

# The effects of L-arabinose on intestinal sucrase activity: dose-response studies in vitro and in humans<sup>1–3</sup>

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## ABSTRACT

**Background:** On the basis of results in cell cultures, rodents, and pigs, L-arabinose may inhibit intestinal sucrase activity and thereby delay sucrose digestion.

**Objective:** The objective was to investigate the dose-response effects of L-arabinose on intestinal sucrase activity in vitro and glucose tolerance, appetite, and energy intake in humans.

**Design:** In vitro, Caco-2 cells were cultured for 21 d, homogenized, and used as an enzyme preparation with sucrose as substrate in concentrations from 7 to 280 mmol/L with 0.84, 1.4, and 2.8 mmol L-arabinose/L as inhibitor. Released glucose was measured after 30 min. In the human studies, 15 healthy men participated in a randomized, double-blind, crossover study. Sucrose beverages (75 g in 300 mL) supplemented with 0%, 1.3%, 2.7%, and 4% by weight of L-arabinose were tested at breakfast. Blood for the measurement of glucose, insulin, C-peptide, incretin hormones, and triacylglycerol was collected under fasting conditions and for 3 h postprandially. Postprandial appetite sensations and energy intake at lunch were registered.

**Results:** In vitro, the addition of L-arabinose resulted in uncompetitive inhibition of sucrase activity. In the human studies, supplementation with 4% L-arabinose produced an 11% lower glucose peak, a 33% lower and delayed insulin peak, a 23% reduction in the incremental area under the curve (iAUC) for insulin, a 23% lower and delayed C-peptide peak, a 9% reduction in the iAUC for C-peptide, a 53% increase in the iAUC for glucagon-like peptide-1 (GLP-1), and a 28% reduction in the iAUC for glucose-dependent insulinotropic polypeptide. No effects on triacylglycerol, gastrointestinal symptoms, appetite ratings, or energy intake were observed.

**Conclusions:** L-Arabinose inhibits sucrase activity from Caco-2 cells; 4% L-arabinose in sucrose beverages reduces postprandial glucose, insulin, and C-peptide responses and enhances the GLP-1 response in humans without gastrointestinal adverse effects. This trial is registered at clinicaltrials.gov as NCT00302302. *Am J Clin Nutr* 2011;94:472–8.

## INTRODUCTION

The relatively high intake of added sugar in industrialized countries and the subsequent health consequences are currently under debate (1–6). Several authorities and research groups have recommended that <10% of the daily energy intake should be derived from added sugar (7–10). However, in past decades added sugar intake has increased, particularly in children (11–13). A high intake of added sugar may contribute to excess energy intake, which leads to overweight and is one of the factors involved

in the development of the metabolic syndrome and thereby increase the risk of developing type 2 diabetes and cardiovascular diseases (14).

L-Arabinose is a naturally occurring pentose and is widely distributed as a component of complex nonstarch polysaccharides in plant cell walls, including maize, wheat, rye, rice, sugar beets, and plant gums. In vitro studies, performed with porcine intestinal mucosa, indicated an inhibitory effect of L-arabinose on intestinal sucrase activity (15). In vivo studies investigating the effects of L-arabinose by feeding mice (15) and rats (16) sucrose and sucrose in combination with L-arabinose resulted in suppressed blood glucose and insulin responses. Furthermore, liver triacylglycerol concentrations increased when the rats were fed sucrose, but additional feeding of L-arabinose prevented this increase (16). In addition, 2 studies investigating the acute (test meal) (17) and subchronic (twice a day for 9 wk) (18) effects of L-arabinose after sucrose feeding in rats and pigs showed suppressed blood glucose in both cases. Accordingly, potential nutritional advantages of consuming L-arabinose in combination with sucrose may be the delayed digestion of sucrose and consequently a slower absorption of glucose, resulting in delayed and decreased blood glucose and insulin responses. Animal studies using chicks, roosters, and pigs as models indicate that the metabolizable energy of L-arabinose is significantly less than that of D-glucose (19–21). Effects of consumption of L-arabinose on plasma glucose and insulin response and possible gastrointestinal adverse effects have been studied in human subjects to a very limited degree. Only one abstract from a Japanese study is reported in the literature (22). This study showed that increases in plasma glucose and serum insulin were reduced in both healthy and type 2 diabetic subjects after consumption of sucrose with 4% added L-arabinose. Possible digestive implications as increased flatulence, diarrhea, or stomach pain were not reported.

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The human cell line Caco-2 has been widely used as a predictor of in vivo metabolism by the intestinal mucosa in humans. In culture, Caco-2 cells exhibit typical enterocyte-like differentiation, formation of a confluent monolayer, expression of tight junctions, and brush border enzymes such as sucrase (19). These characteristics make the Caco-2 cell model ideal for enzyme kinetic studies on sucrase activity with L-arabinose as inhibitor.

Accordingly, the objective of this study was to investigate the dose-response effects of L-arabinose on intestinal sucrase activity in vitro in the Caco-2 cell model and to evaluate the effects of L-arabinose on postprandial glucose, insulin, C-peptide, incretin hormones, triacylglycerol, and appetite responses at breakfast as well as energy intake at a subsequent lunch in healthy men.

