Defense Mechanisms against Cadmium Toxicity
II. Effects of Pretreatment with a Small Oral Dose of Cadmium on Absorption, Distribution and Excretion of Cadmium after a Large Oral Dose in Mice

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Accepted February 29, 1984

Abstract—Uptake of Cd\(^{2+}\) by the liver and kidney of female mice 24 hr after challenge with a large dose of Cd\(^{2+}\) (100 mg Cd\(^{2+}\)/kg, p.o.) was greatly reduced by pretreatment with a small dose of the cation (15 mg Cd\(^{2+}\)/kg, p.o.) at 24 hr (for liver) and at 6, 24 or 48 hr (for kidney) prior to the challenge dose. The hepatic concentration of Zn\(^{2+}\) tended to be increased by the Cd\(^{2+}\) challenge and was increased further by pretreatment. The renal concentration of Zn\(^{2+}\) was not influenced by Cd\(^{2+}\) administration. The retention rate of Cd\(^{2+}\) in the stomach and its contents 24 hr after the Cd\(^{2+}\) challenge was decreased by pretreatment. In addition, the excretion rate of Cd\(^{2+}\) into the feces 24 hr after the Cd\(^{2+}\) challenge was increased by pretreatment at 6 to 24 hr prior to the challenge dose. Consequently, the absorption rate of Cd\(^{2+}\) 24 hr after the Cd\(^{2+}\) challenge was markedly reduced by pretreatment at 24 hr prior to the challenge dose. The urinary and biliary excretion of Cd\(^{2+}\) was very low. The motility of the small intestine was stimulated 6 hr after a small dose of Cd\(^{2+}\), but returned to normal within 24 hr. The motility tended to be reduced 4 hr after the Cd\(^{2+}\) challenge, but conversely, it was facilitated at 24 hr. Pretreatment at 6 or 24 hr prior to the challenge dose prevented the reduction of the motility 4 hr after the Cd\(^{2+}\) challenge.

Cadmium pretreatment protects animals against the acute toxicity of subsequent larger doses of the metal (1–4). In studies using parenteral administration of Cd\(^{2+}\), such protection occurs in spite of the fact that Cd\(^{2+}\) uptake by the liver after the Cd\(^{2+}\) challenge is increased by pretreatment (2, 5). The explanation is that Cd\(^{2+}\) pretreatment induces hepatic metallothionein (MT) synthesis, which leads to increased binding of subsequently administered Cd\(^{2+}\), thus reducing its toxicity (1, 2, 6, 7). On the other hand, there are few reports on the oral administration of Cd\(^{2+}\) which consider the effects of pretreatment on Cd\(^{2+}\) uptake by the tissues after a subsequent larger dose. Nevertheless, the results with oral Cd\(^{2+}\) administration to rats (8) are similar to those with parenteral Cd\(^{2+}\) administration.

In contrast, as shown in this report, pretreatment of mice with a small oral dose of Cd\(^{2+}\) reduced the cation uptake by the liver and kidney after a large oral dose. Thus, the present study was undertaken to examine the effects of Cd\(^{2+}\) pretreatment on the hepatic and renal concentrations of Cd\(^{2+}\) and Zn\(^{2+}\) and the absorption and excretion of Cd\(^{2+}\) in mice challenged with a large oral dose of Cd\(^{2+}\).

Materials and Methods

Animals and treatment: Female ICR mice (Clea Japan Inc., Osaka), 5 to 6 weeks old, were used. They were kept in an air-conditioned room (temperature: 23±2°C, relative humidity: 60±10%). Standard laboratory chow (MF, Oriental Yeast Co., Ltd., Tokyo) and water were freely available throughout the study. Mice were given cadmium chloride,
dissolved in deionized water, by the oral route. The small dose of Cd\(^{2+}\) employed was 15 mg Cd\(^{2+}\)/kg, while the large dose was 100 mg Cd\(^{2+}\)/kg. Mice were pretreated with a small dose of Cd\(^{2+}\) at 6, 24, 48 or 72 hr prior to challenge with a large dose of the cation. Control mice received the same volume of deionized water (10 ml/kg) instead of Cd\(^{2+}\) solution.

