ELSEVIER

Forensic Science International 133 (2003) 152–158

Forensic Science International

www.elsevier.com/locate/forsciint

Post-mortem analysis of formic acid disposition in acute methanol intoxication

Luis A. Ferrari[a,b], Miriam G. Arado[a,b], Cesar A. Nardo[a], Leda Giannuzzi[c,*]

[a]*Laboratorio de Toxicología y Química Legal, Dirección General de Asesorías Periciales-Suprema Corte de Justicia Provincia de Buenos Aires, Calle 41 y 119 (1900) La Plata, Argentina*

[b]*Cátedra de Toxicología, Universidad de Morón, Cabildo 134 (1708) Moron, Argentina*

[c]*Cátedra Toxicología y Química Legal, Facultad de Ciencias Exactas, Universidad Nacional de La Plata, Calle 47 y 115 (1900), La Plata, Argentina*

Received 23 August 2002; received in revised form 5 February 2003; accepted 5 February 2003

Abstract

Fifteen cases of fatal massive methanol intoxication have been investigated. Victims received either no treatment or ethanol therapeutic treatment. Methanol poisoning cases were classified in three groups according to survival time: more than 3 days (group 1), up to 3 days (group 2) and few hours (group 3). Body distribution of methanol and formic acid, as the main metabolite, was analyzed in blood and in different organs (brain, kidney, lung and liver). Relationships between formic acid concentration in the different tissues, survival time and type of treatment applied to victims were studied. Formic acid in blood and tissues was analyzed by head space gas chromatography (head space-GC) with FID detector, previous transformation in methyl formate, essentially as described by Abolin. Formic acid concentration was between 0.03 and 1.10 g/l in the samples under study. A good correlation between blood and brain, but poor between blood and the remaining tissues was found. Obtained data suggested that the use of blood and brain could help to improve the analysis of formic acid intoxication. The best correlation among organs was found between lung and kidney for all groups ($r^2 = 0.91$, 0.84 and 0.87, corresponding to groups 1, 2 and 3, respectively). Lethality index was defined as LI = (concentration of formic acid in blood in (g/l)/0.5) × 100, taking into account that 0.5 g/l is the concentration reported by Mahieu in severe methanol poisoning. LI parameter was used to estimate formic acid incidence on the lethality of methanol poisoning cases. LI showed a good correlation with total formic acid concentration of the different tissues analyzed ($r^2 = 0.80$). Furthermore, LI allowed us to discriminate between individuals that received therapeutic treatment and survived different periods. LI > 100 indicated a severe intoxication and short survival time if the victim was assisted with ethanol therapy and hemodialysis was not applied. With regard to victims who received no therapeutic treatment and died in few hours, LI was in the range 40–100. LI was below 40 for individuals that survived more than 3 days and hemodyalisis was not performed. Results showed the importance of performing formic acid analysis to diagnose severe methanol intoxication in post-mortem cases.

Keywords: Methanol intoxication; Formic acid; Poisoning index; Post-mortem analysis; Tissue concentration

1. Introduction

Methanol poisoning by ingestion is a world-wide problem, and in some regions it is connected with high morbidity and mortality [1–3]. Methanol or wood alcohol finds extensive use in industry. It is present as a contaminant in many commercial wines and other alcoholic drinks in low concentration [4,5]. Inhalation, topical exposure and oral routes absorb methyl alcohol rapidly and well. It is metabolized to formaldehyde and formic acid by hepatic dehydrogenases and toxic effects appear to be related to high levels of formic acid [6]. Severe anion gap metabolic acidosis in

* Corresponding author.
E-mail addresses: laferrari@unimoron.edu.ar (L.A. Ferrari), leda@biol.unlp.edu.ar (L. Giannuzzi).