## SUBJECTS AND METHODS

### In vitro studies

Caco-2 cells from passage numbers 38 and 39 were seeded onto polycarbonate membranes and cultured at 37°C and 5/95% CO<sub>2</sub>/air. At day 21, the Caco-2 cells were rinsed with phosphate-buffered saline, scraped off the membranes, and homogenized by sonication. For the kinetic studies, a sucrase assay was established with Caco-2 cell homogenates corresponding to 2.2 mg protein/mL and sucrose solutions at final concentrations of 7, 14, 28, 70, 140, and 280 mmol/L in 0.1 mol maleate buffer/L (pH = 6.0). As inhibitor, L-arabinose was used at final concentrations of 0.84, 1.4, and 2.8 mmol/L. Sucrose was analytic grade and was obtained from Sigma-Aldrich; L-arabinose had a purity of >98% and was obtained from Nordic Sugar. The amount of glucose released by the enzymatic reaction was linear up to 60 min, so a 30-min reaction time was used and released glucose was measured by using a gluco-quant Glucose/HK kit (GLU Roche/Hitachi 1447513; Roche Diagnostics GmbH, Mannheim, Germany) with a Cobas Mira Plus Spectrophotometer (Roche Diagnostic Systems, F Hoffmann-La Roche, Basel, Switzerland)

### Human studies

#### Subjects

In planning the study, it was estimated that 15 subjects would be sufficient to detect significant differences in glucose, insulin, and C-peptide peak changes. The inclusion criteria were as follows: healthy males, 18–30 y of age, normal to slightly overweight (body mass index; in kg/m<sup>2</sup>; 18.5–26), no blood donation within the past 3 mo before entering the study, no diabetes, no hypertension, no chronic diseases, no use of dietary supplements, alcohol intake <21 drinks/wk, no smoking, no regular use of medicine, not an elite athlete, and no change in physical activity during the study. Males were recruited from the area of Copenhagen by Internet advertisement, and flyers were posted at local universities. Approximately 40 men responded by telephone or e-mail, and 18 of these men went through a screening test with the measurement of body weight, height, and blood pressure and an interview regarding general health and drinking and smoking habits. One man did not meet the inclusion criteria, 1 declined to participate, and 1 fainted during the first test day and dropped out.

Fifteen healthy men (age 18–30 y) of normal weight or slightly overweight (body mass index < 26) and no sign of gastrointestinal disorders were included. Body weight was registered

before each meal, and height was measured on the first morning. After having received verbal and written information about the study, all subjects gave their written consent. This study was approved by the Regional Ethical Committee to be in accordance with the Helsinki-II declaration (KF) 01 270121. The study was carried out at the Department of Human Nutrition, University of Copenhagen. The baseline characteristics of the 15 subjects are given in **Table 1**.

### Experimental design

This dose-response study had a randomized double-blinded crossover design based on 4 single tests, with increasing doses of arabinose consumed at 0800. The test periods were separated by 1-wk washout periods.

The evening before each test, the subjects received a standardized meal consisting of rice, meat sauce, and juice to be eaten before 2000. The energy content of the meal was 4.5 MJ, and 50% of energy was from carbohydrate, 13% of energy was from protein, and 37% of energy was from fat; the dietary fiber content was 0.4 g/100 g. After fasting overnight, the subjects ingested sucrose beverages containing 75 g sucrose dissolved in 300 mL water. The dose-response effects of L-arabinose were studied by adding 0, 1, 2, and 3 g L-arabinose (corresponding to 0%, 1.3%, 2.7%, and 4% by weight) to the 4 sucrose beverages, respectively. The test beverages were prepared by Nordic Sugar, who blinded them as A, B, C, and D, respectively.

The ad libitum lunch served was a pasta salad. It consisted of pasta, smoked boiled ham, carrots, peas, sour cream, mayonnaise, olive oil, and basil. The distribution of energy was as follows: 54.5% of energy as carbohydrates, 15.5% of energy as protein, and 30.0% of energy as fat. The dietary fiber content of the lunch was 1.5 g/100 g. All nutrient calculations of the meals were done by using the computer database of foods from The National Food Agency of Denmark (Dankost 2000) (23).

### Measurements

The subjects were not allowed to drink alcohol or perform strenuous physical activity 24 h before each test day. On the test day, the fasting subjects traveled to the department by car, bus, or train (the least strenuous means of transportation). On arrival, they were weighed and, after 10 min of rest, blood pressure was measured and an intravenous catheter was placed in the antecubital vein. After an additional 10 min of rest, fasting blood samples were collected at –15 min and again at 0 min and then the sucrose beverage, which had to be consumed within 15 min, was served. Blood samples for

**TABLE 1**  
Subject characteristics (n = 15)<sup>1</sup>

Variable	Value
Age (y)	25 ± 3.2
Weight (kg)	76.1 ± 8.9
Height (m)	1.83 ± 0.07
BMI (kg/m <sup>2</sup> )	22.8 ± 2.1
Blood pressure (mm Hg)	
Systolic	125 ± 7
Diastolic	71 ± 5
Heart rate (beats/min)	61 ± 11