**Determination of Cd\(^{2+}\) and Zn\(^{2+}\) concentrations in the tissues, contents of gastrointestinal tract, feces and urine:** Following Cd\(^{2+}\) administration, mice were housed individually in metabolic cages during the entire experimental period until sacrifice to collect the feces and urine. At autopsy, the liver, kidney, stomach, small intestine and large intestine were removed. The contents of the stomach, small intestine and large intestine were collected separately. These samples, except the urine, were ashed at 450°C and then digested with nitric acid and perchloric acid (1:1). The residues were dissolved in 1 N-hydrochloric acid. The urine samples were diluted with deionized water. Concentrations of Cd\(^{2+}\) and Zn\(^{2+}\) in the liver and kidney were determined by atomic absorption, and atomic absorption was also used to determine the concentration of Cd\(^{2+}\) alone in the stomach, small intestine, large intestine, their contents, feces and urine.

**Determination of small intestinal motility:** Small intestinal motility was determined by the modified method of Lauener and Pogge (9). At time intervals of 4, 6, 24, 48 or 72 hr after Cd\(^{2+}\) administration, mice were orally given charcoal meal (10 ml/kg) which consisted of 5% charcoal powder in 10% arabic gum suspension. Thirty min after charcoal meal administration, mice were sacrificed, and the total length of the small intestine and the length of the small intestine traversed by the meal were immediately measured. The propulsive motility of the small intestine was expressed as a percentage of the total length of the intestine.

**Statistical analysis:** The data were analyzed by Student’s t-test.

**Results**

**Concentrations of Cd\(^{2+}\) and Zn\(^{2+}\) in the liver and kidney:** The uptake of Cd\(^{2+}\) by the liver 24 hr after challenge with a large dose of Cd\(^{2+}\) was greatly reduced by pretreatment with a small dose of the cation at 24 hr prior to the challenge dose, but not by pretreatment at other times (group Cd(S)-Cd(L), Table 1). The hepatic uptake of Cd\(^{2+}\) remained constant between 6 and 72 hr after a small dose of the cation (group Cd(S)).

The concentration of Zn\(^{2+}\) in the liver tended to be increased to the same degree both in the group given a low dose of Cd\(^{2+}\) (group Cd(S)) and that given a high dose (group Aq-Cd(L)) and was further increased in the Cd\(^{2+}\)-pretreated group (group Cd(S)-Cd(L), Table 2).

On the other hand, the renal uptake of Cd\(^{2+}\) 24 hr after a challenge dose of Cd\(^{2+}\) was strikingly reduced by pretreatment at 6, 24 or 48 hr prior to the challenge dose (group Cd(S)-Cd(L), Table 3). The Cd\(^{2+}\) uptake in the kidney remained constant between 6 and 72 hr after a small dose of the cation (group Cd(S)).

On the whole, the renal concentration of Zn\(^{2+}\) was unaffected by the administration of Cd\(^{2+}\) (Table 4).

**Gastrointestinal absorption, retention and excretion of Cd\(^{2+}\):** The retention rate of Cd\(^{2+}\) in the stomach and its contents, but not in the intestine and its contents, 24 hr after challenge with a large dose of Cd\(^{2+}\) was greater in the water-pretreated mice (group Aq-Cd(L)) than in the Cd\(^{2+}\)-pretreated mice (group Cd(S)-Cd(L), Table 5). In contrast, the excretion rate of Cd\(^{2+}\) into the feces 24 hr after the Cd\(^{2+}\) challenge was greater in the Cd\(^{2+}\)-pretreated animals. These findings suggested that pretreatment with a small dose of Cd\(^{2+}\) stimulated Cd\(^{2+}\) excretion after a subsequent challenge dose. This stimulative effect was greatly induced by pretreatment at 6 or 24 hr prior to the challenge dose.

