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## Effects of 4-week continuous ingestion of champignon extract on halitosis and body and fecal odor

Jun Nishihira<sup>a,\*</sup>, Mie Nishimura<sup>a</sup>, Aiko Tanaka<sup>a</sup>, Akihiro Yamaguchi<sup>b</sup>, Toshio Taira<sup>b</sup><sup>a</sup> Department of Medical Management and Informatics, Hokkaido Information University, Hokkaido, Japan<sup>b</sup> Primary Cell Division, Cosmo Bio Co., Ltd., Sapporo, Japan

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## ABSTRACT

This was placebo-controlled double-blind parallel-group comparative clinical trial targeting 80 men and women aged 50–79 years with halitosis and body and fecal odor. We investigated whether daily champignon extract ingestion for 4 weeks improved these conditions. Subjects were divided into four groups: a placebo group and 50, 500, and 1000 mg/day ingestion groups. No severe adverse events or side effects were noted during the study period. Compared with the placebo group, all champignon extract ingestion groups showed improvement or tendency toward improvement in halitosis and body and fecal odor. Furthermore, our results suggested that the effectiveness of champignon extract in alleviating odors is dose-dependent, i.e., it increases with the dosage.

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### 1. Introduction

*Kareishu* (literally, distinctive body odor of the middle-aged or elderly), which is believed to frequently occur in men aged  $\geq 40$  years,<sup>1</sup> is considered to be more common in modern society because of high animal fat-containing modern diets and high levels of stress. Halitosis and body and fecal odor negatively affect peoples' quality of life, and they are increasingly becoming a serious problem in caregiving settings in Japan, with the rapidly increasing proportion of the elderly in the Japanese society.

To manufacture champignon extract, an extract boiled from the mushroom *Agaricus bisporus* (*tsukuritake* or champignon mushroom) is mixed with dextrin and then spray-dried it into a powder. The extract contains amino acids, polyphenols, polysaccharides, flavonoids, vitamins, and minerals. Currently, this extract has been patented and is available for sale in Japan, six European countries, South Korea, the United States, and Canada. The most significant feature of champignon extract is deodorization within the intestinal tract.

Conditions such as halitosis and body and fecal odor are believed to be caused by certain toxic substances produced

within the intestinal tract; champignon extract directly inhibits their production. Thus, champignon extract is widely used in candy, jellies, drinks, and other food products for maintaining both personal esthetics and health. Champignon extract shows a deodorizing effect by inhibiting ammonia nitrogen generation.<sup>2</sup> Furthermore, in an *in vitro* trial using gas chromatography to investigate its effects on the methyl mercaptan component of halitosis, champignon extract was found to increase the molecular weight of methyl mercaptan, thereby inhibiting its odor and causing overall decreased odor. This was because it converts the mercapto group of methyl mercaptan to sulfo group. Studies regarding the function of champignon extract and the utility of its characteristic effects against methyl mercaptan are underway.<sup>2</sup>

In a clinical trial targeting elderly inpatients (70 subjects; mean age: males 73.5 years, females 78.6 years) and involving ingestion of 2 g of champignon extract daily for 30 days, the extract was shown to not affect gastrointestinal symptoms but improved stool characteristics, reduced fecal and body odor, and decreased blood ammonia content. In another trial involving 24 residents of an elderly care facility who ingested jelly candies containing 300 mg of champignon extract daily for 30 days, favorable effects such as improved "stool characteristics" and "life satisfaction" were observed.

Here we aimed to investigate whether daily champignon extract ingestion for 4 weeks improved halitosis and body and fecal odor.

\* Corresponding author. Hokkaido Information University, Department of Medical Management and Informatics, Ebetsu City, Hokkaido 069-8585, Japan. Fax: +81 11 385 4411.

E-mail address: [nishihira@do-johodai.ac.jp](mailto:nishihira@do-johodai.ac.jp) (J. Nishihira).

## 2. Material and methods

### 2.1. Study design

We included 80 men and women aged 50–79 years with problematic halitosis and body and fecal odor in this placebo-controlled double-blind parallel-group comparative study. It involved four study groups: consisting of 3 test foods and a placebo, as shown in Table 1. (1) a placebo group ( $n = 20$ ), (2) 50 mg/day champignon ingestion group ( $n = 20$ ), (3) 500 mg/day champignon ingestion group ( $n = 20$ ), and (4) 1000 mg/day champignon ingestion group ( $n = 20$ ). The ingestion period was 4 weeks. With regard to ingestion volume and ingestion method, the test food was ingested as one packet (2 g) per day. However, we did not implement any restrictions regarding method or frequency of ingestion. The Champignon Extract BX100FPD (Ricom Corporation, Tokyo, Japan) used for ingestion.

Each subject underwent a medical interview and blood test on the day of starting ingestion of the test food and after 2 and 4 weeks of ingestion. The week before starting ingestion was set as the pre-observation period (washout period).