0379-0738/03/$ – see front matter
doi:10.1016/S0379-0738(03)00071-9

the marker of the poisoning is primarily the result of the increase of formic acid concentration [7–9]. The accumulation of formic acid has been detected in many human methanol poisoning cases [10–13]. Two pathways have been suggested for the disposition of formic acid: oxidation either through the catalase-peroxidative system or through the one-carbon pool. The catalase system appears to be poor in rats and monkeys probably due to the low level of peroxidative capacity of the hepatic system and the low-level activity of peroxide-generating oxidases. An alternative pathway for the metabolism of formic acid is a tetrahydrofolic acid (THF)-dependent one-carbon pool. Formic acid enters this pool by combining with tetrahydrofolic acid to 10-formyl-THF. The ATP-dependent reaction is catalyzed by 10-formyl-THF synthetase, a ubiquitous enzyme in mammalian tissues. Thus, two mechanisms may be operative in explaining slow formic acid oxidation causing accumulation of the acid in humans, mainly low hepatic THF levels and reduced hepatic 10-formyl-THF dehydrogenase activity [14,15].

Apart from severe metabolic acidosis, renal insufficiency and respiratory failure characterize methanol poisoning [16]. An understanding of the mechanism of toxicity, treatment protocol, and clinical course is essential. Successful patient outcomes depend on calculation of anion and osmolar gaps and quantitative analysis of methanol and formic acid in a biological fluid such as serum or plasma.

Analysis of blood and tissue formic acid concentration in post-mortem cases would be relevant in assessing methanol poisoning, principally methanol is absent when therapeutic treatment is carried out during several days before death [17].

There are a few published reports on the determination of formic acid levels resulting from methanol intoxication [13,18–21]. Neither are contributions that considered the relationship between formic acid concentration in different organs, survival time and existence or absence of a therapeutic treatment.

This work is aimed at determining the distribution of formic acid in blood and tissue of 15 out of 47 fatal victims who died due to a massive intoxication as a result of methanol adulterated white wine ingestion, during February and September 1993 in Argentina. The correlation between formic acid and survival time of victims was analyzed as well.

2. Material and methods

2.1. Samples

Fifteen fatal victims were studied. All autopsies involved major organs analysis together with a review of clinical histories. Blood, brain, lung and kidney were analyzed to determine methanol and formic acid. Blood specimens were taken from the femoral region and were placed in tubes without preservative at 4 °C. The viscera were stored frozen until analyzed.

2.2. Methods

Determinations of methanol and formic acid were performed using head space gas chromatography (head space-GC) method with FID detector. Methanol dosage was performed according to our routine methodology. Either 1 ml blood or 1 g chopped viscera were received in 1 ml saturated solution of potassium carbonate and 1 ml isopropyl alcohol (1‰ final concentration) used as an internal standard. The samples were placed in 5 ml glass vials covered with rubber caps and sealed. Firstly, they were incubated at 30 °C for 30 min and then at 60 °C for another 45 min. Afterwards, 0.4–0.6 ml of the air space phase were withdrawn with a disposable syringe.

Formic acid dosage was determined previous transformation in methyl formate according to Abolin et al. [22], using a different column that improved separation. Either 500 μl blood or 0.5 g of tissue were placed in a tube with 250 μl of concentrate sulfuric acid (EM-Science-Merck, SX-1244-S, Germany); the tube was sealed with laboratory film. It was shaken and incubated for 20 min, cooled at room temperature avoiding contact of the acid with the plastic material. Then, 15 μl acetonitrile solution (Merck-Schuchardt, 5-16-27-44, Munchen; 0.197 M), as internal standard, and 15 μl methanol were added. After mixing, preparation was incubated for 20 min at room temperature and gently shaken. Finally, 0.4–0.6 ml of airspace were injected in a Shimadzu GC 14 gas chromatograph.

For methanol, a stainless steel column (2 m long, 3 mm internal diameter), packed with 0.3% carbowax and 1500-graphapack 60/80 (EMQ-All Tech) was used at isothermal conditions (100 °C). FID detector connected to a Shimadzu C-R4A Chromatopac integrator was used. Both injector and detector operated at 150 °C. N_2 was the gas carrier with a constant flow of 40 ml/min. Both air and H_2 pressure in the detector were 5 psi. The whole equipment was calibrated using standard methanol (EM-Science-Merck, MX-0475-1, Germany) solutions within the range 0–4 g/100 g. Formic acid was run in a megabore column, DB-WAX (J & W), 30 m long and 0.53 mm internal diameter. The oven temperature was held firstly, at 35 °C for 1 min and then raised at 10 °C/min to the final temperature, 100 °C.

