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Renal Effects of Low-dose Methoxyflurane with Cardiopulmonary Bypass

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In a randomized prospective study of ten patients anesthetized with small doses of methoxyflurane (mean arterial concentration 7.8 ± 1.0 mg/100 ml) for operations involving cardiopulmonary bypass, no changes in renal function were found. Results were compared with those in a control group of ten patients anesthetized with morphine–halothane. Mean peak serum inorganic fluoride concentration, although increased from 1.7 ± 0.3 μmol/l to 45.7 ± 4.2 μmol/l, was only 25–50 per cent of that previously found to produce renal dysfunction. These findings support the hypothesis that renal dysfunction associated with methoxyflurane anesthesia is dose-related. (Key words: Methoxyflurane; Nephrotoxicity; Cardiopulmonary bypass; Inorganic fluoride; Metabolism.)

IN A RECENT CONTROLLED STUDY, renal dysfunction was clinically evident in half of a group of surgical patients who received methoxyflurane anesthesia.[1] The degree of dysfunction correlated with increased serum and urinary concentrations of two methoxyflurane metabolites, inorganic fluoride and oxalic acid, suggesting a dose-related renal lesion.[2] Contrary to this hypothesis, end-alveolar methoxyflurane concentrations did not correlate with the development of polyuric nephropathy. Because patients undergoing operations with cardiopulmonary bypass need only relatively small total doses of anesthetic, this situation presented an opportunity to study the renal effects of measured small doses of methoxyflurane. Methoxyflurane is administered as the major anesthetic agent for about half of our approximately 800 patients who annually undergo operations with cardiopulmonary bypass. In view of the controversy over its use, it was important to determine its renal effects in this group.

Methods

Twenty male patients scheduled for cardiac valve replacement or coronary artery–saphenous vein bypass graft were randomly divided into two groups: ten patients received methoxyflurane and oxygen; the control group received morphine, 1.0–1.5 mg/kg, iv, with minimal halothane supplementation (0.3–0.5 per cent). All patients were A.S.A. physical status 4. Patients given methoxyflurane–oxygen anesthesia received diazepam, 10 mg, and atropine, 0.4 mg, im, as premedication; patients given morphine–oxygen anesthesia received morphine, 10 mg, and atropine, 0.4 mg. Induction of anesthesia in patients given methoxyflurane was by mask, with the addition of thiopental, 50–150 mg, iv, if excitement occurred. The vaporizer setting was 1.0 per cent for induction, the duration of which rarely exceeded ten minutes, and 0.1 to 0.3 per cent for maintenance. Succinylcholine, 1.0 mg/kg, was administered to facilitate endotracheal intubation, and *d*-tubocurarine, 0.3–0.6 mg/kg, for intraoperative muscle relaxation.

Control patients breathed 100 per cent oxygen while morphine, 1.0–1.5 mg/kg, was administered intravenously during a 10–15-minute period. If the patient was still conscious at the end of this time, 0.3–0.5 per cent halothane was introduced. Succinylcholine and *d*-tubocurarine were used for intubation

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TABLE 1. Mean Serum Values ± SEM

	Na^+ (mEq/l)	K^+ (mEq/l)	Creatinine (mg/100 ml)	Urea Nitrogen (mg/100 ml)	Uric Acid (mg/100 ml)	Osmolality (mOsm/kg)	F^- (μmol/l)
Methoxyflurane							
Preoperative	139.4 ±0.4	4.19 ±0.04	1.19 ±0.08	19.4 ±1.8	6.49 ±0.43	292.2 ±1.3	1.70 ±0.27
Postoperative days 1–4	136.8 ±0.5	4.32 ±0.07	1.12 ±0.05	21.3 ±2.0	5.69 ±0.71	287.5 ±1.8	31.87 ±3.99
Morphine							
Preoperative	139.8 ±0.6	4.12 ±0.17	1.03 ±0.03	15.3 ±1.6	6.13 ±0.52	290.3 ±1.3	1.2 ±0.1
Postoperative days 1–4	137.7 ±0.7	4.27 ±0.07	1.10 ±0.04	21.0 ±1.9	5.64 ±0.59	290.7 ±1.8	1.3 ±0.1
P,* methoxyflurane *vs.* morphine	>0.4	>0.5	<0.1	>0.1	>0.4	>0.1	<0.001

* Preanesthetic–postanesthetic differences.

and intraoperative muscle relaxation, respectively, as described above.

