

# Effect of vegetable juice consumption prior to eating rice on postprandial blood glucose and insulin levels

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**Abstract.** Vegetable juice has been demonstrated to attenuate the elevation of postprandial blood glucose when consumed prior to meals. The present study aimed to investigate the effect of pre-meal consumption of vegetable juice on blood glucose and insulin levels. A total of 10 healthy volunteers aged 20-29 years ingested 200 ml of either water, a sugar solution with the same sugar composition as the vegetable juice or vegetable juice 30 min prior to consuming the cooked rice, and their blood glucose and insulin levels were measured. At the time of rice consumption and 15 min thereafter, blood glucose and plasma insulin levels tended to be lower in the vegetable juice intake group compared with those in the sugar solution intake group. However, there were no significant differences in the kinetic parameters (incremental area under the glucose curve and maximum change in glucose concentration) between these two groups. These results suggest that the sugars contained in vegetable juice account for the suppression of postprandial hyperglycemia.

## Introduction

Complications of diabetes include neuropathy, retinopathy, nephropathy and, most notably, macroangiopathy (1,2), which is considered to be the major cause of death among patients with diabetes (3). Type 2 diabetes worsens as it progresses from the postprandial hyperglycemic stage to the fasting hyperglycemic stage. Postprandial hyperglycemia promotes

the onset and progression of diabetes (4) and, by inducing oxidative stress, likely causes macroangiopathy (5,6). Hence, suppression of postprandial hyperglycemia may help prevent diabetes, its progression and the occurrence of its complications.

Numerous diverse factors affect postprandial blood glucose levels, including insulin, the incretins [gastric inhibitory polypeptide (GIP) and glucagon-like peptide-1 (GLP-1)], the gastric emptying rate, the glycemic index and digestive enzymes. Inadequate secretion of insulin and the incretins elevates postprandial blood glucose levels (7), and insulin secretion is particularly low in Japanese patients with diabetes (8). Recent studies suggest that the order in which food is consumed limits postprandial glucose excursions. Protein and olive oil promote the secretion of incretins (9-12), while dietary fiber delays gastric emptying (13). Changing the order in which protein or olive oil and fiber are ingested or ingesting them prior to meals may curb postprandial glucose excursions.

Previous studies by our group reported that the consumption of a green salad, which has a low glycemic index, prior to meals limits postprandial glucose excursions (14,15). A further study by our group then focused on vegetable juice, which is easier to prepare and ingest than a green salad, demonstrating that the consumption of 200 ml vegetable juice 30 min prior to the consumption rice significantly attenuated increases in blood glucose levels (16). However, the mechanism has remained elusive.

The present study focused on the sugar content of vegetable juice and investigated the effect of the sugars on blood glucose and insulin levels when ingested prior to a standard meal.

## Materials and methods

**Test foods.** For the present study, as in the previous study by our group (16), a commercially available brand of vegetable juice (Yasai Ichinichi Kore Ippon; Kagome Co., Ltd.) was used. Additional test substances were glucose (Yoshida Pharmaceutical Company, Ltd.), D(-)-fructose and sucrose (Fujifilm Wako Pure Chemical Corp.), and pre-packaged cooked rice (Sato No Gohan; Sato Foods Co., Ltd.). The content of sugar (glucose, fructose, sucrose) of the vegetable juice is presented in Table I. The amount of carbohydrates per 200 ml was 14.6 g, whereas the total amount of sugar determined was 11.4 g, as described previously (16). A sugar solution with the same glucose, fructose and sucrose sugar compositions as the vegetable juice was prepared.

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**Abbreviations:** GIP, gastric inhibitory polypeptide; GLP-1, glucagon-like peptide-1; IAUC, incremental area under the curve;  $\Delta C_{max}$ , maximum change in concentration; SS group, sugar solution group; VJ group, vegetable juice group

**Key words:** preprandial, vegetable juice, postprandial blood glucose, insulin, fructose

**Subjects.** Subjects were recruited from Josai University in November 2014 and the study was conducted at the Laboratory of Drug Safety Management of Josai University. The study cohort comprised 10 healthy individuals (7 males and 3 females) aged 20–29 years. The sample size was determined based on a previous study by our group (16), and as in that study, none of the subjects exhibited any impaired glucose tolerance in tests performed during the previous year (Table II). Subjects participated in all three trials with a one-week washout period between each trial.