2.3. Classification of victims in groups according to methanol poisoning

A preliminary classification of methanol poisoning cases was established considering whether victims received or not treatment and survival time informed in clinical history. The therapeutic treatment consisted in the administration of 100 mg ethanol, 5% dextrose solution and bicarbonate.

Three groups were defined as follows: group 1, those who received treatment and survived more than 3 days

(five cases), group 2, those who received the specific therapy and survived up to 3 days (four cases) and group 3, those who received no treatment and died in few hours (six cases).

3. Results and discussion

3.1. Body distribution of methanol and formic acid in man

Fig. 1 shows the average concentration of methanol and formic acid in different biological matrixes (blood, liver, lung, brain and kidney) in the three groups of victims analyzed. Bars indicate the range of concentration found. The highest methanol concentrations were observed in victims of group 3, followed by those of group 2; no methanol was detected in victims of group 1, regardless of the analyzed matrix. In decreasing order, average methanol concentrations in brain, blood, kidney and lung were 1.98, 1.75, 1.68 and 1.41 g/l, respectively. The concentration of methanol in the different organs and blood informed in the present work compared well with those reported for other methanol fatalities [23–25]. Fatal levels of methanol in blood have been reported over a wide range (0.2–6.3 g/l) [24] although the major toxic effects are primarily due to the matabolite formic acid and not unchanged methanol. Other authors informed that blood methanol concentration in 20 fatal cases averaged 1.9 g/l within the same range, 0.2–6.3 g/l [25,26]. In this work, the high concentration found in brain (higher than blood values) is greater than its theoretical value, considering its content in water. This indicates that methanol distribution is not similar to that reported for ethanol [26]. Pla et al. [24] arrived to a similar conclusion for liver. In the present work the highest level of methanol in viscera was detected in brain followed by kidney, lung and liver. It agrees with methanol distribution in rats reported by Barlett using ^{14}C-labeled methanol [27,28]. He found the highest concentrations in kidney, liver and gastrointestinal tract.

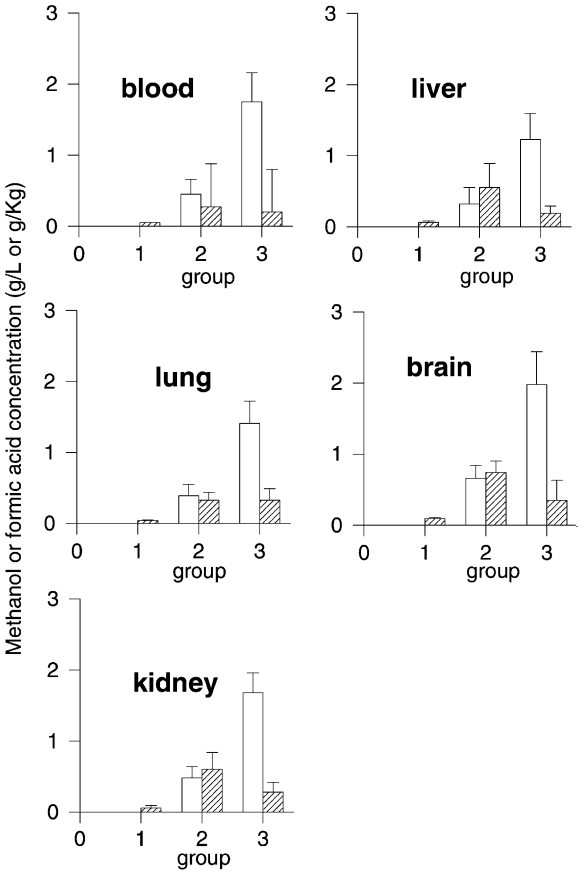

Fig. 1. Methanol (□) and formic acid concentration (▨) (g/l or g/kg) in groups 1–3 in blood, liver, lung, brain and kidney.

Fig. 2 shows ratios of methanol concentrations in blood to the different organs for groups 2 and 3. For group 1, no methanol was detected in any tissue or blood. The good correlation between the two groups, with a correlation coefficient $r^2 = 0.996$, indicates that equilibrium distribution has been achieved, and allowed to compare methanol concentration in different biological matrixes.

