

Original article

The effects of food materials on postprandial hyperglycemia

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Abstract

Purpose: One of the methods for reducing glycation stress is suppression of postprandial hyperglycemia (PPHG). In this study, we picked up food materials with nutrition, *i.e.*, protein, lipids, acetic acid, and dietary fiber, which were reported to have the suppressing effect on PPHG, and their effects were verified.

Methods: The subjects were 20 healthy young men and women (8 men, 12 women, 23.3 ± 1.8 years). As the standard food, 200 g of cooked rice (carbohydrate: 67.8 g) + 2.5 g of seasoning was used. The model food, rich in specific nutrition, included A: Salad chicken (protein: low dose (L) 11.5 g/ high dose (H) 23.0 g), B: olive oil (lipid: 14 g/28 g), C: grain vinegar (acetic acid: 0.63 g/1.26 g), D: indigestible dextrin (dietary fiber: 4.2 g/8.4 g), and E: cabbage (dietary fiber: 0.9 g/1.8 g). The complex foods include F: vinegar rice, G: fried chicken, H: fried chicken + lemon juice, and I: Japanese dumplings (gyoza) + ponzu soy sauce. The test food was ingested for 10 min after starting the test, and in the cases of rice + the model food or rice + complex food, these were ingested prior to rice. FreeStyle Libre Pro (Abbott) was used to continuously measure blood glucose levels in the tissue interstitial fluid. The effect on PPHG was evaluated by the maximum blood glucose change (ΔC_{max}) and the incremental area under the curve (iAUC). This study was approved by the Ethics Review Committee of Doshisha University.

Results: When the model foods A, B, C, and E were ingested in high doses (H), the iAUC tended to be lower than that of the standard food. In particular, the ΔC_{max} and iAUC in test food C were significantly decreased ($p < 0.05$) and the effect was in a dose dependent manner. On the other hand, the model food D did not have a suppressing effect on PPHG. As compared to the standard food, the complex food F tended to lower ΔC_{max} and iAUC, while the complex foods G, H, and I significantly reduced the ΔC_{max} and iAUC ($p < 0.05$).

Conclusion: The suppression of PPHG was strongly influenced by protein and acetic acid in food, and weakly influenced by lipids and dietary fiber in the young subjects. It was suggested that the content of protein and acetic acid in complex foods may be important when expecting side dishes to suppress PPHG.

KEY WORDS: postprandial hyperglycemia, protein, lipids, acetic acid, dietary fiber

Introduction

In the body, reducing sugars, *i.e.* glucose, bind to proteins non-enzymatically, and glycation end products (AGEs), which are the causative factors of aging, are produced and accumulated; these reactions are called “glycation stress”. Glycation stress is one of the risk factors for aging and is a factor that promotes skin aging and diabetic complications^{1,2)}. Methods for reducing glycation stress include suppression of

postprandial hyperglycemia (PPHG), recently referred to as “glucose spikes”, suppression of AGE formation, and promotion of AGE decomposition/excretion. Among these, suppression of PPHG is a measure that can be easily incorporated into a daily diet.

Methods to reduce PPHG include reduction of sugar intake (sugar restriction)³⁾, intake of low glycemic index (GI) foods⁴⁾, intake of specified health foods, *i.e.*, indigestible dextrin⁵⁾, Salacia^{6,7)}, and eating vegetables first^{8,9)}. However,

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the dietary method of eating low GI foods or vegetables first sometimes reduces mental enjoyment due to mental stress on appetite. On the other hand, continuous intake of foods for specified health foods as a daily diet poses a cost problem.

It was already shown that ingestion of beef bowl was effective in suppressing PPHG, and that its action was suggested to be due to the meat containing protein and lipids ^{10, 11}. The pre-meal intake of lemon juice was reported to have a suppressing effect of PPHG, and the action was due to citric acid and polyphenol contained in lemon juice ¹². Also the pre-meal intake of vinegar ¹³ suppressed blood glucose elevation, and this action was possibly due to the acetic acid contained in vinegar. Since general food is a complex combination of various materials and nutritional ingredients, it is difficult to estimate how much food intake affects the increase in postprandial glucose level.

Based on these things, the purpose of this study was to compare and evaluate the effects of nutritional components on PPHG by co-eating various foods rich in protein, lipid, acetic acid, and dietary fibers. By studying the effect of each nutritional component on blood glucose, this study was designed so that we can propose a method for reducing glycative stress from the daily diet menu in a different way from dietary restrictions.

Methods

Subjects

There were 20 subjects who met the following selection criteria ([Table 1](#)). Men and women between the ages of 20 and 29 at the time of obtaining consent to participate in the study. Study participants were healthy persons without

chronic illness. Persons with the ability to give consent after receiving an adequate explanation of the purpose and content of the study, and who volunteered to participate of their own accord after proper understanding and who provided a written consent to participate in this study. Persons who can attend the designated examination date to undergo examination. Persons determined to be suitable as a subject of this study by the principal investigator.

Survey items and inspection contents

As a subject background survey, the subjects themselves completed a blood test along with filling out a questionnaire regarding their age, medical history, and food allergies ([Table 2](#)). For the test, FreeStyle Libre Pro (Abbott Laboratories, Chicago, USA) was used, and the glucose concentration in the interstitial fluid measured during the test period was used as the blood glucose level ¹⁴.

Test protocol

As in the previous reports ¹⁰⁻¹², this study was conducted according to the uniform protocol by the Japan Glycemic Index (GI) Study Group ¹⁵.

Subjects were instructed to observe the following items during the test period. Avoid irregular life, *i.e.*, sleep deprivation, overeating, excessive drinking, and live as usual. It is prohibited to start taking health foods or supplements. Other than that, it is prohibited to affect the test results.

Instructions were given on the day before and on the day of the test so that the following items were observed. Excessive exercise is prohibited. Sleep 6 hours or more the day before the test. Alcohol drinking is prohibited from the day before the test until the end of the test on the day of the

Table 1. Subject profile.

	Unit	Total	Male	Total
Number of subjects		20	8	12
Age	years	23.3 ± 1.8	24.3 ± 2.0	22.6 ± 1.3
Body height	cm	162.7 ± 9.3	170.4 ± 4.5	157.5 ± 7.9
Body weight	kg	57.0 ± 11.2	65.0 ± 12.1	51.7 ± 7.0
BMI		21.4 ± 2.4	22.3 ± 3.3	20.8 ± 1.4

Results are expressed as mean ± standard deviation. BMI, body mass index.

Table 2. Result of the blood chemistry test.

