Effect of Small Doses of Fructose and Allulose on Postprandial Glucose Metabolism in Type 2 Diabetes: A Double-blind, Randomized, Controlled, Acute Feeding Equivalence Trial

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ABSTRACT

Aims: Fructose shows adverse effects at hypercaloric doses, but potential glycemic advantages at small doses (≤10-g/meal). Whether allulose, a low-calorie C-3 epimer of fructose, shares its advantages without concerns brought by its calories is unclear. Our aim was to assess and compare the effect of small doses of fructose and allulose on postprandial blood glucose regulation in type 2 diabetes.

Material and Methods: We conducted a double-blind, multiple-crossover, randomized, controlled, acute feeding equivalence trial in 24 participants with type 2 diabetes. Each participant was randomly assigned 6 treatments separated by ≥1-week washout. Treatments consisted of fructose or allulose at 0g (control), 5g or 10g added to a 75g glucose solution. A standard 75g-oral glucose tolerance test (75g-OGTT) protocol was followed with blood samples at -30, 0, 30, 60, 90, and 120-minutes. The primary outcome measure was plasma glucose incremental area under the curve (iAUC).

Results: Allulose significantly reduced plasma glucose iAUC by 8% at 10g compared with 0g (717.4±38.3 versus 777.5±39.9 mmol·min/L, p=0.015) with a linear dose response gradient between the reduction in plasma glucose iAUC and dose (p=0.016). Allulose also significantly reduced several related secondary and exploratory outcome measures at 5g (plasma glucose absolute mean and total AUC) and 10g (plasma glucose absolute mean, absolute and incremental maximum concentration [C_{max}], and total AUC) ($p<0.0125$). There was no effect of fructose at any dose. Although allulose showed statistically significant reductions in plasma glucose iAUC compared with fructose at 5g, 10g and pooled doses, these reductions were within the pre-specified equivalence margins of ±20%.

Conclusions: Allulose, but not fructose, led to modest reductions in the postprandial blood glucose response to oral glucose in individuals with type 2 diabetes. There is a need for long-term randomized trials to confirm the sustainability of these improvements.

INTRODUCTION

Sugars have emerged as the dominant nutrient of concern in the epidemics of obesity and diabetes. The fructose moiety in particular has been implicated as a potent driver of type 2 diabetes due to its unique set of biochemical, metabolic and endocrine responses[1, 2].

A less appreciated body of research suggests that small doses ($\leq$ 10g/meal) of fructose, at a level obtainable from fruit, may elicit a 'catalytic' effect on hepatic glucose metabolism by increasing glycogen synthesis as shown by ^{13}C nuclear magnetic resonance (NMR) under euglycemic conditions in people without diabetes[3] and decreasing hepatic glucose output under hyperglycemic clamp conditions in people with type 2 diabetes [4]. Clinical translation of these findings has shown that small doses of fructose decrease the postprandial blood glucose response to oral glucose in people with[5] and without type 2 diabetes [6]. Under chronic feeding conditions, fructose in exchange for other carbohydrates has further been shown to decrease HbA_{1c} in systematic reviews and meta-analyses of controlled feeding trials[7, 8]. This apparent benefit, however, is tempered by evidence that fructose providing excess calories has an adverse effect on body weight[9], fasting blood glucose levels and insulin sensitivity[8], fasting[10, 11] and postprandial[12] triglycerides, uric acid[13], and markers of non-alcoholic fatty liver disease (NAFLD)[14].

Identifying low-calorie alternatives to fructose that share its advantages without its adverse effects is of interest. Allulose is a low-calorie (< 0.2 kcal/g) C-3 epimer of fructose found

naturally in small amounts in dried fruits, brown sugar and maple syrup that shares many of its functional and sensory properties and is generally regarded as safe (GRAS) as a sugar substitute by the Food and Drug Administration (FDA) [15-18]. It has shown similar 'catalytic' effects on hepatic glucose metabolism in cultured hepatocytes[19, 20] and animal models[21, 22]. Small doses of allulose have also shown to reduce the postprandial blood glucose response to high glycemic index carbohydrate meals in people who are otherwise healthy[23] or have prediabetes[24]. Whether these effects of allulose are reproducible and are equivalent to those of fructose in people with type 2 diabetes is untested. The minimum dose at which improvements in glucose metabolism are observed also remains to be determined for both fructose and allulose in people with type 2 diabetes. The aim of this randomized, controlled, acute feeding trial was to assess and compare the effects of small 'catalytic' doses (5g, 10g) of fructose and allulose on postprandial glucose regulation in response to a 75g-oral glucose tolerance test (75g-OGTT) in individuals with type 2 diabetes.

MATERIAL AND METHODS

Participants

Recruitment took place from November 2015 to July 2016. Participants were included in the study if they met the following eligibility criteria: age 18 – 75 years, non-pregnant, non-smoker, BMI 18.5 – 35 kg/m^2, well-controlled type 2 diabetes ($HbA_{1c} \leq 58$ mmol/mol [7.5%]) on diet and/or antihyperglycemic agents, not taking insulin and free of other major illnesses. Eligible participants provided informed consent and received a financial reward for their participation. The study protocol was approved by the St. Michael's Hospital Ethics Review Board and registered on ClinicalTrials.gov (NCT02459834).