It is well known that the main metabolic pathway is the hepatic one, where the enzyme alcohol dehydrogenase oxidizes 60–70% of the toxic into formaldehyde, which is, in turn, enzymatically transformed into formic acid. Both lung and kidney are important disposal pathways, excreting 10–20 and 5–7% methanol, respectively [29].

Clinical observations of patients with methanol intoxication have revealed that the onset of symptoms and the development of metabolic acidosis began approximately 12–24 h after ingestion [9]. No relationship between metha-

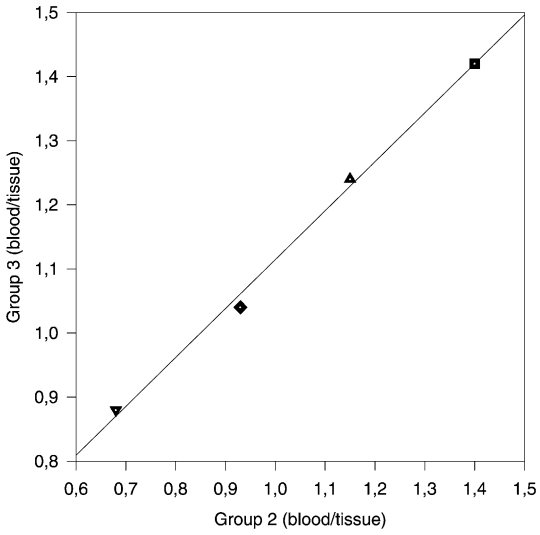

Fig. 2. Ratios of average concentrations of methanol in blood to different tissues for groups 2 and 3: (▼) blood/brain; (▲) blood/lung; (◆) blood/kidney; (■) blood/liver.

nol concentration in blood and the degree of toxicity was found. It is misleading to state that a specific quantity of methanol is a fatal dose. This is because the major toxic manifestations of methanol ingestion are due to the metabolite formic acid that is found in widely varying concentrations after consumption of a specific amount of methanol [30,31]. Thus, methanol should not be considered as a survival prognostic parameter. In cases of group 1 methanol was not found in blood nor in organs, however formic acid was present in all matrixes studied (Fig. 1). This is in agreement with Baselt and Cravey [4], who informed that blood methanol concentration is not necessarily a good prognostic index. That is why, a formic acid rather than methanol itself is the main toxic agent reported by different authors [2,6,7,13]. The highest levels of formic acid were found in victims belonging to group 2 for all viscera studied and brain was the predominant matrix (0.74 g/l). Therefore, the study of formic acid distribution in each victim became necessary.

3.2. *Post-mortem analysis of formic acid disposition in acute methanol intoxication*

Previous reports concerning formic acid measurement have been, mainly, confined to blood concentration. Reported concentrations were 0.31 g/l by Shahangian et al. [13]; 2.5–104 mg/ml by Fraser and MacNeil [18] and 0.015–0.19 mg/ml by Mahieu et al. [19]. Table 1 shows the post-mortem distribution of formic acid (g/l) in blood, liver, kidney, brain and lung. Besides, Table 1 includes total formic acid found in liver, lung, brain and kidney of each of the 15 victims (classified in three groups) after lethal methanol intoxication.

Formic acid concentration in blood was correlated with its concentration in the different studied organs. The best correlation corresponded to blood versus brain concentrations with $r^2 = 0.86$ (Fig. 3a). Correlation coefficients between blood and the other tissues were blood versus liver $r^2 = 0.69$, blood versus kidney $r^2 = 0.68$ and blood versus lung $r^2 = 0.56$.

The best correlation between tissues was found for lung versus kidney for each group and correlation coefficients for groups 1, 2 and 3 were 0.91, 0.84 and 0.87, respectively (Fig. 3b). Other tissue relationships (lung versus liver, lung versus brain, and brain versus liver) did not show a good correlation. These data suggested that concentrations in blood and brain can improve the interpretation of formic acid analysis; and moreover, the relationship between organs (lung and kidney) may help in the interpretation of methanol intoxication in post-mortem investigation.

