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# Increasing Effect of Nori on the Fecal Excretion of Dioxin by Rats

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**The effects of nori (*Porphyra yezoensis*), a kind of red alga, on the gastrointestinal absorption and reabsorption of 17 types of dioxin were investigated in male Wistar rats. The rats were fed with 4 g of the control diet or 4 g of the nori diet containing a standard dioxin solution (233 ngTEQ/kg of body weight) for five consecutive days. In the group fed with the 10% nori diet, the fecal excretion of dioxin from days 1 to 5 was higher ($p<0.01$) than that of the control group by 5.5-fold for 2,3,7,8-TCDD, 6.6-fold for 1,2,3,7,8-pentaCDD, and 6.0-fold for 2,3,4,7,8-pentaCDF. In another experiment, the rats were fed with 4 g of the control diet containing a standard dioxin solution (2991 ngTEQ/kg of body weight) on the first day of the experiment and then given the control diet for 7 consecutive days, before being given either the control diet or the nori diet for 28 consecutive days more. In the group fed with the 10% Nori diet, the fecal excretion of dioxin during the period from days 8 to 35 was higher ($p<0.01$ or $p<0.05$) than that of the control group by 2.4-fold for 2,3,7,8-TCDD, 2.3-fold for 1,2,3,7,8-pentaCDD, and 2.4-fold for 2,3,4,7,8-pentaCDF. These results suggest that the administration of nori prevented dioxin from being efficiently absorbed and reabsorbed from the gastrointestinal tract, and might be useful for protecting humans exposed to dioxin from ill effects.**

**Key words:** dioxin; polychlorinated dibenzo-*p*-dioxin; polychlorinated dibenzofuran; nori; rats

Dioxin, which is the most toxic substance among synthetic chemicals, is a potent pollutant of the environment and can be detected worldwide. It is well known for dioxin to be generated by an incinerator when rubbish is destroyed by fire. Human beings generally ingest polychlorinated dibenzo-*p*-dioxin (PCDD) and polychlorinated dibenzofuran (PCDF) from various foods.[1–6] The daily intake of PCDD and PCDF congeners is approximately 2 pg toxic equivalent (TEQ)[7]/kg of body weight per day, and this level is considered to be almost the same among many countries in the world.[3] In Japan, the intake of PCDD and PCDF congeners had been reduced from 3.75 pg TEQ/kg of body weight per day (1977) to 0.92 (1998).[6] According to reports published in Germany (1994–1995), the excreted amount of PCDD and PCDF congeners (98 pg TEQ/day) was greater than their intake (49 pg TEQ/day).[8] Their concentrations in the serum have been reported to have been reduced from 42.7 pg TEQ/g of liquid (1991) to 20.7 (1996).[9]

PCDD and PCDF congeners are easily absorbed *via* the digestive tract due to their lipophilic nature and distributed to every tissue in the body by way of blood circulation following the lymphatic systems,[10] before finally being accumulated mostly in the adipose tissue and liver. The 2,3,7,8-substituted PCDD and PCDF congeners in particular remain for a long time in the tissues of humans and animals, because they cannot be easily metabolized.[11–14] The rate of excretion of the PCDD and PCDF congeners from the human body *via* the urine and feces tends to be considerably slower than that from other mammals.[15] It has been proved that the PCDD, PCDF, and polychlorinated biphenyl (PCB) congeners are excreted through the intestinal wall into the digestive tract.[16–18] This seems to be their main elimination route in humans.[18]

Nori is a traditional Japanese marine foodstuff prepared from *Porphyra yezoensis* UEDA, which is a kind of seaweed belonging to the genus *Porphyra, Rhodophyceae*. Nori is well known to contain a large quantity of protein and dietary fiber, the respective amounts of which are 43.6% and 44.4% on a dry weight basis. Nori is also rich in minerals, vitamins, and chlorophyll as shown in Table 1.

In order to prevent humans from health damage by foods containing dioxin, it is necessary to eat in such a way that the absorption of dioxin *via* the digestive tract can be suppressed and its intake quantity can be reduced. In addition, it is also important for humans exposed to dioxin to develop a method by which the excretion of dioxin into the feces can be accelerated and its accumulation in the body can be decreased.[19–22] We investigated in this study the effects of nori on the gastrointestinal absorption and reabsorption of the PCDD and PCDF congeners and on acceleration of its excretion into the feces.