<sup>1</sup> All values are means ± SDs.

measurement of glucose, insulin, and C-peptide were taken 15 and 0 min before the test beverage was consumed to ensure proper baseline values and again 15, 30, 45, 60, 90, 120, and 180 min after the test beverage. Blood samples for the measurement of triacylglycerols, glucose-dependent insulinotropic polypeptide (GIP) and glucagon-like peptide-1 (GLP-1) were collected 0, 30, 60, 90, 120, and 180 min after ingestion of the test beverage. During the postprandial measurements, the subjects could watch television or videos or read (light entertainment); toilet visits were allowed when necessary. Water (200 mL) was offered at 90 min, and intake was registered and repeated on every test day. After removal of the intravenous catheter, the subject was walked to the lunch room, where lunch was served at  $\approx$ 195 min after the test beverages.

### Appetite and energy intake

Visual analogue scales (VAS) (24) expressing the most positive and the most negative ratings were used to assess hunger, satiety, fullness, prospective food consumption, thirst, comfort, and the desire to eat something fatty, salty, or sweet or meat/fish. The questionnaires were presented as a small booklet with one question per page. Ratings of subjects' appetite sensations were performed before and 30, 60, 90, 120, 150, and 180 min after the test beverage was served.

The ad libitum energy intake at lunch was registered, as was the ad libitum water intake on the first test day. The VAS was used to assess the palatability (appearance, smell, taste, aftertaste, and overall palatability) of the ad libitum lunch.

### After study

Gastrointestinal symptoms (heartburn, distension, nausea, vomiting, stomachache, rumbling in the gut, flatulence, and diarrhea) were registered 21 h after the tests by asking the subjects to fill out a questionnaire by rating symptom severity on a 5-level scale.

### Laboratory analysis

Blood was drawn without stasis through the indwelling catheter. Blood samples for glucose, insulin, and C-peptide analyses were collected into plain tubes; within 30 min of collection, the samples were centrifuged for 15 min at  $2800 \times g$  and  $4^\circ\text{C}$ , and the serum was stored at  $-20^\circ\text{C}$  until analyzed. Blood for the measurement of triacylglycerol, plasma GIP, and GLP-1 was collected in iced tubes containing EDTA. Within 30 min, samples were centrifuged for 15 min at  $2800 \times g$  and  $4^\circ\text{C}$ , and the plasma was stored at  $-20^\circ\text{C}$  until analyzed.

Glucose and triacylglycerol were measured with the use of an enzymatic colorimetric method on a Cobas Mira Plus. Serum glucose concentrations were measured by using a gluco-quant Glucose/HK kit (GLU Roche/Hitachi 1447513; Roche Diagnostics GmbH, Mannheim, Germany) (25). Plasma triacylglycerol concentrations were measured by using the triacylglycerols GPOPAP kit (TAG Roche/Hitachi 2016648; Roche Diagnostics GmbH) (26). Serum insulin concentrations were determined by using an enzyme-linked immunosorbent assay technique (AutoDELFIA Insulin kit B080-101; Wallac Oy, Turku, Finland) on an assay system (AutoDELFIA 1235-514; Wallac Oy) (27). Serum C-peptide concentrations were measured by using a solid-phase, competitive

chemiluminescent enzyme immunoassay on an Immulite 1000 Analyzer (Siemens). GIP and GLP-1 concentrations in plasma were measured after extraction of plasma with 70% ethanol (vol: vol, final concentration). The GIP radioimmunoassay used the C-terminally directed antiserum R65, which cross-reacts fully with human GIP but not with the so-called GIP 8000, whose chemical nature and relation to GIP secretion is uncertain. Human GIP and  $^{125}\text{I}$  human GIP (70 Mbq/nmol) were used for standards and tracer. The plasma concentrations of GLP-1 were measured against standards of synthetic GLP-1 7-36 amide by using antiserum code no. 89390, which is specific for the amidated C-terminus of GLP-1 and therefore mainly reacts with secreted GLP-1 of intestinal origin.

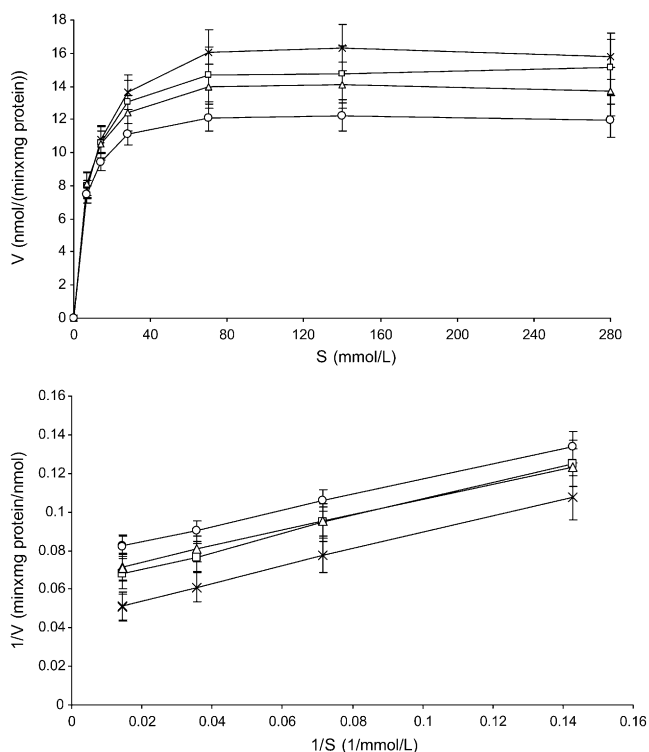
### Statistical analysis and calculations

