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The Effects of DMSA and DMPS on Arsenate Excretion in Mice

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Summary

Previous studies have shown that DMSA (meso-2, 3-dimercaptosuccinic acid, succimer) and DMPS (2, 3-dimercapto-1-propane sulfonic acid, Na salt, unithiol) are effective antidotes for pentavalent arsenate and trivalent arsenite because they reduce the mortality of mice poisoned with these arsenic compounds. Studies have also shown that in mice treated with arsenite, DMPS and DMSA promote excretion of arsenic into the bile and urine, respectively. This study was conducted to determine whether DMSA and DMPS promote arsenate excretion to the same extent as previously reported for arsenite. The results demonstrated that DMPS antagonized arsenate toxicity by improving excretion of arsenic, although to a lesser extent than in studies with arsenite, while DMSA showed a slight but statistically insignificant promotion of early arsenic excretion in the urine.

Introduction

DMPS (2, 3-dimercapto-1-propane sulfonic acid, Na salt, unithiol) as well as DMSA (meso-2,3-dimercaptosuccinic acid, succimer) are promising antidotes for poisoning by heavy metals such as arsenic, mercury or lead[1,2,3,4,5,6]. In the US, DMSA has already been approved as a drug of choice for children suffering from heavy lead poisoning [7]. DMPS, however , has not yet been approved.

One of the present authors[3] previously reported that in experiments using mice, DMPS and DMSA showed promoted excretion of arsenic into the bile and into the urine, respectively.

Inorganic arsenic, when given to animals, is biotransformed into organic arsenic compounds[8,9,10]. In general, arsenate is first converted to arsenite, then methylated to monomethylarsonate and further converted to diemthylarsinate. These biotransformations can be considered as a process of intoxication, that is, a conversion to less toxic arsenic compounds in the animal body.

In acute tests[2,4], antagonizing effects of both DMSA and DMPS against toxicity of arsenic compounds were observed for trivalent arsenic, pentavalent arsenate and monomethylarsonate (in decreasing order) in contrast to BAL (dimercaprol) which is an effective antidote for arsenite, but not for arsenate.

We examined whether DMSA and DMPS promote arsenate excretion into the mouse urine or feces to the same extent as previously reported for arsenite[3].

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Materials and Methods

Materials : Sodium arsenate (Na_2HAsO_4) and DMSA were obtained from Nacalai Tesque, Inc. (Kyoto, Japan), and DMPS from Sigma Chemical Co. (St. Louis, MO). Both sodium arsenate and DMPS were dissolved in physiological saline. DMSA was dissolved in 5% $NaHCO_3$ and diluted with physiological saline. All solutions were prepared immediately before use.

Animals and treatments : This study was performed according to the guidelines of our school for animal experiments. ICR strain male mice were obtained from Japan SLC Inc. (Shizuoka). They were used at 5 weeks of age, after a week quarantine, weighing 25–30g. The animals were housed in a temperature-controlled room at 22 ± 2℃ with a 12 hr light/dark cycle and allowed free access to a commercial diet (MF, Oriental Yeast Co., Ltd. Tokyo) and distilled water during the study. The animals were divided into three groups of 15 mice, and as three mice were housed together in a one metabolic cage during the experiment, each group comprised 5 cages. One group was given a single dose of sodium arsenate (5mg, As/kg), subcutaneously. Two other groups were given DMSA (100mg/kg) or DMPS (100 mg/kg) intraperioneally, immediately after subcutaneous administration of the arsenate (5mg, As/kg).

Sample collections and arsenate determination : Urine collections were made at 12hr, 14hr and 48hr after the treatment. Feces were collected at 24hr intervals over 2 days. Arsenate contents in the urine and feces were determined by atomic absorption spectrometry method. In this method[11], since a process of reduction of arsenate into arsenite in the samples was included, all arsenic compounds were determined to be trivalent arsenic.

Statistical methods : Statistical significance was evaluated by Student's t test. Only $p \leqq 0.05$ was considered significant. Values were expressed as means ± S.D.

Results

Table 1 shows arsenic excretion in the urine and feces after a single administration of sodium arsenate with and without DMSA or DMPS in mice.

Table 1 : Urinary and fecal excretion of arsenic after a single administration of sodium arsenate with DMSA or DMPS in mice (% of dose, Mean ± S.D.)

