Bull. Environ. Contam. Toxicol. (1988) 40:280–286

Bulletin of Environmental Contamination and Toxicology

Metabolism and Excretion of Orally and Intraperitoneally Administered Methylarsonic Acid in the Hamster

Hiroshi Yamauchi, Naohisa Yamato, and Yukio Yamamura

Department of Public Health, St. Marianna University School of Medicine, 2-16-1, Sugao, Miyamae-ku, Kawasaki, 213 Japan

A number of investigators have demonstrated that when inorganic arsenic is administered to humans and experimental animals, methylarsonic acid (MAA) is formed *in vivo* (Yamauchi and Yamamura 1979a,b,1985; Buchet et al. 1981; Vahter 1981). Low concentrations of MAA have been detected in human organs (Yamauchi and Yamamura 1983) and urine (Yamauchi and Yamamura 1979a,b; Buchet et al. 1981). Compounds containing MAA are used as arsenic agrochemicals (Abdelghani et al. 1986), and subacute arsenic poisoning has been reported in a farmer exposed to a large quantity of MAA (Hessle and Berman 1982). Few studies of the metabolism and elimination of MAA have been published. Following administration of a single oral dose of MAA to human subjects, Buchet et al.(1981) reported that MAA was rapidly metabolized to dimethylarsinic acid (DMAA) *in vivo* and excreted in urine. While the elimination of MAA has been investigated experimentally in animals (Shariatpanshi and Anderson 1984), nothing is known of MAA metabolism and distribution *in vivo*.

In the present study, the metabolism of MAA was investigated following its administration to hamsters. Arsenic species deposited in selected organs and blood, and the amounts and chemical species of arsenic excreted in urine and feces were determined.

MATERIALS AND METHODS

Male syrian golden hamsters, weighing 95.7±7.0 g, were used in groups of 5. The hamsters had free access to a pellet feed manufactured by Japan CLEA, Tokyo, and distilled water. The hamsters received a single dose of 5, 50 or 250 mg/kg body weight of MAA (Ventron Corp.,

Send reprint requests to H. Yamauchi at the above address

Danvers, Mass., U.S.A.) through a stomach tube, or a single intraperitoneal dose of 50 mg/kg body weight. The sample of MAA used contained 0.5% inorganic arsenic. The hamsters were killed before, and 1, 6, 12, 24, 72 or 120 hr after the administration of MAA. Urine and feces were collected separately for each 24 hours by housing the animals in individual plastic metabolic cages. All assay materials were preserved frozen at -20 ^{o}C until assay.

Hair was washed with distilled water, ethanol and acetone. For arsenic analysis, 0.5 to 1.0 g of tissue or feces, and 0.5 to 1.0 ml of blood or urine were transferred into 10-ml polymethylpentene test tubes. After addition of 5 ml of 2 N NaOH, the specimens were heated in a heating block (YAMATO model HF-41) at 95^{o}C for 3 hr. In preliminary experiments, neither MAA, DMAA nor trimethylarsenic compound (TMA) degraded into any other chemical species of arsenic even when heated at 95^{o}C in 2 N NaOH. Inorganic arsenic, MAA, DMAA and TMA were determined by liquid nitrogen trapping - arsine generation - atomic absorptiometry (Yamauchi and Yamamura 1984a). The detection limits of the 4 chemical species of arsenic by the respective methods were 0.5 ng, with the coefficient of variation being less than 5%.

From the determined amount of inorganic, MAA, DMAA and TMA in urine and feces following the administration of MAA the respective background values described in the Results were deducted (mean values before the administration of MAA).

RESULTS AND DISCUSSION

Table 1 shows the concentrations of chemical species of arsenic in the tissues following the oral administration of a single 50 mg/kg body weight dose of MAA. The peak total arsenic concentration was highest in the kidney, followed in order of decreasing peak concentration by spleen, lung, skin, liver, muscle, hair and brain. The pattern of deposition indicated that a large fraction of the MAA is accumulated in the kidney within 6 hr of administration. It has been reported that the administration of arsenic trioxide to hamsters leads to the *in vivo* formation of MAA (Yamauchi and Yamamura 1985), and that this MAA is deposited in the kidney at high levels. The pattern of MAA deposition in the present study was in accord with those findings. The MAA concentrations in all tissues, other than the MAA undergoing clearance, rapidly decreased with only a trace of MAA detected at 120 hr.