Descriptive data are reported as means  $\pm$  SDs, and the results are reported as means  $\pm$  SEMs. Results were considered significant when  $P < 0.05$ . The Lineweaver-Burk double reciprocal plot represents the inverse values of substrate concentration plotted against the inverse reaction velocity and is used to determine the kind of enzyme inhibition, and parallel lines indicate uncompetitive inhibition. The incremental area under the curve (iAUC) or above the curve (iAOC), ignoring the area beneath (or above in the case of iAOC) the fasting concentration was calculated geometrically by applying the trapezoid rule. The iAUC or iAOC was calculated for the entire test period from 0 to 180 min. Postprandial response curves were evaluated by comparing peak values and iAUC by using analysis of covariance, with fasting values as covariates and time to peak values by using analysis of variance. Furthermore, appetite ratings were evaluated with the 3-h mean and iAUC or iAOC values for ratings with increasing or decreasing ratings, respectively. In these evaluations, a mixed-model analysis of covariance with fasting values as covariates and subjects included as random effects was performed. In addition the curves of glucose, insulin, C-peptide, and GLP-1 were analyzed by using repeated-measures analysis of covariance. Residual plots of data were examined to consider homogeneity of variance and Shapiro-Wilk test performed for normal distribution of data and logarithmic transformation was used when required. A significant result was followed by using a Tukey-Kramer test for post hoc analysis. All statistical analyses were performed by using Statistical Analysis Package (version 8.02; SAS Institute, Cary, NC).

## RESULTS

### In vitro studies

Reaction velocity ( $V$ ) plotted against substrate concentrations showed classic Michaelis-Menten kinetics and significant inhibition with increasing amounts of L-arabinose (**Figure 1**). The identification of uncompetitive inhibition was based on Lineweaver-Burk analyses, which showed parallel lines for the different inhibitor concentrations (Figure 1). Maximum  $V$  ( $V_{\text{max}}$ ) decreased from 19.8 through 14.7 and 14.1 to 12.2 nmol/(min  $\cdot$  mg protein), and  $K_m$  decreased from 9.8 through 7.3 and 6.1 to 5.3 mmol/L, when the inhibitor concentrations increased from 0 through 0.84 and 1.4 to 2.8 mmol L-arabinose/L. Thus, the addition of 0.84, 1.4, and 2.8 mmol L-arabinose/L resulted in



**FIGURE 1.** Michaelis-Menten curves (top) and Lineweaver-Burk plot (bottom). Shown are substrate (S; sucrose) concentrations of 0–280 mmol/L and different L-arabinose concentrations [0.0 mmol/L (x), 0.84 mmol/L (□), 1.4 mmol/L (△), and 2.8 mmol/L (○)] as inhibitor of sucrase. Data are means ± SEMs. *n* = 6. *V* is the velocity of the reaction.

25%, 29%, and 38% inhibition of sucrase activity, respectively, at  $V_{max}$ . The apparent  $K_i$  was calculated to  $2.8 \pm 0.3$  mmol/L (mean ± SEM; *n* = 3) from the Lineweaver-Burke plots.