Test item	Unit	Measured value	Reference range
FBG	mg/dL	80.9 ± 6.0	70 - 109
HbA1c	%	5.3 ± 0.2	4.6 - 6.2
Insulin	μU/mL	6.2 ± 2.1	1.7 - 10.4
Total cholesterol	mg/dL	185.9 ± 33.9	120 - 219
TG	mg/dL	83.2 ± 45.9	30 - 149
HDL-C	mg/dL	63.9 ± 13.5	40 - 85
LDL-C	mg/dL	105.7 ± 31.3	65 - 139

Results are expressed as mean ± standard deviation. FBG, fasting blood glucose; TG, triglyceride; HDL-C, high-density lipoprotein cholesterol; LDL-C, low-density lipoprotein cholesterol.

test. For dinner on the day before the pretest and the test, avoid high-lipid foods and do not ingest anything other than water after 22:00. On the day of the test, physical activity that may cause exercise and sweating is prohibited until the end of the test. For women, the test is not conducted during the menstrual period.

During the test, the subjects stayed in a sitting position and were prohibited from making phone calls, sleeping, excessive brain activity, *i.e.*, e-mail, computer, and physical activity.

Subjects self-attached the Libre Pro sensor to the outside of the upper arm two days before the test. During the wearing period of the sensor, no restrictions were placed on bathing, swimming, and exercise. During the test, the standard or test food was ingested for 10 min at 10:00. Subjects then watched a DVD in a sitting position, allowing them to remain relaxed until 12:00, when the test ended.

The standard and test foods were swallowed after chewing 30 times or more. Blood glucose levels were measured 15 min after starting the test food (1st time), 15

min after starting the test (2nd time), 30 min (3rd time), 45 min (4th time), 60 min (5th time), and 90 min (6th time) and 120 min (7th time).

Test food

The nutritional components of the test foods used in the study were calculated using the values indicated on each food (**Table 3-5**). Cooked (steamed) rice 200 g (Sato no Gohan Koshihikari: Sato Foods, Niigata, Japan) + 2.5 g of seasoning (Noritama: Marumiya Food Industry, Suginami-ku, Tokyo, Japan) was used as the standard food.

For the test foods, 5 model foods containing a large amount of nutrition, which were reported to have an effect of reducing PPHG, 4 complex foods containing some or all of those nutrition, and cooked rice were used.

The model foods include A: Salad chicken (Awaji Island Alga salt domestic salad chicken: Prima Ham, Shinagawa-ku, Tokyo), B: Olive oil (Garcia extra virgin olive oil: Tominaga Trading, Kobe, Hyogo, Japan), C: Grain vinegar (Mitsukan,

Table 3. Nutrition facts of test food (Standard food and model foods A-E).

Test food	Serving unit (g)	Energy (Kcal)	Protein (g)	Lipids (g)	Acetic acid (g)	Carbohydrate (g)	Dietary fiber (g)
Standard food	200	294	4.2	0	0	67.8	60.6
AL/AH	255/310	352/410	15.7/27.2	0.95/1.9	uncalculated	68.5/69.2	0.65/0.7
BL/BH	214/228	420/546	4.2/4.2	14/28	0	0/0	0/0
CL/CH	215/230	297.8/301.6	4.25/4.3	0/0	0.63/1.26	68.9/70	0/0
DL/DH	205.2/210.4	298.9 to 301.5/ 303.8 to 309	0/0	0/0	uncalculated	72.4/77	4.8/9
EL/EH	250/300	306/318	4.9/5.6	0/0	0	70.4/73	1.5/2.4

Standard food; cooked rice 200 g, AL; salad chicken 55 g before cooked rice 200 g, AH; salad chicken 110 g before cooked rice 200 g, BL; olive oil 14 g before cooked rice 200 g, BH; olive oil 28 g before cooked rice 200 g, CL; grain vinegar 15 g before cooked rice 200 g, CH; grain vinegar 30 g before cooked rice 200 g, DL; indigestible dextrin 5.2 g before cooked rice 200 g, DH; indigestible dextrin 10.4 g before cooked rice 200 g, EL; cabbage 50g before cooked rice 200 g, EH; cabbage 100g before cooked rice 200 g

Table 4. Nutrition facts of test food (Complex food F, I).

Test food	Serving unit (g)	Energy (Kcal)	Protein (g)	Lipids (g)	Acetic acid (g)	Carbohydrate (g)	Dietary fiber (g)
F	198	290.4	3.7	0	0.6	67.7	0.5
I	282	469.1	12.3	16.2	0.3	67.7	2.6

F, vinegared rice (cooked rice 177 g + sushi vinegar 21 g); I, Japanese dumplings (gyoza) 138 g + ponzu soy sauce 15 g before cooked rice 129 g

Table 5. Nutrition facts of test food (Complex food G, H).

Test food	Serving unit (g)	Energy (Kcal)	Protein (g)	Lipids (g)	Citric acid (g)	Carbohydrate (g)	Dietary fiber (g)
G	284	479.6	20.8	13.4	0	67.8	1.5
H	295	478.7	20.7	13.4	1	67.8	1.5

G, fried chicken 135 g before cooked rice 149 g; H, fried chicken 135 g + lemon juice 15 g before cooked rice 145 g

Handa, Aichi, Japan), D: Indigestible dextrin (Easy Fiber Tokuhō: Kobayashi Pharmaceutical, Chuo-ku, Osaka, Japan), and E: Cabbage (Salad Club shredded cabbage: Salad Club, Chofu, Tokyo).

The complex foods include F: vinegared rice (Sato no Gohan) assorted by sushi vinegar with kelp soup stock (Mitsukan), G: 4 pieces of fried chicken (Tokukara: Nichirei Foods, Chuo-ku, Tokyo), H: 4 pieces of fried chicken (Tokukara) + lemon juice (Pokka lemon 100: Pokka Sapporo Food & Beverage, Shibuya-ku, Tokyo), I: 6 pieces of Japanese dumplings (gyoza) + ponzu soy sauce (Frozen Gyoza: Ajinomoto, Chuo-ku, Tokyo, Ajipon: Mitsukan). When the complex food was eaten together with rice, the total carbohydrate amount was adjusted to the standard food (rice 200 g, carbohydrates: 67.8 g).