The following equation was defined in order to study the degree of lethality due to formic acid in cases of methanol poisoning taking into account the survival time:

$$\mathrm{LI} = (\text{concentration of formic acid in blood in } (\mathrm{g/l})/0.5) \times 100$$

LI is defined as the lethality index and 0.5 is the level of formic acid reported in cases of methanol poisoning. Mahieu et al. [19] defined as criteria for predicting severe methanol poisoning a blood formic acid level above 0.5 g/l. LI is a good parameter to define the degree of lethality of formic acid in blood in cases of methanol poisoning. Additionally, the ratio of formic acid in different organs was investigated to study the post-mortem distribution of formic acid in different organs.

Table 1
Formic acid concentration (g/l or g/kg)) in different biological matrixes of 15 victims of methanol poisoning with wines

Victim	Blood	Liver	Lung	Brain	Kidney	Liver + lung + brain + kidney
Group 1: survival time more than 3 days						
1	0.08	0.09	0.05	0.11	0.06	0.31
2	0.07	0.07	0.03	0.10	0.03	0.23
3	0.12	0.03	0.04	0.09	0.04	0.20
4	0.08	0.08	0.05	0.12	0.07	0.32
5	0.06	0.08	0.03	0.07	0.04	0.22
Group 2: survival time up to 3 days						
6	0.56	0.88	0.47	0.50	0.86	2.71
7	0.56	0.15	0.19	0.74	0.29	1.28
8	0.91	0.62	0.40	0.82	0.60	2.44
9	0.69	0.52	0.38	1.10	0.82	2.82
Group 3: without treatment						
10	0.61	0.34	0.58	0.67	0.49	2.08
11	0.07	0.06	0.08	0.09	0.08	0.31
12	0.25	0.25	0.32	0.29	0.32	1.18
13	0.32	0.38	0.35	0.38	0.22	1.33
14	0.21	0.19	0.42	0.16	0.45	1.22
15	0.26	0.09	0.20	0.50	0.17	0.96

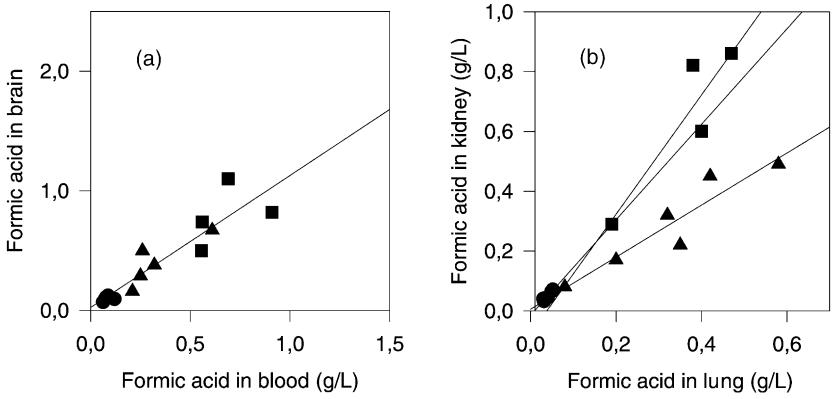

Fig. 3. Scattergram showing correlation between (a) brain formic acid and the corresponding blood concentration ($r^2 = 0.86$, $n = 15$); (b) kidney formic acid and the corresponding lung concentration. (●) Group 1; (■) group 2; (▲) group 3.

Fig. 4a–c shows the plot of LI versus different ratios of formic acid in kidney, brain, liver and lung corresponding to the 15 victims of the three informed groups and Tanaka et al. [20] data. Fig. 4 was divided in three zones depending on LI value, as follows: zone I with LI < 40, zone II with $40 <$ LI < 100 and zone III with LI > 100.