Trial Design

The trial followed a randomized, double-blind, multiple-crossover, acute feeding, equivalence design with a ≥1-week washout period. Sequence randomization of the six treatments was performed using a random sequence generator[25]. The study statistician who performed this randomization was blinded to the identity of participants and did not have contact with the participants or the data. There were two levels of allocation concealment. First, the manufacturer of the treatments (Tate & Lyle Ingredients Americas LLC, Hoffman Estates, IL, USA) provided unique codes for each of the six treatments. Second, the statistician who was blinded to the identity of these codes used the codes to label the packaging of the six treatments so that the treatments were only distinguishable by the participant number and the visit number to which they corresponded based on the randomization. The participants, study staff, investigators and outcome assessors were blinded to the identity of these treatment sequences. The two sets of blinding codes for each participant were not broken until all participants had completed the study and all analyses were completed.

Treatments

Participants received a total of six treatment drinks (provided and manufactured by Tate & Lyle Ingredients Americas LLC, Hoffman Estates, IL, USA) in random order: two control drinks and four test drinks. Treatments consisted of fructose or allulose at 0g (control), 5g or 10g added to a 75g glucose solution dissolved in 500 ml of water. The drinks were matched for appearance, sweetness, texture and packaging. Flavor and color enhancements were used to mask any differences.

Protocol

The protocol followed the World Health Organization guidelines for the administration of an OGTT[26]. This study was conducted in an outpatient setting at the Clinical Nutrition and Risk Factor Modification Centre in St. Michael's Hospital (Toronto, Canada). Participants arrived at the study center on six separate mornings following a 10-12h overnight fast. They were instructed to consume a minimum of 150g of carbohydrates each day over the three days prior to the study visit, and maintain their regular dietary, exercise, and medication patterns the evening before each study visit. Antihyperglycemic medication use was discontinued on the morning of each study visit. To ensure that fasting blood glucose was similar on each day, participants provided a finger prick blood sample for the measurement of fasting blood glucose using a point of care glucometer (Contour®Next EZ blood glucose monitor, Bayer, NJ, USA). If the fasting glucose value fell outside $\pm$ 2 mmol/L of their initial screening value or the average value of all previous study visits for those who had attended two or more visits, then participants were asked to return for another visit the following week[27]. If fasting blood glucose was acceptable, a Registered Nurse inserted a catheter into a forearm vein; the catheter was secured by tape and kept patent by saline. Two samples were collected in the fasting state: one at -30min and the other at 0min. One of the six treatment drinks was then administered in random order with instructions to consume it at a constant rate over 5 minutes. Additional blood samples were drawn at 30, 60, 90 and 120 minutes after the start of the treatment.

Outcome Measures

The pre-specified primary outcome measure was the incremental area under the curve (iAUC) for plasma glucose. Pre-specified secondary outcome measures included plasma insulin iAUC, plasma glucose and insulin absolute maximum concentrations (C_{max}), time of maximum concentrations (T_{max}), and mean incremental concentrations; the Matsuda whole body insulin

sensitivity index (Matsuda ISI_{OGTT}); and the early insulin secretion index ($\Delta PI_{30-0}/\Delta PG_{30-0}$). Exploratory outcome measures which were not pre-specified included plasma glucose and insulin total AUC, incremental C_{max}, and mean absolute concentrations; and the insulin secretion-sensitivity index-2 (ISSI-2).

Plasma Glucose and Insulin Analyses

Blood samples for glucose and insulin were collected in fluoride oxalate and EDTA tubes respectively, with plasma separated by centrifuge and immediately frozen at -72^{0}C. *Mount Sinai Services Inc.* performed analyses of plasma glucose using the hexokinase method[28, 29] and plasma insulin using the electrochemiluminescence immunoassay[30].

Calculations

Plasma glucose and insulin concentrations at -30min and 0min were averaged to provide a single measurement of fasting glucose and fasting insulin. Total AUC and iAUC (which ignored values below the fasting value) were calculated geometrically using the trapezoidal rule for plasma glucose and insulin for each participant[31]. The early insulin secretion index ($\Delta PI_{30-0}/\Delta PG_{30-0}$) is a measure of insulin secretion derived from the early period of the OGTT. It was calculated as the change in plasma insulin (PI) from 0 minutes to 30 minutes divided by the change in plasma glucose (PG) over the same period[32]. The Matsuda Insulin Sensitivity Index (Matsuda ISI_{OGTT}) is an OGTT-derived measure of whole-body insulin sensitivity that has been validated against the euglycemic insulin clamp technique[33]. It was calculated using the 75g-OGTT PG and PI concentrations as follows: $\sqrt{}$ (fasting PG x fasting PI x mean PG x mean PI), where PG was expressed in mg/dL (1/18 mmol/L) and PI in μU/ml (6 pmol/L). ISSI-2 is an OGTT-derived measure of β-cell function that has been validated against the disposition index from the

frequently sampled intravenous glucose tolerance[34]. It was calculated by taking the product of 1) insulin secretion as measured by the ratio of total area-under-the-insulin-curve (AUC_{ins}) to the total-area-under-the-glucose curve (AUC_{glu}) and 2) insulin sensitivity as measured by the Matsuda ISI_{OGTT}. ISSI-2 was calculated using SI units for AUC_{ins}, AUC_{glu} and Matsuda ISI_{OGTT}, such that ISSI-2 = total $AUC_{ins/glu}$ x Matsuda ISI_{OGTT}.