No volatile agent was administered during cardiopulmonary bypass; when excessive movement occurred, *d*-tubocurarine was given to patients anesthetized with methoxyflurane; morphine and *d*-tubocurarine to those anesthetized with morphine and halothane. A disposable bubble-type oxygenator with semi-occlusive roller pumps, primed with 5 per cent dextrose in Ringer's lactate solution, was used for cardiopulmonary bypass. The perfusion rate, initially 2.5–3.5 l/min, was adjusted to maintain a mean arterial pressure of approximately 80 mm Hg. Mannitol, 12.5 g, was added to the prime solution at the beginning of cardiopulmonary bypass and repeated at 60-minute intervals for as long as artificial circulation was in use. Body temperature was reduced to 33 C during cardiopulmonary bypass, then returned to normal prior to electrical defibrillation of the heart. Following cardiopulmonary bypass, 0.1–0.3 per cent methoxyflurane was administered intermittently to patients anesthetized with methoxyflurane and 0.3–0.5 per cent halothane to control patients if they appeared to be too lightly anesthetized.

Fluids administered intraoperatively consisted of 500–1,000 ml of 5 per cent dextrose in 0.2 per cent saline solution and whole blood as needed. Postoperatively, patients received approximately 2 l/day of 5 per cent dextrose in 0.2 per cent saline solution with 60 to 100 mEq of potassium chloride supplementation.

Electrocardiogram, pulse rate, direct arterial blood pressure, central venous pressure, esophageal temperature, and expiratory concentration of volatile anesthetic agent[3] were continuously monitored in every patient. Methoxyflurane concentrations in arterial blood immediately before cardiopulmonary bypass and just before the end of operation were determined by a direct-injection gas chromatographic method.[4] Arterial *p*H, P_{O_2}, and P_{CO_2} were measured at frequent intervals with an Instrumentation Laboratory Blood Gas Analyzer, Model 113.

Preoperative laboratory tests included determination of serum and 24-hour urinary sodium, potassium, urea nitrogen, creatinine, osmolality, uric acid, and inorganic fluoride levels[5] and urinary oxalic acid excretion.[6] Intake and output of fluids and body weight were measured daily. Creatinine, uric acid, and osmolal clearances were calculated, as were daily sodium, potassium, osmolal and fluoride excretions. Values for at least two preoperative days were obtained for each patient. Postoperatively, biochemical tests were repeated daily for a week. However, oxalic

TABLE 2. Mean Urinary Values ± SEM

	Na^+ (mEq/l)	K^+ (mEq/l)	Creatinine (mg/100 ml)	Urea Nitrogen (mg/100 ml)	Uric Acid (mg/100 ml)	Osmolality (mOsm/kg)	F^- (μmol/l)	Urine Volume (l/Day)
Methoxyflurane								
Preoperative	55.9 ±12.3	55.5 ±6.2	114.7 ±10.9	900.0 ±117.3	57.7 ±7.5	507.4 ±46.5	26.33 ±2.36	1.49 ±0.16
Postoperative days 1–4	50.9 ±7.4	81.9 ±5.0	117.7 ±8.3	1044.3 ±107.6	85.6 ±8.6	657.0 ±36.4	1686.17 ±296.92	1.41 ±0.11
Morphine								
Preoperative	60.8 ±13.7	55.6 ±8.4	119.9 ±14.4	865.8 ±124.9	53.2 ±6.0	538.9 ±66.1	41.45 ±5.31	1.37 ±0.23
Postoperative days 1–4	55.1 ±4.9	77.8 ±5.9	126.1 ±17.6	992.3 ±101.5	83.1 ±14.9	644.0 ±53.2	36.14 ±4.77	1.47 ±0.11
P^* methoxyflurane *vs.* morphine	>0.5	>0.5	>0.5	>0.5	>0.5	>0.3	<0.001	>0.3

* Preanesthetic–postanesthetic differences.

acid determinations were done only on the first two postoperative days because of the cost and time needed for analyses.