### 2.2. Study procedure

This study was approved by the Bioethics Committee of Hokkaido Information University. After providing the subjects with written and oral explanations of the study contents in accordance with the Declaration of Helsinki, we obtained written consent to participate from the subjects. After initially screening, this study included 80 consenting subjects, who fulfilled the selection criteria and not the exclusion criteria, and were believed to be suitable to participate by the principal investigator. For group allocation, an allocation manager from a third-party data center (Media Education Center, Hokkaido Institute of Information Technology, Ebetsu, Hokkaido) referred to the subject list and equally divided the subjects via stratified randomization into four groups considering age composition, male-to-female ratio, and odor questionnaire scores. The allocation manager carefully stored the allocation-related documents containing personal information of the subjects in a locked cabinet. Subjects were then notified of the date, time, and place for the clinical trial. Because three subjects quit the study owing to personal reasons before the trial started, only 77 subjects were finally included.

Subjects ingested one packet (2 g) per day of test food or placebo for 4 weeks from the day of starting ingestion. Subjects arrived at the assigned facilities on the day of starting ingestion and after 2 and 4 weeks of ingestion and underwent testing regarding prescribed items. The tests were conducted at the Health Center, Hokkaido Information University, Ebetsu Internal Medicine Clinic, Takahashi Internal Medicine 3Ban-dori Clinic, and Taguchi Clinic (all in Ebetsu, Hokkaido). Subjects began recording information in their lifestyle journals (daily condition, whether the test food was

ingested, etc.) and also documented their bowel movements 1 week before starting ingestion and then throughout the 4-week study period until the completion of stool sampling. The journals were submitted on each clinic day and every time feces were sampled.

For disclosure (display of allocation list), when all relevant test results and analysis data had been prepared, the allocation manager displayed the subject allocation list. The ingestion rate was calculated with the following formula: Ingestion rate (%) = (number of test foods actually ingested/number of test foods planned to be ingested)  $\times$  100.

### 2.3. Ethics committee

All subjects provided written informed consent before undergoing any study-related tests, and the study protocol was approved by the Ethics Committee of Hokkaido Information University (a certificate number; 2014-04). The study protocol conformed to the Helsinki Declaration and was registered at the UMIN Clinical Trial Registration System (certificate number UMIN000014256).

### 2.4. Statistical analysis

The Wilcoxon rank sum test was used for intragroup comparisons and the Mann–Whitney U and chi-square tests were used for intergroup comparisons.

### 2.5. Study items

#### 2.5.1. Visual analogue scale (VAS) questionnaire

For VAS questionnaire, 100-mm lines were prepared for each item with the left and right edges indicating worst and best states, respectively. We evaluated how subjects felt about their own state at the time of the questionnaire by having them mark an “X” on the relevant part of each line. The questionnaire results were scaled by measuring the length from the left edge to the “X” mark.

Subjects and cooperating people (those living along with the subjects) were asked to answer the questionnaire regarding halitosis and body and fecal odor using the prescribed method. Subjects were instructed to bathe and brush their teeth the night before and sleep using a clean pillow cover (or to cover the pillow with a towel) while wearing freshly laundered pajamas. The cooperating person evaluated halitosis after standing one handbreadth away from the subject and speaking to the subject for 1–2 min. For body odor evaluation, the odor of the pillow cover (or towel) and pajamas was evaluated by the subject and cooperating person.

For the questionnaire items regarding bowel movements, subjects evaluated the bowel movement status during the days between two clinic visits. However, if subjects could not record results at the time of awakening on the test day, they were allowed to include results from up to 2 days before the test day. We investigated the following four items: (1) fecal odor, (2) bowel movement

**Table 1**  
Composition of placebo and test food materials.

Composition of materials	Quantity contained (mg)			
	Placebo	Test food		
		50 mg/day champignon	500 mg/day champignon	1000 mg/day champignon
Champignon extract BX100FPD	0	50	500	1000.00
Dextrin	1503.5	1458.1	1050.1	596.7
Glucose	400	400	400	400
Malic acid	90	85.5	45	0
Caramel (food color)	6.5	6.4	4.9	3.3

regularity, (3) strain during bowel movements, and (4) sensation of residual stools. To evaluate fecal odor, the cooperating person evaluated the smell after the subject used the toilet.

### 2.5.2. Bowel movement and lifestyle journals

The following characteristics regarding bowel movement were recorded on a daily basis: (a) bowel movement frequency, (b) bowel movement volume (visual estimate of number of sample containers the stools would fill), (c) stool shape, (d) stool color, (e) stool odor, and (f) sensation of having completely evacuated after each bowel movement. The evaluation periods were the 1-week pre-observation period (non-ingestion period) and 4-week test food ingestion period. On days when multiple bowel movements were experienced, subjects recorded items from (b) through (f) for the first bowel movement only. In the lifestyle journal, subjects recorded meal content, physical condition, and whether they ingested the test food daily.

### 2.5.3. Physical measurements and blood testing

Blood samples were taken for testing on the starting day and after 4 weeks of ingestion. In addition to a medical interview by a doctor, height, weight, body mass index (BMI), and percent body fat were measured. Vital signs (blood pressure upon arrival, pulse rate, body temperature) were also taken. General blood tests included complete blood count (WBC, RBC, Hgb, Hct, and platelets), liver function test (AST, ALT,  $\gamma$ -GTP, ALP, and LDH), kidney function (BUN, Cre, and UA), blood lipid profile (total cholesterol, TG, LDL-cholesterol, and HDL-cholesterol), and blood sugar test (blood sugar and HbA1c).