Statistical Analysis

Statistical analyses were performed using STATA 13.1 (StataCorp LP, College Station, TX, USA). We needed to recruit 25 participants to achieve a final sample size of n=20 (based on 20% attrition) to detect a difference in iAUC plasma glucose of 160 mmol·min/L (based on a 20% reduction from 800 mmol·min/L) assuming a standard deviation of 130 mmol·min/L with 90% power (1-β=90%)[35]. The sample size also provided 80% power (1-β=80%) to detect equivalence in the iAUC plasma glucose differences between fructose and allulose using margins of $\pm$ 20% assuming a standard deviation of 16.25% (130/800 mmol·min/L*100%)[36]. The 20% difference and equivalence margins were based on the minimally important difference proposed by Health Canada to support postprandial blood glucose response reduction claims[36]. Participants were excluded from analysis if fasting plasma glucose values at one or more study visits fell outside of the pre-specified tolerance limit of $\pm$ 2 mmol/L of the baseline fasting plasma glucose value (defined as the mean of all six study visits).

Separate analyses were conducted for fructose and allulose with the data averaged for the two controls (0g) for comparisons with the two other doses (5g, 10g). Linear mixed-effects models were used to assess differences in all outcome measures with unstructured covariance for repeated measures within subjects. Although we had pre-specified the use of repeated measures

ANOVA with the Dunnett's test to adjust for the pairwise comparisons between each dose (5g, 10g) and the mean of the two controls (0g) for fructose and allulose, we selected linear mixed-effects models as they allowed for the handling of missing data, fitting of the correlation between repeated measures in the same subject, and modelling of time, sequence, and carryover effects[37, 38]. We assessed the interactive effects of treatment and time (0, 30, 60, 90 and 120 mins) on mean incremental changes in plasma glucose and insulin. Significant interactions were explored at individual time points. Linear dose-response relationships were assessed using a continuous exposure variable in the mixed-effects model, while departures from linearity were assessed by comparing the linear dose model with the categorical dose model using a likelihood ratio test. Equivalence testing was conducted using the two one-sided test (TOST) procedure by determining whether the upper and lower bounds of the 90% CI for the effect of allulose on iAUC for plasma glucose fell within the equivalence margins ($\pm$ δ) set at $\pm$ 20% [36]. An equivalence test was chosen instead of a traditional comparative test to allow us to assess whether any differences between allulose and fructose were not just statistically significant but clinically significant based on the minimally important difference set by Health Canada to support postprandial blood glucose response reduction claims [36]. Subgroup analyses were conducted using linear mixed-effects models with interaction terms. Significance for the primary outcome measure was established at $p<0.05$. To reduce the false discovery rate, secondary and exploratory outcome measures were evaluated at $p<0.0125$. This alpha level was chosen by dividing $\alpha=0.05/4$ to adjust for the multiplicity of testing across the four broad domains of our secondary and exploratory outcomes (glucose response, insulin response, insulin resistance, and insulin secretion) within which results would be expected to be correlated. All data are presented as mean $\pm$ standard error of the mean (SEM), unless specified otherwise.

RESULTS

Flow of Study Participants

Supplementary Figure 1 shows the flow of study participants. Two hundred and thirty-eight participants were assessed for eligibility of whom 27 were randomized. Of these, 24 participants were included in the final analysis as two participants were unable to complete the trial due to work conflict and one participant was excluded from analysis due to fasting plasma glucose values at one or more study visits exceeding $\pm$ 2 mmol/L of their average value from all six study visits.

Participant Characteristics

Table 1 outlines the participant characteristics. 24 participants with type 2 diabetes (age, 66$\pm$1.2 years; BMI, 27.0$\pm$0.9 kg/m^2; diabetes duration, 11.3$\pm$1.7 years; HbA_{1c}, 50.0$\pm$1.3 mmol/mol [6.7$\pm$0.1%]) were analyzed. Diabetes was managed with diet alone (n=5), metformin (n=8), or metformin plus a second-line therapy (n=11). Second-line therapies included DPP-4 inhibitors (n=6), sulfonylureas (n=3), thiazolidinediones (n=1) and SGLT-2 inhibitors (n=1).

Primary Outcome Measure

Figure 1A and **Supplemental Figure 4A** show the effect of fructose at 0g (control), 5g, and 10g on the postprandial plasma glucose iAUC response to a 75g-OGTT. Pairwise comparisons showed that fructose at 5g and 10g did not have a significant effect on the plasma glucose iAUC response ($p>0.05$) compared with 0g (control). No significant linear or non-linear dose responses were identified ($p>0.05$).