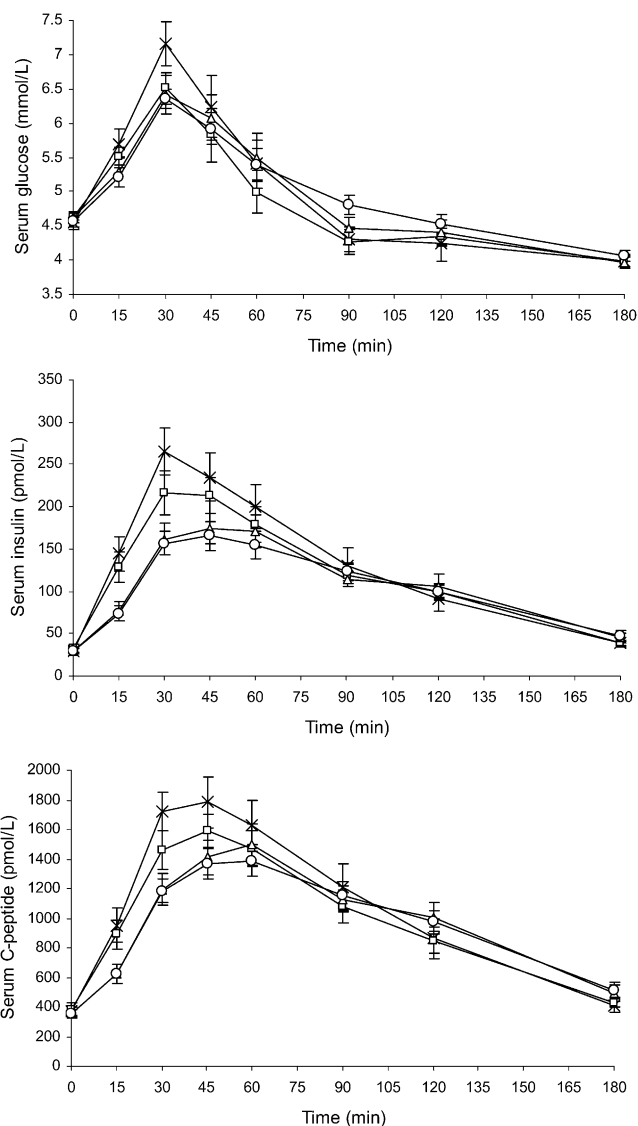
**Human studies**

*Glucose*

Postprandial serum glucose had a higher peak value after treatment with 0 g L-arabinose ( $7.31 \pm 0.32$  mmol/L) than after treatment with 1 g ( $6.65 \pm 0.27$  mmol/L; *P* < 0.01), 2 g ( $6.57 \pm 0.28$  mmol/L; *P* < 0.01), and 3 g L-arabinose ( $6.47 \pm 0.14$  mmol/L; *P* < 0.01) (Figure 2). Time to peak and iAUC were not different between treatments. Repeated-measures analyses from 0 to 180 min showed no difference between treatments (*P* = 0.31); however, analysis of the curves from 0 to 120 min showed a significant dose effect (*P* = 0.03), and post hoc adjustment showed a difference between treatments with 0 g and 1 g L-arabinose (*P* = 0.04).

*Insulin*

The treatment with 0 g L-arabinose increased the postprandial serum insulin peak ( $281.4 \pm 28.2$  pmol/L) in comparison with the 3 other treatments (1 g L-arabinose:  $235.8 \pm 25.9$  pmol/L, *P* < 0.01; 2 g L-arabinose:  $196.9 \pm 19.8$  pmol/L, *P* < 0.01; 3 g L-arabinose:  $186.2 \pm 15.3$  pmol/L, *P* < 0.01; Figure 2), and the peak occurred earlier than with the treatments with 2 and 3 g L-arabinose (*P* < 0.05). iAUC was significantly different between treatments with 0 and 3 g L-arabinose (*P* = 0.02) and was highest after the treatment with 0 g L-arabinose. Repeated-measures anal-



**FIGURE 2.** Mean (±SEM) serum concentrations of glucose, insulin, and C-peptide in 15 normal-weight to slightly overweight men after intake of sucrose beverages supplemented with 0 (x), 1 (□), 2 (△), or 3 (○) g L-arabinose. Glucose [peak, *P* = 0.001; time to peak, *P* = 0.133; incremental area under the curve (iAUC), *P* = 0.245], insulin (peak, *P* < 0.0001; time to peak, *P* = 0.002; iAUC, *P* = 0.017), C-peptide (peak, *P* < 0.0001; time to peak, *P* = 0.07; iAUC, *P* = 0.36). Peak values and iAUC were evaluated by using ANCOVA, with fasting values as covariates; time to peak values were evaluated by using ANOVA. Postprandial response curves were evaluated by repeated-measures ANCOVA.

ysis at 0–180 min showed a significant dose effect (*P* = 0.039), and post hoc adjustment showed that the difference was between treatments with 0 and 2 g L-arabinose (*P* = 0.04). Analysis of the curves from 0 to 120 min showed a significant dose effect (*P* < 0.001), and post hoc adjustment showed the difference to be between 0 and 2 g L-arabinose (*P* = 0.002), 0 and 3 g L-arabinose (*P* = 0.01), and 1 and 2 g L-arabinose (*P* = 0.04); the lowest doses of L-arabinose (0 and 1 g) had the highest response curves.

*C-peptide*

Postprandial serum C-peptide showed a higher peak value after treatment with 0 g L-arabinose ( $1974 \pm 152$  pmol/L) than after

treatment with 1 g ( $1690 \pm 128$  pmol/L;  $P < 0.01$ ), 2 g ( $1579 \pm 133$  pmol/L;  $P < 0.01$ ), and 3 g ( $1520 \pm 93$  pmol/L;  $P < 0.01$ ) L-arabinose (Figure 2), and a tendency for a delayed peak after treatment with L-arabinose was observed but was not statistically significant ( $P = 0.08$ ). No differences between treatments were found in repeated-measurement analyses or in iAUC values.

### Triacylglycerol

Repeated-measures analysis showed no significant dose effects (Figure 3).

