Daily Intake of Trehalose Is Effective in the Prevention of Lifestyle-Related Diseases in Individuals with Risk Factors for Metabolic Syndrome

Akiko MIZOTE, Mika YAMADA, Chiyo YOSHIKANE, Norie ARAI,
Kazuhiko MARUTA, Shigeyuki ARAI, Shin ENDO, Rieko OGAWA,
Hitoshi MITSUZUMI, Toshio ARIYASU and Shigeharu FUKUDA

Hayashibara Co. Ltd., 675–1 Fujisaki, Naka-ku, Okayama 702–8006, Japan

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Summary We previously performed animal studies that suggested that trehalose potentially prevents the development of metabolic syndrome in humans. To evaluate this possibility, we examined whether trehalose suppressed the progression of insulin resistance in a placebo-controlled, double-blind trial in 34 subjects with a body mass index (BMI) ≥ 23 . The subjects were divided into two groups and were assigned to ingest either 10 g/d of trehalose or sucrose with meals for 12 wk. During the study, body composition and blood biochemical parameters were measured at week 0, 8, and 12. These parameters were also measured 4 wk after the end of intake to confirm the washout of test substances. In the trehalose group, blood glucose concentrations after a 2-h oral glucose tolerance test significantly decreased following 12 wk of intake in comparison with baseline values (0 wk). When a stratified analysis was performed in the subjects whose percentage of truncal fat approached the high end of the normal range, the change in body weight, waist circumference, and systolic blood pressure were significantly lower in the trehalose group than in the sucrose group. Our data indicated that a daily intake of 10 g of trehalose improved glucose tolerance and progress to insulin resistance. Furthermore, these results suggested that trehalose can potentially reduce the development of metabolic syndrome and associated lifestyle-related diseases, such as type 2 diabetes.

Key Words trehalose, glucose tolerance, insulin resistance, lifestyle-related diseases, humans

Trehalose is a nonreducing disaccharide composed of two D-glucose residues. In humans, the main dietary sources of naturally occurring trehalose are mushrooms, baker's and brewer's yeast, and certain types of shrimp (1). Our previous studies on metabolic syndrome using a mouse model showed that trehalose intake in mice fed with a high-fat diet (HFD) suppressed mesenteric adipocyte hypertrophy and decreased impaired glucose tolerance in an oral glucose tolerance test (OGTT) (2). In the mice fed with HFD, we also found that trehalose intake mitigated insulin resistance, reduced mRNA expression of plasminogen activator inhibitor-1 (PAI-1) (2), and increased serum high molecular weight (HMW) adiponectin (3). These effects were also evident in an established obese mouse model when 0.3% (weight/volume) trehalose was consumed via its addition to drinking water (3). The effective dose was 0.2 g/kg of body weight/d and translated to 10 g/d in humans weighing

50 kg (3). Thus, these findings suggested that ingestion of 10 g/d trehalose is effective for reducing the risk of onset of metabolic syndrome in humans. Furthermore, we have shown that trehalose exhibits a protective effect on the pancreatic islets of mice fed with HFD (4). Therefore, trehalose is considered a unique functional disaccharide that potentially provides a novel dietary approach for humans at risk of or with metabolic syndrome.

Metabolic syndrome is defined based on abdominal obesity plus two or more metabolic risk factors, including impaired glucose tolerance, high blood pressure, and dyslipidemia, associated with the presence of type 2 diabetes and arteriosclerosis (5). It is well known that the symptoms of metabolic syndrome are most frequently attributed to visceral fat accumulation, which is closely associated with adipocyte hypertrophy (6, 7). Hypertrophic adipocytes have been shown to reduce insulin sensitivity via the release of adipocytokines, such as tumor necrosis factor- α (TNF- α) and PAI-1, and the decrease of adiponectin (8). Excessive secretion of these adipocytokines can induce insulin resistance and result in the development of impaired glucose tolerance (8). Therefore, mitigation of insulin resistance is very important for the prevention of lifestyle-related diseases, such as type 2 diabetes.

E-mail address: akiko.mizote@hb.nagase.co.jp

Abbreviations: BMI, body mass index; HbA1c, hemoglobin A1c; HFD, high-fat diet; HMW adiponectin, high molecular weight adiponectin; HOMA-IR, homeostasis model assessment as an index of insulin resistance; ND, no data; OGTT, oral glucose tolerance test; PAI-1, plasminogen activator inhibitor-1; SD, standard deviation; TNF- α , tumor necrosis factor- α ; WO, washout.