### 2.5.4. Food frequency questionnaire

For the food frequency questionnaire, we used FFQg Ver4 (Kenpakusha, Tokyo, Japan). We used a questionnaire composed of 29 food groups and 10 types based on food classifications from the National Health and Nutrition Survey. With 1-week units, we estimated volume of intake by food group and nutrient intake based on portion size and food frequency. We calculated mean values and standard deviation for the intake volume of nutrients (calories, protein, fat, carbohydrates, dietary fiber, and salt) on the day of starting ingestion and after 4 weeks of ingestion.

## 3. Results

Table 2 presents the male-to-female ratio, mean age, weight, BMI, and percent body fat of the four subject groups: placebo group ( $n = 19$ ), 50 mg/day ingestion group ( $n = 18$ ), 500 mg/day ingestion group ( $n = 20$ ), and 1000 mg/day ingestion group ( $n = 20$ ). For the continuous variables (age, weight, BMI, and percent body fat), we performed the Student's *t*-test with two independent samples. We used Fisher's exact test to calculate the number and proportion of male subjects in each group for the categorical variable of sex. With

regard to the analysis groups, no statistically significant differences were observed for ingestion rates between the placebo group and all ingestion groups (Table 3).

### 3.1. VAS questionnaire for subjects

Mean scores and standard deviation were calculated for each group for the results of the VAS questionnaire regarding the seven items of halitosis, pillow odor, pajama odor, fecal odor, bowel movement regularity, strain during bowel movements, and sensation of residual stools. We then calculated the amount of change in mean scores and standard deviation for each group for each measurement time point (evaluation time point) from before starting ingestion (Table 4). With regard to the amount of change, we also calculated frequency distribution after dividing these results into five levels (<20, 20–39, 40–59, 60–79, and  $\geq 80$  changes expression in mm). The results for each of these seven items are shown below.

#### 3.1.1. Halitosis

The respective amount of change in VAS questionnaire scores regarding halitosis from before starting ingestion to after 2 weeks of ingestion and from before starting ingestion to after 4 weeks of ingestion was as follows: placebo group,  $10.95 \pm 16.99$  mm and  $19.11 \pm 18.89$  mm; 50 mg/day group,  $19.33 \pm 20.40$  mm and  $22.44 \pm 22.59$  mm; 500 mg/day group,  $13.75 \pm 17.59$  mm and  $17.85 \pm 20.78$  mm; 1000 mg/day group,  $20.00 \pm 19.22$  mm and  $29.32 \pm 25.56$  mm. When test multiplicity was adjusted and the level of significance for each test was set at 2.5%, halitosis significantly improved after 2 and 4 weeks of ingestion compared with that before starting ingestion ( $p = 0.001$ ,  $p = 0.001$ ). After 2 weeks of ingestion, differences were also observed between all ingestion groups and the placebo group. After 4 weeks of ingestion, differences were observed between the 500 and 1000 mg/day ingestion groups and the placebo group.

#### 3.1.2. Pillow odor

The respective amount of change in VAS questionnaire scores regarding pillow odor from before starting ingestion to after 2 weeks of ingestion and from before starting ingestion to after 4 weeks of ingestion was as follows: placebo group,  $9.00 \pm 18.10$  mm and  $10.82 \pm 22.36$  mm; 50 mg/day group,  $11.11 \pm 10.53$  mm and  $12.11 \pm 17.78$  mm; 500 mg/day group,  $19.05 \pm 22.02$  mm and  $21.10 \pm 26.00$  mm; 1000 mg/day group,  $12.20 \pm 16.47$  mm and  $17.50 \pm 22.99$  mm. When test multiplicity was adjusted and the level of significance for each test was set at 2.5%, pillow odor significantly improved after 2 and 4 weeks of ingestion compared with that before starting ingestion ( $p = 0.004$ ,  $p = 0.003$ ). After 2 weeks of ingestion, differences were also observed between all ingestion groups and the placebo group. After 4 weeks of ingestion,

**Table 2**  
Demographic data for subjects.

Group	Placebo	50 mg/day champignon ingestion group	500 mg/day champignon ingestion group	1000 mg/day champignon ingestion group
Number of subjects	n = 19	n = 18	n = 20	n = 20
	Ave. $\pm$ SD	Ave. $\pm$ SD	Ave. $\pm$ SD	Ave. $\pm$ SD
Age(years old)	64.4 $\pm$ 7.8	64.8 $\pm$ 7.3	64.0 $\pm$ 7.9	63.2 $\pm$ 6.8
Body weight(kg)	60.63 $\pm$ 9.95	57.84 $\pm$ 9.55	60.12 $\pm$ 9.03	59.19 $\pm$ 11.31
BMI	23.54 $\pm$ 2.72	23.12 $\pm$ 2.62	23.18 $\pm$ 2.72	22.83 $\pm$ 2.81
Body fat (%)	26.93 $\pm$ 6.64	25.78 $\pm$ 7.37	26.78 $\pm$ 6.85	27.02 $\pm$ 5.14
Number of male subjects	9	9	11	9
Proportion of male (%)	47.4	50.0	55.0	45.0

**Table 3**  
Ingestion rates for placebo and test foods.

Group	Placebo	50 mg/day champignon	500 mg/day champignon	1000 mg/day champignon
Number	n = 19	n = 18	n = 20	n = 20
Ingestion rate (%)	Ave. $\pm$ SD 98.31 $\pm$ 3.44	Ave. $\pm$ SD 99.21 $\pm$ 1.96	Ave. $\pm$ SD 98.57 $\pm$ 3.36	Ave. $\pm$ SD 98.57 $\pm$ 2.93

differences were observed between the 50 and 500 mg/day ingestion groups and the placebo group.