STATISTICAL METHODS

For each patient, the means of the data obtained on the first four postoperative days were calculated and compared with the individual's preoperative means. Only the values for the first four days were included because most patients were receiving oral alimentation after that time. In addition, mean preoperative–postoperative differences for each group were calculated and compared. Student's t test was used for statistical analyses.

Results

Preoperatively, there were no significant differences between the two groups (tables 1–3). Patients' ages, types and durations of operations, and durations of cardiopulmonary bypass were also similar (table 4).

Postoperatively, there was no unexpected morbidity or mortality in either group, nor were there any differences in renal function. No polyuria, hypernatremia, serum hyperosmolality, hyperuricemia, or significant weight loss was seen. The only significant differences were in those variables which reflect methoxyflurane metabolism: serum inorganic fluoride concentration and urinary inorganic fluoride and oxalic acid excretion (tables 1–3). Considerable increases were found in patients anesthetized with methoxyflurane, while there were no changes in patients anesthetized with morphine–halothane. Determinations on the first postoperative day after methoxyflurane anesthesia showed increases in mean serum inorganic fluoride concentration from 1.7 ± 0.3 to 45.7 ± 4.2 μmol/l, urinary inorganic fluoride excretion from 39.0 ± 5.1 to 3,122 ± 509 μmol/24 hours, and urinary oxalic acid excretion from 30.0 ± 2.9 to 97.1 ± 14.2 mg/24 hours. These values declined exponentially during the postoperative period. Figure 1 shows the results for a patient anesthetized with methoxyflurane. In this patient only, oxalic acid determinations were continued for seven days.

Mean arterial methoxyflurane concentration before cardiopulmonary bypass was 7.8 ± 1.0 mg/100 ml; after bypass it was 4.7 ± 0.9 mg/100 ml. Minimum alveolar concentration (MAC) for methoxyflurane, 0.16 vol per cent,[7] corresponds to a blood level of 13.4 mg /100 ml in young healthy volunteers.[8]

Discussion

It is now generally accepted that renal dysfunction may be associated with administration of methoxyflurane.[1, 9] The degrees of dysfunction correlate with increased concen-

trations of two methoxyflurane metabolites, inorganic fluoride and oxalic acid[2]; patients with the greatest renal impairment have the greatest increases in these metabolites, suggesting a dose-related renal lesion.

The direct cause of this lesion has not been conclusively established. However, there is now strong evidence in the experimental animal that inorganic fluoride is the nephrotoxic substance. It has been reported that polyuric renal dysfunction can be produced in rats by chronic high intake of dietary sodium fluoride[10] or by intravenous or intraperitoneal injection of sodium fluoride.[11] In our laboratory we have recently produced acute polyuric renal dysfunction in rats with methoxyflurane anesthesia and produced a syndrome biochemically and morphologically similar with intraperitoneal injection of sodium fluoride.‡ Since inorganic fluoride is a result of methoxyflurane metabolism,[2, 12, 13] its serum concentration is most closely related to total methoxyflurane dosage. Although it has been suggested that increased metabolism of halothane[14] and fluroxene § occurs at low concentrations, this has not been reported for methoxyflurane.

Total anesthetic dose, determined by anesthetic concentration times duration of administration, was of particular concern in the present study. In our previous study, end-alveolar methoxyflurane concentration, 0.6–1.9 MAC (mean 1.4), indicated only moderate anesthetic depth. This measurement describes anesthetic depth at the moment of sampling but does not take into account duration of administration. In fact, it is likely that total doses in these patients were large. This probably resulted from the use of an inhalation agent for induction and omission of barbiturates, narcotics, and nitrous oxide in procedures lasting more than four hours. In the present study patients were generally so ill that anesthetic requirements were minimal. This explains the very low mean arterial methoxyflurane concentration, 7.8 ± 1.0 mg/100 ml, found in this group. No anesthetic was used during cardiopulmonary bypass, and only intermittent administration of methoxy-

‡Mazze, R. I., and Cousins, M. J., unpublished data.

§ Sawyer, D. C., personal communication.