When Cd\(^{2+}\) absorption was expressed as the total amount of Cd\(^{2+}\) administered minus the amount of Cd\(^{2+}\) retained in the gastrointestinal tract and its contents and excreted into the feces, the absorption rate of Cd\(^{2+}\) 24 hr after the Cd\(^{2+}\) challenge was markedly reduced by pretreatment at 24 hr prior to the challenge dose (Table 5).

Although not shown in the table, the urinary excretion of Cd\(^{2+}\) was very low.
### Table 1. Concentration of Cd$^{2+}$ in the liver after a challenge dose of Cd$^{2+}$ in mice pretreated with a small dose of Cd$^{2+}$

| Group  | Cd$^{2+}$ in liver (µg/g wet wt.) | Time after treatment (hr) | 6     | 24     | 48     | 72     |
|--------|----------------------------------|---------------------------|-------|--------|--------|--------|
| A: Aq  | trace                            |                           | trace | trace | trace | trace |
| B: Cd(S)| 1.29±0.24<sup>A</sup> (10)       |                           | 1.69±0.36<sup>A</sup> (10) | 1.23±0.12<sup>A</sup> (10) | 1.52±0.24<sup>A</sup> (10) |

| Time interval between pretreatment and challenge (hr) | 6     | 24     | 48     | 72     |
|-------------------------------------------------------|-------|--------|--------|--------|
| A: Aq–Cd(L)                                           | 29.50±4.36<sup>AB</sup> (10) | 26.37±1.83<sup>AB</sup> (15) | 24.40±1.14<sup>AB</sup> (8) | 27.19±2.05<sup>AB</sup> (11) |
| B: Cd(S)–Cd(L)                                        | 24.26±2.42<sup>AB</sup> (15) | 17.41±2.35<sup>AB</sup> (14) | 27.26±3.03<sup>AB</sup> (13) | 29.59±3.40<sup>AB</sup> (13) |

Aq, Cd(S): Mice were treated with either deionized water (10 ml/kg, p.o.) or Cd$^{2+}$ (15 mg/kg, p.o.) at 6, 24, 48 or 72 hr prior to sacrifice. Aq–Cd(L), Cd(S)–Cd(L): 6, 24, 48 or 72 hr after pretreatment with either deionized water (10 ml/kg, p.o.) or Cd$^{2+}$ (15 mg/kg, p.o.) mice were challenged with Cd$^{2+}$ (100 mg/kg, p.o.) and sacrificed after a further 24 hr. Each value represents the mean±S.E. (No. of mice). Alphabetical superscript represents a significant difference from the corresponding alphabet group at P<0.01 (capital letter) or P<0.05 (small letter).

### Table 2. Concentration of Zn$^{2+}$ in the liver after a challenge dose of Cd$^{2+}$ in mice pretreated with a small dose of Cd$^{2+}$

| Group  | Zn$^{2+}$ in liver (µg/g wet wt.) | Time after treatment (hr) | 6     | 24     | 48     | 72     |
|--------|----------------------------------|---------------------------|-------|--------|--------|--------|
| A: Aq  | 29.57±1.47 (5)                   |                           | 27.33±1.50 (5) | 28.34±1.02 (5) | 30.45±1.35 (5) |
| B: Cd(S)| 33.08±1.93 (10)                  |                           | 30.30±1.33 (10) | 29.33±1.47 (10) | 32.50±1.59 (10) |

| Time interval between pretreatment and challenge (hr) | 6     | 24     | 48     | 72     |
|-------------------------------------------------------|-------|--------|--------|--------|
| A: Aq–Cd(L)                                           | 31.97±2.56 (10) | 30.75±1.00 (15) | 28.67±1.77 (8) | 33.24±1.26 (11) |
| B: Cd(S)–Cd(L)                                        | 40.02±1.02<sup>AB</sup> (15) | 35.78±0.94<sup>AB</sup> (14) | 37.21±1.49<sup>AB</sup> (13) | 36.18±1.28<sup>A</sup> (13) |