### 3.1.3. Pajama odor

The respective amount of change in VAS questionnaire scores regarding pajama odor from before starting ingestion to after 2 weeks of ingestion and from before starting ingestion to after 4 weeks of ingestion was as follows: placebo group, 5.05  $\pm$  11.13 mm and 9.37  $\pm$  17.36 mm; 50 mg/day group, 8.06  $\pm$  13.33 mm and 7.00  $\pm$  17.23 mm; 500 mg/day group, 20.75  $\pm$  22.19 mm and 21.70  $\pm$  28.25 mm; 1000 mg/day group, 12.15  $\pm$  17.33 mm and 15.00  $\pm$  20.48 mm. When test multiplicity was adjusted and the level of significance for each test was set at 2.5%, pajama odor significantly improved after 2 and 4 weeks of ingestion compared with that before starting ingestion ( $p = 0.005$ ,  $p = 0.004$ ). After 2 weeks of ingestion, differences were also observed between all ingestion groups and the placebo group. After 4 weeks of ingestion, differences were observed between the 500 and 1000 mg/day ingestion groups and the placebo group.

### 3.1.4. Fecal odor

The respective amount of change in VAS questionnaire scores regarding fecal odor from before starting ingestion to after 2 weeks of ingestion and from before starting ingestion to after 4 weeks of ingestion was as follows: placebo group, 12.95  $\pm$  19.35 mm and 21.63  $\pm$  23.03 mm; 50 mg/day group, 11.22  $\pm$  15.34 mm and 15.56  $\pm$  19.57 mm; 500 mg/day group, 19.26  $\pm$  14.30 mm and 27.10  $\pm$  21.27 mm; 1000 mg/day group, 18.95  $\pm$  21.98 mm and 30.95  $\pm$  22.14 mm. When test multiplicity was adjusted and the level of significance for each test was set at 2.5%, fecal odor significantly improved after 2 and 4 weeks of ingestion compared with that before starting ingestion ( $p = 0.001$ ,  $p = 0.001$ ). After 2 and 4 weeks of ingestion, differences were also observed between all ingestion groups and the placebo group.

### 3.1.5. Bowel movement regularity

The respective amount of change in VAS questionnaire scores regarding bowel movement regularity from before starting ingestion to after 2 weeks of ingestion and from before starting ingestion to after 4 weeks of ingestion was as follows: placebo group, 3.26  $\pm$  14.67 mm and 8.74  $\pm$  19.38 mm; 50 mg/day group, -1.76  $\pm$  21.60 mm and 6.89  $\pm$  13.42 mm; 500 mg/day group, 5.00  $\pm$  16.06 mm and 1.85  $\pm$  11.80 mm; 1000 mg/day group, -8.22  $\pm$  21.51 mm and -8.82  $\pm$  30.52 mm. When test multiplicity was adjusted and the level of significance for each test was set at 2.5%, no significant differences were observed between values at the day of starting ingestion and at each measurement time point.

### 3.1.6. Strain during bowel movements

The respective amount of change in VAS questionnaire scores regarding strain during bowel movements from before starting ingestion to after 2 weeks of ingestion and from before starting ingestion to after 4 weeks of ingestion was as follows: placebo group, -5.79  $\pm$  30.44 mm and 0.16  $\pm$  17.05 mm; 50 mg/day group, 6.11  $\pm$  32.04 mm and 11.61  $\pm$  16.99 mm; 500 mg/day group,

0.70  $\pm$  13.19 mm and 3.70  $\pm$  11.62 mm; 1000 mg/day group, 8.68  $\pm$  11.89 mm and 5.89  $\pm$  27.58 mm. When test multiplicity was adjusted and the level of significance for each test was set at 2.5%, strain during bowel movements significantly decreased after 2 weeks of ingestion compared with that before starting ingestion ( $p = 0.005$ ).

### 3.1.7. Sensation of residual stools

The respective amount of change in VAS questionnaire scores regarding sensation of residual stools from before starting ingestion to after 2 weeks of ingestion and from before starting ingestion to after 4 weeks of ingestion was as follows: placebo group, -2.21  $\pm$  15.37 mm and 2.00  $\pm$  13.59 mm; 50 mg/day group, 5.89  $\pm$  19.58 mm and 2.94  $\pm$  14.74 mm; 500 mg/day group, 1.45  $\pm$  14.25 mm and -0.45  $\pm$  21.81 mm; 1000 mg/day group, 6.74  $\pm$  13.01 mm and 2.78  $\pm$  24.18 mm. When test multiplicity was adjusted and the level of significance for each test was set at 2.5%, no significant differences were observed between values at the day of starting ingestion and at each measurement time point.