- Standard food: rice 200 g + seasoning 2.5 g
- Test food AL (salad chicken low dose + cooked rice): Salad chicken 55 g + rice 200 g + seasoning 2.5 g
- Test meal AH (salad chicken high dose + cooked rice): Salad chicken 110 g + rice 200 g + seasoning 2.5 g
- Test food BL (low dose of olive oil + cooked rice): Olive oil 14 g + rice 200 g + seasoning 2.5 g
- Test food BH (high-dose olive oil + cooked rice): olive oil 28 g + rice 200 g + seasoning 2.5 g
- Test meal CL (low-dose grain vinegar + cooked rice): Grain vinegar 15 g + rice 200 g + seasoning 2.5 g
- Test food CH (high-dose grain vinegar + cooked rice): Grain vinegar 30 g + rice 200 g + seasoning 2.5 g
- Test food DL (low-dose indigestible dextrin + cooked rice): Indigestible dextrin 5.2 g + rice 200 g + seasoning 2.5 g
- Test food DH (high dose of indigestible dextrin + cooked rice): Indigestible dextrin 10.4 g + rice 200 g + seasoning 2.5 g
- Test food EL (low dose cabbage + cooked rice): cabbage 50 g + rice 200 g + seasoning 2.5 g
- Test food EH (high dose cabbage + cooked rice): cabbage 100 g + rice 200 g + seasoning 2.5 g
- Complex food F (vinegared rice): sushi vinegar 21 g + rice 177 g
- Complex food G: fried chicken 135g+rice 149g+ seasoning 2.5 g
- Complex food H: fried chicken 135 g + lemon juice 15 g + rice 145 g + seasoning 2.5 g
- Complex food I: Gyoza 138 g + ponzu soy sauce 15 g + rice 129 g + 2.5 g seasoning

The standard and test foods were consumed 10 min after the start of the test. When ingesting the model food or complex food and the standard food, the model food or complex food was ingested in the first 5 minutes, and then the standard food was ingested. Grain vinegar and indigestible dextrin were ingested after diluting them in 150 mL of water. The complex food F (vinegared rice) was prepared by mixing sushi vinegar with cooked rice and ingesting it 10 min after the start.

Selection of subjects for efficacy analysis

Regarding the efficacy analysis, from the subjects who completed all the prescribed tests and contents of the trials, subject to the following exclusion criteria were excluded.

A person whose behavior impairs the reliability of the test results was remarkably observed. Persons who were found to have met the exclusion criteria or were unable to comply with the restrictions.

Since there were no subjects that met the exclusion criteria, 20 participants were all included in the efficacy analysis.

Statistical analysis

For the efficacy analysis, the below values were measured in the selected subjects; change in blood glucose (Δ blood glucose; Δ BG) obtained by subtracting the value before the test food was ingested (first time; 0 min value) from the measured values after the test food was ingested, the maximum value (Δ Cmax) and incremental area under curve (iAUC) according to the uniform protocol of the Japan Glycemic Index (GI) Study Group.

IMB SPSS Statics 26 (IMB Japan, Tokyo, Japan) was used for the statistical analysis. The paired-t test was performed for comparison between two groups, while Bonferroni test was used for multiple analyses, and it was judged as a significant difference when the risk rate was less than 5% ($p < 0.05$) in a two-sided test. Results were expressed as the average value \pm standard error (SE).

Ethical standards

This study was conducted in compliance with the Declaration of Helsinki (revised at the 2013 WMA Fortaleza General Assembly) and the ethical guidelines for human-based medical research (notification by Ministry of Education, Culture, Sports, Science and Technology [MEXT] and Ministry of Health, Labour and Welfare [MHLW]). The test content was fully explained to the subjects in advance. The test was implemented after the applicant requested participation in the test and received a voluntary consent form. This study was conducted under the deliberation and approval of the Ethics Review Committee of Doshisha University (application No.18039). This test was pre-registered in the public database established by the National University Hospital Meeting (UMIN Test ID: 000034009).

Results

Salad chicken intake and PPHG suppression

Out of 20 total subjects, 10 subjects (4 males, 6 females, 23.3 ± 2.2 years, 161.8 ± 8.0 cm, 55.2 ± 11.2 kg, body mass index: BMI] 20.9 ± 2.6 kg/m²) participated in the test. The test food was salad chicken (A).

The change in Δ BG after starting the test is shown in *Fig. 1-a*. No difference was observed in the fasting blood glucose level (0 min). Blood glucose increased after ingesting each test food, reached the maximum value after 45 min, and then decreased until 120 min later. There was slight difference in the blood glucose level at each time among the subjects, it was not significant. The average value of Δ BG at each time was lower in the model foods AL and AH than in the standard food, but it was not significant.

