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Original article

Effects of highly cross-linked distarch phosphate-containing food on glucose spikes

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Abstract

***Purpose*:** It is becoming increasingly clear that hyperglycaemia (blood glucose spike) causes an aldehyde spark, resulting in the causes of tissue disorders including vascular ebdothelial cell disorder, carbonylation of proteins, and production of advanced glycation end products (AGEs). Distarch phosphate with a very high level of crosslinking is a non-biodegradable starch. Here, the blood glucose spike mitigation action of distarch phosphate blended in bread was verified.

***Methods*:** Thirty-two healthy males and females under the age of 60 (13 students and 19 company employees) were selected as subjects for an experiment using rolls made of wheat blended with 50% wheat starch as the control food. The subjects underwent a blood glucose level test after ingestion of the the control bread and the test bread which were blended with distarch phosphate (NutraStar RA-900, Sanwa Starch Co.,Ltd.) by 50% (on the different day for each bread.) Blood glucose level was measured 15, 30, 45, 60, 90, 120 minutes after the start of ingestion of test foods by use of self blood glucose meters to analyze the incremental area under the curve (iAUC) and the maximum value of glucose concentration change (ΔCmax). At the same time, the safety level was analyzed.

***Results*:** In the total analysis, postprandial glucose levels were kept low during the time of ingestion of test food (breads including distarch phosphate), while iAUC (–11.4%) and ΔCmax were in a decreasing trend ($p < 0.1$). As a result of the subclass analysis which compares a group of company employees (36.9 ± 9.3 years) and a group of college students (22.6 ± 1.3 years), the former showed a remarkable decline of iAUC (–16.7%, $p < 0.05$), while in the latter, a more modest decline was observed (–3.8%. $p > 0.1$). During the period of observation, no harmful event occured.

***Conclusions*:** It was confirmed that the blood glucose level spike after ingestion was mitigated by ingesting breads including distarch phosphate (test food), and the effect was much more pronounced in the case of middle-aged groups compared to the young, while the safety of test foods was verified. Based on the fact that the blood glucose spike follwed by the aldehyde spark are mitigated by ingestion of test food, resulting in a decrease of glycative stress, there is a possibility that distarch phosphate contributes to human health.

***KEY WORDS*:** cross-linked starch phosphate, post-prandial hyperglycemia, wheat starch, insulin

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Introduction

A phenomenon of reducing sugar non-enzymatically combining with protein to produce advanced glycation end products (AGEs) through the process of glycated protein is called "glycation" [1, 2]. The stress reducing sugar or aldehyde given to the living organism is called "glycated stress" which is one of the risk factors that promotes aging in the body. To reduce glycative stress, it is recommended to suppress postprandial hyperglycemia, to suppress saccharification, and to promote disassembly/excretion of glycation endproducts.

Postprandial hyperglycemia differs depending on individuals and food content. When Cmax is 140 mg/dL, it is called "glucose spike" [3]. Though blood glucose has a cyclic structure in 99% of cases, some have a straight-chain structure with the aldehyde functional group being exposed. There is a phenomenon where at the peak of a blood glucose spike where aldehyde from straight-chain glucose structures react with blood monosaccharide or sugar chains on the surface of protein/tissues, concurrently causing multiple kinds of aldehyde to be produced. We named this phenomenon an "aldehyde spark" [4, 5]. The reaction of aldehyde and protein was so strong that it was considered that carbonylation protein or AGEs might be formed causing various tissue injuries including vascular endothelial dysfunction. Seen from the viewpoint of glycative stress, by preventing blood sugar spikes from occurring, it is possible to avoid aldehyde sparks, which is considered much more important than has been previously discussed.

To prevent glucose spikes, the following methods are considered: to select low glycemic index foods (low GI food), or low glycemic foods (low GL food); to ingest dietary fibers, yogurts, and vinegars before carbohydrates; to ingest foods including a-glucosidase inhibitory component; and to restrict carbohydrates [2].

In this test, we examined humans who ingested test foods blended with the processed starch to analyze their blood glucose fluctuation. The test was conducted using the rolls made of wheat blended with 50% wheat starch. Healthy males and females were given a single-ingestion of sample breads which were blended with distarch phosphate. Thus, the blood glucose increase inhibitory effect was verified in the open test.

Method

Subjects

The subjects of this study were healthy males and females 20 years or older and 60 years or younger, and they had submitted letters of consent for participation in this study. The test food was a table roll, bread blended with highly cross-linked distarch phosphate (NutraStar RA-900; Sanwa Starch Co., Ltd., Kashihara, Nara, Japan). The inhibition effect of the test food on postprandial hyperglycemia was verified by a single ingestion. The subjects are the same as in the previous report [4].