Group	Sample	Time of sample collection			Total arsenic in urine or feces	Total arsenic in urine and feces
		12 hr	24 hr	48 hr		
arsenate[1]	Urine	63.9±7.9	1.8±3.3	1.4±0.5	67.1±9.7	
	Feces		8.8±4.6	0.4±0.5	9.2±4.6	76.3±9.2
arsenate and DMSA[2]	Urine	72.2±3.4	0.8±0.7	1.0±1.1	74.0±4.0	
	Feces		2.2±1.5	0.3±0.6	2.5±1.4*	76.5±3.4
aesenate and DMPS[3]	Urine	73.0±5.5	1.8±0.6	1.5±2.3	76.3±4.4	
	Feces		13.7±1.8	0.1±0.1	13.8±1.8	90.1±5.7*

1) Sodium arsenate (5mg, As/kg) was given by subcutaneous injection.
2) DMSA (100mg/kg) was given by intraperitoneal injection, immediately after the administration of sodium arsenate (5mg, As/kg).
3) DMPS (100mg/kg) was given by intraperitoneal injection, immediately after the administration of sodium arsenate (5mg, As/kg).

*$p \leqq 0.05$, significantly different from respective value of arsenate group. N=5.

In the first group given only sodium arsenate (5mg, As/kg, s.c.), 63.9% of the dose was excreted in the urine within 12hr after the treatment, and 67.1% was observed as cumulative excretion until 48 hr after the administration. Arsenic in the feces was detected as 8.8% of the dose after 24hr and 9.2% within 48hr. Total arsenic excreted through the urine and feces was 76.3% within 48hr.

In the second group, given sodium arsenate (5mg, As/kg, s.c.) and DMSA (100 mg/kg, i.p.), 72.2% and 74.0% of the dose were excreted in the urine 12hr and 48hr after the treatment, respectively. Although arsenic contents in the urine, collected at 12hr and cumulatively until 48hr after the treatment, exceeded the control value, both of these values were not significantly different from the respective control (the first group) values. But the total amout of arsenic excreted in feces during the period showed a significant decrease.

In the third group, given arsenate (5mg, As/kg, s.c.) and DMPS (100 mg/kg, i.p.), 73.0%, 74.8% and 76.3% of the dose were excreted in the urine at 12hr, 24hr and 48 hr after the treatment, respectively. None of these values was statistically significant. Arsenic detected in the feces within 24hr after the treatment was 13.7%, but this was also not statistically significant. Althouth no statistical differences were evident, total arsenic excretion rates including urine and feces in the third group was 90.1% and this was significantly different ($p \leqq 0.05$) from the control value (76.3%).

Improved excretion of arsenate by the administraion of DMPS was thus more evident as compared with DMSA.

Discussion

Many investigations[1,2,3,4,5,6] have been reported concerning the effectiveness of DMSA and DMPS on antagonizing the toxicity of heavy metals. In the acute toxicity tests in mice[2], both DMSA and DMPS protected well against inorganic trivalent arsenic poisoning, reducing mortality. Although the effect in reducing of mortality in mice poisoned with inorganic pentavalent arsenate was slightly less than that in the case of the trivalent arsenite, these drugs were considered as useful antagonists against arsenate as well as arsenite. This is in contrast with BAL which is the only approved antidote for arsenic poisoning in Japan, and which does not reduce the mortality of mice poisoned by pentavalent arsenate.

In the present study, arsenic excretion by DMSA in the urine until 12hr after the administration of arsenate appeared to be improved, although without statistical significance, while values in DMPS group exceeded the control value of arsenic content in urine collected 0–12hr after the administration, as well as in feces collected during 0–24hr. Therefore, total arsenic excreted in the urine and feces within 48hr after the administration in the DMPS group was the most significant amount among the three groups.

In this study DMPS appeared to improve the arsenic excretion via feces, presumably through the bile, in addition to excretion into the urine. In the DMSA group and the control group, the total amount of arsenic excreted during 48hr after the administration was the same level as about 76% of the dose, suggesting that 24% of the dose still remained in the body 48hr after the administration. This contrasts with the case of DMPS where much greater amout of administered arsenic (more than 90%) was excreted within 48hr.

In summary, this study evidenced that DMPS antagonized the arsenate toxicity by improving the excretion of arsenic, while DMSA appeared to show only a slight promotion of early excretion of arsenic in the urine, although without statistical difference.

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抄録：有害金属拮抗薬 DMSA および DMPS のヒ酸ナトリウムの排泄に対する作用

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有害金属拮抗薬の DMSA（meso-2, 3-dimercatosuccinic acid, succimer）および DMPS（2, 3,-dimercapto-1-propane sulfonic acid, Na salt, unithiol）はともに無機の3価ヒ素の排泄をきわめて著明に促進する効果があることをすでに報告した．また急性投与によるヒ酸ナトリウム（5価ヒ素）に対しても解毒効果があった．そこでヒ酸ナトリウムについても3価ヒ素と同様に排泄促進効果があるかどうかをマウスを用いて調べた．その結果，DMPS はおもに12時間までの尿中，および24時間までの糞中への排泄増加があり，48時間までの総排泄量に有意の排泄促進が認められた．これに対して DMSA 投与の場合は投与12時間までの尿中排泄を増加させるようであったが，48時間までの総排泄量には有意差はみられなかった．