The present study was approved by the Life Sciences Research Ethics Committee of Josai University (Sakadoshi, Japan; date of approval, October 20, 2014; approval no. H26-5) and informed consent was obtained from all subjects.

**Experimental protocol.** The subjects consumed a 106.2 g cooked rice (sugar content, 35.4 g) 30 min after drinking 200 ml of either water (control group), the sugar solution (SS group) or vegetable juice (VJ group). The study was performed using the randomized crossover method. A random participant table was created using a computer program and the order of the tests was randomly determined. The nutritional content of the test meal and the beverages is provided in Table III.

The subjects were prohibited from eating and drinking anything but water from 9 pm on the night prior to the experiment until the start of the experiment. All tests were conducted at 9 am. It took <3 min for all subjects to consume the test beverage and <5 min to eat the cooked rice.

Blood was sampled nine times: Immediately prior to drinking the test beverage (–30 min), prior to eating the rice (0 min), and 15, 30, 45, 60, 90, 120 and 180 min after eating the rice. Blood was collected from a fingertip using a lancet (Medisafe FineTouch; Terumo Corp.). Blood glucose levels were measured enzymatically using a self-testing blood glucose monitor (Glutest Neo Alpha; Sanwa Kagaku Kenkyusho, Co., Ltd.). For the measurement of insulin levels, 100  $\mu$ l of blood was transferred from a capillary tube (Hematlon-L®; Minato Medical Co., Ltd.) to a microtube; the blood was centrifuged (2,610  $\times$  g, 4°C, 5 min), and 25  $\mu$ l of the supernatant (plasma) was immediately frozen at –80°C. Insulin levels were later measured using an insulin kit (YK060 Insulin ELISA kit®; Yanaihara Institute Inc.) according to the manufacturer's protocol.

**Data analysis.** Changes in blood glucose and insulin levels were calculated by subtracting the blood glucose and plasma insulin values measured prior to intake of the test beverages from the values measured at the various time-points after the subjects consumed the cooked rice. The maximum change in concentration, which may be at any time-point up to 180 min after consumption of the test beverages, was defined as  $\Delta C_{\max}$ . Incremental areas under the curves (IAUCs) were calculated using the trapezoid formula to assess the kinetics of the changes in blood glucose and insulin levels.

**Statistical analysis.** Statistical analysis was performed using Statsel3® software (OMS Publishing Inc.). Differences in  $\Delta C_{\max}$ , IAUCs, and changes in postprandial blood glucose and insulin levels were examined using one-way repeated analysis of variance. F-tests with significant results were followed by a Tukey-Kramer test.  $P < 0.05$  was considered to indicate a statistically significant difference.

Table I. Nutritional content of the vegetable juice used in the present study (per 200 ml).

Parameters	Value
Energy (kcal)	71
Protein (g)	2.1
Fat (g)	0
Carbohydrates (g)	14.6
Sugars, total (g)	11.4
Sucrose (g)	5.6
Glucose (g)	2.8
Fructose (g)	3.0
Dietary fiber (g)	1.9
Calcium (mg)	53
Potassium (mg)	830
Citric acid (mg)	1,190
Malic acid (mg)	514
Total polyphenols (mg GAE)	130

GAE, gallic acid equivalents.

Table II. Descriptive characteristics of the subjects.

Item	Total (n=10)	Males (n=7)	Females (n=3)
Age (years)	22.9 $\pm$ 1.6	23.0 $\pm$ 2.1	22.7 $\pm$ 0.9
Body height (cm)	169.5 $\pm$ 7.0	173.1 $\pm$ 4.6	161.0 $\pm$ 3.6
Body weight (kg)	62.1 $\pm$ 10.1	67.5 $\pm$ 6.6	49.3 $\pm$ 2.5
BMI (kg/m <sup>2</sup> )	21.5 $\pm$ 2.1	22.5 $\pm$ 1.7	19.0 $\pm$ 0.2
HbA1c (%)	5.2 $\pm$ 0.3	5.2 $\pm$ 0.3	5.2 $\pm$ 0.1

Values are expressed as the mean  $\pm$  standard deviation. BMI, body mass index; HbA1c, glycosylated hemoglobin.

## Results

**General characteristics.** There were no dropouts in any test group and no side effects were reported. There were no differences in blood glucose and plasma insulin levels between males and females.