Food Materials and Postprandial Hyperglycemia

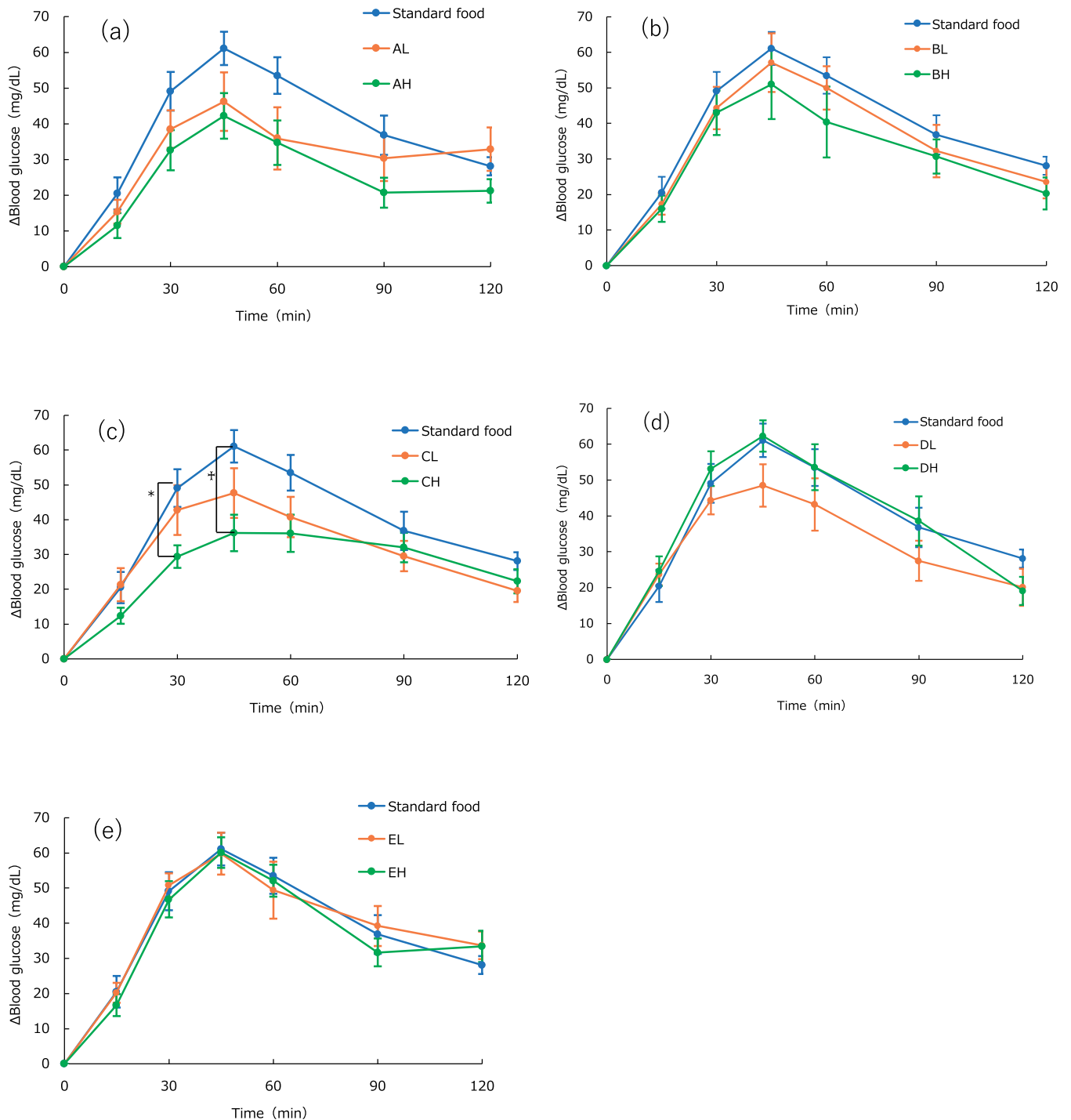


Fig.1. Fluctuation of the blood glucose level at the time of intaking test food ahead of rice.

a) AL and AH, **b)** BL and BH, **c)** CL and CH, **d)** DL and DH, **e)** EL and EH. Results are expressed as mean \pm standard error, $n = 10$, $\dagger p < 0.1$, $* p < 0.05$ vs standard food, Bonferroni test. Standard food, cooked rice 200 g; AL, salad chicken 55 g before cooked rice 200 g; AH, salad chicken 110 g before cooked rice 200 g; BL, olive oil 14 g before cooked rice 200 g; BH, olive oil 28 g before cooked rice 200 g; CL, grain vinegar 15 g before cooked rice 200 g; CH, grain vinegar 30 g before cooked rice 200 g; DL, indigestible dextrin 5.2 g before cooked rice 200 g; DH, indigestible dextrin 10.4 g before cooked rice 200 g; EL, cabbage 50 g before cooked rice 200 g; EH, cabbage 100g before cooked rice 200 g.

ΔC_{max} was 62.7 ± 4.3 mg/dL for the standard food, 54.2 ± 4.2 mg/dL for AL, 46.2 ± 5.7 mg/dL for AH, and no differences were observed (Fig. 2-a). The iAUC was standard food: $4,690 \pm 433$ mg/dL·min, AL: $3,712 \pm 590$ mg/dL·min, and AH: $3,014 \pm 454$ mg/dL·min (Fig. 2-b). The iAUC tended to lower the model food AH by 35.7% compared to the standard food ($p < 0.1$).

Olive oil intake and PPHG suppression

The subjects were the same as participants in the salad chicken test. The test food was olive oil (B). The change in ΔBG after starting the test is shown in Fig. 1-b. The average values of ΔBG from 45 to 120 min were lower in the model foods BL and BH than in the standard food, but it was not significant.

The ΔC_{max} was 62.7 ± 4.3 mg/dL for the standard food, 62.8 ± 5.0 mg/dL for BL, and 56.0 ± 7.7 mg/dL for BH, and no differences were observed (Fig. 2-a). The iAUC was standard food: $4,690 \pm 433$ mg/dL·min, BL: $4,085 \pm 400$ mg/dL·min, BH: $3,811.5 \pm 549$ mg/dL·min, and no difference was observed (Fig. 2-b).

Grain vinegar intake and PPHG suppression

The subjects were the same as the two study participants described above. The test food was grain vinegar (C). The change in ΔBG after starting the test is shown in Fig. 1-c. The average values of ΔBG from 30 to 120 min were lower in the model foods CL and CH than in the standard food. Particularly, the ΔBG at 30 min in CH was 29.0 ± 3.2 mg/dL, which was 20.1 mg/dL lower ($p < 0.05$) compared to that of the standard food (49.1 ± 5.4 mg/dL), also the ΔBG at 45 min tended to be lower ($p < 0.01$).

ΔC_{max} was 62.7 ± 4.3 mg/dL for the standard food, 54.8 ± 3.9 mg/dL for CL, and 41.2 ± 3.9 mg/dL for CH (Fig. 2-a). The iAUC was standard food: $4,690 \pm 433$ mg/dL·min,

CL: $3,780 \pm 371$ mg/dL·min, CH: $3,280 \pm 358$ mg/dL·min (Fig. 2-b). The iAUC of CH was lower by 30.1% than that of the standard food ($p < 0.05$).

Indigestible dextrin intake and PPHG suppression

The subjects were the same as the three study participants described above. The test food was indigestible dextrin (D). The change in ΔBG after starting the test is shown in Fig. 1-d. The average values of ΔBG at each time were almost the same as that of the standard food when the model food DL was ingested. The average values of ΔBG from 30 to 120 min were lower in the model food DH than in the standard food, but it was not significant.

ΔC_{max} was 62.7 ± 4.3 mg/dL for the standard food, 53.0 ± 5.6 mg/dL for DL, and 64.0 ± 4.2 mg/dL for DH, and no difference was observed (Fig. 2-a). The iAUC was standard food: $4,690 \pm 433$ mg/dL·min, DL: $3,848 \pm 503$ mg/dL·min, and DH: $4,694 \pm 407$ mg/dL·min (Fig. 2-b).

Cabbage intake and PPHG suppression

The subjects were the same as the four study participants described above. The model food was cabbage (E). The change in ΔBG after starting test is shown in Fig. 1-e. There was no significant difference in ΔBG from 0 to 120 min between groups.

ΔC_{max} was 62.7 ± 4.3 mg/dL for the standard food, DL: 63.6 ± 5.3 mg/dL for DL, and 61.3 ± 4.0 mg/dL for DH (Fig. 2-a). The iAUC was standard food: $4,690 \pm 433$ mg/dL·min, DL: $4,755 \pm 499$ mg/dL·min, and DH: $4,478 \pm 353$ mg/dL·min (Fig. 2-b).