## 3.2. VAS questionnaire for the cooperating people

We implemented a VAS questionnaire regarding odor for cooperating people as well. It comprised the four items of subject halitosis, pillow odor, pajama odor, and fecal odor (odor of feces after subject used the toilet). We then calculated the amount of change in mean scores and standard deviation for each group for each measurement time point (evaluation time point) from before starting ingestion (Table 5). With regard to the amount of change, we also calculated frequency distribution after dividing subjects into five levels (<20, 20–39, 40–59, 60–79, and  $\geq$ 80 years). The results for each of these four items are shown below.

### 3.2.1. Halitosis

The respective amount of change in VAS questionnaire scores regarding halitosis from before starting ingestion to after 2 weeks of ingestion and from before starting ingestion to after 4 weeks of ingestion was as follows: placebo group, 6.58  $\pm$  20.46 mm and 11.58  $\pm$  22.47 mm; 50 mg/day group, -0.53  $\pm$  26.42 mm and 6.94  $\pm$  27.92 mm; 500 mg/day group, 12.50  $\pm$  33.37 mm and 19.75  $\pm$  17.54 mm; 1000 mg/day group, 11.05  $\pm$  27.97 mm and 21.53  $\pm$  29.79 mm. When test multiplicity was adjusted and the level of significance for each test was set at 2.5%, halitosis significantly improved after 2 weeks of ingestion compared with that before starting ingestion ( $p = 0.005$ ). After 2 weeks of ingestion, differences were also observed in the 50 and 500 mg/day ingestion groups compared with the placebo group. After 4 weeks of ingestion, differences were observed in all ingestion groups compared with the placebo group.

### 3.2.2. Pillow odor

The respective amount of change in VAS questionnaire scores regarding pillow odor from before starting ingestion to after 2 weeks of ingestion and from before starting ingestion to after 4 weeks of ingestion was as follows: placebo group, 5.95  $\pm$  14.05 mm and 8.63  $\pm$  20.35 mm; 50 mg/day group, 10.81  $\pm$  22.81 mm and

**Table 4**  
VAS questionnaire for subjects on odor.

	Halitosis			Pillow odor			Pajama odor			Fecal odor		
	Before ingestion	After 2 weeks of ingestion	After 4 weeks of ingestion	Before ingestion	After 2 weeks of ingestion	After 4 weeks of ingestion	Before ingestion	After 2 weeks of ingestion	After 4 weeks of ingestion	Before ingestion	After 2 weeks of ingestion	After 4 weeks of ingestion
Placebo group	52.53 ± 24.74	63.47 ± 23.72 <sup>a</sup>	71.63 ± 18.64 <sup>a</sup>	61.88 ± 20.32	72.32 ± 17.39	75.05 ± 16.00	66.95 ± 21.57	72.00 ± 17.07	76.32 ± 16.75	47.53 ± 26.75	60.47 ± 21.59 <sup>a</sup>	69.16 ± 18.79
Amount of change		10.95 ± 16.99	19.11 ± 18.89		9.00 ± 18.10	10.82 ± 22.36		5.05 ± 11.13	9.37 ± 17.36		12.95 ± 19.35	21.63 ± 23.03
50 mg/day champignon ingestion group	48.39 ± 25.60	67.72 ± 21.87 <sup>a</sup>	70.83 ± 18.55 <sup>a</sup>	61.06 ± 24.90	72.17 ± 19.45 <sup>a</sup>	73.17 ± 20.62 <sup>a</sup>	64.44 ± 24.15	72.50 ± 20.07 <sup>a</sup>	71.44 ± 22.16	47.72 ± 17.91	58.94 ± 22.14 <sup>a</sup>	63.28 ± 20.76 <sup>a</sup>
Amount of change		19.33 ± 20.40 <sup>c</sup>	22.44 ± 22.59		11.11 ± 10.53 <sup>c</sup>	12.11 ± 17.78 <sup>c</sup>		8.06 ± 13.33 <sup>c</sup>	7.00 ± 17.23		11.22 ± 15.34 <sup>c</sup>	15.56 ± 19.57 <sup>c</sup>
500 mg/day champignon ingestion group	46.30 ± 15.26	60.05 ± 16.66 <sup>a</sup>	64.15 ± 18.15 <sup>a</sup>	53.75 ± 23.19	72.80 ± 17.33 <sup>a</sup>	74.85 ± 17.24 <sup>a</sup>	53.20 ± 23.44	73.95 ± 16.04 <sup>a</sup>	74.90 ± 18.67 <sup>a</sup>	38.35 ± 13.43	58.16 ± 15.43 <sup>a</sup>	65.45 ± 18.60 <sup>a</sup>
Amount of change		13.75 ± 17.59 <sup>c</sup>	17.85 ± 20.78 <sup>c</sup>		19.05 ± 22.02 <sup>c</sup>	21.10 ± 26.00 <sup>c</sup>		20.75 ± 22.19 <sup>c</sup>	21.70 ± 28.25 <sup>c</sup>		19.26 ± 14.30 <sup>c</sup>	27.10 ± 21.27 <sup>c</sup>
1000 mg/day champignon ingestion group	44.79 ± 20.44	62.40 ± 19.43 <sup>a</sup>	71.95 ± 16.46 <sup>a</sup>	62.65 ± 25.84	74.85 ± 16.53 <sup>a</sup>	80.15 ± 12.09 <sup>a</sup>	64.85 ± 23.03	77.00 ± 16.06 <sup>a</sup>	79.85 ± 12.18 <sup>a</sup>	37.63 ± 22.27	56.55 ± 21.41 <sup>a</sup>	69.30 ± 14.44 <sup>a</sup>
Amount of change		20.00 ± 19.22 <sup>c</sup>	29.32 ± 25.56 <sup>c</sup>		12.20 ± 16.47 <sup>c</sup>	17.50 ± 22.99		12.15 ± 17.33 <sup>c</sup>	15.00 ± 20.48 <sup>c</sup>		18.95 ± 21.98 <sup>c</sup>	30.95 ± 22.14 <sup>c</sup>