Victims of group 1 showed low LI values (zone I), low concentration of formic acid in liver and lung and higher concentrations in brain (Fig. 4a–c). Victims of group 3 fell in the intermediate zone with IL between 100 and 40 (zone II). The data of formic acid in blood and viscera reported by Tanaka et al. [20], corresponding to victims without therapeutic treatment, were close to group 3 data of the present work. For this group, the higher concentrations of formic acid were detected in brain and lung (Fig. 4b and c). Victims of group 2 presented values of IL > 100 (zone III) and higher concentrations of formic acid in brain (Fig. 4c). The analysis of formic acid ratios between different organs indicated that brain showed the highest concentrations among the three groups of victims, demonstrating the relevance of this organ in post-mortem investigations of methanol intoxications (Fig. 4). Besides, lung/kidney ratio allowed dividing the three

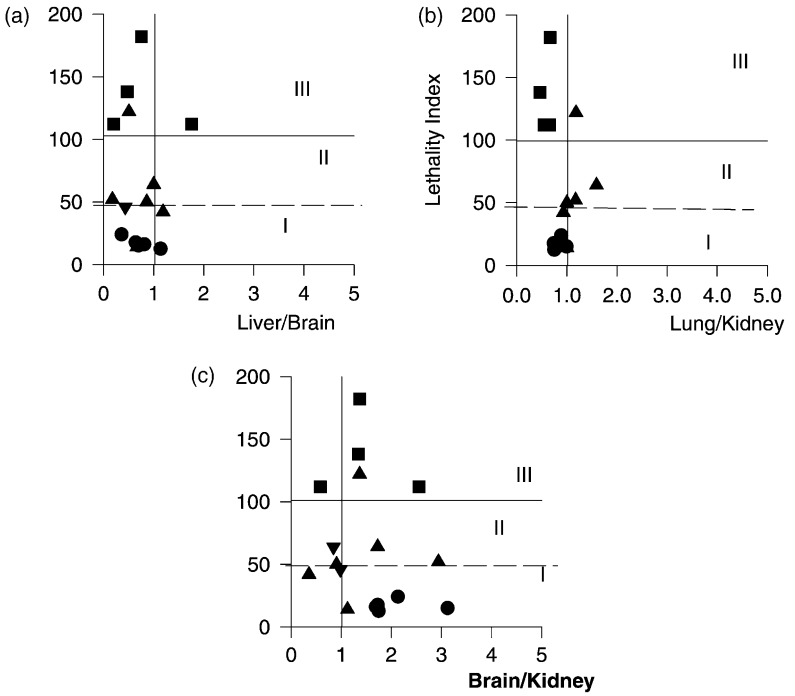

Fig. 4. Lethality index as a function of formic acid concentration ratios in different tissues: (●) group 1; (■) group 2; (▲) group 3; (▼) Tanaka data.

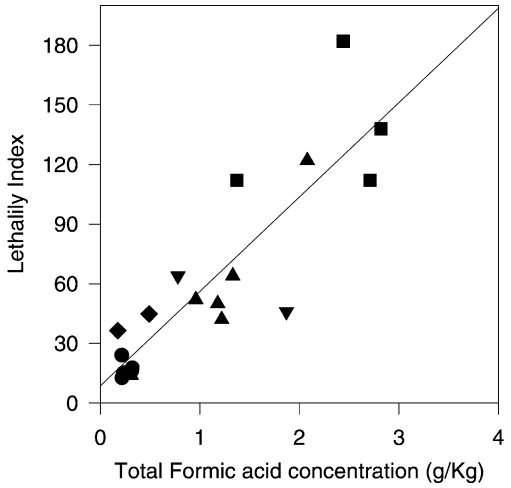

Fig. 5. Correlation between lethality index vs. total formic acid concentration (mg/l): (●) group 1; (■) group 2; (▲) group 3; (▼) Tanaka data; (◆) Hantson data.

groups of victims (according to their survival time) in well defined zones: group 1 with LI < 40 and lung/kidney < 1, group 2 with LI > 100 and lung/kidney < 1 and group 3 with $40 <$ LI < 100 and lung/kidney > 1. Considering the previous analysis where the best correlation was lung versus kidney (Fig. 3b), together with LI coefficient, a better interpretation of formic acid distribution in post-mortem processes according to survival time and type of therapeutic treatment applied was obtained. However, it should be taken into account that application of hemodialysis to patients markedly modified correlations between tissues.