A briefing for the explanation of the test was conducted after obtaining the approval of the ethical committee concerning "studies involving human subjects" of Doshisha University, and 32 subjects whose consent had been obtained and who did not conflict with the following exclusion criteria were incorporated in this study:

1. Individuals who are using medical products for any disease
2. Individuals who are under treatment or have a history of impaired glucose tolerance, mental disabilities, sleep disorder, hypertension, diabetes, dyslipidemia or other serious dysfunctions
3. Individuals who used a drug for treatment in the past 1 month (one-shot medicine for headache, menstrual pain and colds are excluded)
4. Individuals who are under treatment of or have a history of serious liver dysfunction, kidney damage, heart disease or blood disease
5. Individuals who are under treatment of or have a history of digestive organ disease (a history of appendicitis is excluded)
6. Individuals whose body mass index (BMI) is over 30 kg/m^2
7. Individuals who donated blood over 200 mL in the past 1 month or over 400 mL in the past 3 months
8. Individuals with serious anemia
9. Individuals who are sensitive to test products or individuals who have the fear of severe allergy in other foods and medical supplies
10. Individuals who are pregnant or breastfeeding or individuals who could possibly become pregnant
11. Individuals who had a habit to continuously ingest health-promoting foods advocating glycometabolism or healthy foods in the past 3 month or will ingest those foods during the test period (ingest for the purpose of health maintenance is acceptable)
12. Individuals who were judged inappropriate as the subjects for this study by a responsible doctor

Test Design

This test was an open-label test with a control.

A table roll made with highly cross-linked distarch phosphate blended with 50% replacement of wheat flour (bread containing highly cross-linked distarch phosphate) was used as the test food (***Table 1***). A table roll made with wheat starch blended 50% replacement (bread containing wheat starch) was used as the control food.

The subjects were instructed to avoid excessive exercise, eat a specified dinner menu and sleep longer than 6 hours on the day before the test. The menus of the dinner on the day before the test are shown in ***Table 2***. Alcohol intake was prohibited from the previous day of the test to the completion of the day of the test.

The subjects selected their dinner from ① and ② of their free will and decided to take the same menu both times.

On the day of the test, they were waiting quietly in sitting position, while DVDs of animation and travel shows were televised, and they were prohibited from telephoning, sleeping, excessive brain activities and physical activities with a possibility of sweating until the end of the test. After the ingestion of the test food, they were only allowed to take water.

On the day of the first test, after waiting quietly, the subjects underwent physical measurements and, by collecting blood samples, hematologic test and blood biochemical test. At the same time, they conducted self-monitoring of blood glucose (GLU). (the first time). After that, they ingested three

Table 1. Composition of table roll bread (produced by the straight method).

Table roll bread	Control food	Test food
Bread Flour	48.5	48.5
Wheat starch	50	—
Distarch phosphate (NutraStar RA-900)	—	50
Vital gluten	1.5	1.5
Yeast	2	2
Yeast food	0.1	0.1
White soft sugar	12	12
Salt	1.8	1.8
Non-fat dry milk	3	3
Margarine	12	12
Water	56.6	61.6

Table 2. The dinner menu selection on the day before the test.

	Selection ①	Selection ②
Steamed rice	200 g	300 g
"Gyudon" pack	1 pack	1 pack
"Gyudon" mini pack	—	1 pack
Freeze-dried miso soup	1 pack	1 pack
Energy	668 Kcal	1,014 Kcal
PFC ratio	12 : 39 : 48	12 : 41 : 46

Steamed rice, Papatto Rice Koshihikari, Hagoromo Foods Corporation, Shizuoka, Japan; "Gyudon" pack, "Gyudon" mini pack: Yoshinoya Co.,Ltd., Tokyo, Japan. "Gyudon" is a rice bowl topped with beef; Freeze-dried miso soup, Yoshinoya Co.,Ltd.; PFC, protein fat carbohydrate.

pieces of control food (the dough was divided and baked so that the carbohydrate amount was 75 g). A piece of control food was cut into six pieces for the subjects to chew a piece 10 times and swallow. Food ingest time was from 9 to 10 minutes. The subjects conducted self-monitoring of GLU 15 minutes (the 2nd time), 30 minutes (the 3rd time), 45 minutes (the 4th time), 60 minutes (the 5th time), 90 minutes (the 6th time) and 120 minutes (the 7th time) after starting ingestion. On the day of the second test, the subjects conducted self-monitoring of GLU after waiting quietly (the 1st time). Then they ingested three pieces of test food together with 200 mL of water. The ingestion method, the way of waiting quietly during self-monitoring of GLU and the test implementation time were the same as those of the first test.

The test period was from August 2017 to December 2017.

Self-monitoring of GLU

The subjects conducted self-monitoring of GLU using a self-monitoring blood glucose meter (Glucocard G Black: GT-1830, Arkray Co., Kyoto, Japan). The measurement was conducted twice and the average value was used. When the difference between the two values was more than 10%, a 3rd value was measured and the average value of the two values with the least difference was used.

Physical measurements

Body height, body weight, body fat percentage, BMI, systolic and diastolic blood pressures and pulse rate were measured.

Blood tests

Peripheral blood tests and biochemical tests were conducted using blood samples. The test items for this analysis were white blood count (WBC), red blood count (RBC), hemoglobin content (Hb), hematocrit value (Ht), mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC), platelet count (PLT), blood urea nitrogen (BUN),

creatinine (CRE), uric acid (UA), aspartate aminotransferase (AST), alanine aminotransferase (ALT), gamma-glutamyl transpeptidase (γ-GTP), total cholesterol (TC), triglyceride (TG), low-density-lipoprotein cholesterol (LDL-C), high-density lipoprotein cholesterol (HDL-C), HbA1c, fasting plasma glucose (HPG), insulin (imunoreactive insulin: IRI) and glucagon. The measurements using blood samples were conducted at LSI Medience Corporation (Tokyo, Japan).

Statistical analysis

Analytical items were GLU measured value, incremental area under the curve (iAUC) of GLU level changes, amount of change from initial value (Δblood glucose value), Cmax and change value of Cmax (ΔCmax), and fundamental statistics (average value, standard deviation and 95% confidence range) were calculated.