See the legend of Table 1.
### Table 3. Concentration of Cd²⁺ in the kidney after a challenge dose of Cd²⁺ in mice pre-treated with a small dose of Cd²⁺.

| Group | A: Cd(S) | B: Cd(S) | C: Cd(S)–Cd(L) | D: Cd(S)–Cd(L) |
|-------|----------|----------|----------------|----------------|
|       | 6        | 24       | 6              | 24             |
|       | Time after treatment (hr) | Time interval between pretreatment and challenge (hr) | Trace | Trace |
|       | 0.54±0.09A (10) | 0.66±0.09A (10) | 0.65±0.07A (10) | 0.68±0.10A (10) |
|       | 12.83±3.32A (10) | 9.73±1.84A (15) | 14.29±2.91A (15) | 7.41±0.68A (13) |
|       | 4.72±0.43A (15) | 4.34±0.49A (14) | 6.40±0.58A (10) | 20.06±0.49A (13) |

See the legend of Table 1.

### Table 4. Concentration of Zn²⁺ in the kidney after a challenge dose of Cd²⁺ in mice pre-treated with a small dose of Cd²⁺.

| Group | A: Cd(S) | B: Cd(S) | C: Cd(S)–Cd(L) | D: Cd(S)–Cd(L) |
|-------|----------|----------|----------------|----------------|
|       | 6        | 24       | 6              | 24             |
|       | Time after treatment (hr) | Time interval between pretreatment and challenge (hr) | Trace | Trace |
|       | 22.17±1.17 (5) | 23.01±1.79 (5) | 20.22±1.07 (5) | 19.75±0.88 (11) |
|       | 18.47±1.02B (10) | 19.78±0.68A (10) | 20.02±0.65 (8) | 20.13±1.74 (13) |
|       | 22.08±1.08b (10) | 21.53±1.48 (14) | 21.51±0.90 (15) | 20.06±0.65 (8) |

See the legend of Table 1.
Table 5. Cd²⁺ retention in the gastrointestinal tract and its fecal excretion after a challenge dose of Cd²⁺ in mice pretreated with a small dose of Cd²⁺

| Group          | Time interval | Stomach (hr) | Stomach contents | Small intestine | Small intestinal contents (percent of dose) | Large intestine | Large intestinal contents | Feces | Absorption |
|----------------|---------------|--------------|------------------|----------------|---------------------------------------------|----------------|--------------------------|-------|------------|
| Aq–Cd(L)       | 6             | 7.6±2.1      | 29.4±5.3         | 1.8±0.1        | 1.8±0.7                                     | 1.5±0.5       | 32.3±4.5                 | 13.8±4.8 | 11.8±0.5   |
| Cd(S)–Cd(L)    | 6             | 1.5±0.3**    | 11.1±1.1**       | 1.6±0.1        | 1.6±0.4                                     | 2.3±0.6       | 27.4±8.6                 | 42.1±8.3* | 12.4±3.5   |
| Aq–Cd(L)       | 24            | 6.7±2.9      | 46.3±2.6         | 1.1±0.2        | 1.8±0.8                                     | 0.7±0.3       | 11.9±5.4                 | 19.9±8.0 | 11.6±2.2   |
| Cd(S)–Cd(L)    | 24            | 1.9±0.3      | 21.5±1.5**       | 1.6±0.1*       | 1.3±0.4                                     | 0.3±0.0       | 10.5±5.5                 | 57.2±5.4** | 5.7±1.0*   |
| Aq–Cd(L)       | 48            | 7.4±3.4      | 50.2±2.3         | 1.4±0.2        | 1.5±0.5                                     | 0.5±0.2       | 10.4±3.1                 | 16.4±7.5 | 12.2±3.5   |
| Cd(S)–Cd(L)    | 48            | 2.9±0.3      | 34.2±2.9**       | 1.2±0.2        | 1.6±0.6                                     | 0.9±0.3       | 13.9±4.0                 | 38.7±7.9 | 6.6±2.5    |
| Aq–Cd(L)       | 72            | 5.7±2.4      | 49.9±3.4         | 0.7±0.1        | 1.1±0.5                                     | 0.3±0.1       | 10.8±1.6                 | 21.4±6.0 | 10.1±2.1   |
| Cd(S)–Cd(L)    | 72            | 1.9±0.4      | 33.9±3.0**       | 1.2±0.1*       | 1.1±0.4                                     | 0.3±0.0       | 10.0±1.1                 | 42.6±7.3 | 9.0±1.4    |