Vinegared rice intake and PPHG suppression

Out of 20 total participants, 11 subjects (2 males, 9 females, 23.1 ± 1.3 years, 159.7 ± 8.9 cm, 52.3 ± 7.0 kg,

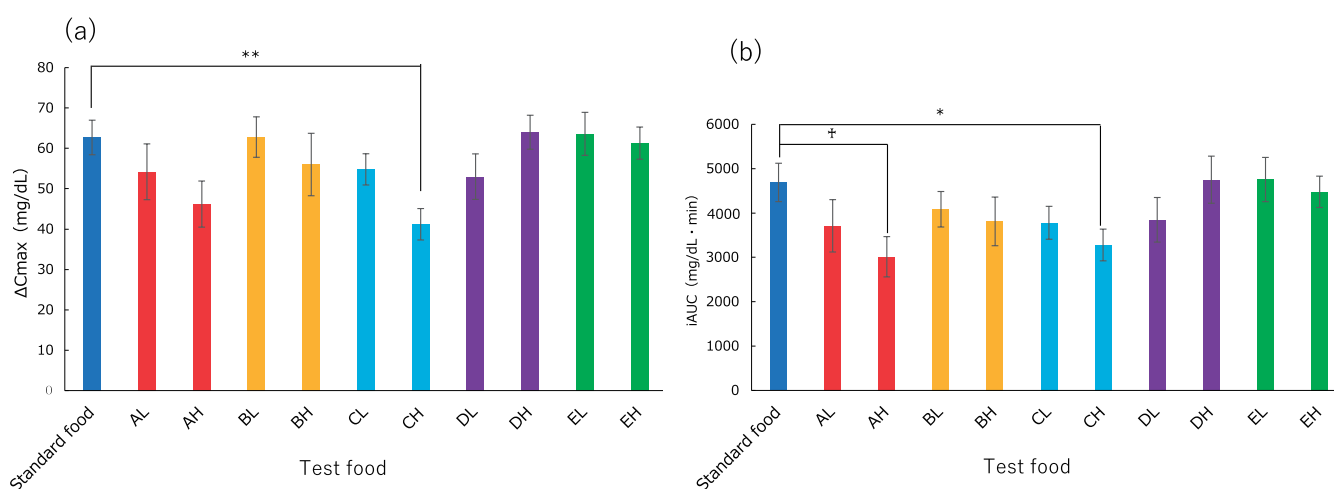


Fig. 2. The amount of ΔC_{max} (a) and iAUC (b) after intaking test food.

Results are expressed as mean \pm standard error, $n = 10$, † $p < 0.1$, * $p < 0.05$, ** $p < 0.01$ vs standard food, Bonferroni test. ΔC_{max} , maximum blood glucose level change; iAUC, incremental area under the curve of blood glucose level change. See Figure 1 for group names.

BMI $20.5 \pm 1.6 \text{ kg/m}^2$) participated in the study. The test food was vinegared rice (F). The change in ΔBG after the starting test is shown in **Fig. 3-a**. The values of ΔBG in F were significantly lower by 27% at 45 min and by 28% at 60 min ($p < 0.05$) compared to the standard food. ΔCmax was significantly lower by 19.3% ($p < 0.01$) and iAUC tended to be lower by 19.7% ($p < 0.1$) in F.

Fried chicken intake with or without lemon juice and PPHG suppression

Out of 20 total participants, 14 subjects (5 men, 9 women, 23.0 ± 1.3 years, 162.5 ± 9.9 cm, 56.3 ± 10.7 kg, BMI $21.2 \pm 2.1 \text{ kg/m}^2$) participated in the study. The test foods were fried chicken (G) and fried chicken + lemon juice (H). The change in ΔBG after starting test is shown in **Fig. 3-b**. The average values of ΔBG from 30 to 90 min were lower in the complex foods G and H than in the standard food. Specifically, the values of ΔBG in G and H at 30, 45, 60 min were significantly lower than that in the standard food.

The ΔCmax in G was significantly lower by 36.7% ($p < 0.01$), and the iAUC was significantly lower by 32.0% ($p < 0.05$) than that of the standard food (**Fig. 4-b**). The ΔCmax in H was significantly lower by 34.9% ($p < 0.01$), and the iAUC was significantly lower by 21.8% ($p < 0.05$) than that of the standard food.

Gyoza + Ponzu soy sauce intake and PPHG suppression

The subjects were the same as the study participants in the complex foods G and H described above. The complex food was gyoza + ponzu soy sauce (I). The change in ΔBG after starting test is shown in **Fig. 3-c**. The values of ΔBG in complex food I were significantly lower by 52.0% at 15 min ($p < 0.05$), by 55.6% at 30 min ($p < 0.01$), by 50.4% at 45 min ($p < 0.01$), and by 41.3% at 60 min ($p < 0.01$). The ΔCmax was significantly lower by 44.4%, and the iAUC was significantly lower by 39.7% than that of the standard food ($p < 0.01$, **Fig. 4-c**).