Fig. 1. Experimental design. The study design is depicted in the flowchart above. Thirty-four subjects were selected by two screenings. Each symbol indicates the following measurement parameters: ○, body composition (body weight, waist circumference, percentage of body fat, percentage of truncal fat, and blood pressure); ●, fasting blood glucose, insulin, and HbA1c; □, total PAI-1, HMW adiponectin, and blood glucose after a 2-h oral glucose tolerance test (OGTT).

As previously stated, it was anticipated that trehalose could play a role in preventing the progression of insulin resistance. In this study, we examined the effect of trehalose on the progression of insulin resistance in humans. We aimed to clarify whether this disaccharide could contribute to the improvement of glucose tolerance and prevention of lifestyle-related diseases, such as type 2 diabetes.

MATERIALS AND METHODS

Test substances. TREHA® (Hayashibara Co. Ltd., Okayama, Japan) was used as the trehalose source in this study and was subdivided into 3.7 g doses. The administered dose was equivalent to 3.3 g of anhydrous trehalose because TREHA® contains more than 98.0% of trehalose dihydrate. Extra-fine granulated sugar (Pearl Ace Corp., Tokyo, Japan) was used as the sucrose source and was subdivided into 3.3 g doses and served as the test substance for the control group during this study. In addition, the caloric content of trehalose is the same as that of sucrose at 4 kcal/g. Both test substances were packed in plain silver film bags so that they were indistinguishable.

Subjects. Healthy adult volunteers with a body mass index (BMI) ≥ 23 were recruited, with 43 applicants (41 males and two females) enrolled in this study. The purpose and prospective outcomes of the study were fully explained to each volunteer. Risk markers for metabolic syndrome were considered to be those at values that were at the higher end of the normal-to-mild range, and the following criteria were set for selecting the subjects: 1) body fat percentage, $\geq 23\%$ in males and $\geq 35\%$ in females; 2) percentage of truncal fat, $\geq 23.8\%$ in males

and $\geq 33.0\%$ in females; 3) the homeostasis model assessment as an index of insulin resistance (HOMA-IR), ≥ 1.7 (normal range, <1.6); or 4) fasting blood glucose in the 100–125 mg/dL range (i.e., at the higher end of the normal-to-borderline diabetic range based on the Japan Diabetes Society).

The screening test for applicants was conducted twice, and 34 subjects (33 males and one female) met at least one eligibility criteria and were selected. The subjects were divided into two groups ($n=17$); one group received trehalose and the other received sucrose. Neither group had any significant differences in terms of fasting blood glucose, HOMA-IR, or percentages of body or truncal fat. The average age in the trehalose group (17 males) and sucrose group (one female, 16 males) was 47.9 ± 7.7 y (range 32–58 y) and 47.2 ± 6.0 y (range 38–57 y), respectively.

Experimental design. The study was designed as a double-blind, parallel-group comparison. A single bag of the assigned test substance was taken by each subject at every meal (3 bags/d) for 12 wk. Therefore, daily intake of the test substances was approximately 10 g/d. The manner in which the subjects prepared the test substances for ingestion varied from sprinkling them on meals to dissolving them in drinks. To evaluate the effects of the test substances, body composition and blood biochemical parameters were measured following 0, 8, and 12 wk of intake and after a 4-wk washout (WO) period. The study was performed between August and November 2012. The experimental design is illustrated in Fig. 1.

This study was performed under the supervision of a medical doctor. While the subjects were not given spe-

cific instructions on the amount of diet and exercise, they were instructed not to change their lifestyle habits, including their dietary, exercise, and medication patterns throughout the study period. To document each subject's lifestyle circumstances during the study period, individuals' exercise regimen, subjective symptoms, physical conditions, and medication intake were recorded using a check sheet. All subjects had access to a doctor who could diagnose their condition when necessary during the study period. All subjects provided informed consent before entry into the study. This study was approved by the Ethics Committee of Hayashibara, Co., Ltd. (Approval number 142) and was conducted in accordance with the Declaration of Helsinki.