For the test of significant difference, the comparisons between groups at the time of ingesting the control food and the test food were evaluated using unpaired t test or Bonferroni's multiple test (paired). For the test of significant difference, an appropriate statistical analytical software (Excel Statistics, Social Survey Research Information Co., Ltd., Tokyo, Japan) was used. The levels of significance were assumed to tend to be less than 5% and 10%, respectively. This system was prepared so that various statistical analyses can be done from the viewpoint of exploratory research such as a stratified analysis in responding to subject characteristics and research of correlation from the viewpoint of affectivity.

Ethical review

This study was conducted in compliance with the declaration of Helsinki (revised at 2013 WMA General Assembly, Fortaleza, Brazil) and the ethical guideline regarding the "medical and health research involving human subjects" (announced by the Ministry of Education, Culture, Sports, Science and Technology and the Ministry of Health, Labour and Welfare). For this test, the "Committee for Medical Test involving human subjects" was held at the Ethical Review Committee concerning the "study involving human subjects" of Doshisha University, where the morality and validity of this test was discussed, and approval was granted (Application No. 17054). This test was pre-registered as a clinical trial (UMIN No.000029026).

Results

General background

Thirty-two subjects examined in this test (overall subject) were all healthy adults. None of them were classified as D (requiring treatment) according to the Japan Society of Ningen Dock (***Table 3***). The subjects consisted of 19 males and 13 females who were 30.9 ± 10.7 years.

The 33 subjects were selected from those who met the selection standard and did not meet to the exclusion criteria with an approval of the doctors responsible for the test. A change of the number of test subjects is shown below (***Fig. 1***).

The test subjects consisted of a group of 13 college students (Doshisha University students and undergraduate students; six males and seven females), and a group of 19 company employees (Sanwa Starch' employees; 13 males and six females.) Company employees showed higher ages, weights, BMI, TC, LDL-C, HORMA-IR, compared to the college students ($p < 0.01$). On the other hand, they showed a significantly lower pulse rate and IRI value ($p < 0.01$). The gap of average ages between the two groups was 14.3 years, showing that there was an influence of age on the reslt. In the test for carbohydrate metabolism of a group of company employees, IRI value was lower by 2.7 μ unit/mL (1.8 times lower), and HORMA-IR value was higher by 0.5 point (1.7 times higher) compared to the group of college students. Though the difference of carbohydrate metabolism was within the referene range, it was presumed that the group of company employees has an inclination to show an increasingly high insulin resistance due to aging (***Table 4***).

Table 3. Subject background profile.

		Average		SD
Age		30.9	±	10.7
Height	cm	166.7	±	7.7
Weight	kg	60.0	±	10.8
BMI	–	21.5	±	2.7
Blood pressure (systolic)	mmHg	119.7	±	13.1
(diastolic)	mmHg	70.7	±	10.4
Pulse	/min	66.8	±	14.1

n = 32. BMI, body mass index; SD, standard deviation.

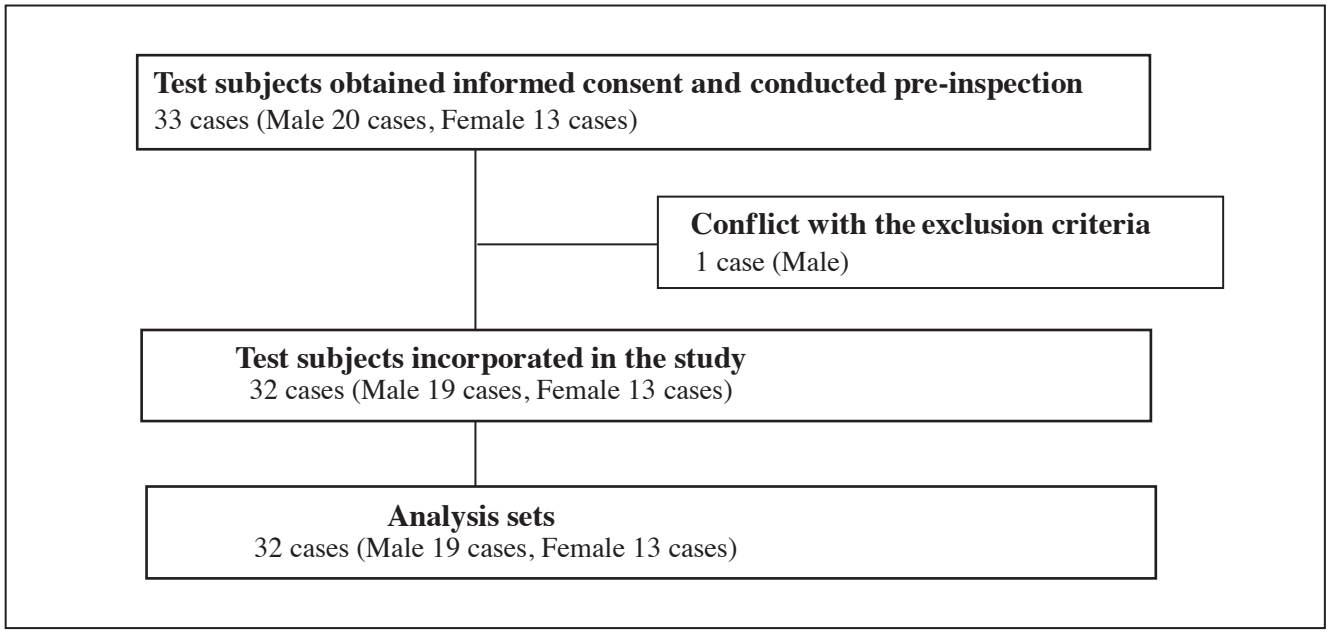

Fig. 1. **The number of analysis sets.**

Table 4. Comparison between College students and Company workers.