### GIP

A significant difference between treatments was found in postprandial plasma concentrations of GIP (Figure 3) in peak ( $P < 0.01$ ) and iAUC ( $P < 0.01$ ) values. A post hoc adjustment showed a significant difference between treatments with 0 and 2 g L-arabinose (peak:  $P < 0.01$ ; iAUC,  $P < 0.01$ ), with 0 g L-arabinose having the highest response, and between treatments with 1 and 2 g L-arabinose (peak:  $P < 0.01$ ; iAUC:  $P = 0.02$ ), with 1 g L-arabinose having the highest response. Repeated-measures analysis showed a significant dose effect ( $P = 0.04$ ), and post hoc adjustment showed that GIP values were higher after treatment with 0 or 1 g L-arabinose than after 2 or 3 g L-arabinose.

### GLP-1

Postprandial plasma GLP-1 increased more (iAUC) after treatment with 3 g L-arabinose than after treatment with 2 g L-arabinose ( $P = 0.04$ ), 1 g L-arabinose ( $P = 0.001$ ), and 0 g L-arabinose ( $P < 0.001$ ) (Figure 3). The peak value was highest after treatment with 3 g L-arabinose and was significantly different from treatment with 1 g L-arabinose ( $P = 0.01$ ). Repeated-measures analysis showed a significant dose effect ( $P < 0.001$ ), and post hoc adjustment showed that the difference was between treatments with 0 g L-arabinose and 1, 2, and 3 g L-arabinose ( $P = 0.03$ ,  $P = 0.003$ , and  $P < 0.001$ , respectively). Furthermore, there was a difference between treatments with 3 g L-arabinose and 1 and 2 g L-arabinose, respectively ( $P \leq 0.001$ ), with the highest response after treatment with 3 g L-arabinose.

### Appetite

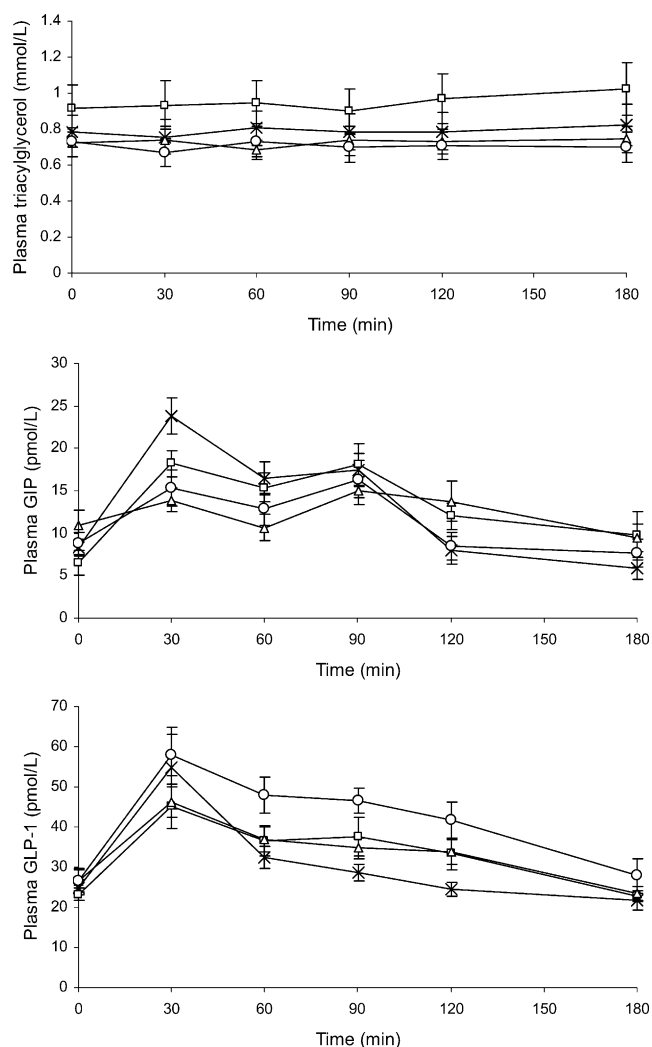
After the treatment with 3 g L-arabinose, subjects postprandially felt a greater desire to eat something fatty than after the treatments with 0 and 2 g L-arabinose (Figure 4). However, for the preference of eating something fat, a significant period effect was also observed ( $P = 0.011$ ).

### Energy intake

No differences were seen in palatability ratings (appearance, smell, taste, aftertaste, and overall palatability) of the ad libitum lunch. No difference in energy intake was observed between treatments ( $P = 0.6$ ; 0 g L-arabinose:  $3467 \pm 261$  kJ; 1 g L-arabinose:  $3345 \pm 265$  kJ; 2 g L-arabinose:  $3253 \pm 202$  kJ; and 3 g L-arabinose:  $3229 \pm 266$  kJ). Adjustment for body weight or metabolic body weight did not change these results.