Aq–Cd(L), Cd(S)–Cd(L): 6, 24, 48 or 72 hr after pretreatment with either deionized water (10 ml/kg, p.o.) or Cd²⁺ (15 mg/kg, p.o.), mice were challenged with Cd²⁺ (100 mg/kg, p.o.) and sacrificed after a further 24 hr. Each value represents the mean±S.E. of 5 to 7 mice. Absorption was expressed as the total amount of Cd²⁺ administered minus the amount of Cd²⁺ retained in the gastrointestinal tract and its contents and excreted into feces. *, **: Significantly different from the corresponding group Aq–Cd(L) at P<0.05 and P<0.01, respectively.
Twenty-four hr after the Cd\textsuperscript{2+} challenge, 0.01 to 0.05% of the dose given was present in the 24 hr urine samples. As for the biliary excretion of Cd\textsuperscript{2+}, there was no significant difference between the Cd\textsuperscript{2+}-pretreated mice (group Cd(S)-Cd(L)) and the water-pretreated controls (group Aq-Cd(L)) in the Cd\textsuperscript{2+} content of the bile between 1 and 5 hr after the Cd\textsuperscript{2+} challenge (0.05 to 0.07% of the dose given in each group).

**Time course of gastrointestinal absorption, retention and excretion of Cd\textsuperscript{2+} after a small or a large dose of the cation:** At 6 hr after administration, the Cd\textsuperscript{2+} retention rate in the gastrointestinal tract and its contents in mice treated with a small dose of Cd\textsuperscript{2+} (group Cd(S)) was approximately the same as that with a large dose (group Cd(L), Fig. 1). At 24 hr, 10.1% and 58.3% of the administered dose were found in the gastrointestinal tract and its contents in mice given a small dose of Cd\textsuperscript{2+} and a large dose, respectively. However, no significant differences were observed between groups in the retention rate of Cd\textsuperscript{2+} in the gastrointestinal tract and its contents at 96 hr and thereafter.

In mice given a small dose of Cd\textsuperscript{2+} (group Cd(S)), the cumulative excretion rate of the cation in the feces increased rapidly to 71.7% of the given dose by 24 hr after administration and increased slowly thereafter (Fig. 1). On the other hand, in mice treated with a large dose of Cd\textsuperscript{2+} (group Cd(L)), the fecal excretion of Cd\textsuperscript{2+} was only 25.8% of the dose at 24 hr after treatment, and it did not reach nearly the same rate as that in mice with a small dose until 96 hr.

The different dose levels of Cd\textsuperscript{2+} did not significantly influence the absorption rate of the cation during the 168-hr period (Fig. 1).

**Small intestinal motility after Cd\textsuperscript{2+} administration:** Effect of Cd\textsuperscript{2+} on the motility of the small intestine was examined by the passage of charcoal meal through the gastrointestinal tract (Table 6). The motility was facilitated at 6 hr after administration of a small dose of Cd\textsuperscript{2+}, but returned to normal within 24 hr (group Cd(S)). On the other hand, the motility tended to be restrained at 4 hr after challenge with a large dose of Cd\textsuperscript{2+}, but conversely to be facilitated at 24 hr, and then returned to normal by 72 hr (group Aq-Cd(L)). Pretreatment with a small dose of Cd\textsuperscript{2+} at 6 or 24 hr prior to the challenge dose prevented the restraint of small intestinal motility observed at 4 hr after the challenge dose (group Cd(S)-Cd(L)).
**Table 6.** Propulsive motility of the small intestine after a challenge dose of Cd$^{2+}$ in mice pretreated with a small dose of Cd$^{2+}$