		College student (n=13)			Company worker (n=19)			p value
Gender	Male	6			13			0.419
	Female	7			6			
Age		22.6	±	1.3	36.9	±	9.3	<0.001
Height	cm	164.1	±	7.1	168.1	±	8.0	0.118
Weight	kg	52.3	±	6.4	64.1	±	9.1	<0.001
BMI	-	19.4	±	1.6	22.6	±	2.1	<0.001
Blood pressure (systolic)	mmHg	124.2	±	13.1	119.1	±	16.4	0.095
(diastolic)	mmHg	70.4	±	9.5	73.9	±	13.8	0.899
Pulse	/min	74.7	±	15.4	61.9	±	11.5	0.005
TC	mg/dL	165.9	±	22.5	196.9	±	39.3	0.004
LDL-C	mg/dL	88.5	±	18.5	107.4	±	34.5	0.004
HDL-C	mg/dL	63.5	±	10.3	75.6	±	17.5	0.500
TG	mg/dL	76.5	±	32.9	72.4	±	39.3	0.782
FPG	mg/dL	82.9	±	6.6	89.9	±	9.4	0.454
HbA1c	%	5.1	±	0.2	5.2	±	0.2	0.138
Insulin (IRI)	μU/mL	5.9	±	2.3	3.2	±	1.1	0.002
Glucagon	pg/mL	107.5	±	10.4	112.8	±	14.2	0.793
HORMA-IR		0.7	±	0.2	1.2	±	0.6	0.004

Data are expressed as mean ± SD, paired t test. BMI, body mass index; TC, total cholesterol; LDL-C, low-density-lipoprotein-cholesterol; HDL-C, high-density-lipoprotein-cholesterol; TG, triglyceride; FPG, fasting plasma glucose; IRI, immunoreactive insulin; HORMA-IR, homeostasis model assessment of insulin resistance; SD, standard deviation.

Postprandial blood glucose change (a total analysis)

The total subjects' postprandial blood glucose change curve for each food shows higher glucose levels in the order of control food and test food. The blood glucose level of the subjects declined significantly 45 minutes after they began ingesting the test food ($p < 0.05$, ***Table 5***, ***Fig. 2***). Regarding iAUC, it showed a decreasing trend in the case of test food ingestion compared to control food ($p < 0.1$, ***Table 6***, ***Fig. 3***). Regarding ΔCmax, it showed a decreasing trend in the case of test food ingestion compared to control food ($p < 0.1$, ***Table 7***, ***Fig. 4***). Based on the above results, it was confirmed that distarch phosphate blended in the test food was effective at mitigating postprandial blood glucose rise.

Postprandial blood sugar change (subclass analysis)

All of the subjects were divided into two groups of 13 college stuldents and 19 company employees for the analysis. In the case of the college students group, postprandial blood sugar level change curve showed higher levels of blood glucose level in the order of control food and test food in the same way as the analysis conducted for the total subjects (***Table 8***, ***Fig. 5***).

In the case of the company employees group, in the same way as the analysis conducted for the total subjects, the postprandial blood sugar level change curve showed higher levels of blood glucose level in the order of control food and test food (***Table 9***, ***Fig. 6***). Postprandial blood sugar level declined significantly 60 minutes after ingestion ($p < 0.05$), while it showed a declining trend at 45 minutes and 120 minutes after the ingestion ($p < 0.1$). Regarding iAUC, it declined more in the case of test food (4,060.0 ± 1,405.7) compared to the case of control food (4,583.9 ± 1,422.1, −11.4%, $p < 0.1$, ***Table 10***, ***Fig. 7***). According to the subclass analysis, in the case of company employees, iAUC was significantly reduced (4,564.9 ± 1,472.0 → 3,801.1 ± 1,453.0, −16.7%, $p < 0.05$), and it did not show a significant change in the case of college students group (4,611.6 ± 1,404.5 → 4,438.3 ± 1,404.5, −3.8%. $p > 0.1$). It was verified that postprandial hyperglycemia suppression action due to distarch phosphate (developed product) blended in the test food was more effective in the company employees group compared to the college students group.

Table 5. Changes in blood glucose level after meals: Total analysis.

	0 min	15 min	30 min	45 min	60 min	90 min	120 min
Control food	0.0 ± 0.0 –	16.3 ± 16.6 5.7	58.8 ± 17.1 5.9	61.9 ± 20.5 7.1	48.1 ± 18.0 6.3	33.5 ± 18.3 6.4	29.5 ± 17.7 6.1
Test food	0.0 ± 0.0 –	19.4 ± 13.7 4.7	55.6 ± 21.4 7.4	53.5 ± 21.2 7.4	41.2 ± 19.6 6.8	28.6 ± 16.1 5.6	23.1 ± 14.9 5.2

Data [unit: mg/dL] are expressed as mean ± SD, 95%CI, n = 32. SD, standard deviation; CI, confidence interval. The blood glucose values at 0 minute are regarded as 0.0 mg/dL.