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**Table 1.** Composition of Nori

| Component | g/100 g |
|---|---|
| Moisture | 8.4 |
| Protein | 39.4 |
| Lipid | 3.7 |
| Carbohydrate | 38.7 |
| Ash | 9.8 |
| Dietary fiber | 31.2 |
| Chlorophyll | 0.680 |
| Carotenoids | 0.025 |
| Vitamin B1 | 0.001 |
| Vitamin B2 | 0.003 |
| Ascorbic acid | 0.108 |

**Table 2.** Composition of the Experimental Diets

| Component | Diet (g/100 g) | | |
|---|---|---|---|
| | Control | 2% Nori | 10% Nori |
| Sucrose | 65 | 63 | 55 |
| Cellulose | 5 | 5 | 5 |
| Casein | 20 | 20 | 20 |
| Corn oil | 5 | 5 | 5 |
| Mineral mixture | 4 | 4 | 4 |
| Vitamin mixture | 0.85 | 0.85 | 0.85 |
| Choline chloride | 0.15 | 0.15 | 0.15 |
| Nori | | 2 | 10 |

## Materials and Methods

*Animals.* Male Wistar rats were purchased from Seac Yoshitomi Co. (Fukuoka, Japan) and kept in the animal facility of the Fukuoka Institute of Health and Environmental Sciences. They were raised in metabolic cages and freely supplied with water and a diet while exposed to a 12/12h light/dark cycle. The composition of the control diet is shown in Table 2. The mineral and vitamin mixtures (Harper combination) and casein were purchased from Oriental Yeast Co. (Tokyo, Japan). Animal care and use conformed to published NIH guidelines.

*Samples and chemicals.* Nori, a marine foodstuff prepared from the thalli of *Porphyra yezoensis*, was provided by Shirako Co. (Tokyo, Japan). Seventeen kinds of native PCDD and PCDF standard solutions (Wellington Laboratories, Guelph, Canada) were dissolved in corn oil (Table 3). Each of the $^{13}C$-labeled PCDD and PCDF standard solutions (Wellington Laboratories) was also dissolved in n-nonane as an internal standard for a quantitative analysis of dioxin. Hexane, acetone, chloroform, methanol, and dichloromethane were purchased from Wako Pure Chemicals Industries Co. (Osaka, Japan). These reagents were of the grade required for determination of the residual agricultural chemicals. All other reagents used were of a special grade. Silica gel of silver nitrate was prepared by dissolving 10 g of silver nitrate 5 ml of $H_2O$ while heating, adding 85 g of Kieselgel 60 (70–230 mesh; Merck & Co. Darmstadt, Germany) to the solution, before mixing well and leaving to stand overnight.

*Animal treatment.* In experiment 1 (Fig. 1), the rats were assigned to three groups of four animals each after acclimation to the experimental conditions for 5 days. After overnight starvation, the animals, with an average body weight of 150 g, were administered with 0.2 ml of the dioxin mixture (standard solution A), which had been added to 4 g of the control diet or nori diet, on the first day of the experiment (Tables 2 and 3). The rats were subsequently fed with the control diet, 2% nori diet, or 10% nori diet for 5 consecutive days. Each diet did not contain the dioxin mixture. The dioxin mixture administered on the first day amounted to 233 ng TEQ/kg of body weight.

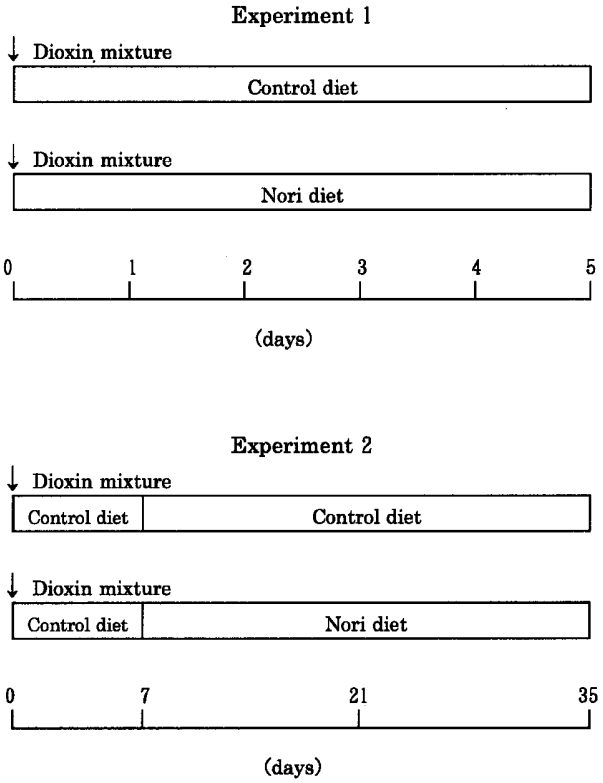


**Fig. 1.** Administration of the Dioxin Mixture and Nori Diet.

In experiment 2 (Fig. 1), the rats with an average body weight of 112 g were administered with 0.2 ml of the dioxin mixture (standard solution B) after overnight starvation, which had been added to 4 g of the control diet on the first day of the experiment (Tables 2 and 3), before being raised on the control diet without the dioxin mixture for 7 consecutive days. After one week, two groups among the three were fed on either the control diet or the 10% nori diet from the 8th day to the 35th day. The dioxin mixture administered in this experiment was 2991 ng