Body composition and blood biochemical parameters. All biochemical measurements were performed after an overnight fast. Body weight and percentage of body fat were measured using body composition monitors, HBF-352 (Omron Healthcare Co., Ltd., Kyoto, Japan). The percentage of truncal fat was measured using an abdominal fat analyzer, AB-140 (Tanita Corp., Tokyo, Japan). Blood pressure was measured using blood pressure monitors, HEM-7020 (Omron Healthcare Co., Ltd.). BMI was calculated using the numerical expression: $\text{BMI} = \text{body weight (kg)} / \text{body height (m)}^2$. Waist circumference was defined as the circumference at the level of the umbilicus.

Blood glucose, insulin, and hemoglobin A1c (HbA1c) were assayed at the Okayama Medical Association test center (Okayama, Japan). Total PAI-1 in the blood was assayed by SRL, Inc. (Tokyo, Japan). Serum HMW adiponectin was measured using an ELISA kit (DHWAD0, R&D Systems, Inc., Minneapolis, MN). HOMA-IR was calculated using the numerical expression for fasting insulin ($\mu\text{U/mL}$) $\times$ fasting glucose (mg/dL) concentrations/405.

OGTT. OGTT was performed using 75 g of glucose at baseline (0 wk) and after 12 wk of substance intake. Blood samples were collected before and 2 h after ingestion of 75 g glucose. Blood glucose and insulin concentrations were measured at the Okayama Medical Association test center. The composite index was calculated as follows: $\{10,000 / \text{square root of} [\text{fasting glucose (mg/dL)} \times \text{fasting insulin (}\mu\text{U/mL)}] \times [\text{2-h glucose (mg/dL)} \times \text{2-h insulin (}\mu\text{U/mL)}]\}$ (9, 10).

Stratified analysis. It is well known that the percentage of truncal fat reflects the visceral adipose tissue area (11). Therefore, we selected subjects whose percentage of truncal fat at week 0 was $\geq 23.8\%$ in males and $\geq 33.0\%$ in females. These selection criteria were based on the lower limit of the high-to-normal range described in the instruction manual of the AB-140 abdominal fat analyzer, which corresponded to the mean $+(1/2)$ standard deviation (SD) for the adult data collected by the manufacturer. Thus, 12 subjects in the trehalose group and 13 subjects in the sucrose group were selected, and the effects of consuming the test substance were analyzed.

Statistical analyses. Data analyses were performed to compare the values measured between the two groups or the changes from the initial values within each

group. The data were expressed as mean $\pm$ SD. Differences between the two groups were determined using an unpaired *t*-test. The changes from the baseline within each group were examined using a paired *t*-test. A *p* value of <0.05 was considered statistically significant.

RESULTS

Subject background

Throughout the study period, each subject ingested their test substance a total of 252 times, and the overall intake was 831.6 g/subject according to the experimental design schedule. The rate of test substance intake was not significantly different between the trehalose group ($97.9 \pm 3.8\%$) and the sucrose group ($94.4 \pm 6.7\%$).

During the study period, no clinical signs related to intake of the test substance were observed in either group. No change in dietary habits was observed, which was analyzed using nutrition analysis software (Excel Eiyoun-kun, version 6.0, Kenpakusya, Tokyo, Japan). When caloric intake was compared, no difference was observed between the sucrose and trehalose groups at either week 0 or week 12. When each caloric intake was analyzed as a percentage of the initial value, no difference was observed between the two groups (trehalose group, $98.3 \pm 24.6\%$; sucrose group, $100.1 \pm 26.4\%$).

Body composition and blood biochemical parameters

Mean changes in body composition and blood biochemical parameters of all subjects are shown in Table 1. The baseline parameters measured were not significantly different between the trehalose and sucrose groups.

To investigate the effect of 10 g/d trehalose intake in each meal on glucose tolerance, blood glucose was measured after a 2-h OGTT (Table 1). Before intake of the test substances (week 0), there was no significant difference in blood glucose after a 2-h OGTT between the trehalose and sucrose groups. However, in the trehalose group, blood glucose after a 2-h OGTT following 12 wk of intake showed a significant decrease in comparison with that from 0 wk. On the other hand, blood glucose after the 2-h OGTT in the sucrose group following 12 wk of intake was not significantly different compared with the 0 wk value.

The other parameters did not significantly differ between the groups or between experimental periods.