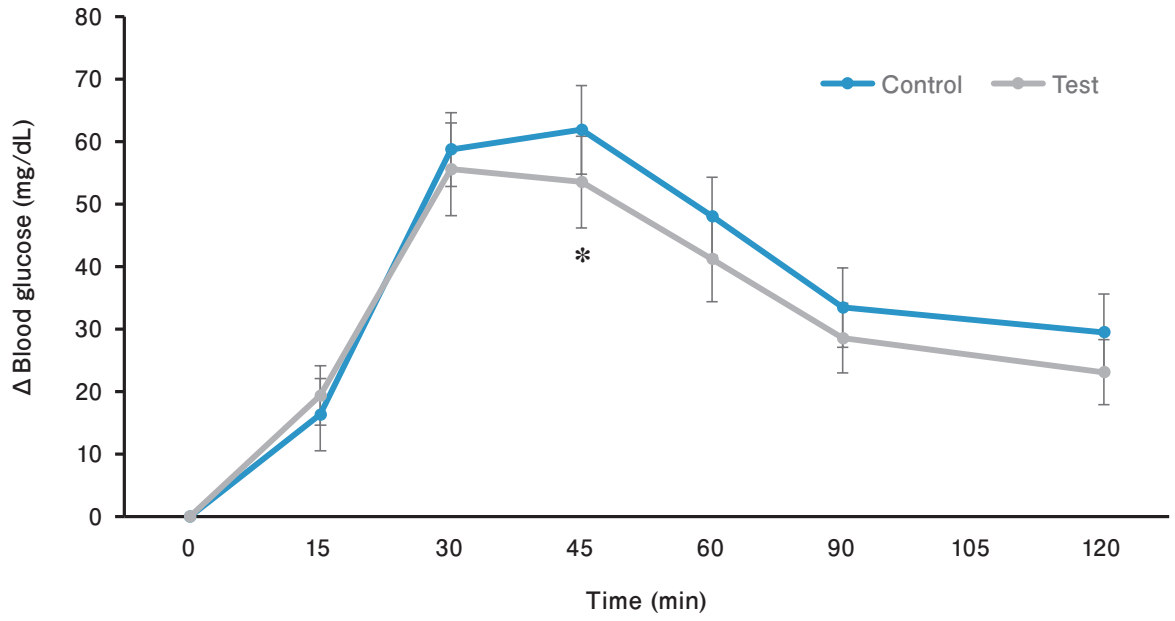

Fig. 2. **Changes in Δblood glucose level after meals: Total analysis.**

Data are expressed as mean ± 95%CI, n = 32, *p < 0.05 vs control by Bonferroni multiple comparison analysis with related data. ΔBlood glucose, changes in blood glucose; CI, confidence interval.

Table 6. iAUC, Cmax and ΔCmax: Total analysis.

	iAUC	Cmax (mg/dL)	ΔCmax (mg/dL)
Control food	4583.9 ± 1422.1 492.7	162.5 ± 22.2 7.7	68.5 ± 17.6 6.1
Test food	4060.0 ± 1405.7 487.1	158.0 ± 20.9 7.2	62.2 ± 20.7 7.2

Data are expresees as mean ± SD, 95%CI, n = 32. In ΔMax blood glucose level, the blood glucose values at 0 minute are regarded as 0.0. iAUC, incremental area under the curve; Cmax, maximum value of glucose concentration; ΔCmax, maximum value of glucose concentration change; SD, standard deviation; CI, confidence interval.

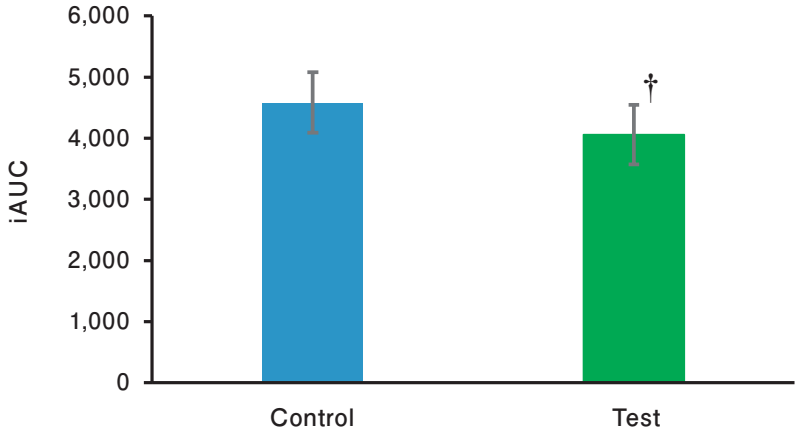

Fig. 3. **Comparison of iAUC between test group and control: Total analysis.**

Data are expressed as mean ± 95%CI, n = 32, † p < 0.1 vs control by Bonferroni multiple comparison analysis with related data. iAUC, incremental area under the curve; CI, confidence interval.

Table 7. Multiple comparison analysis by Bonferroni: Total analysis.

Level 2	Level 2	Changes in blood glucose level (min)							iAUC	Cmax	ΔCmax
		0	15	30	45	60	90	120			
Control	Test	-	0.926	1.000	0.041	0.107	0.292	0.148	0.085	0.386	0.083

Data shows p values by Bonferroni analysis, n = 32. iAUC, incremental area under the curve; Cmax, maximum value of glucose concentration; ΔCmax, maximum value of glucose concentration change.