**Table 3.** Concentrations of PCDD and PCDF Congeners in Dioxin Mixtures Dissolved in Corn Oil

| Dioxin | Concentration ($\mu$g/l) | |
|---|---|---|
| | A | B |
| 2,3,7,8-TCDD | 50 | 500 |
| 1,2,3,7,8-PentaCDD | 50 | 500 |
| 1,2,3,4,7,8-HexaCDD | 50 | 500 |
| 1,2,3,6,7,8-HexaCDD | 50 | 500 |
| 1,2,3,7,8,9-HexaCDD | 50 | 500 |
| 1,2,3,4,6,7,8-HeptaCDD | 50 | 500 |
| OctaCDD | 100 | 1000 |
| 2,3,7,8-TCDF | 50 | 500 |
| 1,2,3,7,8-PentaCDF | 50 | 500 |
| 2,3,4,7,8-PentaCDF | 50 | 500 |
| 1,2,3,4,7,8-HexaCDF | 50 | 500 |
| 1,2,3,6,7,8-HexaCDF | 50 | 500 |
| 1,2,3,7,8,9-HexaCDF | 50 | 500 |
| 2,3,4,6,7,8-HexaCDF | 50 | 500 |
| 1,2,3,4,6,7,8-HeptaCDF | 50 | 500 |
| 1,2,3,4,7,8,9-HeptaCDF | 50 | 500 |
| OctaCDF | 100 | 1000 |
| Total TEQ | 169.02 | 1690.2 |

TEQ/kg of body weight. In experiments 1 and 2, the rats were housed individually in metabolic cages, the diet and water were provided *ad libitum*. The body weight, food intake, and fecal weight of each animal were measured. Feces were dried overnight at 70°C. At the end of each experiment, the rats were anesthetized with ether, and the whole bodies were homogenized in a vertical cutter mixer (R-3 plus; FMI Co., Osaka, Japan). Each resulting homogenate was stored at −20°C until the dioxin content was determined.

*Analysis of PCDD and PCDF congeners.* Fecal samples from each rat were homogenized and quantitatively extracted with 150 ml of chloroform-methanol (2:1, v/v) in a cylindrical glass-fiber filter by Soxhlet extraction. The extract of each sample was concentrated to approximately 5 ml by evaporation and then diluted with chloroform to a final volume of 50 ml. To analyze the dioxin level in each fecal sample, 10–30 ml of the extract was put into a test tube (50 ml), concentrated and dried. After adding 200–1000 pg of a $^{13}$C-labeled internal standard (Wellington Laboratories), 10 ml of 1 M KOH in ethanol was added to each sample, before hydrolyzing overnight at room temperature. After adding 10 ml of hexane and 5 ml of $H_2O$, the alkaline hydrolysate of each sample was shaken, centrifuged at 2500 rpm for 10 min, and the hexane layer was then collected. The aqueous layer was extracted twice with 10 ml of hexane, and all the hexane extracts were combined. The collected hexane layer was washed with 5 ml of $H_2O$ and concentrated to approximately 20 ml. After being washed 4 times with 10 ml of conc. $H_2SO_4$, the hexane extract was concentrated to 2 ml, applied to a column containing 0.8 g of silver nitrate (7 mm in diameter) and eluted from the column with 8 ml of hexane, the eluate then being concentrated to 1 ml. The resulting eluate was applied to a column containing 0.6 g of Florisil (7 mm in diameter; U.S. Silica Company, New York, NY, USA), and the PCDD and PCDF congeners were eluted with 8 ml of dichloromethane after being washed with 4 ml of hexane. The eluate from the column was dried and then dissolved in 50 $\mu$l of n-nonane.

Approximately 10 g of the homogenate from the whole body of each rat was put into a test tube (50 ml) for centrifugation, and then 200–1000 pg of a $^{13}$C-labeled internal standard (Wellington Laboratories) was added. A 10-ml amount of 1.5 M KOH in ethanol was then added, and the mixture hydrolyzed overnight at room temperature. The subsequent procedure for analysis was carried out in the same way as that for the fecal samples. The PCDD and PCDF congeners were analyzed by gas chromatography-mass spectrometry (AutoSpec-Ultima; Micromass, Manchester, England) with a capillary column (0.25 mm × 60 m, BPX 5; SGE Co., Yokohama, Japan), setting the resolution mode at 10,000. The PCDD and PCDF congeners in each sample were quantitatively determined in the selected ion monitoring (SIM) mode.

*Statistics.* Differences between the control group and each test group fed on a nori diet were statistically tested with the Student-t test, and a p-value of less than 0.05 is considered significant.

## Results

*Effects of nori on the food intake, body weight, and fecal quantity*

In experiments 1 and 2, there was no significant difference in the body weight gain and food intake between the control group and the test group after administering the dioxin mixture (Table 4). However, the fecal amount from each test group was significantly higher than that of the control group ($p<0.01$ or $p<0.05$).