Figure 1B and **Supplemental Figure 4B** show the effect of allulose at 0g (control), 5g, and 10g on the postprandial plasma glucose iAUC response to a 75g-OGTT. Pairwise comparisons showed that allulose at 10g significantly reduced the plasma glucose iAUC response to the 75g-OGTT by 8% compared with 0g (control) (717.4±38.3 vs 777.5±39.9 mmol·min/L, p=0.015), while the 5g dose was of borderline significance (p=0.051). A significant linear dose response gradient was shown between the reduction in plasma glucose iAUC and dose (p=0.016). No significant non-linear dose threshold was identified (p>0.05).

Secondary and Exploratory Outcome Measures

Figure 2A, Supplementary Table 1 and Supplemental Figures 5-7 show the effect of fructose at 0g (control), 5g, and 10g on the 75g-OGTT derived secondary and exploratory outcome measures. Pairwise comparisons did not show a significant effect of fructose and no significant linear or non-linear dose responses were identified for any of the secondary or exploratory outcome measures (p>0.0125).

Figure 2B, Supplemental Table 2 and Supplementary Figures 8-10 show the effect of allulose at 0g (control), 5g, and 10g on the 75g-OGTT derived secondary and exploratory outcome measures. Pairwise comparisons showed that allulose significantly reduced plasma glucose absolute mean (13.0±0.6 vs 13.6±0.5 mmol/L, p=0.002) and total AUC (1615.7±67.6 vs 1694.1±57.8 mmol·min/L, p=0.003) at 5g and plasma glucose absolute mean (12.9±0.5 vs 13.6±0.5 mmol/L, p=0.001), absolute (16.1±0.7 vs 17.5±0.6 mmol/L, p<0.001) and incremental (8.7±0.5 vs 9.8±0.5 mmol/L, p<0.001) C_{max} and total AUC (1607.7±59.3 vs 1694.1±57.8 mmol·min/L) at 10g compared with 0g (control) (P<0.0125). A significant linear dose response gradient was shown for plasma glucose absolute (p<0.0001) and incremental (p<0.0001) C_{max},

total AUC (p=0.002) and absolute mean (p=0.001). No significant non-linear dose thresholds were identified (p>0.0125).

Equivalence Assessment

Figure 3 shows results of the equivalence test comparing the effect of allulose with fructose on iAUC for plasma glucose. Although allulose showed statistically significant reductions compared with fructose at 5g (MD = -7.47% [90% CI: -13.02% to -1.93%]), 10g (MD = -7.36% [90% CI: -14.32% to -0.40%] and pooled doses (MD = -7.42% [90% CI: -11.91% to -2.92%]), these reductions were within the pre-specified equivalence margins of ±20%.

Subgroup Analyses

Supplementary Figure 11 and 12 show the sub-group analyses of the pooled effect of fructose and allulose, respectively, on plasma glucose iAUC compared to control (0g). Self-reported ethnicity was a significant effect modifier of the effect of fructose (p=0.02), and baseline 2h-plasma glucose (2hPG) during the 75g-OGTT (p=0.02) and type of background diabetes therapy (p=0.03) were significant effect modifiers of the effect of allulose.

Side effects

Most participants tolerated the treatments well. There was one report of nausea and one report of a slight headache following consumption of the 75g-OGTT + 10g fructose which subsided by the end of the study visit.

DISCUSSION

Summary of Findings

We conducted a double-blind, randomized, controlled, acute feeding equivalence trial of the effect of small 'catalytic' doses (5g and 10g) of fructose and allulose on postprandial blood glucose regulation in response to oral glucose based on a 75g-OGTT in individuals with well-controlled type 2 diabetes. The 10g dose of allulose resulted in a modest lowering in the postprandial blood glucose response to oral glucose with a linear dose-response gradient over 0g to 10g. There was no effect on measures of insulin resistance or secretion. The 5g and 10g doses of fructose did not have a significant effect on any outcome measures of postprandial blood glucose regulation. Although allulose significantly reduced the postprandial blood glucose response at 5g, 10g and pooled doses when compared with fructose, these reductions were within the pre-specified equivalence margins of $\pm$ 20%.

Findings in Context of Previous Literature

We failed to demonstrate the presence of a 'catalytic' effect with fructose in decreasing the postprandial blood glucose response to a glucose load. It was previously shown that 7.5g fructose significantly reduced the 3-h plasma glucose iAUC response to a 75g oral glucose load by 14% in individuals with type 2 diabetes [5]. We were unable to reproduce these findings with 5g and 10g fructose. Potential sources of discrepancy between the previous trial and our trial include: follow-up duration (3-h vs. 2-h), sample size (n=5 vs. n=24), handling of medications (discontinued 5 days prior to treatment vs. on the morning of the treatment), participant age (42 $\pm$ 5 vs. 66 $\pm$ 1.2 years), participant BMI (42 $\pm$ 4 vs. 27 $\pm$ 0.9 kg/m^2) and HbA_{1c} (8.5 $\pm$ 0.5% vs. 6.7 $\pm$ 0.1%). In a study conducted in 11 healthy participants, 7.5g fructose reduced the iAUC plasma glucose response by 19% to a 75g oral glucose challenge[6]. However, a follow-up study which assessed timing of fructose administration in 31 healthy participants failed to demonstrate postprandial blood glucose reduction when 10g fructose was consumed with an instant mashed

potato meal (50g available carbohydrate). Instead, postprandial blood glucose reductions of 25% and 27% were observed only when fructose was consumed 60 or 30 min prior to the meal load, respectively[39]. It could be possible that although we did not observe a 'catalytic' effect from fructose in our trial, fructose administration prior to, instead of with the 75g oral glucose challenge may reduce the postprandial blood glucose response in individuals with type 2 diabetes.