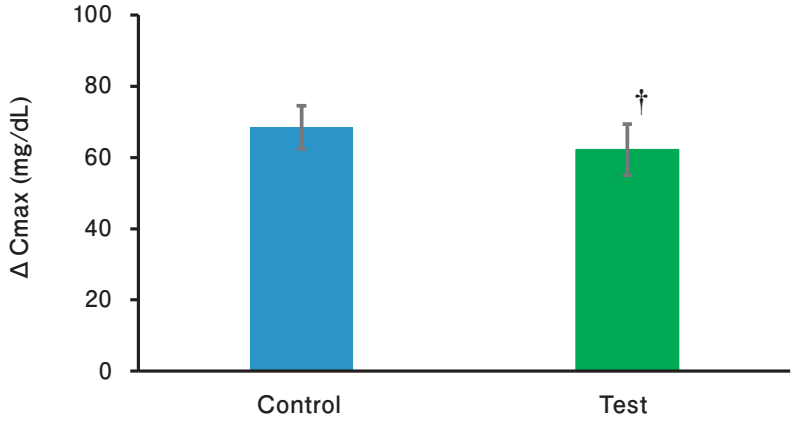

Fig. 4. **Comparison of ΔCmax between test group and control: Total analysis.**

Data are expressed as mean ± 95%CI, n = 32, † p < 0.1 vs control by Bonferroni multiple comparison analysis with related data. ΔCmax, maximum value of glucose concentration change; CI, confidence interval.

Table 8. Changes in blood glucose level after meals in College students.

	0 min	15 min	30 min	45 min	60 min	90 min	120 min
Control food	0.0 ± 0.0 –	17.3 ± 15.7 5.4	62.0 ± 17.0 5.9	64.2 ± 19.1 6.6	46.7 ± 16.7 5.8	31.5 ± 13.3 4.6	30.8 ± 20.2 7.0
Test food	0.0 ± 0.0 –	26.1 ± 12.0 4.1	58.5 ± 14.0 4.8	56.2 ± 22.3 7.7	44.5 ± 16.4 5.7	30.8 ± 16.6 5.7	26.8 ± 11.4 4.0

Data [unit: mg/dL] are expressed as mean ± SD, 95%CI, n = 13. SD, standard deviation; CI, confidence interval. The blood glucose values at 0 minute are regarded as 0.0.

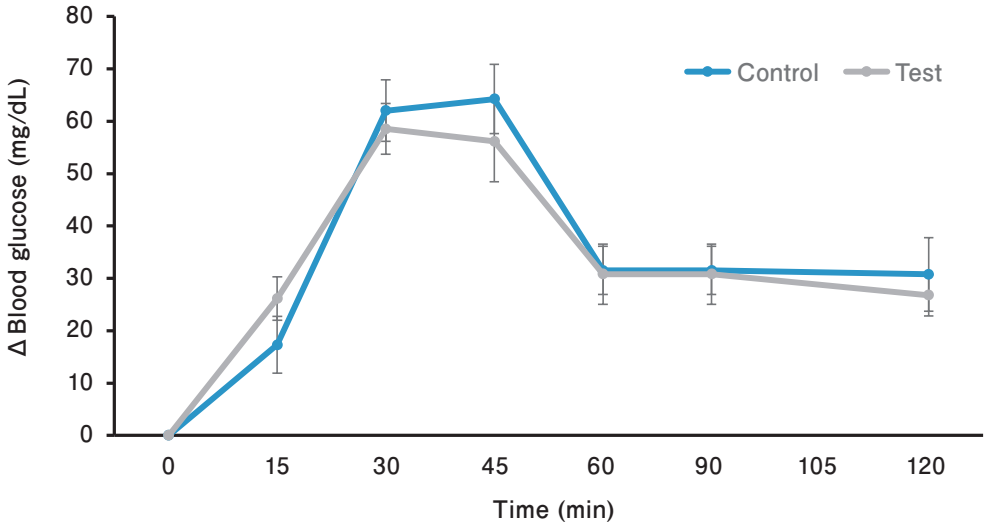

Fig. 5. **Changes in Δblood glucose level after meals in College students.**

Data are expressed as mean ± 95%CI, n = 13. ΔBlood glucose, changes in blood glucose; CI, confidence interval.

Table 9. Changes in blood glucose level after meals in Company workers.

	0 min	15 min	30 min	45 min	60 min	90 min	120 min
Control food	0.0 ± 0.0 –	15.7 ± 17.5 6.1	56.5 ± 17.2 6.0	60.3 ± 21.8 7.6	49.0 ± 19.3 6.7	34.9 ± 21.4 7.4	28.6 ± 16.3 5.7
Test food	0.0 ± 0.0 –	14.8 ± 13.2 4.6	53.6 ± 25.5 8.8	51.8 ± 20.9 7.2	39.0 ± 21.7 7.5	27.0 ± 16.0 5.5	20.7 ± 16.8 5.8

Data [unit: mg/dL] are expressed as mean ± SD, 95%CI, n = 19. SD, standard deviation; CI, confidence interval. The blood glucose values at 0 minute are regarded as 0.0.