LI showed a good correlation ($r^2 = 0.80$) with total formic acid in liver, lung, brain and kidney informed in Table 1 for each victim (Fig. 5), data of Tanaka et al. [20] and Hantson et al. [21] were considered as well. Values corresponding to patients that survived up to 3 days are more dispersed than those of Hantson et al. [21]. However, these authors stressed that the patients were submitted to hemodialysis, what implied a rapid elimination of formic acid and thus, led to very low formic acid levels in post-mortem analysis. In our case, patients were submitted to ethanol therapy but not to hemodialysis. This fact suggests that hemodialysis is a relevant parameter in the quantification of post-mortem findings of individuals that did not survive the therapy.

Thus, LI increases to values close to those informed by Mathieu et al. [19] for severe methanol intoxication. Considering that the cases of Hantson et al. [21] with a survival time between 2 and 3 days fell within our data of group 1 (survival time above 3 days), again we can see that the type of therapy influences formic acid concentration of post-mortem tissues. Also the higher dispersion degree of data of group 2 compared to those of the mentioned authors should be considered.

Group 1, corresponding to individuals that survive more than 3 days (one of them survived 10 days), showed low LI values and similar values of total formic acid in viscera (Fig. 5). This suggests that in cases with long survival time and when a therapy is applied, post-mortem values of formic acid will be very low. Lung/kidney ratio was below 1 opposite to cases of Hantson et al. [21] where the ratio was above 1. The hemodialysis performed in the mentioned cases may explain these differences.

It can be observed that higher formic acid concentration in blood and viscera correlated well with the severity of the effects as seen in our case studies, victims of group 2 received treatment and survived at least 3 days. These results are in agreement with those of Liesivuori and Savolainen, [6], who informed that formic acid concentration in blood and urine correlated well with the severity of effects as seen in experimental and clinical studies. This was not the case for methanol. This makes formic acid a better indicator of methanol poisoning. These results show that it would be necessary to perform formic acid and methanol dosage to asseverate the diagnosis of methanol intoxication both in fatal and non-fatal cases. Formic acid constitutes the best indicator when measured 48 h after methanol ingestion and the therapeutic treatment has been installed.

References

[1] T.A. Gossel, J.D. Bricker, Principles of Clinical Toxicology, Raven, New York, 1984, pp. 65–68.
[2] N.M. Cooper, G.N. Mitchell, I.L. Bennett, F.N. Cary, Methyl alcohol poisoning: an account of the 1951 Atlanta epidemic, J. Am. Med. Assoc. 141 (1952) 48–51.
[3] E.M. Scrimgeour, Outbreak of methanol and isopropanol poisoning in New Britain, Papua New Guinea, Med. J. Aust. 2 (1980) 36–38.
[4] R.C. Baselt, R.H. Cravey, Disposition of Toxic Drugs and Chemical in Man, fourth ed., Chemical Toxicology Institution, Foster City, 1995, pp. 519–521.
[5] M.J. Ellenhorn, D.G. Barceloux, Medical Toxicology: Diagnosis and Treatment of Human Poisoning, Elsevier, New York, 1988, pp. 801–804.
[6] J. Liesivuori, H. Savolainen, Methanol and formic acid toxicity: biochemical mechanism, Pharmacol. Toxicol. 69 (1991) 157–163.
[7] D. Jacobsen, K.E. McMartin, Methanol and ethylene glycol poisoning. Mechanism of toxicity, clinical course, diagnosis and treatment, Med. Toxicol. 1 (1986) 309–334.
[8] J.D. Morrow, H.J. Morgan, A case of methanol ingestion, J. Tenn. Med. Assoc. 81 (4) (1988) 246.
[9] L.C.D.R. King, Acute methanol poisoning: a case study, Heart Lung 21 (3) (1992) 260–264.
[10] K.E. McMartin, J.J. Ambre, T.R. Tephly, Methanol poisoning in human subjects: role of formic acid accumulation in the metabolic acidosis, Am. J. Med. 68 (1980) 414–418.
[11] O.M. Sejersted, D. Jacobsen, S. Ovbrevo, H. Jansen, Formate concentration in plasma from patients poisoned with methanol, Acta Med. Scand. 213 (1983) 105–110.
[12] A. Lund, Excretion of methanol and formic acid in man after methanol consumption, Acta Pharm. Toxicol. 4 (1948) 205–212.

