvent toxicity
146	0.2		0.1	0.2					0.8	Acute solvent toxicity
147	0.0	0.2							0.0	Drug overdose and solvent abuse
148	0.2		0.0						1.3	Acute solvent and ethanol toxicity
149	0.4	0.5	0.5		0.22	0.18	1.70		0.0	Combined toxicity of toluene and methanol
150	0.3	0.3	0.3						2.2	Solvent abuse and drowning
151	0.1	0.3	0.1						0.0	Acute organic solvent toxicity
Formaldehyde poisonings										
152	0.3	0.3	0.2		0.37	0.12	0.08		0.4	Formaldehyde overdose (suicide)
153	0.3	0.4	0.3	0.4	0.72	0.57	0.41	1.87	0.0	Formaldehyde toxicity (suicide)

Vi, vitreous humor; –, specimen not available or analysis not performed; Ur, urine; GSW, gunshot wound; MVA, motor vehicle accident; MEK, methylethylketone; MEBK, methylethylbutyl ketone.

and the presence of methanol and formate is attributed to co-ingestion of methanol (especially case 109). In all other cases, the presence of methanol was presumed to be due to inadvertent contamination. Such contamination may typically be from one of two sources. Many rural medical examiners collect specimens at the local funeral home, and may inadvertently use a syringe contaminated with a residue of embalming fluid. Such as scenario is likely for case 127, where both the blood and vitreous contain methanol, but not formate. Apart from the lack of formate in this case, the distribution of methanol in the blood and vitreous is not consistent with ingestion during life. The second source of methanol, although difficult to prove, is from windshield washer fluid in motor vehicle accidents. In a violent motor vehicle accident involving the front end of a vehicle, it is likely that the windshield washer container would burst and if the firewall is breached can result in windshield washer fluid being sprayed over the driver and passenger. In such cases, the most exposed tissue where methanol could easily be absorbed would be the eye.

However, contamination of other tissue is possible if the chest or abdomen is injured, such as case 108, where methanol and traces of isopropanol were detected in the "urine," liver, and spleen. The front of the body of the victim was massively disrupted, exposing the abdomen. This explained contamination of the liver and spleen (3.0, 1.9 g/kg liver; and 70, 20 g/kg spleen) where the first and second values for each organ were from sampling of the outer and inner surfaces of the tissues, respectively). However, it did not explain how methanol came to be in the urine at such a high concentration (5.3 g/L), until it was revealed that the bladder had been torn open during the collision, and therefore easily contaminated with windshield washer fluid.

In case 113, a death attributed to a massive GI hemorrhage in a chronic alcoholic, it was not possible to determine if methanol was ingested prior to death, as only blood was available for analysis.

In cases 139–151, the presence and distribution of methanol are consistent with the reported history of solvent abuse, mostly lacquer thinner or similar solvents containing low concentrations of methanol, in addition to acetone, toluene, and methylethylketone; determination of formate in case 149 confirms exposure to methanol prior to death.

Cases 152 and 153 are included for completeness. Both had a history that indicated the probable ingestion of formaldehyde solution. Although methanol is present in most commercial formaldehyde solutions as a preservative, the relatively high-formate concentrations in these two cases are consistent with metabolism of ingested formaldehyde to formate prior to death (24).

Conclusions

Formate concentrations in blood or vitreous humor >0.5 g/L are highly correlated with fatal outcome in methanol poisoning cases, unlike methanol concentrations alone. The presence of formate concentrations >0.5 g/L in blood or vitreous humor can strongly indicate that (i) the methanol was ingested during life, (ii) the methanol and formate were at least potentially life-threatening, and therefore that (iii) the methanol was not present as an artifact or an accident or of specimen collection. Similarly, finding methanol in the absence of formate, or where blood or vitreous humor formate concentrations are substantially <0.5 g/L, or finding the presence of methanol but at markedly different concentrations in blood compared with vitreous humor (or *vice versa*), strongly indicates that the methanol may have been introduced into one or more parts of the body as an artifact and is not indicative of ingestion during life.

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Additional information and reprint requests:
Graham R. Jones, Ph.D., D-ABFT
Alberta Medical Examiner's Office
7007-116 Street NW
Edmonton
Alberta T6H 5R8
Canada
E-mail: graham.jones@gov.ab.ca