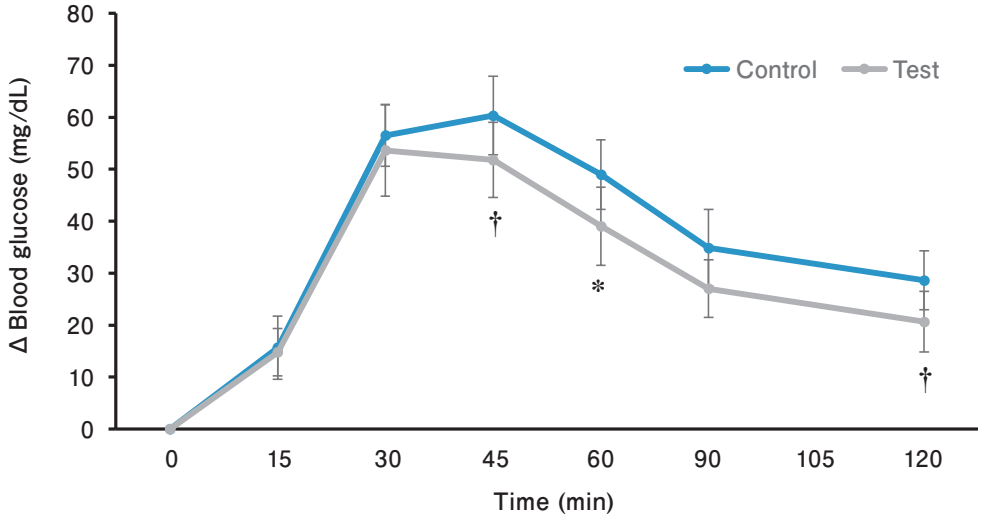

Fig. 6. **Changes in Δblood glucose level after meals in Company workers.**

Data are expressed as mean ± 95%CI, n = 19, * $p < 0.05$, † $p < 0.1$ vs control by Bonferroni multiple comparison analysis with related data. ΔBlood glucose, changes in blood glucose; CI, confidence interval.

Table 10. Multiple comparison analysis by Bonferroni: Total analysis.

Level 2	Level 2	Changes in blood glucose level (min)							iAUC	Cmax	ΔCmax
		0	15	30	45	60	90	120			
Control	Test	-	1.000	1.000	0.098	0.035	0.074	0.097	0.033	0.364	0.171

Data show p values by Bonferroni analysis, n = 19. iAUC, incremental area under the curve; Cmax, maximum value of glucose concentration; ΔCmax, maximum value of glucose concentration change.

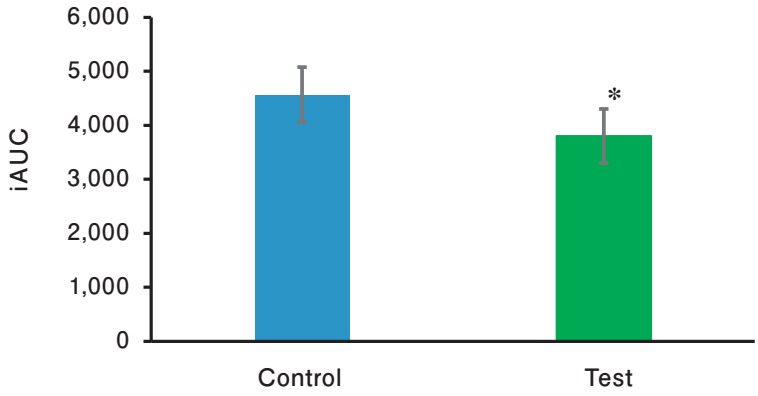

Fig. 7. **Comparison of iAUC between test group and control in Company workers.**

Data are expressed as mean ± 95%CI, n = 19, * p < 0.05 vs control by Bonferroni multiple comparison analysis with related data. iAUC, incremental area under the curve; CI, confidence interval.

Discussion

Crosslinked starch

Crosslinked starch is literally a processed starch whose structure is made stronger by cross-linking the chain of starch. As typical examples of cross-linked starch, there are distarch phosphate, which is cross-linked starch with phosphoric acid, and acetylated adipic acid cross-linked starch produced by use of adipic acid (dicarboxylic acid). Both of them are domestically utilized. Japanese Standard of Food Additives stipulates that phosphorus content for distarch phosphate should be below 0.5%, and adipic acid content for adipic acid cross-linked starch should be below 0.135%. However, in the case of distarch phosphate, it is possible to change the characteristics of starch with quite small amounts of phosphorus content. Therefore, in many cases, distarch phosphate whose phosphorus content is below 0.1% is chosen. The distarch phosphate used in this test contained phosphoric acid by as much as 0.5% so that the cross-linked structure of the starch would be strong enough to become non-biodegradable to amylase. Highly cross-linked distarch phosphate is reported to have an effect to mitigate blood glucose level rise [6, 7].

Summary of results

In this test, it was shown that the ingestion of food blended with distarch phosphate caused a significant decline of blood sugar level 45 minutes after ingestion, as well as a declining trend of iAUC and ΔCmax. The difference between test food and control food is the same as that between conventional starch and distarch phosphate. It is presumed that because distarch phosphate blended with the test food (developed product) was non-biodegradable, a decomposition of starch and a formation of glucose were delayed, which resulted in the mitigation of postprandial blood glucose rise.

As a result of the subclass analysis which compared the young and the middle-aged, the rate of decline of iAUC due to the ingestion of test food was higher in the case of the middle-aged (company employees group: −16.7%) compared to the case of the young (college students group: −3.8%). A blood glucose spike suppression effect obtained by distarch phosphate (developed product) was observed to be more significant in the case of the middle aged.