Stratified analysis by percentage of truncal fat

To determine the effect of trehalose in humans with signs of metabolic syndrome, we selected the subjects whose percentage of truncal fat was at the higher end of the normal range. The measured values of stratified subjects are shown in Table 2. In the stratified subjects, blood glucose after a 2-h OGTT following 12 wk of trehalose intake showed a significant decrease compared with that before intake, which was similar to the analysis results for all subjects. Furthermore, when compared with the percentage of fasting value in blood glucose after a 2-h OGTT at week 12, the trehalose group value was significantly lower than that in the sucrose group.

Individual changes of the measured parameters

To evaluate the suppressive effect of trehalose in each

Table 1. Body composition and blood biochemical parameters of all subjects.

	Trehalose (n=17)					Sucrose (n=17)				
	week 0	week 8	week 12	WO ⁷ week 4	week 0	week 8	week 12	WO week 4	week 0	week 4
Body composition										
Body weight (kg)	77.3±10.1	77.3±10.2	77.2±10.5	76.7±10.7	74.6±8.2	75.1±8.4	75.1±7.9	75.1±8.2	74.6±8.2	75.1±8.2
BMI ¹ (kg/m ²)	26.4±2.8	26.4±2.8	26.3±2.8	26.2±2.9	26.2±2.6	26.3±2.6	26.4±2.5	26.4±2.6	26.2±2.6	26.4±2.6
Body fat (%)	24.2±2.9	25.2±3.1	25.6±3.1	25.9±3.4	24.9±3.5	25.2±3.7	26.5±3.8	26.4±3.2	24.9±3.5	26.4±3.2
Waist circumference (cm)	87.7±8.1	86.7±7.4	87.9±7.4	87.7±8.4	87.5±7.0	87.7±7.6	87.9±8.0	87.6±7.4	87.5±7.0	87.6±7.4
Truncal fat (%)	27.1±5.4	27.8±5.5	26.9±4.2	27.0±5.0	27.6±5.4	28.2±5.7	28.1±5.6	27.8±5.5	27.6±5.4	27.8±5.5
Systolic blood pressure (mmHg)	134.2±14.0	134.6±11.6	131.0±12.2	134.4±12.7	126.5±12.5	132.8±20.2	129.8±19.1	132.5±13.2	126.5±12.5	132.5±13.2
Diastolic blood pressure (mmHg)	85.9±9.8	86.1±11.0	83.6±9.9	86.5±12.1	82.5±12.5	85.1±13.3	81.6±11.9	82.9±10.2	82.5±12.5	82.9±10.2
Blood biochemical parameters										
Fasting blood glucose (mg/dL)	92.6±8.7	92.6±8.7	93.7±10.3	96.2±10.4	92.9±7.3	94.9±10.5	94.1±10.8	98.5±10.7	92.9±7.3	94.1±10.8
Blood glucose after 2-h of OGTT ² (mg/dL)	121.1±27.6	ND ⁶	102.2±22.6*	ND	118.7±30.6	ND	116.3±31.9	ND	118.7±30.6	116.3±31.9
% of fasting value	130.2±21.3	ND	108.9±20.1*	ND	127.8±31.5	ND	123.3±30.4	ND	127.8±31.5	123.3±30.4
Fasting blood insulin (μIU/mL)	7.0±2.9	8.1±5.5	9.0±6.2	8.9±6.8	7.1±3.4	9.4±6.7	7.7±2.6	8.9±5.2	7.1±3.4	7.7±2.6
Blood insulin after 2-h of OGTT (μIU/mL)	50.8±39.2	ND	45.1±32.5	ND	50.1±26.1	ND	55.6±26.3	ND	50.1±26.1	55.6±26.3
% of fasting value	713.4±345.6	ND	538.9±259.5	ND	882.5±636.6	ND	767.9±396.8	ND	882.5±636.6	767.9±396.8
Composite index	6.9±3.8	ND	8.1±6.5	ND	6.5±3.7	ND	5.6±2.4	ND	6.5±3.7	5.6±2.4
HbA1c (%)	5.4±0.2	5.2±0.3	5.3±0.3	5.4±0.3	5.3±0.3	5.1±0.3	5.3±0.2	5.4±0.2	5.3±0.3	5.3±0.2
HOMA-IR ³	1.6±0.7	1.9±1.4	2.1±1.7	2.2±2.0	1.6±0.7	2.2±1.6	1.8±0.7	2.1±1.2	1.6±0.7	1.8±0.7
Total PAI-1 ⁴ (ng/mL)	27.3±13.5	ND	28.4±11.2	ND	22.6±6.8	ND	27.5±9.1	ND	22.6±6.8	27.5±9.1
HMW adiponectin ⁵ (ng/mL)	2,126.2±1,557.7	ND	2,247.7±1,735.7	ND	2,939.9±1,969.0	ND	3,268.8±2,242.9	ND	2,939.9±1,969.0	3,268.8±2,242.9