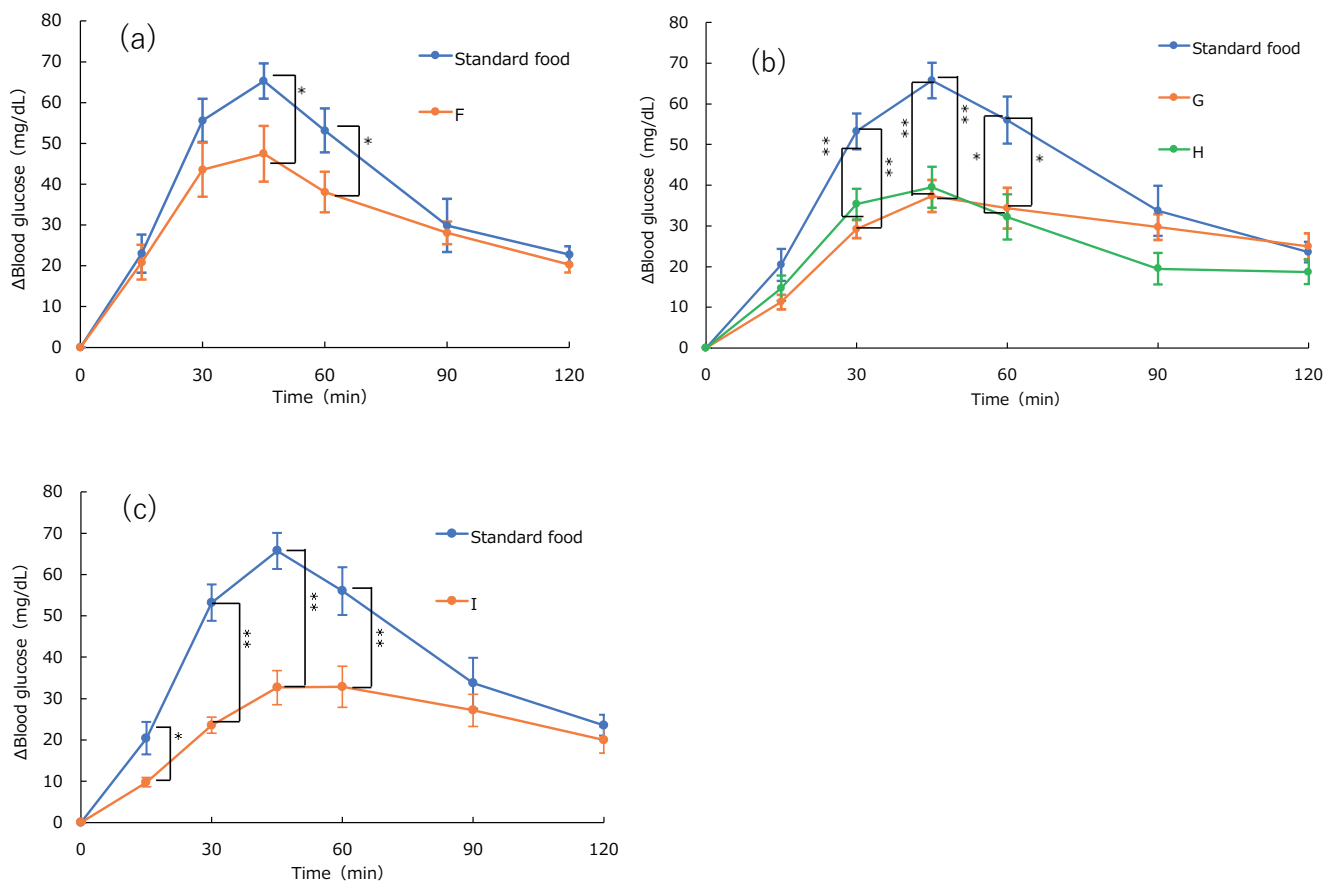


Fig. 3. Fluctuation of the blood glucose level at the time of intaking test food ahead of rice.

a) F. **b)** G and H. **c)** I. Results are expressed as mean \pm standard error, **a)** $n = 11$, paired t-test, **b)** $n = 14$, Bonferroni test, **c)** $n = 14$, paired t-test, * $p < 0.05$, ** $p < 0.01$ vs standard food. Standard food, cooked rice 200 g; F, vinegared rice; G, fried chicken 135 g before cooked rice 149 g; H, fried chicken 135 g + lemon juice 15 g before cooked rice 145 g; I, Japanese dumplings (gyoza) 138 g + ponzu soy sauce 15 g before cooked rice 129 g.

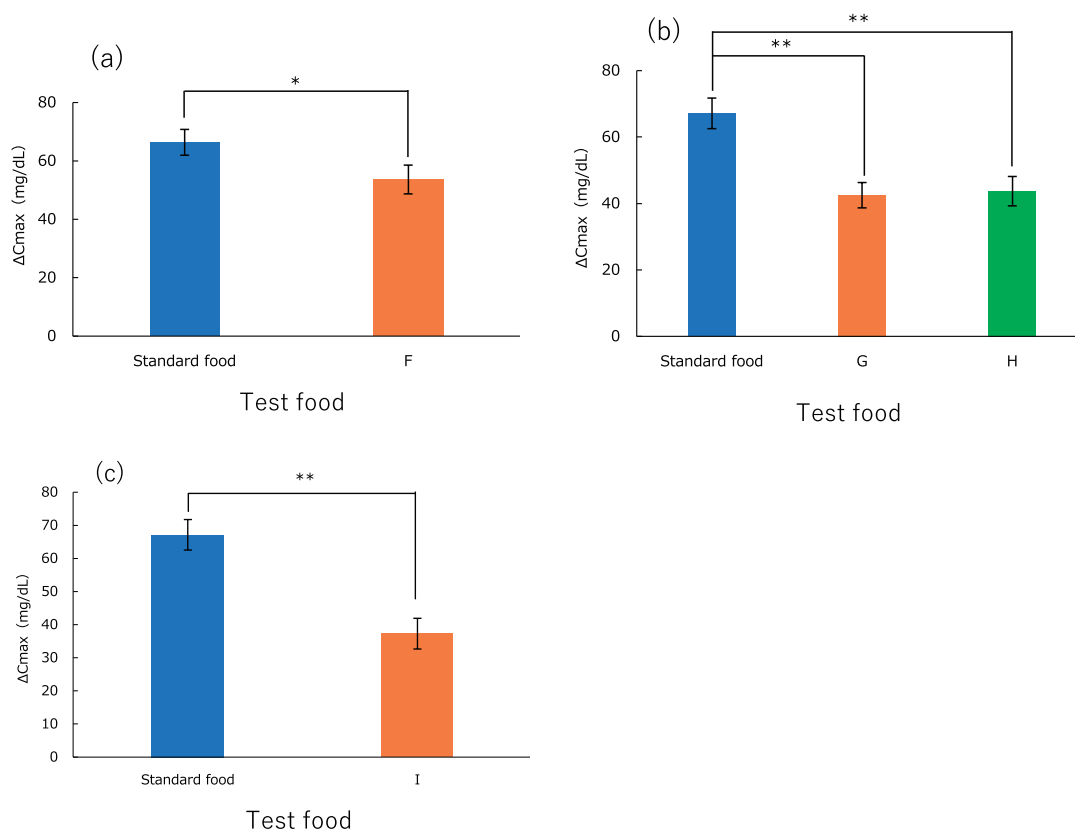


Fig. 4. The amount of ΔC_{max} after intake of test food.

a) F. **b)** G and H. **c)** I. Results are expressed as mean \pm standard error, **a)** $n = 11$, paired t-test, **b)** $n = 14$, Bonferroni test, **c)** $n = 14$, paired t-test, * $p < 0.05$, ** $p < 0.01$ vs standard food. ΔC_{max} , maximum blood glucose level change. See Figure 3 for group names.

Discussion

Protein intake and PPHG

Salad chicken (model food A) was used to examine the suppressive effect of protein intake on PPHG. A decrease in ΔBG , C_{max} , and $iAUC$ was observed depending on the intake of model food A (Fig. 1-a, Fig. 2). The intake of model food AH before the rice significantly suppressed PPHG, which was considered to be an effect of the components contained in model food A. The amount of protein in the test food calculated from model food A intake was 11.5 g in model food AL and 23 g in model food AH (Table 3).

The protein, rich in model food A, are reported to promote secretion of gastrointestinal hormones including GIP (glucose-dependent insulinotropic polypeptide) and GLP-1 (glucagon-like peptide-1)¹⁶. GLP-1 promotes insulin secretion from pancreatic β -cells as an incretin, cooperated with GIP, in a blood glucose-dependent manner, and GIP influences gastric peristalsis, reduces gastric emptying and delays glucose elevation. This study also indicated that protein intake significantly suppressed PPHG, and that its action became stronger as intake increased.

Lipid intake and PPHG

Olive oil (model food B) was used to examine the effect of lipid intake on PPHG. PPHG was not significantly inhibited by intake of the model food B as compared with the standard food (Fig. 1-b, Fig. 2).