*Effects of nori on the fecal excretion and accumulation in the body of the PCDD and PCDF congeners (Experiment 1)*

The amounts of the PCDD and PCDF congeners excreted into the feces during the period from the 1st day to the 5th day after administering the dioxin mixture are shown in Table 5. In the test group fed on the 2% nori diet, the fecal excretion of the PCDD and PCDF congeners was higher ($p<0.01$ and $p<0.05$) by 1.1–2.5-fold than that of the control group. In the group fed on the 10% nori diet, the fecal excretion of the PCDD and PCDF congeners was higher ($p<0.01$

and $p<0.05$) by 1.6–6.6-fold than that of the control group, except for 1,2,3,4,6,7,8,9-octachlorodibenzo-*p*-dioxin (octaCDD). As the amount of nori added to the diet was increased from 2% to 10%, the fecal excretion of the PCDD and PCDF congeners was also increased, indicating that the nori diet significantly inhibited the absorption of the PCDD and PCDF congeners contained in the diet.

Table 6 shows the amounts of the PCDD and PCDF congeners in the body on the 5th day after administering the dioxin mixture to the rats. Compared with the control diet, the 2% nori diet reduced the amounts of the PCDD and PCDF congeners in the body by 7.3–64.0% ($p<0.01$) except for 2,3,7,8-tetrachlorodibenzo-*p*-dioxin (TCDD), 2,3,7,8-tetrachlorodibenzofuran (TCDF), 1,2,3,7,8-pentachlorodibenzofuran (pentaCDF), and 2,3,4,7,8-pentaCDF, while the 10% nori diet reduced the amounts of the PCDD and PCDF congeners in the body by 7.5–85.8% ($p<0.01$), except for TCDF.

Increasing the amount of nori in the diet from 2% to 10% decreased the amounts of the PCDD and PCDF congeners that accumulated in the body. The results of this study clearly show that the nori diets inhibited the absorption and accumulation of the PCDD and PCDF congeners.

**Table 4.** Effect of the Nori Diets on the Food Intake, Body Weight Gain, and Feces Weight During the Period from Days 1 to 5 in Rats Administered with the Dioxin Mixture (Experiment 1) and During the Period from Days 8 to 35 in Rats Administered with the Dioxin Mixture (Experiment 2)

| Diet | Food intake | Body weight gain | Feces weight |
|---|---|---|---|
| | | Experiment 1 | |
| | (g/5 days) | (g/5 days) | (g/5 days) |
| Control | 98.2±5.6 | 54.4±3.4 | 5.0±0.6 |
| 2% Nori | 100.7±3.6 | 53.3±2.1 | 6.0±0.3* |
| 10% Nori | 102.4±2.9 | 57.0±4.7 | 9.8±0.7** |
| | | Experiment 2 | |
| | (g/28 days) | (g/28 days) | (g/28 days) |
| Control | 596.6±35.0 | 170.9±21.7 | 39.1±2.6 |
| 10% Nori | 594.8±31.3 | 175.1±10.6 | 63.7±5.4** |

Each value represents the mean ± SD ($n=4$). *Significantly different from the control group ($p<0.05$). **Significantly different from the control group ($p<0.01$).

*Effects of nori on the fecal excretion of the PCDD and PCDF congeners and their accumulation in the body (Experiment 2)*

The fecal excretion of the PCDD and PCDF congeners during the period from the 8th day to the 35th day after administering the dioxin mixture are shown in Table 7. In the test group fed on the 10% nori diet, the fecal excretion of the PCDD and PCDF congeners was 1.5–2.4-fold higher ($p<0.01$ and $p<0.05$) than that of the control group, except for octaCDD, 1,2,3,7,8-pentaCDF, 1,2,3,7,8,9-hexachlorodibenzofuran (hexaCDF), and octaCDF. These results reveal that the nori diet inhibit the reabsorption of the PCDD and PCDF congeners which had been secreted again into the digestive tract through the intestinal wall, and that the nori diet facilitated their excretion into the feces.

The percentages of the PCDD and PCDF con-

**Table 5.** Effect of the Nori Diets on the Fecal Excretion of PCDD and PCDF Congeners in Rats Administered with the Dioxin Mixture (Experiment 1)