<sup>b</sup>Statistically significant compared with the placebo ( $p < 0.05$ , Mann–Whitney).

<sup>a</sup> Statistically significant vs before taking test foods or placebo ( $p < 0.05$ , Wilcoxon).

<sup>c</sup> Statistically significant compared with the placebo ( $p < 0.05$ , Chi-square).

**Table 5**  
VAS questionnaire for the cooperating people on odor.

	Halitosis			Pillow odor			Pajama odor			Fecal odor		
	Before ingestion	After 2 weeks of ingestion	After 4 weeks of ingestion	Before ingestion	After 2 weeks of ingestion	After 4 weeks of ingestion	Before ingestion	After 2 weeks of ingestion	After 4 weeks of ingestion	Before ingestion	After 2 weeks of ingestion	After 4 weeks of ingestion
Placebo group	60.37 ± 27.66	66.95 ± 23.92	71.89 ± 15.93	62.32 ± 27.66	68.16 ± 24.17	70.89 ± 18.16	64.11 ± 28.12	69.53 ± 21.90	74.89 ± 17.14 <sup>a</sup>	47.37 ± 24.70	54.47 ± 21.71	62.32 ± 19.14 <sup>a</sup>
Amount of change		6.58 ± 20.46	11.58 ± 22.47		5.95 ± 14.05	8.63 ± 20.35		5.53 ± 15.07	10.89 ± 17.72		7.37 ± 21.68	15.05 ± 21.65
50 mg/day champignon ingestion group	66.41 ± 29.40	65.82 ± 27.40	73.29 ± 21.14	59.75 ± 25.35	70.63 ± 28.53	70.00 ± 30.84	64.13 ± 26.08	71.06 ± 28.90	74.94 ± 29.54	49.47 ± 22.33	59.94 ± 30.47	67.12 ± 25.67 <sup>a</sup>
Amount of change		-0.53 ± 26.42 <sup>c</sup>	6.94 ± 27.92 <sup>c</sup>		10.81 ± 22.81	14.63 ± 26.08 <sup>c</sup>		6.94 ± 23.10 <sup>c</sup>	10.81 ± 24.68 <sup>c</sup>		10.35 ± 26.97	17.59 ± 28.05 <sup>c</sup>
500 mg/day champignon ingestion group	44.70 ± 26.07	56.95 ± 22.95	64.50 ± 24.24 <sup>a</sup>	47.05 ± 24.62	62.35 ± 23.55 <sup>a</sup>	64.55 ± 25.79 <sup>a</sup>	49.75 ± 25.05	62.90 ± 22.80 <sup>a</sup>	69.90 ± 22.94 <sup>a</sup>	33.25 ± 18.56	49.60 ± 19.45 <sup>a</sup>	56.55 ± 21.78 <sup>a</sup>
Amount of change		12.50 ± 33.37 <sup>c</sup>	19.75 ± 17.54 <sup>c</sup>		15.45 ± 21.85 <sup>c</sup>	17.50 ± 22.81 <sup>c</sup>		13.20 ± 19.26	20.15 ± 22.00 <sup>c</sup>		16.40 ± 17.97	23.30 ± 18.50 <sup>c</sup>
1000 mg/day champignon ingestion group	54.26 ± 26.64	65.21 ± 20.41	75.79 ± 18.23 <sup>a</sup>	56.00 ± 26.44	70.32 ± 16.34 <sup>a</sup>	77.16 ± 15.67 <sup>a</sup>	58.74 ± 27.00	72.58 ± 19.09 <sup>a</sup>	78.79 ± 15.60 <sup>a</sup>	44.79 ± 23.07	60.79 ± 17.05 <sup>a</sup>	70.26 ± 19.99 <sup>a</sup>
Amount of change		11.05 ± 27.97	21.53 ± 29.79 <sup>c</sup>		14.16 ± 21.88 <sup>c</sup>	21.11 ± 27.06 <sup>c</sup>		13.89 ± 23.25 <sup>c</sup>	20.05 ± 30.24 <sup>c</sup>		16.11 ± 27.49 <sup>c</sup>	25.58 ± 24.49 <sup>c</sup>

<sup>b</sup>Statistically significant compared with the placebo ( $p < 0.05$ , Mann–Whitney).