| Group          | Time interval (hr) | Passage of charcoal meal in 30 min (% traversed) | Time after challenge (hr) |
|----------------|-------------------|-------------------------------------------------|--------------------------|
|                |                   |                                                 | 4           | 6           | 24          | 48          | 72          |
| Aq             | —                 | 64.0±4.1 (7)                                    | 66.1±2.8 (7) | 67.7±4.6 (7)| 68.5±4.5 (7)| 65.2±3.8 (7) |
| Cd(S)          | —                 | 83.5±2.0* (7)                                   |             |             |             |             |             |
| Aq–Cd(L)       | 6                 | 49.4±9.2 (7)                                    |             |             |             |             |             |
| Cd(S)–Cd(L)    | 6                 | 73.2±3.1b (7)                                   |             |             |             |             |             |
| Aq–Cd(L)       | 24                | 54.6±7.4 (7)                                    |             |             |             |             |             |
| Cd(S)–Cd(L)    | 24                | 78.3±3.2b (7)                                   |             |             |             |             |             |
| Aq–Cd(L)       | 48                | 53.5±5.6 (7)                                    |             |             |             |             |             |
| Cd(S)–Cd(L)    | 48                | 52.8±7.0 (7)                                    |             |             |             |             |             |
| Aq–Cd(L)       | 72                | 50.8±6.4 (7)                                    |             |             |             |             |             |
| Cd(S)–Cd(L)    | 72                | 51.7±5.0 (7)                                    |             |             |             |             |             |

Aq, Cd(S): Mice were treated with either deionized water (10 ml/kg, p.o.) or Cd$^{2+}$ (15 mg/kg, p.o.) at 4, 6, 24, 48 or 72 hr prior to charcoal meal administration. Aq–Cd(L), Cd(S)–Cd(L): 6, 24, 48 or 72 hr after pretreatment with either deionized water (10 ml/kg, p.o.) or Cd$^{2+}$ (15 mg/kg, p.o.), mice were challenged with Cd$^{2+}$ (100 mg/kg, p.o.) and given charcoal meal after a further 4, 24, 48 or 72 hr. Each value represents the mean±S.E. (No. of mice). *: Significantly different from the corresponding group Aq at *P*<0.05. **: Significantly different from the corresponding group Aq–Cd(L) at *P*<0.05.
Discussion

It is known that the hepatic uptake of parenterally administered Cd$^{2+}$ is increased by Cd$^{2+}$ pretreatment, but the uptake of Cd$^{2+}$ by the kidney and other organs is unaffected (2, 5). Similarly, according to Squibb et al. (8), in rats pretreated with an oral dose of 20 mg Cd$^{2+}$/kg at 24 hr prior to a larger oral dose of 100 mg Cd$^{2+}$/kg, the Cd$^{2+}$ uptake in the liver, kidney and testis 5 hr after a larger dose is greater than in water-pretreated animals. On the contrary, the present study with mice showed that the Cd$^{2+}$ uptake by the liver and kidney 24 hr after challenge with a large oral dose of Cd$^{2+}$ (100 mg Cd$^{2+}$/kg) was reduced by pretreatment with a small oral dose (15 mg Cd$^{2+}$/kg) at 24 hr and at 6 or 24 hr prior to the challenge dose, respectively. The reason for these differences in observations with the same route of administration is unknown.