Table 1 Concentration of arsenic compounds in blood and tissues after a single oral administration of methylarsonic acid (50 mg/kg).

Sample	Chemical species	Concentration of arsenic (μg As/g wet wt.)						
		Hours after administration						
		C	1	6	12	24	72	120
Whole* blood	InAs	0.07	0.11	0.10	0.17	0.14	0.10	0.11
	MAA	-	0.57	0.91	0.44	0.22	0.06	<0.01
	DMAA	-	-	<0.01	0.03	0.02	<0.01	-
Plasma*	InAs	0.11	0.13	0.10	0.14	0.18	0.13	0.10
	MAA	-	0.61	0.85	0.23	0.08	0.03	<0.01
	DMAA	-	-	0.02	0.08	0.06	<0.01	-
Brain	InAs	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
	MAA	-	<0.01	0.02	0.01	<0.01	<0.01	<0.01
	DMAA	-	-	0.01	0.03	0.03	0.01	<0.01
Hair	InAs	0.05	0.05	0.04	0.03	0.02	0.04	0.04
Kidney	InAs	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
	MAA	-	0.80	1.87	1.74	1.88	2.09	1.47
	DMAA	-	0.01	0.11	0.15	0.10	0.02	0.01
Liver	InAs	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
	MAA	-	0.10	0.17	0.08	0.04	<0.01	<0.01
	DMAA	-	<0.01	0.03	0.04	0.04	0.01	<0.01
Lung	InAs	0.01	0.01	0.01	0.01	0.01	0.01	0.01
	MAA	-	0.27	0.78	0.72	0.41	0.11	0.02
	DMAA	-	0.01	0.16	0.35	0.25	0.10	0.01
Muscle	InAs	0.01	0.01	0.01	0.01	0.01	0.01	0.01
	MAA	-	0.05	0.08	0.10	0.07	0.04	0.03
	DMAA	-	<0.01	0.01	0.02	0.03	0.01	<0.01
Skin	InAs	0.05	0.07	0.05	0.04	0.04	0.05	0.06
	MAA	-	0.17	0.27	0.20	0.13	0.05	0.02
	DMAA	-	-	0.01	0.04	0.05	0.01	-
Spleen	InAs	0.05	0.06	0.06	0.05	0.06	0.07	0.06
	MAA	-	0.25	1.17	1.52	1.24	0.19	0.03
	DMAA	-	-	0.05	0.09	0.08	0.03	-

*, μg As/ml; Mean for five hamsters; C, controls; -, not detected
After the administration of MAA, trimethylarsenic compound (<0.01 μg As/g.wet wt.) was detected in the kidney, liver, lung and muscle.

The MAA concentration in the kidney declined very slowly. The total arsenic concentrations in most tissues, following the oral administration of MAA to hamsters remained low, compared with the result reported in experiments with a similar arsenic dose (about 1.5 mg/body weight) as DMAA (Yamauchi and Yamamura 1984a) and arsenobetaine (Yamauchi et al. 1986). This difference may be caused by a low rate of absorption of MAA from the gastrointestinal tract.

Table 2 Percent excretion of arsenics in urine and feces of hamster after a single oral and intraperitoneal administration of methylarsonic acid (50 mg/kg).

Hours after administration		Percentage of administered dose							
		InAs		MAA		DMAA		TMA	
		oral	ip	oral	ip	oral	ip	oral	ip
Urine	-24	0.02	0.01	26.9	77.7	1.43	0.82	0.07	0.02
	-48	0.02	0.01	1.82	3.00	2.10	0.22	0.18	0.02
	-72	0.02	0.01	0.19	0.41	0.39	0.05	<0.01	0.01
	-96	0.01	0.01	0.06	0.20	0.16	0.03	0.02	0.01
	-120	0.01	<0.01	0.06	0.09	0.11	0.01	0.04	0.01
Feces	-24	0.02	<0.01	56.1	0.74	0.08	0.02	-	-
	-48	0.02	<0.01	4.41	0.12	0.15	<0.01	-	-
	-72	0.01	<0.01	0.07	0.03	0.02	<0.01	-	-
	-96	<0.01	<0.01	0.01	0.01	-	<0.01	-	-
	-120	<0.01	<0.01	0.01	0.01	-	<0.01	-	-