TABLE 3. Results of Miscellaneous Tests (Mean ± SEM)

	Solute Excretion/24 Hours					Clearances (ml/min)			Intake (l/Day)	Output (l/Day)	Weight (kg)
	Na^+ (mEq)	K^+ (mEq)	Osmolality (mOsm)	F^- (μmol)	Oxalic Acid (mg)	Creatinine	Uric Acid	Osmolality			
Methoxyflurane											
Preoperative	77.1 ± 13.7	81.4 ± 13.0	701.0 ±40.4	39.5 ±5.1	30.0 ±2.0	80.6 ±0.7	7.52 ±0.67	1.54 ±0.09	1.98 ±0.20	1.49 ±0.24	76.2 ±2.7
Postoperative days 1–4	78.0 ±17.3	114.0 ±11.8	908.7 ±53.9	2242.1 ±317.1	91.1 ±12.8	93.5 +7.1	10.46 ±2.39	2.11 ±0.11	2.14 ±0.23	1.66 ±0.15	76.0 ±2.5
Morphine											
Preoperative	76.3 +19.2	65.5 ±6.7	661.2 ±75.3	59.4 ±12.6	31.2 ±4.0	90.2 ±11.0	7.44 ±0.93	1.52 ±0.20	1.85 ±0.14	1.10 ±0.14	80.8 ±5.6
Postoperative days 1–4	81.2 ±9.5	108.9 ±3.7	901.0 ±37.5	±50.6 ±5.4	30.6 ±2.6	99.9 ±9.1	15.47 ±2.78	2.08 ±0.12	2.07 ±0.08	1.61 ±0.13	80.3 ±5.6
P^* methoxyflurane *vs.* morphine	>0.5	>0.2	>0.4	<0.001	<0.001	>0.4	>0.5	>0.5	>0.1	>0.3	>0.3

* Preanesthetic–postanesthetic differences.

TABLE 4. Patient Groups and Operative Treatment

	Patient Age (Years)*	Morphine (mg.)*	Duration (Hours-Minutes)*			Operative Procedure
			Methoxyflurane Administration	Cardiopulmonary Bypass	Operation	
Methoxyflurane	49.4 ±2.2	—	2'17" ±12"	1'49" ±16"	4'56" ±20"	Valve replacement, 5† Coronary artery bypass graft, 6†
Morphine	47.4 ±2.3	87.5 ±5.7	—	1'44" ±09"	4'18" ±11"	Valve replacement, 5 Coronary artery bypass graft, 5

* Mean ± SE.
† One patient had aortic valve replacement and coronary artery bypass graft.

flurane was necessary after circulation had been restored. The total periods of methoxyflurane administration averaged 2 hours and 17 minutes, and during much of this time delivered concentrations were only 0.1 to 0.3 per cent. The low blood anesthetic concentrations and the relatively short periods of administration confirmed our clinical impression that this type of operation is accomplished with small total doses of methoxyflurane.

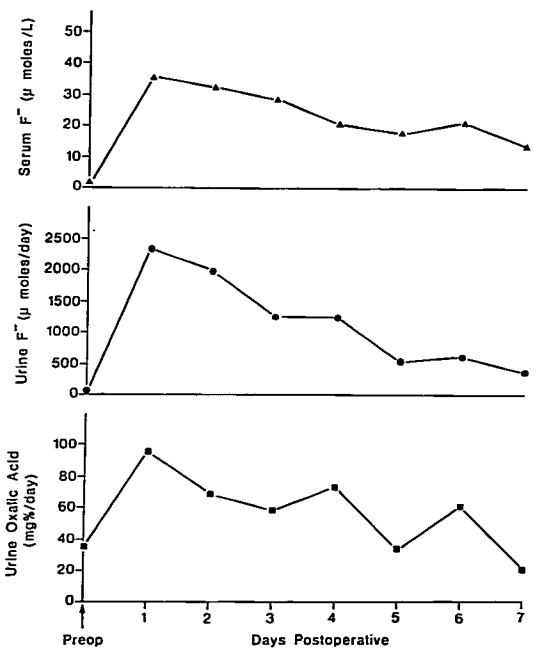

FIG. 1. Daily serum inorganic fluoride and 24-hour urinary inorganic fluoride and oxalic acid excretion of a patient anesthetized with methoxyflurane.