In general, as we grow to become middle-aged or elderly, our capacity for digestion and absorption including gastric acid secretion is lower than in the young [8, 10]. However, at the same time, the presence or absence of infection of Helicobacter pylori and the infectious period [9], stages of atrophic gastritis [9], and immune function such as Th1 immune reaction superiority of gastric mucosa [10] can be considered to be related factors which have a significantly large impact in the case of the elderly. As the progress speed of atrophic gastritis is negatively related to IGF-I concentration which is the second messenger of growth hormone, it is verifiable that an IGF-I low-value person is prone to suffer from atrophic gastritis progression [11].

It is considered that in the case of the young, even if they ingest starch with non-biodegradable characteristics, they are able to decompose it easily due to the strong amylase activity based on their high level gastric acid secretion. As a result, their blood glucose suppression action was less likely to appear than in the middle-aged. This result was observed in the previous report, too. In the test of indigestible dextrin and its postprandial blood glucose action, the effect was not easily verified in the case of the young [12].

Limitations of the study

In this test, a crossover method was not utilized, due to the test food production. Instead, we used control food in the first test, and then test food in the second test, and followed the process in the same order. Therefore, our analysis was conducted based on an assumption that the difference based on the time occasion or environmental factors on the test day might be almost equal.

The level of postprandial hyperglycemia was influenced by the duration of the fasing period [13], blood glucagon value [13], sleep time of the day before [5], and melatonin secretion [14]. In other words, when fasting time becomes longer due to skipping breakfast, postprandial hyperglycemia becomes remarkable, which is verifiably related to glucagon secretion [13]. Quality of sleep greatly influences postprandial blood glucose levels on the following day, while shorter sleep time causes a significant blood glucose spike [5]. A large amount of melatonin secretion mitigates postprandial hyperglycemia [14]. We conducted an analysis on the assumption that "the factors mentioned above were almost equal" if the subject was the same.

Safety

Both during and after the test, no significant harmfull event caused by this test food was observed.

Conclusion

It was shown that the newly-developed distarch phosphate (test food) being indigestible has the effect to delaying postprandial blood glucose rise, and to mitigate postprandial hyperglycemia or blood glucose spikes. Blood glucose spike mitigation action by distarch phosphate was found to have a greater impact on middle-aged adults compared to the young in their early 20's who have a highly active capability in digestion and absorption. At the same time, the test food was verified to be safe. A striking level of postprandial hyperglycemia is called a blood glucose spike and has been shown to cause an aldehyde spark. In conclusion, by use of distarch phosphate in various foods, we are able to mitigate postprandial hyperglycemia and blood glucose spikes, as well as to prevent various glycation stress disorders.

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Conflict of Interest Statement

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Reference

1) Nagai R, Mori T, Yamamoto Y, et al. Significance of advanced glycation end products in aging-related disease. Anti-Aging Med. 2010; 7: 112-119.
2) Ichihashi M, Yagi M, Nomoto K, et al. Glycation stress and photo-aging in skin. Anti-Aging Med. 2011; 8: 23-29.
3) Tanaka M, Ito H. Knowledge: What is a glucose spike? Heart View. 2017; 21: 837-843. (in Japanese)
4) Yagi M, Takabe W, Wickramasinghe U, et al. Effect of heat-moisture-treated high-amylose corn starch-containing food on postprandial blood glucose. Glycative Stress Res. 2018; 5: 151-162.
5) Yonei Y, Yagi M, Takabe W. Prime: International Journal of Aesthetic & Anti-Ageing Medicine. 2018; 8: 19-23.
6) Tachibe M, Oga H, Nishibata T, et al. Resistant starch Type 4, cross-linked phosphate starch and hydroxypropyl distarch phosphate attenuate rapid glycemic response in men. Japanese Pharmacology & Therapeutics (Yakuri to Chiryo). 2010; 38: 731-736.
7) Tachibe M, Ohga H, Nishibata T, et al. Digestibility, fermentability, and energy value of highly cross-linked phosphate tapioca starch in men. J Food Sci. 2011; 76: H152-155.
8) Masaoka I, Yakabi K, Nakamura T. A study on gastric acid secretion in the elderly. Journal of Geriatric Gastroenterology. 1994; 6: 55-59. (in Japanese)
9) Shimatani T, Inoue M, Hprikawa Y, et al. Is the decline of gastric acid secretion in the elderly caused by aging? Journal of the Japanese Conference of Geriatric Gastroenterology. 1999; 1: 42-46. (in Japanese)
10) Fukui H, Chiba T. Influence of *Helicobacter pylori* infection to changes of the stomach function by aging. Proceedings of the Japanese Society of Gastric Secretion Research. 2003; 35: 25-29. (in Japanese)
11) Yonei Y, Maeda N, Inagaki Y, et al. Relation between serum insulin like growth factor-I and the atrophic gastritis with or without liver cirrhosis. Journal of the Japanese Conference of Geriatric Gastroenterology. 2002; 4; 63-67. (in Japanese)
12) Morita Y, Hirano A, Sawanobori M. Indigestible dextrin-containing drink and postprandial blood glucose. Glycative Stress Res. 2017; 4: 93-103.
13) Hayashi S, Takabe W, Ogura M, et al. Effect of breakfast on lunch time postprandial blood glucose. Glycative Stress Res. 2017; 4: 124-131.
14) Ogura M, Okuda F, Hattori A, et al. Effect of melatonin intake on postprandial blood glucose in the breakfast. Glycative Stress Res. 2018; 5: 75-81.