**Blood glucose levels.** The changes in blood glucose levels in the different groups are displayed in Fig. 1 and the  $\Delta C_{\max}$  values and IAUCs are provided in Table IV. In the control group, the blood glucose levels rose rapidly after the subjects consumed the rice, peaking at 45 min. In the SS and VJ groups, the blood glucose levels rose after the subjects consumed their respective beverages (sugar solution or vegetable juice). These groups subsequently had significantly higher values at 0 min (prior to consuming rice) compared with those in the control group ( $P < 0.01$ ) and the blood glucose levels reached a peak at 30 min. Furthermore, the increase tended to be more gradual in the VJ group, and the VJ group had a significantly lower

Table III. Nutritional content in the three groups.

Group	Protein (g)	Fat (g)	Available CHO (g)	Fiber (g)
Control	2.17	0.3	Total, 35.4 Rice, 35.4	0.3
SS	2.17	0.3	Total, 46.8 Rice, 35.4 Sugar solution, 11.4 (glucose, 2.8; fructose, 3.0; sucrose, 5.6)	0.3
VJ	4.27	0.3	Total, 50 Rice, 35.4 Vegetable juice, 14.6	2.2

Groups: SS, sugar solution; VJ, vegetable juice; CHO, carbohydrates.

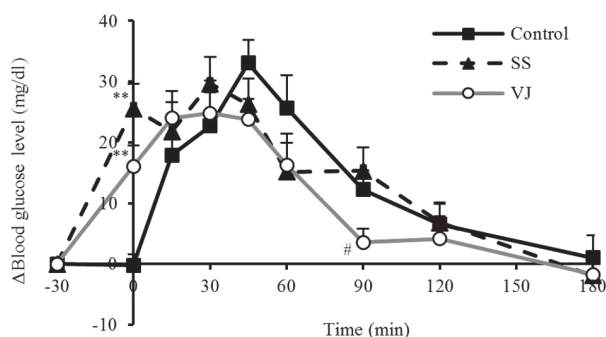


Figure 1. Mean changes in blood glucose levels over time. Values are expressed as the mean  $\pm$  standard error of the mean (n=10). \*\*P<0.01 vs. control; #P<0.05 vs. SS. Groups: SS, sugar solution; VJ, vegetable juice.

value at 90 min compared with that in the SS group (P<0.05). The  $\Delta C_{\max}$  of the VJ group was lower than that of the control and SS groups, but the difference was not significant. The IAUC in the SS group was obviously but insignificantly higher than that in the control group, while the IAUC was identical in the VJ and control groups.

**Plasma insulin levels.** The changes in plasma insulin levels in the different groups are presented in Fig. 2 and the  $\Delta C_{\max}$  values and IAUCs are provided in Table V. In the control group, the plasma insulin levels rose rapidly after the subjects consumed the rice, peaking at 60 min. In the SS group, the plasma insulin levels rose after the subjects consumed the sugar solution, peaking at 15 min. The levels at 0 and 15 min were significantly higher in the SS group compared with those in the control group (P<0.01), whereas the levels at 180 min were significantly lower in the VJ group compared with those in the SS group (P<0.01). In the VJ group, the plasma insulin levels rose after the subjects consumed the vegetable juice, peaking at 45 min. The plasma insulin levels then declined until 90 min and subsequently leveled off. Levels at 0 and 15 min were significantly higher in the VJ group than those in the control group (P<0.01 and P<0.05, respectively), whereas the levels at 180 min were significantly higher in the VJ group compared with those in the SS group (P<0.01). All three groups had essentially the same  $\Delta C_{\max}$ . The SS and VJ group

Table IV. Kinetic parameters of blood glucose.

Group	$\Delta C_{\max}$ (mg/dl)	IAUC (mg $\times$ min/dl)
Control	36.2 $\pm$ 3.2	2287.1 $\pm$ 348.4
SS	36.2 $\pm$ 3.9	3018.2 $\pm$ 466.5
VJ	33.0 $\pm$ 3.6	2331.3 $\pm$ 416.0

Values are expressed as the mean  $\pm$  standard error of the mean (n=10). Groups: SS, sugar solution; VJ, vegetable juice.  $\Delta C_{\max}$ , maximum postprandial blood glucose excursion; IAUC, incremental area under the curve.