We were able to confirm a 'catalytic' effect of allulose in decreasing the postprandial blood glucose response to oral glucose, particularly in individuals with poorer glucose tolerance (75g-OGTT 2hPG $\geq$ 11.1 mmol/L). A study in 20 healthy subjects found that 5g and 7.5g allulose reduced the postprandial blood glucose response by ~22% and ~32%, and insulinemic response by ~28% and ~31%, respectively, to a 75g maltodextrin challenge[23]. In another separate study when 11 healthy participants consumed 5g allulose sweetened tea with a standard meal load, no significant differences were found in postprandial glucose and insulin responses when compared to consumption of the same meal load with 10mg aspartame sweetened tea[24]. However, in this same study when 15 participants with prediabetes were analyzed, 5g allulose-sweetened tea resulted in ~14% reduction in postprandial blood glucose response to the standard meal load compared to aspartame-sweetened tea.

Potential Mechanism of Action

The mechanism by which allulose reduces the postprandial blood glucose response to an oral glucose load is unclear. One possibility is enhanced glucose-stimulated insulin secretion by allulose. This mechanism was not supported by our data as allulose failed to show a significant

effect on plasma insulin iAUC responses, the insulin secretion index ($\Delta PI_{30-0}/\Delta PG_{30-0}$), or the ISSI-2.

Another possibility is reduced intestinal absorption of glucose in the presence of allulose. Glucose and allulose pass through different transporters (SGLT1 and GLUT2, respectively) as they move from the intestinal lumen to the apical membrane of the enterocyte. However, they utilize the same transporter (GLUT2) as they pass from the basolateral membrane of the enterocyte to the portal circulation[40]. It has been suggested that allulose may competitively inhibit the transport of glucose at the basolateral GLUT2 transporter. Support for this hypothesis is provided from experiments conducted in Caco-2 monolayer cell lines where addition of 30 mM allulose to 30 mM glucose reduced glucose permeability by 60% [41]. No studies have been conducted in humans to confirm this mechanism.

There has also been some suggestion that allulose may reduce the postprandial blood glucose response by enhancing hepatic glucose uptake. Hepatic glucokinase activity is decreased in some individuals with type 2 diabetes [42, 43]. Phosphorylation of glucose by glucokinase is a rate-determining step in hepatic glucose metabolism. Glucokinase is inhibited by glucokinase regulatory protein (GKRP), and this action is enhanced in the presence of fructose-6-phosphate. Under fasting conditions, hepatic glucokinase is localized primarily in the nucleus, where it is bound to the glucokinase regulatory protein (GKRP) and fructose-6-phosphate. In the postprandial state (presence of allulose and glucose), allulose is phosphorylated to allulose-1-phosphate by an enzyme called ketohexokinase. Allulose-1-phosphate competes with fructose-6-phosphate from GKRP. This enables the liberated and activated glucokinase to translocate from the nucleus to the cytosol where it can drive hepatic glucose uptake, promote glycogen synthesis, suppress hepatic glucose output and reduce plasma glucose levels[40]. In support of this

hypothesis, immunohistochemical analysis in allulose-fed rats have showed induction of glucokinase translocation from the nucleus to the cytoplasm and increased amount of hepatic glycogen content after glucose loading[22, 44]. No studies have been conducted in humans to confirm this mechanism.

Implications

Implications of these findings are that allulose may be a useful substitute for sugars, especially when consumed as part of high glycemic index carbohydrate foods. Allulose tastes ~70% as sweet as sucrose and contains 90% fewer calories. When consumed alone, allulose does not raise blood glucose and insulin levels in healthy individuals[23]. Our study along with a previous study in participants with prediabetes have shown that addition of small doses of allulose also helps to lower the postprandial blood glucose response to high glycemic index carbohydrate meals (i.e. 75g-OGTT or standard Japanese meal) by ~8-14%[24]. This decrease is modest when compared to an oral antihyperglycemic agent such as acarbose, which has shown reductions of ~31-58% on postprandial glycemia when administered with a meal load[45, 46].

Strengths and Limitations

Our acute trial had several strengths. These included a randomized double-blind controlled design, which provides the best protection against bias; a crossover design, which allows each participant to act as their own control reducing between-subject variation; a reliable estimate of fasting glucose and insulin based on the mean of 2 fasting samples at -30min and 0min; and a reliable estimate of the comparator based on the mean of two separate controls (0g).