<sup>a</sup> Statistically significant vs before taking test foods or placebo ( $p < 0.05$ , Wilcoxon).

<sup>c</sup> Statistically significant compared with the placebo ( $p < 0.05$ , Chi-square).

14.63 ± 26.08 mm; 500 mg/day group, 15.45 ± 21.85 mm and 17.50 ± 22.81 mm; 1000 mg/day group, 14.16 ± 21.88 mm and 21.11 ± 27.06 mm. When test multiplicity was adjusted and the level of significance for each test was set at 2.5%, body odor significantly improved after 2 and 4 weeks of ingestion compared with that before starting ingestion ( $p = 0.011$ ,  $p = 0.003$ ). After 2 weeks of ingestion, differences were also observed in the 500 and 1000 mg/day ingestion groups compared with the placebo group. After 4 weeks of ingestion, differences were observed in all ingestion groups compared with the placebo group.

### 3.2.3. Pajama odor

The respective amount of change in VAS questionnaire scores regarding pajama odor from before starting ingestion to after 2 weeks of ingestion and from before starting ingestion to after 4 weeks of ingestion was as follows: placebo group, 5.53 ± 15.07 mm and 10.89 ± 17.72 mm; 50 mg/day group, 6.94 ± 23.10 mm and 10.81 ± 24.68 mm; 500 mg/day group, 13.20 ± 19.26 mm and 20.15 ± 22.00 mm; 1000 mg/day group, 13.89 ± 23.25 mm and 20.05 ± 30.24 mm. When test multiplicity was adjusted and the level of significance for each test was set at 2.5%, pajama odor significantly improved after 2 and 4 weeks of ingestion compared with that before starting ingestion ( $p = 0.018$ ,  $p = 0.010$ ). After 2 weeks of ingestion, differences were also observed in the 500 and 1000 mg/day ingestion groups compared with the placebo group. After 4 weeks of ingestion, differences were observed in all ingestion groups compared with the placebo group.

### 3.2.4. Fecal odor (odor after using toilet)

The respective amount of change in VAS questionnaire scores regarding fecal odor (odor after using toilet) from before starting ingestion to after 2 weeks of ingestion and from before starting ingestion to after 4 weeks of ingestion was as follows: placebo group, 7.37 ± 21.68 mm and 15.05 ± 21.65 mm; 50 mg/day group, 10.35 ± 26.97 mm and 17.59 ± 28.05 mm; 500 mg/day group, 16.40 ± 17.97 mm and 23.30 ± 18.50 mm; 1000 mg/day group, 16.11 ± 27.49 mm and 25.58 ± 24.49 mm. When test multiplicity was adjusted and the level of significance for each test was set at 2.5%, fecal odor significantly improved after 2 and 4 weeks of ingestion compared with that before starting ingestion ( $p = 0.021$ ,  $p = 0.001$ ). After 2 weeks of ingestion, differences were also observed only in the 1000 mg/day ingestion groups compared with the placebo group. After 4 weeks of ingestion, differences were observed in all ingestion groups compared with the placebo group.

## 3.3. Bowel movement journal

Subjects in the three ingestion groups recorded in diaries information regarding bowel movement frequency, bowel movement regularity, bowel movement volume, stool shape, stool color, stool odor, and sensation of having completely evacuated after each bowel movement.

### 3.3.1. Results for the 50 and 500 mg/day groups vs. placebo group

When test multiplicity was adjusted and the level of significance for each test was set at 5/3 (=1.67%) for the 50 mg/day group and placebo group, the proportion of responses indicating that stool color was “brown, ocher, or yellow” was significantly higher in the placebo group on the day of starting ingestion and after 4 weeks of ingestion ( $p = 0.002$ ,  $p = 0.001$ ). No significant differences were noted for any evaluation items between the 500 mg/day group and placebo group.

### 3.3.2. Results for the 1000 mg/day group vs. placebo group

The following results clearly suggested the superiority of the test food when the 1000 mg/day group and placebo group were compared. First, when test multiplicity was adjusted and the level of significance for each test was set at 2.5% for bowel movement frequency, the amount of change in bowel movement frequency from before starting ingestion to after 2 and 4 weeks of ingestion was significantly higher in the ingestion groups ( $p = 0.016$ ,  $p = 0.006$ ). When test multiplicity was adjusted and the level of significance for each test was set at 5/2 = 2.5% for bowel movement volume, the amount of change in bowel movement volume from before starting ingestion to after 4 weeks of ingestion was significantly higher in the ingestion group (1000 mg/day) than in the placebo group ( $p = 0.014$ ).

When test multiplicity was adjusted and the level of significance for each test was set at 5/3 (=1.67%) for stool color, the proportion of responses indicating “brown, ocher, or yellow” was significantly higher in the placebo group after 4 weeks of ingestion ( $p = 0.001$ ). When test multiplicity was adjusted and the level of significance for each test was set at 5/3 (=1.67%) for sensation of having completely evacuated after each bowel movement, the proportion of responses indicating “smooth or pleasantly smooth” was significantly higher in the all ingestion group after 2 weeks of ingestion ( $p = 0.001$ ).