Mean for five hamsters; ip, intraperitoneal

In the present experiments, MAA was detected in tissues as soon as 1 hr after the administration, and reached a peak in the respective tissues at 6 or 12 hr, DMAA was detected in the liver, kidney, lung and muscle from 1 hr after the oral administration of MAA, and reached a peak in the respective tissues at 12 hr. MAA, when orally administered to hamsters, is methylated *in vivo*, and part of it is converted into DMAA. Neither inorganic arsenic nor TMA appeared in tissues as a result of MAA. No MAA, DMAA or TMA were detected in the hair.

Table 1 shows the concentrations of chemical species of arsenic in the blood following the oral administration of a single 50 mg/kg body weight dose of MAA. The MAA concentration in blood reached a peak 6 hr after dose administration. MAA was present in plasma and blood cells at similair concentration during the first 12 hr, but tended to be associated with the blood cells after 12 hr. The DMAA concentration in plasma reached a peak at 12 hr. Both MAA and DMAA rapidly disappeared from the blood, returning to the respective control values at 120 hr. The inorganic arsenic and TMA concentrations in blood following the administration of MAA did not change significantly, compared with the concentration in control hamsters.

Table 2 shows the patterns of arsenic excretion in urine and feces during 120 hr after the oral or intraperitoneal administration of a single 50 mg/kg of body weight dose of MAA. The background arsenic

concentration prior to the administration of MAA was 0.76±0.2 µg As/day in urine (3% inorganic arsenic, 4% MAA, 24% DMAA and 69% TMA) and 0.35±0.1 µg As/day in feces (34% inorganic arsenic, 6% MAA, 3% DMAA and 57% TMA). The excretion of total arsenic in urine during the 120 hr following the single oral and intraperitoneal doses of MAA amounted to 33.6% and 82.7%, respectively. The 120 hr fecal excretion of total arsenic following the oral and intraperitoneal administration of 50 mg/kg of MAA amounted to 60.9% and 1.0% of the respective dose. The literature indicates that when humans (Buchet et al. 1981) received a single oral dose of MAA, urinary arsenic excretion accounted for 78.3% of the administered dose during the following 4 days. In sheep and goats, an oral dose of 10 mg/kg of MAA resulted in excretion of 93.5% and 89% respectively of the ingested total arsenic (urine + feces) during the following 5 days (Shariatpanahi and Anderson 1984).

The excretory routes of the oral and intraperitoneal MAA dose were different. A large portion of the oral dose was excreted in feces, and most of the intraperitoneal dose was excreted in urine. Since MAA administered orally to goats and sheep was mostly excreted in urine (Shariatpanahi and Anderson 1984), it may be that the excretory pattern is different in different species. From the fact that MAA administered intraperitoneally to hamsters was excreted to a greater extent in urine than in feces, it was judged that only an extremely small amount of MAA would be eliminated in bile. A large portion of the orally and intraperitoneally administered MAA was excreted in unchanged from.

The amount of DMAA excreted in urine during the first 120 hr was equivalent to as little as 4.2±1.8% of the oral dose and 1.2±0.4% of the intraperitoneal dose. Less than 0.2% of the oral or intraperitoneal dose of MAA was excreted as DMAA in feces. Table 3 shows arsenic excretion in urine and feces during the first 24 hr after the oral administration of a single 5, 50 or 250 mg/kg body weight dose of MAA. The urinary DMAA excretion rate accounted for 7.5±3.2% of the 5 mg/kg MAA dose, but for only 0.4% following the administration of the larger 250 mg/kg dose. This indicates that the DMAA formation did not increase directly with the increase in the MAA dose. Less than 1% of the MAA was excreted as DMAA in feces at any of the three doses.

TMA excreted in urine was to 0.3% of the oral dose and 0.1% of the intraperitoneal dose of MAA at 50 mg/kg (Table 2). However, the TMA excreted in urine was 1.9±0.1% of the 5 mg/kg oral dose of MAA (Table 3).

Table 3 Percent excretion of arsenics in urine and feces of hamsters 24 hours after a single oral administration of methylarsonic acid(5,50,250 mg/kg).