Similar to protein and lipids, the main components of model food B promote GLP-1 secretion, thus suppressing gastric motility and PPHG¹⁷. However, another report¹⁸ showed that lipids do not promote insulin secretion despite the increase in GLP-1 concentration and they speculated that the suppressing effect on gastric motility may be too strong to elevate blood glucose and stimulate insulin secretion. In addition, it has been reported that protein intake suppresses PPHG by 2 to 3 times stronger compared to lipids, and there is no synergistic effect between the two¹⁹. In this study, we conducted a test by ingesting lipids alone, but it is possible that the effect of lipids on PPHG was small.

Acetic acid and PPHG

Grain vinegar (model food C) was used to verify the effect of acetic acid on PPHG. A decrease in ΔBG , C_{max} ,

and iAUC was observed depending on the intake amount of model food C (*Fig. 1-c, Fig. 2*). Therefore, it was considered that this suppressive action was the effect of the components contained in the model food C.

The amount of acetic acid in the test food calculated from the model food C was 0.63 g in model food CL and 1.26 g for the model food CH (*Table 3*). The amount of acetic acid useful for suppressing PPHG when ingesting 200 g of rice was estimated to be at least 0.63 g.

Acetic acid contained in vinegar has been confirmed in experiments in a rat model to prolong the gastric detention time and delay digestion and absorption²⁰. In addition, a new physiological action is indicated that acetic acid activates AMP kinase and it leads to a reduction in blood glucose due to inhibition of gluconeogenesis²¹. From the results of the model food C study, acetic acid significantly suppressed PPHG and its action became stronger according to the intake amount.

Soluble dietary fiber and PPHG

Indigestible dextrin (model food D) was used to examine the effect of water-soluble dietary fiber on PPHG.

Even when the model food D was ingested prior the rice intake, no significant suppressing effect on PPHG was observed as compared with the standard food (*Fig. 1-d, Fig. 2*).

Regarding the inhibitory effect on PPHG of water-soluble dietary fiber, which is the main component of model food D, there are detailed studies by Jenkins DJA *et al.*²², Doi K *et al.*²³ and Ebihara K *et al.*²⁴, in which it is considered that water is absorbed in the digestive tract and the gastric contents become viscous, resulting in a longer gastric detention time, thus suppressing PPHG. Despite the low viscosity of indigestible dextrin, it is suggested that it improves glucose tolerance in rats and healthy individuals, and that repeated intake can be expected to correct disorders of glucose metabolism seen in patients with obesity²⁵⁻³¹. Since the glucose tolerance improving effect of indigestible dextrin appears selectively in disaccharides, *i.e.*, sucrose, maltose, it is considered that these actions do not depend only on viscosity^{25, 26, 29}.

Disaccharides are decomposed into monosaccharides by α -glucosidase, a digestive enzyme, and absorbed by blood vessels in the small intestine. When indigestible dextrin binds to disaccharide-linked α -glucosidase as an inhibitor, the inversion of α -glucosidase, containing two hydrolyzed monosaccharides, to the bloodstream side is blocked; later, the bound monosaccharides are released from the intestinal wall to the digestive tract with a delayed decrease in affinity. This reaction is presumed to be a mechanism of PPHG suppression by indigestible dextrin⁵.

Another report showed that, in tests loaded with a starch diet, there was a significant difference in blood glucose level between the control and the test food in the high glucose group with C_{max} in control above the mean value, while there was no significant difference in the low glucose group with C_{max} below the average. In contrast in the sucrose tolerance test, a significant difference was found not only in the high glucose group but also in the low glucose group⁵.

These findings indicate that the affinity of indigestible dextrin is stronger for the sucrose-bound intermediate, as an inhibitor, than for the maltose-bound intermediate. Since

starch (maltose hydrolyzate) is the main component of cooked rice used in this study, the sugar absorption inhibitory effect of indigestible dextrin was perhaps unremarkable, compared with the effect of protein or acetic acid.

Dietary fiber and PPHG

Cabbage (model food E) was used to examine the effect of ingestion of both insoluble dietary fiber and water-soluble dietary fiber on PPHG. When the model food E was ingested before the rice intake, no significant suppressing effect on PPHG was observed as compared with the standard food (*Fig. 1-e, Fig. 2*).

According to the Food Composition Table³², cabbage contains 0.4 g of water-soluble dietary fiber and 1.4 g of insoluble dietary fiber per 100 g of edible portion. Generally, it is the water-soluble dietary fiber that has the suppressing effect on PPHG, while the insoluble dietary fiber is not involved. It is considered that the water-soluble dietary fiber absorbs water in the digestive tract, thus the gastric contents have viscosity and pass slower through the digestive tract, resulting in suppression of PPHG. There is a similar study that verified the suppressing effect on PPHG by the intake sequence of vegetable salad in diabetic patients⁹. In this study, however, the subjects ate rice 10 min after vegetable salad intake. In this experiment, there is a difference in conditions because no time was left between the shredded cabbage and rice.

Furthermore, in the above research, a vegetable salad was ingested with a dressing containing seasonings such as rice vinegar and olive oil. Therefore, it is considered that not the dietary fiber contained in the vegetable salad but the acetic acid and the lipids contained in the dressing were involved in the PPHG suppression. Also, it has been reported that healthy young subjects who ate 50 g of vegetable salad before rice had similar blood glucose levels to those without vegetable salad, and did not suppress PPHG³³. From these results, it was inferred that the PPHG inhibition by dietary fiber was dose-dependent, and it was necessary to ingest a larger amount of dietary fiber in order to obtain moderating effect on PPHG.

It is considered that the intake timing and intake amount of dietary fiber are important in order to exert the action of delaying the decomposition and absorption of carbohydrates.

Effect of complex foods on PPHG

In this study, we examined the PPHG suppressing effect of complex foods containing various nutritional components which include vinegared rice (complex food F), fried chicken (complex food G), fried chicken + lemon juice (complex food G), and gyoza + ponzu soy sauce (complex food I). The results showed that all the complex foods significantly suppressed PPHG (*Fig. 3-5*).

Focusing on iAUC, the complex food F decreased by 19.7% compared to intake of the standard food. Complex food G decreased by 32.0% and complex food H by 21.8%. In addition, complex food I decreased by 39.7%.