| Dioxin | Diet | | |
|---|---|---|---|
| | Control | 2% Nori | 10% Nori |
| 2,3,7,8-TCDD | 2.0±0.3 | 3.5±1.0(175.0)* | 11.0±0.7(550.0)** |
| 1,2,3,7,8-PentaCDD | 4.1±1.2 | 10.6±2.8(258.5)** | 27.2±1.9(663.4)** |
| 1,2,3,4,7,8-HexaCDD | 11.8±3.5 | 28.4±4.9(240.7)** | 51.2±4.2(433.9)** |
| 1,2,3,6,7,8-HexaCDD | 12.9±4.1 | 28.8±4.3(223.3)** | 49.5±3.2(383.7)** |
| 1,2,3,7,8,9-HexaCDD | 24.3±5.6 | 45.4±4.3(186.8)** | 63.7±3.8(262.1)** |
| 1,2,3,4,6,7,8-HeptaCDD | 47.2±5.8 | 71.7±3.2(151.9)** | 79.1±6.0(167.6)** |
| OctaCDD | 79.9±1.4 | 95.1±2.5(119.0)** | 85.8±6.5(107.4) |
| 2,3,7,8-TCDF | 1.2±0.3 | 2.0±0.5(166.7)* | 7.3±0.3(608.3)** |
| 1,2,3,7,8-PentaCDF | 6.1±1.9 | 15.6±3.4(255.7)** | 34.5±2.3(565.6)** |
| 2,3,4,7,8-PentaCDF | 3.8±1.3 | 8.5±2.1(223.7)** | 22.9±1.7(602.6)** |
| 1,2,3,4,7,8-HexaCDF | 16.9±4.6 | 39.3±4.3(232.5)** | 60.5±3.9(358.0)** |
| 1,2,3,6,7,8-HexaCDF | 16.0±4.0 | 36.3±4.4(226.9)** | 56.6±3.5(353.8)** |
| 1,2,3,7,8,9-HexaCDF | 15.7±4.3 | 35.1±4.0(223.6)** | 55.3±2.8(352.2)** |
| 2,3,4,6,7,8-HexaCDF | 18.3±4.7 | 39.7±3.9(216.9)** | 61.2±3.7(334.4)** |
| 1,2,3,4,6,7,8-HeptaCDF | 45.1±5.1 | 67.7±3.2(150.1)** | 74.5±5.4(165.2)** |
| 1,2,3,4,7,8,9-HeptaCDF | 33.7±5.0 | 59.7±4.3(177.2)** | 73.1±5.1(216.9)** |
| OctaCDF | 73.4±1.6 | 92.2±2.0(125.6)** | 85.8±7.4(116.9)* |
| Total TEQ | 6.3±1.6 | 13.8±2.4(219.0)** | 27.9±1.5(442.9)** |

Each value represents the mean of the percentage of dose ± SD ($n=4$) during the period from days 1 to 5 in rats administered with the dioxin mixture; the acceleration index of fecal excretion is shown in parentheses [(percentage fecal excretion of PCDD and PCDF congeners by rats on the nori diet)/(percentage fecal excretion of PCDD and PCDF congeners by rats on the control diet) × 100]. *Significantly different from the control group ($p<0.05$). **Significantly different from the control group ($p<0.01$).

**Table 6.** Effect of the Nori Diets on the Body Burden of PCDD and PCDF Congeners in Rats Administered with the Dioxin Mixture (Experiment 1)

| Dioxin | Diet | | |
|---|---|---|---|
| | Control | 2% Nori | 10% Nori |
| 2,3,7,8-TCDD | 95.9±1.3 | 94.4±1.2 (98.4) | 88.7±3.3(92.5)** |
| 1,2,3,7,8-PentaCDD | 94.3±2.5 | 87.4±1.6 (92.7)** | 70.7±3.4(75.0)** |
| 1,2,3,4,7,8-HexaCDD | 84.8±4.0 | 72.1±3.7 (85.0)** | 48.1±1.0(56.7)** |
| 1,2,3,6,7,8-HexaCDD | 83.8±3.4 | 68.5±4.5 (81.7)** | 46.5±2.6(55.5)** |
| 1,2,3,7,8,9-HexaCDD | 72.8±5.9 | 49.7±4.8 (68.3)** | 28.1±2.1(38.6)** |
| 1,2,3,4,6,7,8-HeptaCDD | 47.9±10.3 | 27.4±3.0 (57.2)** | 14.0±1.3(29.2)** |
| OctaCDD | 21.1±3.1 | 7.6±1.4 (36.0)** | 3.0±0.4(14.2)** |
| 2,3,7,8-TCDF | 19.7±5.7 | 19.8±2.9(100.5) | 16.8±4.9(85.3) |
| 1,2,3,7,8-PentaCDF | 44.5±6.7 | 43.5±2.3 (97.8) | 31.4±1.4(70.6)** |
| 2,3,4,7,8-PentaCDF | 88.8±5.3 | 84.8±2.9 (95.5) | 71.3±3.2(80.3)** |
| 1,2,3,4,7,8-HexaCDF | 80.8±6.2 | 59.5±5.1 (73.6)** | 34.6±1.8(42.8)** |
| 1,2,3,6,7,8-HexaCDF | 78.3±5.9 | 60.0±0.9 (76.6)** | 34.5±1.7(44.1)** |
| 1,2,3,7,8,9-HexaCDF | 69.5±5.3 | 56.3±3.1 (81.0)** | 35.8±1.8(51.5)** |
| 2,3,4,6,7,8-HexaCDF | 79.7±6.7 | 59.1±4.0 (74.2)** | 34.2±1.7(42.9)** |
| 1,2,3,4,6,7,8-HeptaCDF | 41.0±4.8 | 18.7±1.5 (45.6)** | 8.3±0.6(20.2)** |
| 1,2,3,4,7,8,9-HeptaCDF | 58.8±5.5 | 35.9±2.5 (61.1)** | 18.4±1.5(31.3)** |
| OctaCDF | 20.0±3.7 | 7.9±1.9 (39.5)** | 3.1±0.5(15.5)** |
| Total TEQ | 87.3±2.2 | 80.4±0.9 (92.1)** | 66.6±2.6(76.3)** |