Our acute trial also has several limitations. First, the two-hour duration of the OGTTs may not have been long enough to detect meaningful differences in postprandial glucose and insulin responses as individuals with type 2 diabetes typically return to baseline after 3 hours or longer[47-49]. Second, the trial was not designed to examine the mechanism(s) by which allulose reduced the postprandial blood glucose response to an oral glucose load. Third, although we did find a significant linear dose response for allulose, the doses examined may have been too few or insufficient to detect dose-response gradients or thresholds. Finally, the acute design of the trial creates uncertainty as to whether the reductions in the postprandial blood glucose response seen with allulose will manifest as sustainable improvements in glycemic control (i.e. HbA_{1c}) over the long-term.

In conclusion, we demonstrated that allulose, but not fructose, modestly reduced the postprandial blood glucose response to an oral glucose load showing a linear dose-response gradient over 0g to 10g in individuals with type 2 diabetes. There is a need for long-term randomized trials to confirm whether these acute reductions in postprandial blood glucose will lead to sustainable improvements in glycemic control.

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Duality of interest

CWCK has received research support from the Advanced Foods and Material Network, Agrifoods and Agriculture Canada, the Almond Board of California, the American Pistachio Growers, Barilla, the California Strawberry Commission, the Calorie Control Council, CIHR, the Canola Council of Canada, the Coca-Cola Company (investigator initiated, unrestricted grant), Hain Celestial, the International Tree Nut Council Nutrition Research and Education Foundation, Kellogg, Kraft, Loblaw Companies Ltd., Orafti, Pulse Canada, Saskatchewan Pulse Growers, Solae and Unilever. He has received travel funding, consultant fees and/or honoraria from Abbott Laboratories, the Almond Board of California, the American Peanut Council, the American Pistachio Growers, Barilla, Bayer, the Canola Council of Canada, the Coca-Cola Company, Danone, General Mills, the International Tree Nut Council Nutrition Research and Education Foundation, Kellogg, Loblaw Companies Ltd., the Nutrition Foundation of Italy, Oldways Preservation Trust, Orafti, Paramount Farms, the Peanut Institute, PepsiCo, Pulse Canada, Sabra Dipping Co., Saskatchewan Pulse Growers, Solae, Sun-Maid, Tate and Lyle, and Unilever. He is on the Dietary Guidelines Committee for the Diabetes Nutrition Study Group of the European Association for the Study of Diabetes and has served on the scientific advisory board for the Almond Board of California, the International Tree Nut Council, Oldways Preservation Trust, Paramount Farms and Pulse Canada. TMSW is a part owner and the President of Glycemic Index Laboratories, Inc, Toronto, Canada, and has authored several popular diet books on the glycemic index for which he has received royalties from Phillipa Sandall Publishing Services and CABI Publishers. He has received consultant fees, honoraria, travel funding, or research support from or served on the scientific advisory board for CIHR, Diabetes Canada, Dairy Farmers of Canada, McCain Foods, Temasek Polytechnic, Northwestern University, Royal Society of London, Glycemic Index Symbol program, CreaNutrition AG,

McMaster University, Canadian Society for Nutritional Sciences, National Sports and Conditioning Association, Faculty of Public Health and Nutrition—Autonomous University of Nuevo Leon, Diabetes and Nutrition Study Group (DNSG) of the European Association for the Study of Diabetes (EASD). JLS has received research support from the Canadian Institutes of health Research (CIHR), Diabetes Canada, PSI Foundation, Calorie Control Council, Banting and Best Diabetes Centre (BBDC), American Society for Nutrition (ASN), Dr. Pepper Snapple Group (investigator initiated, unrestricted donation), INC International Nut and Dried Fruit Council, and The Tate and Lyle Nutritional Research Fund at the University of Toronto. He has received speaker fees and/or honoraria from Diabetes Canada, Canadian Nutrition Society (CNS), University of Alabama at Birmingham, Abbott Laboratories, Canadian Sugar Institute, Dr. Pepper Snapple Group, The Coca-Cola Company, Dairy Farmers of Canada, Nutrition Foundation of Italy (NFI), C3 Collaborating for Health, WhiteWave Foods, Rippe Lifestyle, mdBriefcase, Alberta Milk, FoodMinds LLC, Memac Ogilvy & Mather LLC, PepsiCo, and Pulse Canada. He has ad hoc consulting arrangements with Winston & Strawn LLP, Perkins Coie LLP, and Tate & Lyle. He is a member of the European Fruit Juice Association Scientific Expert Panel. He is on the Clinical Practice Guidelines Expert Committees of Diabetes Canada, European Association for the study of Diabetes (EASD), and Canadian Cardiovascular Society (CCS), as well as an expert writing panel of the American Society for Nutrition (ASN). He serves as an unpaid scientific advisor for the Food, Nutrition, and Safety Program (FNSP) and the Technical Committee on Carbohydrates of the International Life Science Institute (ILSI) North America. He is a member of the International Carbohydrate Quality Consortium (ICQC), Executive Board Member of the Diabetes and Nutrition Study Group (DNSG) of the EASD, and Director of the Toronto 3D Knowledge Synthesis and Clinical Trials foundation. His wife is an

employee of Unilever Canada. No competing interests were declared by JCN, CRB, AG, TAK, EV, RN, SBM and LAL.