Further evidence of low methoxyflurane dosage was seen in measurements of inorganic fluoride and oxalic acid. Mean peak serum inorganic fluoride concentration, 45.7 ± 4.2 μmol/l, was approximately a quarter of that previously reported in clinically symptomatic patients and less than half that measured in individuals with only laboratory abnormalities in renal function.[2] Urinary inorganic fluoride excretion, $2,242 \pm 317$ μmol/24 hours, a measure of total methoxyflurane metabolism, was also less than 50 per cent of that previously reported.[2]

Although it is most likely that the small total dosage of methoxyflurane accounts for the absence of renal dysfunction, it is possible that hypothermia, mannitol administration, or cardiopulmonary bypass played a part. It has been shown in mice anesthetized with halothane that the greatest amount of metabolism occurs in the first few hours following anesthetic administration.[15] Hypothermia, by retarding enzymatic reactions, could reduce methoxyflurane biodegradation, which would allow a greater proportion of the administered dose to be eliminated by the lungs. However, the considerably greater solubility in blood and fat of methoxyflurane compared with halothane provides a drug depot for postanesthetic metabolism which is significantly larger than that available to halothane. Mannitol protects the kidney from the effects of some nephrotoxins. It is most effective against toxins which act from within the tubular lumen to cause obstruction to urinary flow, such as hemoglobin,[16] and probably of lesser value against substances which have a direct toxic effect upon cell enzyme systems.[17] Furthermore, it is unlikely that high urinary flow rates would offer protection against renal dysfunction associated with administration of methoxyflurane, since polyuria is already a feature of this syndrome. The effects of cardiopulmonary bypass in this situation are not known. The concentration of methoxyflurane in the blood should be less during passage through the oxygenator, while hemolysis would aggravate any tendency towards renal damage. The net result of these divergent factors is difficult to evaluate.

Although operations utilizing cardiopulmonary bypass represent a very specialized situation, the effects of increased inorganic fluoride in these patients may not be substantially different from those in general surgical patients. It appears that low-dose methoxyflurane anesthesia produces only moderate increases in inorganic fluoride concentration which are not associated with renal dysfunction. However, a word of caution is appropriate, since several important questions remain to be answered: Will individual variations in methoxyflurane metabolism or increased sensitivity to inorganic fluoride as a nephrotoxin result in clinical renal toxicity, even at low doses of methoxyflurane? Can enzyme induction due to exposure to various chemical substances such as barbiturates, tranquilizers, insecticides, aerosol sprays, and methoxyflurane itself cause accelerated production of inorganic fluoride? Does repeated administration of low-dose methoxyflurane pose an increased risk of toxicity due to enzyme induction and residual levels of inorganic fluoride? What is the interaction of low-dose methoxyflurane with potentially nephrotoxic therapeutic agents such as tetracycline, gentamicin, and colistin? Finally, the most important question, what is the upper limit of "low-dose" methoxyflurane? Until these questions are answered, the safety of low-dose methoxyflurane remains in doubt.

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References

1. Mazze RI, Shue GL, Jackson SH: Renal dysfunction associated with methoxyflurane anesthesia: A randomized prospective clinical evaluation. JAMA 216:278–288, 1971
2. Mazze RI, Trudell JR, Cousins MJ: Methoxyflurane metabolism and renal dysfunction: Clinical correlation in man. ANESTHESIOLOGY 35:247–252, 1971
3. Lowe HJ, Hagler K: Clinical and laboratory evaluation of an expired anesthetic gas monitor (Narko-Test). ANESTHESIOLOGY 34: 378–382, 1971
4. Cousins MJ, Mazze RI: A rapid direct-injection method for measuring volatile anesthetics in whole blood. ANESTHESIOLOGY 36: 293–296, 1972
5. Fry BW, Taves DR: Serum fluoride analysis with the fluoride electrode. J Lab Clin Med 75:1020–1025, 1970
6. Ribeiro ME, Elliot JS: Direct enzymatic determination of urinary oxalate. Invest Urol 2:78–81, 1964