Each value represents the mean of the percentage of dose±SD ($n=4$) on day 5 in rats administered with the dioxin mixture; the acceleration index of disappearance from the body on day 5 is shown in parentheses [(percentage body burden of PCDD and PCDF congeners by rats on the nori diet)/(percentage body burden of PCDD and PCDF congeners by rats on the control diet)×100]. **Significantly different from the control group ($p<0.01$).

geners in body on the 8th day after administering the dioxin mixture were 85.2% for TCDD, 89.9% for 1,2,3,7,8-pentachlorodibenzo-*p*-dioxin (pentaCDD), 80.3% for 1,2,3,4,7,8-hexachlorodibenzo-*p*-dioxin (hexaCDD), 80.7% 1,2,3,6,7,8-hexaCDD, 63.3% for 1,2,3,7,8,9-hexaCDD, 51.9% for 1,2,3,4,6,7,8-heptachlorodibenzo-*p*-dioxin (heptaCDD), 21.7% for 1,2,3,4,6,7,8,9-octaCDD, 2.8% for 2,3,7,8-TCDF, 21.4% for 1,2,3,7,8-pentaCDF, 85.5% for 2,3,4,7,8-pentaCDF, 76.9% for 1,2,3,4,7,8-hexaCDF, 73.0% for 1,2,3,6,7,8-hexaCDF, 59.7% for 1,2,3,7,8,9-hexaCDF, 75.2% for 2,3,4,6,7,8-hexaCDF, 37.6% for 1,2,3,4,6,7,8-heptachlorodibenzofuran (heptaCDF), 54.8% for 1,2,3,4,7,8,9-heptaCDF, 20.8% for octaCDF, and 80.3% for total TEQ. Since both 2,3,7,8-TCDF and 1,2,3,7,8-pentaCDF are relatively easily metabolized,[23,24] their amounts in the body during the 7 days after administering the dioxin mixture were less than those of the other PCDD and PCDF congeners.

The amounts of the PCDD and PCDF congeners in the body 35 days after their administration are shown in Table 8. In the group fed on the control diet and 10% nori diet, the fecal excretion of the PCDD and PCDF congeners from the 8th day to the 35th day after administering the dioxin mixture was less than 4.29% for all the congeners (Table 7). We infer from this result that the PCDD and PCDF amounts stored in the body 35 days after the administration were little different between the control diet and 10% nori diet. However, the amounts of 2,3,7,8-TCDF and 1,2,3,7,8-pentaCDF in the body were reduced to 23.5% and 52.8% ($p<0.01$ and $p<0.05$), respectively, of the amounts for the control group. This indicates that the nori diet may have had the effect of promoting the metabolism of the PCDD and PCDF congeners or of promoting the fecal excretion of the

**Table 7.** Effect of the Nori Diets on the Fecal Excretion of PCDD and PCDF Congeners in Rats Administered with the Dioxin Mixture (Experiment 2)

| Dioxin | Diet | |
|---|---|---|
| | Control | 10% Nori |
| 2,3,7,8-TCDD | 1.19±0.20 | 2.91±1.03 (244.5)* |
| 1,2,3,7,8-PentaCDD | 1.13±0.22 | 2.63±0.76 (232.7)* |
| 1,2,3,4,7,8-HexaCDD | 1.04±0.13 | 2.32±0.46 (223.1)** |
| 1,2,3,6,7,8-HexaCDD | 1.02±0.14 | 2.14±0.35 (209.8)** |
| 1,2,3,7,8,9-HexaCDD | 1.30±0.13 | 2.23±0.35 (171.5)** |
| 1,2,3,4,6,7,8-HeptaCDD | 1.94±0.39 | 3.09±0.43 (159.3)** |
| OctaCDD | 3.24±0.24 | 4.29±1.04 (132.4) |
| 2,3,7,8-TCDF | 0.007±0.002 | 0.016±0.003(228.6)** |
| 1,2,3,7,8-PentaCDF | 0.12±0.03 | 0.20±0.09 (166.7) |
| 2,3,4,7,8-PentaCDF | 0.27±0.06 | 0.66±0.09 (244.4)** |
| 1,2,3,4,7,8-HexaCDF | 0.73±0.08 | 1.59±0.10 (217.8)** |
| 1,2,3,6,7,8-HexaCDF | 0.65±0.09 | 1.34±0.10 (206.2)** |
| 1,2,3,7,8,9-HexaCDF | 0.91±0.21 | 1.40±0.71 (153.8) |
| 2,3,4,6,7,8-HexaCDF | 0.67±0.08 | 1.20±0.09 (179.1)** |
| 1,2,3,4,6,7,8-HeptaCDF | 1.40±0.31 | 2.14±0.40 (152.9)* |
| 1,2,3,4,7,8,9-HeptaCDF | 1.12±0.16 | 2.06±0.27 (183.9)** |
| OctaCDF | 1.99±0.55 | 2.83±0.80 (142.2) |
| Total TEQ | 0.92±0.15 | 2.12±0.60 (230.4)** |