### Gastrointestinal symptoms

One of 15 subjects reported symptoms (mild distension) after treatment with 0 g L-arabinose and 1 of 15 subjects reported

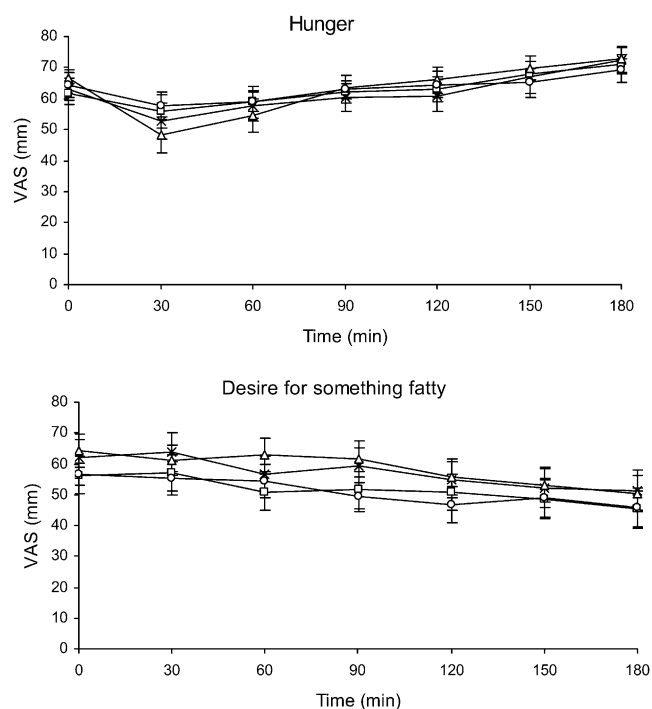


**FIGURE 3.** Mean ( $\pm$ SEM) plasma concentrations of triacylglycerol, glucose-dependent insulinotropic polypeptide (GIP), and glucagon-like peptide-1 (GLP-1) in 15 normal-weight to slightly overweight men after intake of sucrose beverages supplemented with 0 (x), 1 (□), 2 (△), or 3 (○) g L-arabinose. Triacylglycerol [peak,  $P = 0.532$ ; time to peak,  $P = 0.245$ ; incremental area under the curve (iAUC),  $P = 0.854$ ], GIP (peak,  $P = 0.0025$ ; time to peak,  $P = 0.211$ ; iAUC,  $P = 0.0056$ ), GLP-1 (peak,  $P = 0.0172$ ; time to peak,  $P = 0.492$ ; iAUC,  $P < 0.0001$ ). Peak values and iAUC were evaluated by using ANCOVA, with fasting values as covariates; time to peak values were evaluated by using ANOVA. Postprandial response curves were evaluated by repeated-measures ANCOVA.

symptoms (mild nausea) after treatment with 1 g L-arabinose. Two of 15 subjects reported symptoms after treatment with 2 g L-arabinose; one subject reported mild diarrhea and the second mild distension and a severe stomachache and diarrhea in the evening. Two of 15 subjects reported symptoms after treatment with 3 g L-arabinose; the first subject reported mild flatulence, and the second mild heartburn, moderate nausea, and a mild stomachache after the meal. These differences were not statistically significant ( $P = 0.88$ ).

### DISCUSSION

The *in vitro* studies presented here clearly showed that L-arabinose inhibited the brush border enzyme sucrase in an uncompetitive dose-dependent manner. The consequences of



**FIGURE 4.** Mean ( $\pm$ SEM) subjective appetite ratings on 100-mm visual analog scales (VAS) for hunger (“I have never been more hungry”) and desire for something fatty in 15 normal to slightly overweight men after intake of sucrose drinks supplemented with 0 ( $\times$ ), 1 ( $\square$ ), 2 ( $\triangle$ ), or 3 ( $\circ$ ) g L-arabinose. Hunger [incremental area over the curve (iAOC),  $P = 0.013$ ; 3-h mean,  $P = 0.15$ ], fatty (iAOC,  $P = 0.008$ ; 3-h mean,  $P = 0.44$ ). The 3-h mean and iAOC were evaluated by using mixed-model ANCOVA, with fasting values as covariates.

this inhibition were further elucidated in the human study, in which L-arabinose suppressed the increases in blood glucose, insulin, and C-peptide concentrations in plasma after sucrose ingestion and also augmented the postprandial increase in the GLP-1 response.

The *in vitro* study was designed to reveal the kinetics of sucrase activity and to examine the dose-response effects. The lowered  $K_m$  and the decreased maximum enzyme activity ( $V_{max}$ ) indicated uncompetitive inhibition, which may be explained by the binding of L-arabinose to the complex formed between sucrase and sucrose. This increases the apparent affinity of sucrase for sucrose so that the binding period for sucrose or the product (glucose + fructose) to the active site is increased. The inhibiting effect of L-arabinose on sucrase was also shown by Seri et al (15) *in vitro* in porcine intestinal mucosa. Importantly, because the Caco-2 cell line is of human origin, the results are relevant for human physiology, suggesting that arabinose intake would result in decreases in postprandial glucose concentrations.