Values are the means±SD. Statistical analysis was performed using a paired *t*-test. * $p<0.05$ vs. week 0 value. ¹BMI, body mass index; ²OGTT, oral glucose tolerance test; ³HOMA-IR, homeostasis model assessment as an index of insulin resistance; ⁴PAI-1, plasminogen activator inhibitor-1; ⁵HMW adiponectin, high molecular weight adiponectin; ⁶ND, no data; ⁷WO, wash out.

Table 2. Body composition and blood biochemical parameters in subjects with signs of metabolic syndrome.

	Trehalose (n=12)				Sucrose (n=13)			
	week 0	week 8	week 12	WO ⁷ week 4	week 0	week 8	week 12	WO week 4
Body composition								
Body weight (kg)	80.6±10.4	80.1±10.9	80.0±11.3	79.5±11.4	75.2±9.1	75.9±9.4	76.1±8.8	76.1±9.2
BMI ¹ (kg/m ²)	27.3±2.8	27.1±2.9	27.0±3.0	26.9±3.0	26.5±2.9	26.7±2.9	26.8±2.8	26.8±2.9
Body fat (%)	25.0±3.1	26.0±3.4	26.0±3.6	26.5±3.7	25.9±3.2	26.4±3.5	27.7±3.6	27.6±2.6
Waist circumference (cm)	90.5±7.9	88.8±7.6	89.8±8.1	90.1±8.7	88.7±7.6	89.7±7.8	89.8±8.3	89.6±7.4
Truncal fat (%)	29.0±5.3	29.8±5.4	28.3±4.2	28.5±5.2	29.3±4.8	30.0±5.2	30.0±4.8	29.6±5.0
Systolic blood pressure (mmHg)	135.8±16.0	134.9±12.3	130.5±13.2	136.3±14.7	127.0±13.7	136.4±22.0	134.5±19.2	134.2±14.4
Diastolic blood pressure (mmHg)	85.4±9.7	87.0±11.7	82.7±10.1	88.4±12.0	82.8±12.4	87.6±14.2	84.5±12.1	84.5±10.0
Blood biochemical parameters								
Fasting blood glucose (mg/dL)	90.3±7.2	92.4±8.8	92.3±10.5	95.5±11.0	92.7±8.1	95.5±11.6	94.4±12.3	99.2±11.8
Blood glucose after 2-h of OGTT ² (mg/dL)	117.2±16.4	ND ⁶	98.3±22.4*	ND	121.1±32.9	ND	122.0±34.4	ND
% of fasting value	129.7±14.0	ND	106.1±19.1*†	ND	130.7±33.6	ND	129.0±32.5	ND
Fasting blood insulin (μIU/mL)	7.4±3.0	9.3±6.1	10.4±6.9	10.2±7.6	7.6±3.8	10.4±7.4	8.4±2.5	10.0±5.5
Blood insulin after 2-h of OGTT (μIU/mL)	53.8±46.3	ND	45.5±37.9	ND	54.0±26.8	ND	61.8±25.8	ND
% of fasting value	671.2±345.1	ND	438.1±212.0	ND	922.1±677.6	ND	794.5±406.3	ND
Composite index	7.1±4.3	ND	8.7±7.6	ND	5.6±2.0	ND	4.7±1.6	ND
HbA1c (%)	5.4±0.3	5.1±0.3	5.2±0.3	5.4±0.3	5.3±0.3	5.1±0.3	5.3±0.3	5.4±0.2
HOMA-IR ³	1.7±0.8	2.2±1.6	2.5±1.9	2.6±2.4	1.7±0.8	2.5±1.7	2.0±0.7	2.4±1.2
Total PAI-1 ⁴ (ng/mL)	27.0±9.1	ND	28.5±11.8	ND	22.7±7.8	ND	29.7±8.9	ND
HMW adiponectin ⁵ (ng/mL)	2,318.2±1,764.9	ND	2,504.9±1,924.3	ND	2,473.8±2,007.0	ND	2,722.3±2,093.9	ND