Contribution Statement

Each author had full access to all the data in the study and takes responsibility for the integrity of the data and the accuracy of the data analysis. Study concept and design: JLS. Acquisition, analysis and interpretation of the data: JCN, CRB, AG, TAK, EV, SBM, RN, CWCK, TMSW, LAL and JLS. Writing of the manuscript: JCN and JLS. Critical revision of the manuscript for important intellectual content: JCN, CRB, AG, TAK, EV, SBM, RN, CWCK, TMSW, LAL and JLS. Statistical analysis: JCN and TAK. Study guarantor: JLS.

REFERENCES

[1] Lustig RH, Schmidt LA, Brindis CD. Public health: The toxic truth about sugar. Nature. 2012; **482**: 27-29

[2] DiNicolantonio JJ, O'Keefe JH, Lucan SC. Added fructose: a principal driver of type 2 diabetes mellitus and its consequences. Mayo Clin Proc. 2015; **90**: 372-381

[3] Petersen KF, Laurent D, Yu C, Cline GW, Shulman GI. Stimulating effects of low-dose fructose on insulin-stimulated hepatic glycogen synthesis in humans. Diabetes. 2001; **50**: 1263-1268

[4] Hawkins M, Gabriely I, Wozniak R, Vilcu C, Shamoon H, Rossetti L. Fructose improves the ability of hyperglycemia per se to regulate glucose production in type 2 diabetes. Diabetes. 2002; **51**: 606-614

[5] Moore MC, Davis SN, Mann SL, Cherrington AD. Acute fructose administration improves oral glucose tolerance in adults with type 2 diabetes. Diabetes Care. 2001; **24**: 1882-1887

[6] Moore MC, Cherrington AD, Mann SL, Davis SN. Acute fructose administration decreases the glycemic response to an oral glucose tolerance test in normal adults. J Clin Endocrinol Metab. 2000; **85**: 4515-4519

[7] Sievenpiper JL, Chiavaroli L, de Souza RJ, *et al.* 'Catalytic' doses of fructose may benefit glycaemic control without harming cardiometabolic risk factors: a small meta-analysis of randomised controlled feeding trials. The British journal of nutrition. 2012; **108**: 418-423

[8] Cozma AI, Sievenpiper JL, de Souza RJ, *et al.* Effect of fructose on glycemic control in diabetes: a systematic review and meta-analysis of controlled feeding trials. Diabetes Care. 2012; **35**: 1611-1620

[9] Sievenpiper JL, de Souza RJ, Mirrahimi A, *et al.* Effect of fructose on body weight in controlled feeding trials: a systematic review and meta-analysis. Ann Intern Med. 2012; **156**: 291-304

[10] Chiavaroli L, de Souza RJ, Ha V, *et al.* Effect of Fructose on Established Lipid Targets: A Systematic Review and Meta-Analysis of Controlled Feeding Trials. J Am Heart Assoc. 2015; **4**: e001700

[11] Sievenpiper JL, Carleton AJ, Chatha S, *et al.* Heterogeneous effects of fructose on blood lipids in individuals with type 2 diabetes: systematic review and meta-analysis of experimental trials in humans. Diabetes Care. 2009; **32**: 1930-1937

[12] David Wang D, Sievenpiper JL, de Souza RJ, *et al.* Effect of fructose on postprandial triglycerides: a systematic review and meta-analysis of controlled feeding trials. Atherosclerosis. 2014; **232**: 125-133

[13] Wang DD, Sievenpiper JL, de Souza RJ, *et al.* The effects of fructose intake on serum uric acid vary among controlled dietary trials. The Journal of nutrition. 2012; **142**: 916-923

[14] Chiu S, Sievenpiper JL, de Souza RJ, *et al.* Effect of fructose on markers of non-alcoholic fatty liver disease (NAFLD): a systematic review and meta-analysis of controlled feeding trials. European journal of clinical nutrition. 2014; **68**: 416-423

[15] Oshima H, Kimura I, Izumori K. Psicose Contents in Various Food Products and its Origin. Food Science and Technology Research. 2006; **12**: 137-143

[16] GRAS Notice (GRN) for D-Psicose No. 693. U.S. Food and Drug Administration. URL: https://www.accessdata.fda.gov/scripts/fdcc/index.cfm?set=GRASNotices&id=693&sort=GRN_No&order=DESC&startrow=1&type=basic&search=psicose. 2017

[17] GRAS Notice (GRN) for D-Psicose No. 498. U.S. Food and Drug Administration. URL: https://www.accessdata.fda.gov/scripts/fdcc/index.cfm?set=GRASNotices&id=498&sort=GRN_No&order=DESC&startrow=1&type=basic&search=psicose. 2014

[18] GRAS Notice (GRN) for D-Psicose No. 400. U.S. Food and Drug Administration. URL: https://www.accessdata.fda.gov/scripts/fdcc/index.cfm?set=GRASNotices&id=400&sort=GRN_No&order=DESC&startrow=1&type=basic&search=psicose. 2012