On the basis of the results from these *in vitro* studies, a human study with 3 different doses of L-arabinose in sucrose beverages was designed. The main findings were postprandial decreased blood glucose, insulin and C-peptide concentrations and an increase in incretin hormone GLP-1 concentrations after all 3 doses of L-arabinose in a dose-dependent manner. The addition of L-arabinose altered the glucose response to a smaller peak value and a slower decline, but did not change the iAUC. The effect was much more pronounced for the insulin response, for which there was a smaller (23% reduction) and later peak after the highest dose of

L-arabinose. The current results can be explained by a delay in glucose absorption without a reduction in the absorbed amount of glucose. The effects on blood glucose are comparable with those obtained with soluble dietary fibers (28). In one instance we found that the glucose response decreased significantly with 1 g L-arabinose, but not with higher amounts. The small but significant difference observed was probably a consequence of the multiple statistical tests performed and not of physiologic significance in itself.

In addition, we measured the incretin hormones GIP and GLP-1, both of which are strongly insulinotropic and with increased secretion in response to carbohydrate intake (29). GIP is secreted in the upper gastrointestinal tract from endocrine K cells in response to absorbable carbohydrates and lipids and potentiates insulin secretion from pancreatic  $\beta$  cells (29). GLP-1 is mainly produced in the lower gastrointestinal tract from endocrine L cells in response to nutrients in the lumen, and its biological activities include stimulation of glucose-dependent insulin secretion and insulin biosynthesis, inhibition of glucagon secretion, gastric emptying, and food intake (29). Repeated-measures analysis showed a lower response of GIP secretion after the 2 highest doses of L-arabinose and the greatest increase in GLP-1 secretion after the 2 highest doses of L-arabinose (53% after the highest dose of L-arabinose). Sucrase inhibition probably delays the digestion of sucrose in the upper part of the small intestine, with sucrose being transferred to a more distal part of the small intestine, which could explain the decreased GIP response and the augmented GLP-1 response after L-arabinose treatment. The explanation for the concentrations of GLP-1, GIP, and insulin responses is of course speculative. The GLP-1-responses were fairly convincing, with a trend of a dose-related response. At the same time, GIP concentrations decreased, and, because both peptides stimulate glucose-induced insulin secretion, the net effect is unpredictable. When blood glucose is decreasing, the potentiating effects of the incretin hormones disappear.

The GLP-1 response was not sufficient to produce a change in appetite ratings between treatments, or, after subsequent food intake, because the plasma concentration increased only by  $\approx 30\%$ . In experiments documenting the effect, the increase has been 200–400% (30). L-Arabinose added to solid food in a mixed meal may change these results.

Sucrase inhibition may also be expected to increase the amount of carbohydrate entering the colon, causing increased fermentation by colonic bacteria and subsequently gastrointestinal discomfort as a result of hydrogen production (31). However no differences in the reported gastrointestinal symptoms were observed.

As mentioned in the Introduction, only one study has been conducted in human subjects and reported by a Japanese research group (22). The study was performed in both healthy subjects and in patients with type 2 diabetes and investigated the effects of the addition of L-arabinose to a sucrose-containing beverage or meal. In the first experiment, sucrose beverages with and without 2% L-arabinose resulted in a decrease in blood glucose in healthy subjects (crossover design;  $n = 8$ ). Second, the dose response was evaluated in 40 healthy subjects given sucrose jelly with and without 2%, 3%, and 4% added L-arabinose. The addition of 3% and 4% L-arabinose resulted in a decrease in blood glucose (parallel design;  $n = 10$ ). In the third experiment, plasma glucose in patients with type 2 diabetes was similarly suppressed with L-arabinose at a concentration of 3% (22). These results agree with the results obtained in the current study.

Several conditions need to be addressed through improved postprandial blood glucose control to prevent or delay the development of lifestyle diseases. Prediabetes (impaired glucose tolerance) and insulin resistance, and even slightly elevated postprandial blood glucose concentrations, are conditions related to clearly elevated risks of cardiovascular disease (32).

According to the World Health Organization, a diabetes epidemic is ongoing; >250 million people worldwide have diabetes, and by 2025 this total is expected to increase to >380 million people. Type 2 diabetes is responsible for 90–95% of the cases (33). Not only is fasting blood glucose important in the development of type 2 diabetes, but postprandial hyperglycemia is crucial too. In 2007, The International Diabetes Federation published *Guideline for Management of Postmeal Glucose* (32). The guideline states that postmeal hyperglycemia, defined as a plasma glucose concentration >7.8 mmol/L 2 h after ingestion of food, is harmful and should be addressed. Postmeal hyperglycemia is associated with an increased risk of developing cardiovascular disease, retinopathy, cognitive dysfunction in elderly people with type 2 diabetes, and some cancer types (32). In addition the United Nations has just passed a resolution recognizing the global threat of the diabetes epidemic (33). This highlights the importance of developing new strategies in the fight against diabetes, prevention, and delay of the disease. A healthy lifestyle with changes in diet and an increased physical activity level is the most important and sustainable strategy. However, other novel strategies may be additive, such as the one suggested in the current study, wherein the addition of L-arabinose to a sugar beverage was shown to reduce postprandial blood glucose fluctuations. Additional studies are needed to explore whether the same results are obtainable in other food matrixes and to explore the long-term health consequences.

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