Values are the means±SD. Statistical difference between two groups in the same week was calculated by an unpaired *t*-test (**p*<0.05) and between week 0 value and week 12 value was calculated by a paired *t*-test (**p*<0.05). ¹BMI, body mass index; ²OGTT, oral glucose tolerance test; ³HOMA-IR, homeostasis model assessment as an index of insulin resistance; ⁴PAI-1, plasminogen activator inhibitor-1; ⁵HMW adiponectin, high molecular weight adiponectin; ⁶ND, no data; ⁷WO, wash out.

Table 3. Change in values from baseline for each parameter after 12 wk of intake.

	All subjects		Stratified subjects	
	Trehalose (n=17)	Sucrose (n=17)	Trehalose (n=12)	Sucrose (n=13)
Body composition				
Body weight (kg)	-0.2±1.6	0.5±1.5	-0.6±1.7 [†]	0.9±1.0
BMI ¹ (kg/m ²)	-0.1±0.6	0.2±0.5	-0.2±0.6 [†]	0.3±0.4
Body fat (%)	1.4±1.1	1.7±1.2	1.0±1.1	1.8±1.3
Waist circumference (cm)	0.3±2.5	0.4±2.2	-0.8±2.1 [†]	1.2±2.0
Truncal fat (%)	-0.2±2.2	0.5±1.2	-0.7±2.1	0.7±1.1
Systolic blood pressure (mmHg)	-3.2±10.0	3.2±15.1	-5.3±10.7 [†]	7.5±12.9
Diastolic blood pressure (mmHg)	-2.3±5.1	-0.9±10.3	-2.8±5.7	1.7±7.5
Blood biochemical parameters				
Fasting blood glucose (mg/dL)	1.1±7.3	1.2±6.9	1.9±7.6	1.7±7.8
Blood glucose after 2-h of OGTT ² (mg/dL)	-18.9±27.4	-2.4±27.0	-18.8±29.0	0.9±29.3
% of fasting value	-21.4±25.9	-4.5±25.7	-23.5±28.1	-1.7±27.4
Fasting blood insulin (μIU/mL)	2.0±4.2	0.6±4.0	3.0±4.6	0.8±4.6
Blood insulin after 2 h of OGTT (μIU/mL)	-5.7±26.7	5.4±27.3	-8.3±31.4	7.8±30.7
% of fasting value	-174.5±407.8	-114.6±723.1	-233.2±441.4	-127.6±830.8
Composite index	1.3±4.1	-0.9±2.8	1.7±4.8	-0.9±1.9
HbA1c (%)	-0.1±0.1	0.0±0.2	-0.1±0.1	0.0±0.2
HOMA-IR ³	0.5±1.1	0.2±0.9	0.8±1.2	0.3±1.0
Total PAI-1 ⁴ (ng/mL)	1.1±11.3	4.9±9.4	1.5±7.8	7.0±9.4
HMW adiponectin ⁵ (ng/mL)	121.5±421.1	328.9±586.5	155.6±462.9	217.4±546.2

Values are the means±SD. Statistical difference between two groups in the same week was performed unpaired *t*-test ([†]*p*<0.05). ¹ BMI, body mass index; ² OGTT, oral glucose tolerance test; ³ HOMA-IR, homeostasis model assessment as an index of insulin resistance; ⁴ PAI-1, plasminogen activator inhibitor-1; ⁵ HMW Adiponectin, high molecular weight adiponectin.

subject, we analyzed the change of each parameter after 12 wk of intake (Table 3). When all the subjects were analyzed, significant differences were not observed among the two groups. However, in the stratified subjects, body weight, waist circumference, and systolic blood pressure were significantly lower in the trehalose group than in the sucrose group.

DISCUSSION

Using a metabolic syndrome mouse model, we have previously shown that trehalose intake mitigates insulin resistance and decreases impaired glucose tolerance as assessed using OGTT, and these effects are attributed to the suppression of adipocyte hypertrophy (2, 3). In this study, to assess the preventive effect of trehalose on the development of metabolic syndrome, we examined whether this disaccharide suppresses the progression of insulin resistance in humans. Based on our results, it was confirmed that a 12-wk intake of trehalose improved glucose tolerance in the subjects who had a BMI of ≥23. The American Diabetes Association indicates that Asian Americans with a BMI of ≥23 have a high risk of developing type 2 diabetes. Therefore, our finding indicates that trehalose contributes to reducing risk factors for lifestyle-related diseases, such as type 2 diabetes.