Each value represents the mean of the percentage of dose±SD ($n=4$) during the period from days 8 to 35 in rats administered with the dioxin mixture; the acceleration index of fecal excretion is shown in parentheses [(percentage fecal excretion of PCDD and PCDF congeners by rats on the nori diet)/(percentage fecal excretion of PCDD and PCDF congeners by rats on the control diet)×100]. *Significantly different from the control group ($p<0.05$). **Significantly different from the control group ($p<0.01$).

**Table 8.** Effect of the Nori Diets on the Body Burden of PCDD and PCDF Congeners in Rats Administered with the Dioxin Mixture (Experiment 2)

| Dioxin | Diet | |
|---|---|---|
| | Control | 10% Nori |
| 2,3,7,8-TCDD | 52.1±7.7 | 35.8±13.9 (68.7) |
| 1,2,3,7,8-PentaCDD | 65.3±4.3 | 51.9±12.4 (79.5) |
| 1,2,3,4,7,8-HexaCDD | 63.7±3.4 | 60.7±8.9 (95.3) |
| 1,2,3,6,7,8-HexaCDD | 76.8±4.7 | 75.6±5.3 (98.4) |
| 1,2,3,7,8,9-HexaCDD | 49.8±5.1 | 48.1±8.2 (96.6) |
| 1,2,3,4,6,7,8-HeptaCDD | 46.1±5.4 | 45.6±2.2 (98.9) |
| OctaCDD | 15.9±2.5 | 16.5±1.3 (103.8) |
| 2,3,7,8-TCDF | 0.68±0.07 | 0.52±0.04 (76.5)** |
| 1,2,3,7,8-PentaCDF | 3.6±0.1 | 1.7±0.8 (47.2)* |
| 2,3,4,7,8-PentaCDF | 79.6±2.6 | 78.3±6.1 (98.4) |
| 1,2,3,4,7,8-HexaCDF | 74.8±6.2 | 74.1±4.2 (99.1) |
| 1,2,3,6,7,8-HexaCDF | 71.1±3.2 | 70.0±3.7 (98.5) |
| 1,2,3,7,8,9-HexaCDF | 25.7±3.5 | 13.9±11.3 (54.1) |
| 2,3,4,6,7,8-HexaCDF | 69.6±5.8 | 69.0±7.4 (99.1) |
| 1,2,3,4,6,7,8-HeptaCDF | 35.8±6.0 | 35.5±1.6 (99.2) |
| 1,2,3,4,7,8,9-HeptaCDF | 51.3±6.5 | 51.5±0.8 (100.4) |
| OctaCDF | 11.9±2.3 | 14.0±2.1 (117.6) |
| Total TEQ | 59.7±4.2 | 50.1±9.9 (83.9) |

Each value represents the mean of the percentage of dose ± SD ($n=4$) on day 35 in rats administered with the dioxin mixture; the acceleration index of disappearance from the body on day 35 is shown in parentheses [(percentage body burden of PCDD and PCDF congeners by rats on the nori diet)/(percentage body burden of PCDD and PCDF congeners by rats on the control diet)×100]. *Significantly different from the control group ($p<0.05$). **Significantly different from the control group ($p<0.01$).

PCDD and PCDF metabolites.

## Discussion

We investigated in this study how efficiently a nori diet inhibited the absorption of the PCDD and PCDF congeners contained in food through the digestive tract and promoted their excretion into the feces (Table 5). When the rats were given either the 2% nori diet or the 10% nori diet, the fecal excretion of highly toxic TCDD, 1,2,3,7,8-pentaCDD, and 2,3,4,7,8-pentaCDF was increased by 1.7–5.5-, 2.5–6.6-, and 2.2–6.0-fold, respectively, compared with the control diet ($p<0.0$l and $p<0.05$). By using rats that had accumulated the PCDD and PCDF congeners in the body beforehand, we also investigated whether or not a nori diet could inhibit the reabsorption of the PCDD and PCDF congeners which had been excreted into the digestive tract through the intestinal wall[15,16] and accelerate their fecal excretion (Table 7). The rats fed on the 10% nori diet showed increased fecal excretion of highly toxic TCDD, 1,2,3,7,8-pentaCDD, and 2,3,4,7,8-pentaCDF by 2.4-, 2.3-, and 2.4-fold, respectively, compared with the case of the control diet ($p<0.01$ and $p<0.05$).