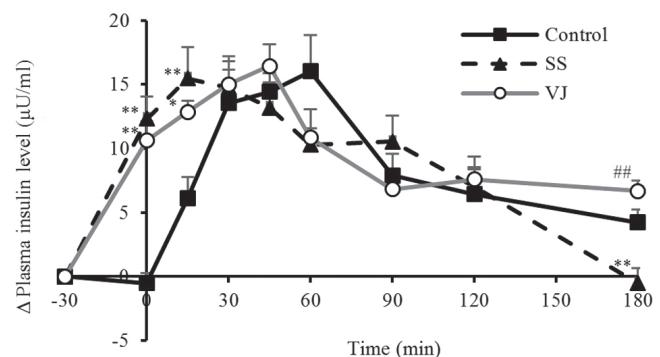


Figure 2. Mean changes in plasma insulin levels over time. Values are expressed as the means  $\pm$  standard error of the mean (n=10). \*P<0.05, \*\*P<0.01 vs. control; #P<0.05 vs. SS. Groups: SS, sugar solution; VJ, vegetable juice.

had higher IAUCs than the control group, but the differences were not significant.

## Discussion

In the present study, the effects of pre-prandial vegetable juice ingestion vs. sugars alone on postprandial blood glucose and insulin levels were assessed and the effect of the sugar contained in vegetable juice was clarified.

A previous study by our group suggests that vegetable juice intake and vegetable salad intake suppress postprandial

Table V. Kinetic parameters of plasma insulin.

Group	$\Delta C_{\max}$ (mg/dl)	IAUC (mg x min/dl)
Control	21.2 $\pm$ 3.0	1534.9 $\pm$ 174.4
SS	20.3 $\pm$ 1.8	1820.7 $\pm$ 130.8
VJ	19.0 $\pm$ 1.9	1895.4 $\pm$ 181.6

Values are expressed as the mean  $\pm$  standard error of the mean (n=10). Groups: SS, sugar solution; VJ, vegetable juice.  $\Delta C_{\max}$ , maximum postprandial plasma insulin excursion; IAUC, incremental area under the curve.

hyperglycemia via different mechanisms (16). The major potential mechanisms of suppression are as follows: i) Enhanced secretion of insulin, GIP and GLP-1 by sugars; ii) reduced digestion and absorption of the sugars by dietary fibers; and iii) reduced digestion and absorption of the sugars by organic acids and polyphenols. Among these mechanisms, the latter two are peculiar to vegetable juice. In the manufacturing process of the vegetable juice used in the present study, insoluble dietary fiber is removed as pomace; hence, most of the dietary fiber in the juice (1.9 g) is presumably water-soluble.

The indigestible dextrin component of water-soluble dietary fiber has been reported to suppress postprandial blood hyperglycemia in several ways (17-20). For instance, it inhibits glucose absorption after sucrose or maltose intake via the disaccharidase-associated transport system, and it mechanically stimulates the digestive tract to induce the secretion of GLP-1. Secreted GLP-1 lowers blood glucose levels by promoting insulin secretion from pancreatic  $\beta$ -cells and simultaneously inhibiting glucagon secretion (21). The microchorionic membrane disaccharide-degrading enzyme is a component of the disaccharidase-associated transport system. Organic acids and polyphenols suppress the activity of digestive enzymes, including  $\alpha$ -amylase and  $\alpha$ -glucosidase (22,23). Polyphenols also inhibit the activity of sodium-glucose transport protein 1 (24-26).

In the present study, blood glucose levels rose within 30 min after the consumption of the sugar solution or vegetable juice (0 min). However, the increase was less pronounced in the VJ group than in the SS group, presumably owing to the dietary fiber, organic acids and polyphenols in the vegetable juice. The rapid increase in blood glucose levels in the SS and VJ groups may be responsible for the significantly higher plasma insulin levels in these groups compared with those in the control group at 15 min; at this time-point, the plasma insulin levels tended to be higher in the SS group compared with those in the VJ group.

At 90 min, the blood glucose level was significantly lower in the VJ group compared with that in the SS group. Moreover, at 180 min, the plasma insulin level was significantly higher in the VJ group compared with that in the SS group. These results are different from those of a previous study by our group (16), where the group equivalent to the VJ group had blood glucose levels that declined from 60 to 120 min, and the 180-min insulin level was the same as the level after fasting. This result may also reflect differences in the amount or types

of intestinal microflora in the study populations. Dietary fiber promoting insulin secretion via a mechanism involving the intestinal microflora has become a popular concept in recent years. Previous studies have indicated that the intake of a small amount of dietary fiber increases the microflora-mediated production of short-chain fatty acids (27), which in turn promote insulin secretion via secretion of GLP-1 (28,29). Although the present study and the previous study by our group (16) assessed healthy adults with no glucose intolerance, neither of them examined the intestinal microflora. Hence, further studies assessing the involvement of the microflora are required in the future.