The blood glucose concentrations of subjects after a 2-h OGTT were higher than their fasting value, suggest-

ing that their glucose tolerance was slightly impaired. In these subjects, trehalose significantly decreased blood glucose after a 2-h OGTT following 12 wk of intake compared with the baseline value. On the other hand, the 12 wk intake of sucrose failed to decrease the same parameter. Fasting blood glucose was not different before and after the period of substance intake in either group. These results suggest that regular intake of trehalose increased insulin sensitivity after OGTT and this increase probably caused the improvement in glucose tolerance. Our present observation was consistent with the results obtained in our previous mouse study. Therefore, regular intake of trehalose exhibited effects on humans, as well as mice. Impaired glucose tolerance is recognized as the early stage of type 2 diabetes. Based on the results of our human and mouse studies, there is a possibility that trehalose reduces the risk of type 2 diabetes development.

Trehalose has been shown to induce lower insulin secretion than glucose in oral saccharide tolerance tests in humans (12). This property of trehalose is considered to result in the suppression of adipocyte hypertrophy. Insulin promotes storage of lipids in adipocytes, and its excessive secretion can cause hypertrophy of these cells (13, 14). Thus, the results of this study suggest the possibility that excessive insulin secretion can be reduced by ingesting trehalose with meals. To investigate this possibility, we are presently considering in vivo experiments

for monitoring insulin secretion after the ingestion of a meal containing trehalose.

It is considered that adipocyte hypertrophy is closely associated with the increase of visceral fat amount (7). Thus, we analyzed the effects of trehalose intake on the subjects with slightly increased visceral fat accumulation, based on their percentage of truncal fat. We found that body weight, waist circumference, and systolic blood pressure were significantly lower in the trehalose group compared with the sucrose group after 12 wk of intake. It has been reported that adipocyte hypertrophy induces insulin resistance and this aberration is closely related to hypertension (15). Because the sample size of this study was small, significant differences in HOMA-IR or composite index, which indicates the degree of insulin resistance, were not observed. However, it seems likely that trehalose suppresses elevated blood pressure by improving insulin resistance in the subjects with signs of increased visceral fat accumulation. To clarify this possibility, we are considering an investigation with an increased number of subjects.

Trehalose is hydrolyzed to two glucose molecules by the trehalase in the intestine; hence, the concentration of blood glucose after trehalose intake depends on the trehalase activity. Oku and Nakamura have described how Japanese subjects are divided into two groups, those with low and those with high trehalase activity (12). These findings imply that the degree of trehalase activity would affect glucose tolerance in our study subjects. To examine this inference, we conducted a trehalose loading test in the same subjects (data not shown). The subjects were divided into two groups as reported by Oku and Nakamura, those with a lower and higher blood glucose elevation after trehalose loading. As a result, glucose tolerance was similarly improved after consumption of trehalose for 12 wk in both groups. There exists a possibility that trehalose exhibits its effects in the intestinal tract before it is digested to glucose.

Furthermore, we considered that daily trehalose intake may affect the functions of the intestine, such as nutrition absorption, secretion of hormones, inflammatory responses, and composition of bacterial flora. The disaccharide is well known to interact with nutrient components, such as fats and carbohydrates (16, 17), and it is expected to influence nutritional absorption. On the other hand, the glucose-dependent insulinotropic polypeptide, known as an intestinal hormone, stimulates insulin secretion and is involved in adipocyte hypertrophy (18–20). Furthermore, our previous mouse study showed that trehalose ingestion caused a significant decrease in the total number of Peyer's patch lymphocytes and a spontaneous release of the inflammatory cytokine interleukin-6 (21). A recent study has shown that some sweeteners induce impaired glucose tolerance by altering the intestinal microbiota (22). These possibilities must be resolved in the future.

In conclusion, we showed that consuming 10 g/d trehalose over 12 wk improved glucose tolerance in subjects with signs of metabolic syndrome. Furthermore, these results suggested that trehalose has the ability to

reduce the risk of developing lifestyle-related diseases induced by insulin resistance associated with metabolic syndrome.

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