[13] S. Shahangian, V.. Robinson, T.A. Jennison, Formate concentrations in a case of methanol ingestion, Clin. Chem. 30 (1984) 1413–1414.
[14] H.R. Witheley, The distribution of formate-activating enzyme involving tetrahydrofolic acid in animal tissue, Comp. Biochem. Physiol. 1 (1960) 222–247.
[15] A.B. Makar, T.R. Thephly, G. Sahin, G. Osweiler, Formate metabolism in young swine, Toxicol. Appl. Pharmacol. 105 (1990) 315–320.
[16] R.B. Naik, W.P. Stephens, D.J. Wilson, A. Walker, H.A. Lee, Ingestion of Formic acid containing agents-report of three fatal cases, J. Postgrad. Med. 56 (1980) 451–456.
[17] L.A. Ferrari, R.R. Nieto, M.G. Arado, Z. Wamba, Blood and tissue distribution of methanol and formic acid in victims of a massive intoxication due to the ingestion of adulterate wine, in: Proceedings of the TIAFT Meeting, Padova, Italy, 1997, pp. 26–32.
[18] A.D. Fraser, W. MacNeil, Gas chromatographic analysis of methyl formate and its application in methanol poisoning cases, J. Anal. Toxicol. 13 (1989) 73–76.
[19] P. Mahieu, A. Hassoun, R. Lauwerys, Predictors of methanol intoxication with unfavorable outcome, Hum. Toxicol. 8 (1989) 135–137.
[20] E. Tanaka, K. Honda, H. Horiguchi, S. Misawa, Postmortem determination of the biological distribution of formic acid in methanol intoxication, J. Forensic Sci. 36 (3) (1990) 936–938.
[21] P. Hantson, V. Haufroid, P. Mahieu, Determination of formic acid tissue and fluid concentrations in three fatalities due to methanol poisoning, Am. J. Forensic Med. Pathol. 21 (4) (2000) 335–338.
[22] C. Abolin, J.A. McRae, T.M. Tozer, S. Takki, Gas chromatographic headspace assay of formic acid as methyl formate in biologica fluids: potential application to methanol poisoning, Biochem. Med. 23 (1980) 209–218.
[23] I. Bennet Jr., F.H. Cary, G.L. Mitchell, M. Cooper, Acute methyl alcohol poisoning: a review based on experiences in an outbreak of 323 cases, Medicine 32 (1953) 431–463.
[24] A. Pla, A.F. Hernandez, F. Gil, M. Garcia Alonso, E. Villanueva, A fatal case of oral ingestión of methanol distribution in postmortem tissues and fluids including pericardial fluid and vitreous humor, Forensic Sci. Int. 49 (1991) 193–196.
[25] R.L. Kane, W. Talbert, J. Harlan, G. Sizemore, S. Cataland, A methanol poisoning outbreak in Kentucky, Arch. Environ. Health 17 (1968) 119–129.
[26] S.H.E. Tonkabony, Postmortem blood concentration in 17 cases of fatal poisoning from contraband vodka, J. Forensic. Sci. 6 (1975) 1–3.
[27] R.N. Harger, S.L. Johnson, E.G. Bridwell, Detection and estimation of methanol with results in human cases of methanol poisoning, J. Biol. Chem. 123 (Suppl.) (1938) 50–51.
[28] G.R. Barlett, Combustion of ^{14}C labeled methanol in intact rat and its isolated tissues, Am. J. Physiol. 163 (1950) 614.
[29] J.C. Mani, R. Pietruszko, H. Theorell, Methanol activity of alcohol dehydrogenases from human liver, horse liver and yeast, Arch. Biochem. Biophys. 140 (1970) 52–59.
[30] G. Martin-Amat, K.E. McMartin, S.S. Hayreh, T.R. Tephly, Methanol poisoning: ocular toxicity produced by formate, Toxicol. Appl. Pharmacol. 45 (1978) 201–208.
[31] J.D. Osterlok, S.M. Pond, S. Grady, C.E. Becker, Serum formate concentrations in methanol intoxication as a criterion for hemodyalisis, Ann. Intern. Med. 104 (1986) 200–203.