The reduction of the hepatic and renal uptakes of orally administered Cd$^{2+}$ induced by pretreatment was due to changes in the gastrointestinal absorption and excretion of the cation. Pretreatment, which was more effective at 6 or 24 hr prior to the challenge dose, decreased the Cd$^{2+}$ concentration in the stomach and its contents 24 hr after the challenge dose and increased that in the feces. These observations suggested that pretreatment prevented the restraint of gastrointestinal motility after the Cd$^{2+}$ challenge, thus reducing the retention time of the stomach contents and increasing the fecal excretion of Cd$^{2+}$. Consequently, the reduced Cd$^{2+}$ absorption from the gastrointestinal tract within 24 hr after a large dose of the cation was induced by pretreatment 24 hr prior to the challenge dose. Interestingly, in considering defense mechanisms against Cd$^{2+}$ toxicity, the motility of the small intestine tended to be restrained at 4 hr after a large dose of Cd$^{2+}$, but conversely facilitated thereafter. Further, intestinal metallothionein (MT) may play a role in the reduced Cd$^{2+}$ absorption by pretreatment with a small dose of Cd$^{2+}$. It has been proposed that intestinal MT prevents Cd$^{2+}$ absorption by sequestering the cation in the mucosal cells during chronic low level exposure of Cd$^{2+}$ to rats (8, 10), although it was not observed when an acute dose of Cd$^{2+}$ was given to rats (8).

The principal route of Cd$^{2+}$ excretion after an i.v. administration of the cation to rats is the gastrointestinal tract (11–13). Within 24 hr after administration of Cd$^{2+}$ to rats (67 μg Cd$^{2+}$/rat, i.v.). 0.83% of the dose is excreted in the bile and 5.31% in the entire gastrointestinal tract and feces (12). Approximately 10% of the administered dose (1 mg Cd$^{2+}$/kg, i.v.) is excreted into the feces within 24 hr after Cd$^{2+}$ administration to rats and less than 0.5% of the dose into the urine during the next 7 days (13). Also in the present study, the urinary and biliary excretion of Cd$^{2+}$ was very small. Within 24 hr after a large dose of Cd$^{2+}$. 0.5% of the given dose was excreted in the urine and at most 0.5% in the bile. No effect of pretreatment with a small dose of Cd$^{2+}$ was observed on Cd$^{2+}$ excretion in the urine and bile after the challenge dose. Although the Cd$^{2+}$ excreted into the feces within 24 hr after the challenge dose partly contained the Cd$^{2+}$ once absorbed and then secreted into the gastrointestinal tract, the amount of the latter was considered small.

The hepatic concentration of Zn$^{2+}$ tended to be increased by both a small oral dose of Cd$^{2+}$ and a large oral dose, and further increased by pretreatment. However, the renal concentration of Zn$^{2+}$ was not influenced by Cd$^{2+}$ administration. These results are in agreement with those of others (14–17). The increase in the hepatic concentration of Zn$^{2+}$ is due to the induction of MT by Cd$^{2+}$ (14, 17).

Several investigators (6–8, 18–21) have reported on the relationship between Cd$^{2+}$ uptake and MT synthesis in the liver and kidney after Cd$^{2+}$ administration. Cd$^{2+}$ taken up by the liver and kidney stimulates MT synthesis and enhances the uptake of a subsequent larger dose of the cation. The intracellular binding of Cd$^{2+}$ with MT in these organs protects animals against the toxic effect of the cation. The experimental data and discussion on the relationship between MT synthesis and the reduction of the Cd$^{2+}$ uptake in these organs after a large oral dose of Cd$^{2+}$ by pretreatment with a small oral dose will be in the next report.

The relatively rapid excretion of Cd$^{2+}$ into
the feces occurred after a small dose of the cation, while the fecal excretion of Cd\textsuperscript{2+} after a large dose was extremely delayed. This might be partly due to the reduction of gastrointestinal motility. The different dose levels of Cd\textsuperscript{2+} did not appear to influence the absorption rate significantly in this study. However, according to Moore et al. (22), the absorption rate of Cd\textsuperscript{2+} is greater at low concentrations than at higher concentrations.

The present results indicate that the protective effect of Cd\textsuperscript{2+} pretreatment of mice against the acute oral toxicity of the cation described in the previous study (4) is attributed in part to the decreased absorption and increased fecal excretion of Cd\textsuperscript{2+} induced by pretreatment.

Acknowledgment: The author wishes to thank Prof. H. Iwata, Department of Pharmacology, Faculty of Pharmaceutical Sciences, Osaka University, for his advice on this manuscript.

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