### 3.4. Food frequency questionnaire (glycometabolism-related items)

We conducted a survey using the food frequency questionnaire to confirm that there was no bias related to food ingestion during the trial. No significant differences related to caloric intake, carbohydrates, protein, fat, dietary fiber, or salt were observed. We also did not observe any significant differences with regards to blood sugar or HbA1c levels during fasting.

## 4. Discussion

Champignon mushrooms, which have originated from Europe and North America, are currently grown in over 70 countries worldwide. They are the most widely eaten of mushroom variety and are known to have many beneficial nutritional properties. First, they contain large amounts of vitamin D and ergosterol, which are important for bone metabolism. Therefore, they are effective for preventing onset and improving symptoms of osteoporosis.<sup>3,4</sup> They are also rich in minerals such as sodium, calcium, and phosphorus and contain linoleic acid and antioxidants, which have been associated with prevention of arteriosclerosis.<sup>5,6</sup> The foaming components of champignon mushrooms show aromatase-inhibiting<sup>7,8</sup> and anticarcinogenic<sup>9,10</sup> effects and can activate natural killer cells, thus promoting the innate immune system.<sup>11</sup> *In vitro* studies have also reported that champignon mushrooms promote dendritic cell function.<sup>12</sup> Moreover, they show anti-inflammatory properties<sup>13</sup> and have been suggested to inhibit inflammation and cancer cell proliferation through macrophages.<sup>14</sup> Notably, because they inhibit the development of fatty liver,<sup>15</sup> champignon mushrooms can possibly prevent lifestyle-related diseases.

In the present placebo-controlled double-blind parallel-group comparative study targeting 80 men and women aged 50–79 years with problematic halitosis and body and fecal odor, the effects of 4-week daily ingestion of 50, 500, and 1000 mg/day champignon extract on the aforementioned conditions were investigated. Our results demonstrated the efficacy of champignon extract. We also conducted a questionnaire survey regarding bowel movements and found that champignon extract also exhibited bowel movement-related effects. Moreover, no marked differences were observed between the placebo group and all ingestion groups with regard to vital signs, blood test findings, liver function, kidney function, lipid

profile, blood glucose levels, or physical measurement during the ingestion of low, medium, or high doses of champignon extract (data not shown). Furthermore, because most findings were within the normal range, this trial was considered safe.

The following conclusions were drawn from the results of the VAS questionnaire regarding halitosis and body and fecal odor. For each of the champignon extract ingestion groups, improvement or improvement tendencies were observed for halitosis and body and fecal odor compared with the placebo group, even in case of cooperating people's data.

Considering the bowel movement journals, for the 50 mg/day ingestion and placebo groups, the proportion of responses indicating that stool color was "brown, ocher, or yellow" was significantly higher in the placebo group after 2 and 4 weeks of ingestion.

Bowel movement volume significantly improved in the 1000 mg/day group after 4 weeks of ingestion. For sensation of having completely evacuated after each bowel movement, there was a significantly high proportion of responses indicating "cleared out or very cleared out" in the 1000 mg/day group after 2 weeks of ingestion.

These results demonstrate that although no clear improvement was observed in bowel movements with 50 mg/day champignon extract ingestion, whereas they clearly improved with a 500 mg/day ingestion. In particular, results showed that bowel movement improvement could be anticipated by the ingestion of a high dose of champignon extract. Our results strongly suggest that the ingestion of 50–1000 mg/day of our test food may improve halitosis and body and fecal odor. In particular, ingestion of the high dose of 1000 mg/day may result in reduction in the odor and favorable intestinal tract environment.

Conducting component analysis of champignon extract to identify the main components causing the deodorizing effects and determining the optimal dosage and other variables in detail is necessary. VAS questionnaire regarding bowel movements also showed that ingestion of a high dose of champignon extract showed clear improvements. Thus, efficacy in a wide range of areas, from body odor to bowel movements, can be anticipated. Although the safety of champignon mushrooms has been highly reported, there have been no reports on their adverse or toxic effects in humans.<sup>16</sup>

*Kareishu* appears with increasing age, but its incidence is increasing in Japan as a result of the higher animal fat intake through increasingly westernized diets as well as increase social stress. Halitosis and body and fecal odor not only decrease a person's quality of life but also can become serious problems in caregiving settings in Japan because the proportion of the elderly population in Japanese society is rapidly increasing.

In conclusion, champignon extract is efficacious against halitosis and body and fecal odor, and it can also improve the intestinal environment. These findings not only provide extremely helpful data that can contribute to the maintenance and improvement of

health of the people of Japan but also encourage further development of champignon extract as a dietary supplement.

## 5. Conclusion

Champignon extract is efficacious against halitosis and body and fecal odor, and it can also improve the intestinal environment. These findings not only provide extremely helpful data that can contribute to the maintenance and improvement of health of the people of Japan but also encourage further development of champignon extract as a dietary supplement.

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