The sushi vinegar used to cook Complex food F contained 0.6 g of acetic acid (*Table 4*). This acetic acid content is compatible with the dose (0.63 g) useful for suppressing PPHG when ingesting 200 g of rice in the model food CL. The concurrent intake of rice and acetic acid as

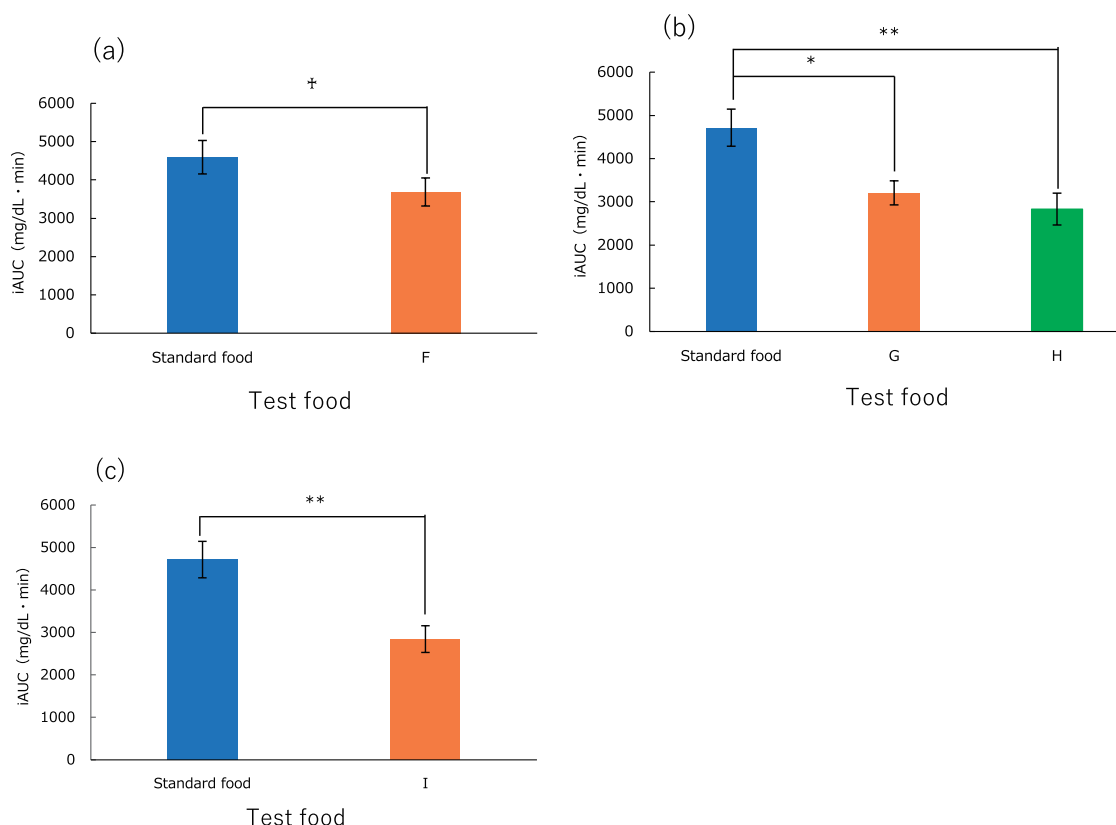


Fig. 5. iAUC after intake of test food.

a) F. b) G and H. c) I. Results are expressed as mean \pm standard error, **a)** $n = 11$, paired t-test, **b)** $n = 14$, Bonferroni test, **c)** $n = 14$, paired t-test, † $p < 0.1$, * $p < 0.05$, ** $p < 0.01$ vs standard food. iAUC, incremental area under the curve of blood glucose level change. See Figure 3 for group names.

vinegared rice significantly suppressed PPHG in this study as well as in the study of the concurrent intake of rice and beef bowl ingredients, rich in protein and lipids, which also showed PPHG suppression^{10, 11}. This indicates that even if carbohydrates and acetic acid are concurrently ingested as a complex food, the actions of delaying digestion/absorption and suppressing gluconeogenesis due to acetic acid exhibit similar effects as those of prior ingestion.

The complex food G contains 16.6 g more protein and 13.4 g more lipids than in standard food (**Table 5**). Even if the model food AL (protein: 11.5 g) and the model food BL (lipid: 14 g) were individually taken before rice, there was no significant difference in blood glucose compared to the standard food. By ingesting protein and lipids as a complex food such as fried chicken, it may be possible that the nutritional content of each synergistically contributed to PPHG suppression. On the other hand, no difference was observed in the effect between the complex foods G and H. Regarding the lemon juice, 30 g was effective, but not 15 g¹². Lemon is rich in organic acid, mainly citric acid, and polyphenols, which may play a role in PPHG suppression. In this study, however, the effect of lemon juice seemed unclear possibly due to the effect of fried chicken alone being strong enough to mask the synergistic action, if any was induced by 15 g lemon juice.

The complex food I contains 8.1 g of protein, 16.2 g

of lipids, 0.3 g of acetic acid, and 2 g of dietary fiber more than in the standard food (**Table 4**). It is possible that each nutrition component synergistically contributed to PPHG suppression by ingesting a complex food with well-balanced nutrition in a similar manner as prior intake of fried chicken.

In order to control blood sugar levels, it is necessary to combine diet therapy, exercise therapy, and drug therapy among which a diet therapy is the most basic for type 2 diabetes. It is necessary to respond flexibly while respecting individual eating habits and preferences for the purpose of a successful long-term diet. In the “Diabetes Practice Guideline 2019”, a guideline for diabetes treatment issued by the Japan Diabetes Society³⁾, there are descriptions about energy intake, distribution of three major nutrients, salt intake, and dietary fiber intake. However, almost no mention is made of the specific dietary content or the nutritional interaction. The purpose of the diabetic diet is to reduce blood glucose to a target level and its daily fluctuation, *i.e.*, glucose spike, to prevent complications, therefore, a diet that is balanced in desirable energy intake and nutrition is important. Complex foods such as fried chicken and gyoza have strong control actions on PPHG, as shown in this study. Additionally, they contain a variety of nutrition, and are easy to incorporate even when eating out. Therefore, we consider that these are side dishes that can be expected to suppress PPHG easily in the daily diet.

Research limitation

The subjects of this study were aged 23.3 ± 1.8 years and whose glucose tolerance was within the standard range. It has been reported that young people do not have large PPHG and are less likely to exhibit the suppressive effect of functional foods than middle-aged people³⁴. Therefore, the strength of food actions evaluated may change depending on the age of the subject. Also, since only the fasting insulin was measured before the test, it was not possible to verify the changes in the hormone secretion induced by intake of the test food. In the future, it is necessary to evaluate the chronological changes of hormone secretion concurrently with blood glucose.

Conclusion

The present study showed that healthy young men and women had significantly suppressed PPHG by consuming salad chicken and cereal vinegar as test foods before eating

rice (standard food), which had a more pronounced effect as the intake amounts of the test foods were increased. It was suggested that these phenomena involved the action of protein to promote incretin secretion and the action of acetic acid to delay digestion and absorption, and to suppress gluconeogenesis, and that the effect depends on the intake amount. On the other hands, if olive oil, indigestible dextrin, and cabbage were ingested prior to rice, no significant difference was observed in PPHG in the young subjects.

Conflict of Interest Statement

The authors claim no conflict of interest in this study.

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