The vegetable juice and sugar solution used in the present study contained fructose and sucrose. In the study by Yau *et al* (30), healthy subjects consumed sugar solutions containing glucose (39.6 g), fructose (36.0 g), sucrose (36.0 g), or glucose (19.8 g) plus fructose (18.0 g), and the levels of blood glucose, insulin, GIP and GLP-1 were measured after ingestion. The levels of insulin, GIP and GLP-1 peaked at 20-30 min after the intake of each sugar solution. In the present study, the high plasma insulin level at 0 min in the VJ group may have suppressed the rise in the blood glucose level following rice consumption. Furthermore, Moore *et al* (31) reported that low-dose fructose improves the glycemic response to an oral glucose load in normal adults without significantly enhancing the insulin or triglyceride response. Regarding the potential mechanism, fructose may activate glucokinase and promote glucose uptake in the liver (32,33). The vegetable juice and sugar solution used in the present study each contained 3 g of fructose, and it is possible that the fructose may have inhibited, at least in part, the postprandial increase in blood glucose levels in the SS and VJ groups. It may therefore be suggested that fructose increased glucose uptake by the liver, consequently stimulating glucose metabolism.

Although the VJ and SS groups consumed sugar (14.6 and 11.4 g, respectively), whereas the control group did not,  $\Delta C_{\max}$  was low in all groups with no significant inter-group difference, and the IAUCs for blood glucose were also similar. These results are reminiscent of the blood sugar transition indicated in the previous study by our group (16) and highly support the contention that vegetable juice suppresses postprandial hyperglycemia. Given the similar effects in the VJ and SS groups, it is likely that the sugars contained in the vegetable juice were mainly responsible for its suppressive effects on blood glucose levels following rice consumption.

The present study was performed to determine the mechanism whereby vegetable juice attenuates the increase in postprandial glucose levels. Blood glucose and plasma insulin levels were compared after intake of vegetable juice or a sugar solution with the same sugar composition as the vegetable juice. Blood glucose kinetic parameters had similar values despite the fact that the VJ group consumed more sugar than the SS group. In the VJ group of the present study, the same postprandial blood sugar rise inhibitory effect as that in the previous study by our group (16) was observed. The VJ group exhibited similar changes in blood glucose levels to those in the SS group, and there were no significant differences in blood glucose kinetic parameters, although the VJ group had a somewhat low value. From this, it may be deduced



that the effect of the sugar contained in vegetable juice is largely responsible for the suppression of the postprandial increase in blood glucose in the VJ group. Furthermore, since the insulin level in the VJ group and the SS group also exhibited the same transitional trend, it may be indicated that the change in the insulin secretion pattern due to the intake of glucose contributes to the suppression of postprandial blood glucose elevation. However, in the present study, the blood glucose level in the VJ group was significantly lower when compared with the SS group at 90 min. This result differed to those obtained in a previous study, where no significant difference was observed (16). This may be due to the influence of the subjects' intestinal environment and insulin secretion ability. In addition, the present study was performed on healthy individuals, and thus, the effect on diabetic patients remains elusive. Based on the results of the study, vegetable juice may contribute to the prevention of diabetes.

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### Availability of data and materials

The datasets used and/or analyzed during the present study are available from the corresponding author on reasonable request.

### Authors' contributions

YI, IM and IK conceived the current study and provided technical support. NK, NI and IK were responsible for the acquisition, analysis and interpretation of the data and contributed to the drafting of the manuscript and its critical revision for important intellectual content. All authors read and approved the final manuscript.

### Ethics approval and consent to participate

The present study was approved by the Life Sciences Research Ethics Committee of Josai University (Sakadoshi, Japan; date of approval: October 20, 2014; approval no. H26-5). All procedures were performed in accordance with the ethical standards of the responsible committee on human experimentation (institutional and national) and/or with the Helsinki Declaration of 1964 and its later versions. Informed consent was obtained from all subjects prior to their inclusion in the study.

### Patient consent for publication

Not applicable.

### Competing interests

The authors declare that they have no competing interests.

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