These results clearly indicated that a nori diet was effective for inhibiting the absorption of the PCDD and PCDF congeners contained in the diet *via* the digestive tract and for preventing their accumulation in the body. A nori diet was also effective for inhibiting the reabsorption of the PCDD and PCDF congeners which are secreted again into the digestive tract *via* internal elimination systems, and for facilitating their elimination from the body.

The mechanism for their metabolism and excretion is assumed to be the conclusive point for applying the results obtained from rats to the human case. In the case of rats, the PCDD and PCDF congeners are metabolized to hydrophilic metabolites and excreted into both the feces and urine.[15,24–29] The amounts of the PCDD and PCDF congeners excreted into the urine, however, are much less than those into the feces, so it is considered that fecal excretion of the PCDD and PCDF congeners is the main route in rats.[15,24–29] The ratio of the amounts of non-metabolites (parent compounds) in the feces of rats to total elimination of the PCDD and PCDF congeners was 9.3% for TCDD, 1.0% for 2,3,7,8-TCDF, and 9.9% for 2,3,4,7,8-pentaCDF.[15] The amounts of the parent compounds excreted were much less than those of their metabolites.

In contrast, the excretory process for the PCDD and PCDF congeners in humans is not yet clear. The ratio of parent compounds in the feces to total elimination of the PCDD and PCDF congeners has been reported to be 37–90% in human beings.[18] In the case of TCDD being dissolved in corn oil and given to humans, the ratio of parent compounds excreted into the feces to the total amount of TCDD excreted into the feces was about 50%.[29] The excretion of parent compounds into the feces is therefore assumed to be the main route for excretion of the PCDD and PCDF congeners from the human body.

The results of the present study show that the excreted amounts of parent compounds during the period from the 8th day to the 35th day after administering the dioxin mixture were increased by 2.4-fold for TCDD ($p<0.05$), 2.3-fold for 1,2,3,7,8-pentaCDD ($p<0.05$), and 2.4-fold for 2,3,4,7,8-pentaCDF ($p<0.01$) when compared to the case of the control diet. If the ratio of the fecal excretion of parent compounds to total elimination of the PCDD and PCDF congeners in humans is 90%, a 10% nori diet is suggested to reduce the biological half-life of TCDD from 7.5 years[11,12] to 3.3 years, that of 1,2,3,7,8-pentaCDD from 15.7 years[14] to 7.2 years, and that of 2,3,4,7,8-pentaCDF from 19.6 years[13] to 8.6 years.

We have previously investigated whether or not the excretion of PCDF congeners from the body could be accelerated by administering rice-bran fiber to rats and humans. With rats, a 10% rice-bran fiber diet increased the fecal excretion of 2,3,4,7,8-pentaCDF by 4.5-fold ($p<0.01$) more than the case of the control diet.[19] Although the administration of cholestyramine (12 g/day) did not increase the excretion of 2,3,4,7,8-pentaCDF from the human body,[30]

the administration of cholestyramine (12 g/day) together with rice-bran fiber (18 g/day) increased the excretion of 2,3,4,7,8-pentaCDF by 1.6–2.6-fold more than that before the administration.[31] These results made it clear that rice-bran fiber was useful for accelerating the excretion of PCDF congeners into the feces for both rats and humans.

A recent experiment carried out to investigate the acceleration of TCDD excretion in humans[32] has indicated that Olestra (sucrose polyester, 66 g/day) increased the fecal excretion of TCDD by 8–10-fold more than before giving Olestra and that this was sufficient to reduce the normally observed elimination half-life of TCDD from about 7 years to 1–2 years.

In order to prevent human beings from health damage in health by dioxin, it is important to suppress the gastrointestinal absorption of dioxin contained in foods, to facilitate its excretion into the feces, and ultimately to reduce the intake of dioxin by the human body. Since the biological half-lives of dioxin compounds are rather long,[11–14] it is also important for humans exposed to dioxin to be prevented from reabsorbing it, whereby it is directly excreted again into the digestive tract from the gastrointestinal wall, to increase its fecal excretion, and finally to reduce the amount accumulated in the body.[19–22]

According to the results of experiments on rats, dietary fiber[19] and chlorophyll[22] efficiently accelerated the excretion of the PCDD and PCDF congeners into the feces, so it may be expected that nori, which is rich in dietary fiber and chlorophyll, would have the advantage of accelerating the excretion of the PCDD and PCDF congeners from the human body.

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