7. Saidman LJ, Eger EI II, Munson ES, et al: Minimum alveolar concentrations of methoxyflurane, halothane, ether and cyclopropane in man: Correlation with theories of anesthesia. ANESTHESIOLOGY 28:994–1002, 1967
8. Eger EI II, Shargel R: The solubility of methoxyflurane in human blood and tissue homogenates. ANESTHESIOLOGY 24:625–627, 1963
9. Statement regarding the role of methoxyflurane in the production of renal dysfunction. Committee on Anesthesia, National Academy of Sciences–National Research Council. ANESTHESIOLOGY 34:505–509, 1971
10. Bond AM, Murray MM: Kidney function and structure in chronic fluorosis. Br J Exp Pathol 33:168–176, 1952
11. Taylor JM, Scott JK, Maynard EA, et al: Toxic effects of fluoride on the rat kidney. 1. Acute injury from single large doses. Toxicol Appl Pharmacol 3:278–289, 1961
12. Holaday DA, Rudofsky S, Treuhaft PS: The metabolic degradation of methoxyflurane in man. ANESTHESIOLOGY 33:579–593, 1970
13. Taves DR, Fry BW, Freeman RB et al: Toxicity following methoxyflurane anesthesia. II. Fluoride concentration in nephrotoxicity. JAMA 214:91–95, 1970
14. Sawyer DC, Eger EI II, Bahlman SH, et al: Concentration dependence of hepatic halothane metabolism. ANESTHESIOLOGY 34:230–235, 1971
15. Cohen EN: Metabolism of halothane-2^{14}C in the mouse. ANESTHESIOLOGY 31:560–565, 1969
16. Cirksena WJ, Bastian RC, Malloy JP, et al: Use of mannitol in exogenous and endogenous intoxications. N Engl J Med 270:161–166, 1964
17. Barry KG, Mazze RI, Malloy JP: Therapy of Acute Renal Failure in Man. Mannitol Symposium, Walter Reed Army Institute of Research, Washington, D. C., February 3, 1962, pp 23–24

Respiration

HUMAN ALVEOLAR MACROPHAGE Although the alveolar macrophage has been implicated as the most vital mechanism for protecting the lungs from damage by microorganisms, very few studies of the function of this cell have been published. Human alveolar macrophages were lavaged from surgically resected lungs and also from lungs of normal subjects. Cells that had been purified by adherence to glass were maintained by tissue culture for as long as 54 days. After three to four weeks *in vitro*, transformation into multinucleated giant cells was apparent, at which time the cells had more than 30 times the phagocytic capacity than after one day *in vitro*. Phagocytosis of heat-killed *Candida albicans* was inhibited by: iodoacetate, sodium fluoride, potassium cyanide, and low partial pressures of oxygen; ancillary evidence suggests that these cells need both oxidative and glycolytic energy sources for optimal particle ingestion. Alveolar macrophages killed *Listeria monocytogenes*, as did monocyte-derived macrophages in a similarly efficient manner, but neutrophils were more efficient than either. Clearing of bacteria is probably not dependent upon myeloperoxidase in the monocyte-derived macrophage or in the alveolar macrophage, since histochemical staining for peroxidase was negative. During four hours of observation, *C. albicans* blastospores were killed by neutrophils and monocytes containing myeloperoxidase, but not by human alveolar macrophages. Large cells with supernormal phagocytic capacity were recovered from patients who had acute pneumonia of infectious origin. This suggests that disease alter macrophage function. Human alveolar macrophages are unique phagocytes, in that they depend on an environmental P_{O_2} greater than 25 torr for maximal activity. A P_{CO_2} as high as 70 torr did not alter this function when the *p*H was constant. These data suggest that patients with chronic bronchitis or atelectasis may have suboptimal macrophage action in areas of the lung where P_{O_2} is abnormally low. Since bronchial clearance of particulate matter is impaired and the lower respiratory tract is frequently infected, defense of the lungs by macrophages assumes an important role in the clinical course of chronic infection. (*Cohen, A. B., and Cline, M. J.: The Human Alveolar Macrophage: Isolation, Cultivation in Vitro, and Studies of Morphologic and Functional Characteristics, J. Clin. Invest. 50:1390, 1971.*)