Dose mg/kg	Percentage of administered dose InAs	MAA	DMAA	TMA
Urine				
5	<0.1	28.6± 5.2	8.4±3.0	1.9±0.1
50	<0.1	26.9±10.3	1.4	<0.1
250	<0.1	33.8± 4.7	0.4	-
Feces				
5	<0.1	50.5± 8.3	1.1±0.1	-
50	<0.1	56.0± 7.4	<0.1	-
250	<0.1	46.0±10.6	<0.1	-

Mean±S.D. for five hamster, -,Not detected

These results indicate that MAA may be converted into TMA to a small extent *in vivo*. In any event, it appears that MAA is less likely to be methylated than DMAA (Yamauchi and Yamamura 1984a) *in vivo*.

It was previously been thought that DMAA (Stevens et al. 1977; Vahter et al. 1984; Yamauchi and Yamamura 1984a) and arsenobetaine (Vahter et al. 1983; Yamauchi et al. 1986) were not degraded into inorganic arsenic *in vivo*. The amounts of inorganic arsenic excreted in urine and in feces accounted for as little as 0.2% of the oral dose and 0.1% of the intraperitoneal dose, respectively. These amounts did not exceed the inorganic arsenic content of the administered sample of MAA. Therefore it is extremely unlikely that any of the administered MAA was demethylated *in vivo*.
It is concluded that MAA, compared to inorganic arsenic and DMAA, is unlikely to be either methylated or demethylated *in vivo*.

REFERENCES

Abdelghani AA, Anderson AC, Jaghabir M, Mather F (1986) Arsenic levels in blood, urine, and hair of workers applying monosodium methanarsonate(MSMA). Arch Environ Health 41:163-169

Buchet JP, Lauwerys R, Roels H (1981) Comparison of the urinary excretion of arsenic metabolites after a single oral dose of sodium arsenite, monomethylarsonate, or dimethylarsinate in man. Int Arch Occup Environ Health 48:71-79

Hessl SM, Berman E (1982) Severe peripheral neuropathy after exposure to monosodium methylarsonate.

J Toxicol Clin Toxicol 19:281-287
Shariatpanshi M, Anderson AC (1984) Distribution and toxicity of monosodium methanearsonate following oral administration of the herbicide to dairy sheep and goats. J Environ Sci Health B19:427-439
Stevens JT, Halle LL, Farmer JD, DiPasqual LC, Chernoff N, Durham WF (1977) Disposition of ^{14}C and/or ^{74}As-cacodylic acid in rats after intravenous, intratracheal, or peroral administration. Environ Health Perspect 19:151-157
Vahter M (1981) Biotransformation of trivalent and pentavalent inorganic arsenic in mice and rats. Environ Res 25:286-293
Vahter M, Marafante E, Denker L (1983) Metabolism of arsenobetaine in mice, rats and rabbits. Sci Total Environ 30:197-211
Vahter M, Marafante E, Denker L (1984) Tissue distribution and retention of ^{74}As-dimethylarsinic acid in mice and rats. Arch Environ Contam Toxicol 13:259-264
Yamauchi H, Yamamura Y (1979a) Dynamic change of inorganic arsenic and methylarsenic compounds in human urine after oral intake as arsenic trioxide. Ind Health 17:79-83
Yamauchi H, Yamamura Y (1979b) Urinary inorganic arsenic and methylarsenic excretion following arsenate-rich seaweed ingestion. Jpn J Ind Health 21:49-54
Yamauchi H, Yamamura Y (1983) Concentration and chemical species of arsenic in human tissue. Bull Environ Contam Toxicol 31:267-270
Yamauchi H, Yamamura Y (1984a) Metabolism and excretion of orally administered dimethylarsinic acid in the hamster. Toxicol Appl Pharmacol 74:134-140
Yamauchi H, Yamamura Y (1984b) Metabolism and excretion of orally ingested trimethylarsenic in man. Bull Environ Contam Toxicol 32:682-687
Yamauchi H, Yamamura Y (1985) Metabolism and excretion of orally administered arsenic trioxide in the hamster. Toxicology 34:113-121
Yamauchi H, Kaise T, Yamamura Y (1986) Metabolism and excretion of orally administered arsenobetaine in the hamster. Bull Environ Contam Toxicol 36:350-355
Received July 2, 1987; accepted October 30, 1987.