[19] Detheux M, Vandercammen A, Van Schaftingen E. Effectors of the regulatory protein acting on liver glucokinase: a kinetic investigation. Eur J Biochem. 1991; **200**: 553-561

[20] Vandercammen A, Detheux M, Van Schaftingen E. Binding of sorbitol 6-phosphate and of fructose 1-phosphate to the regulatory protein of liver glucokinase. Biochem J. 1992; **286 (Pt 1)**: 253-256

[21] Toyoda Y, Mori, S., Umemura, N., Futamura, Y., Inoue, H., Hata, T., Miwa, I., Mura,o K., Nishiyama, A. and Tokuda, M. . Suppression of blood glucose levels by D-psicose in glucose tolerance test in diabetic rats. Japanese Pharmacology and Therapeutics. 2010; **38**: 261-269

[22] Hossain MA, Kitagaki S, Nakano D, *et al.* Rare sugar D-psicose improves insulin sensitivity and glucose tolerance in type 2 diabetes Otsuka Long-Evans Tokushima Fatty (OLETF) rats. Biochem Biophys Res Commun. 2011; **405**: 7-12

[23] Iida T, Kishimoto Y, Yoshikawa Y, *et al.* Acute D-psicose administration decreases the glycemic responses to an oral maltodextrin tolerance test in normal adults. J Nutr Sci Vitaminol (Tokyo). 2008; **54**: 511-514

[24] Hayashi N, Iida T, Yamada T, *et al.* Study on the postprandial blood glucose suppression effect of D-psicose in borderline diabetes and the safety of long-term ingestion by normal human subjects. Biosci Biotechnol Biochem. 2010; **74**: 510-519

[25] Urbaniak, G. C., & Plous, S. Research Randomizer (Version 4.0) [Computer software]. 2013. URL: http://www.randomizer.org/.

[26] World Health Organization Study Group. Diabetes Mellitus: Report of a WHO Study Group. Geneva, World Health Organization, 1985, p 99

[27] Wolever TMS. Determining the GI of Foods - Methodological Considerations. Glycaemic Index: A Physiological Classification of Dietary Carbohydrate. UK: CABI, 2006:32

[28] Peterson JI, Young DS. Evaluation of the hexokinase-glucose-6-phosphate dehydrogenase method of determination of glucose in urine. Analytical biochemistry. 1968; **23**: 301-316

[29] Schmidt FH. [Enzymatic determination of glucose and fructose simultaneously]. Klinische Wochenschrift. 1961; **39**: 1244-1247

[30] Livesey JH, Hodgkinson SC, Roud HR, Donald RA. Effect of time, temperature and freezing on the stability of immunoreactive LH, FSH, TSH, growth hormone, prolactin and insulin in plasma. Clinical biochemistry. 1980; **13**: 151-155

[31] Wolever TM, Jenkins DJ, Jenkins AL, Josse RG. The glycemic index: methodology and clinical implications. Am J Clin Nutr. 1991; **54**: 846-854

[32] Phillips DI, Clark PM, Hales CN, Osmond C. Understanding oral glucose tolerance: comparison of glucose or insulin measurements during the oral glucose tolerance test with specific measurements of insulin resistance and insulin secretion. Diabet Med. 1994; **11**: 286-292

[33] Matsuda M, DeFronzo RA. Insulin sensitivity indices obtained from oral glucose tolerance testing: comparison with the euglycemic insulin clamp. Diabetes Care. 1999; **22**: 1462-1470

[34] Kramer CK, Choi H, Zinman B, Retnakaran R. Glycemic variability in patients with early type 2 diabetes: the impact of improvement in beta-cell function. Diabetes Care. 2014; **37**: 1116-1123

[35] Sievenpiper JL, Jenkins DJ, Josse RG, Vuksan V. Dilution of the 75-g oral glucose tolerance test increases postprandial glycemia: implications for diagnostic criteria. CMAJ. 2000; **162**: 993-996

[36] Bureau of Nutritional Sciences, Food Directorate, Health Products and Food Branch, Health Canada. 2013. Draft Guidance Document on Food Health Claims Related to the Reduction in Post-Prandial Glycaemic Response. Retrieved from: https://chfa.ca/images/uploads/2012/08/Post-Prandial-Glycaemic-Response-Draft-Guidance.pdf.

[37] Williams JD. A Multiple Regression Approach to Multiple Comparisons for Comparing Several Treatments with a Control. Journal of experimental education. 1971; **39**: 93–96

[38] Detry MA, Ma Y. Analyzing Repeated Measurements Using Mixed Models. Jama. 2016; **315**: 407-408

[39] Heacock PM, Hertzler SR, Wolf BW. Fructose prefeeding reduces the glycemic response to a high-glycemic index, starchy food in humans. The Journal of nutrition. 2002; **132**: 2601-2604

[40] Hossain A, Yamaguchi F, Matsuo T, *et al.* Rare sugar D-allulose: Potential role and therapeutic monitoring in maintaining obesity and type 2 diabetes mellitus. Pharmacol Ther. 2015; **155**: 49-59

[41] Hishiike T, Ogawa M, Hayakawa S, *et al.* Transepithelial transports of rare sugar D-psicose in human intestine. Journal of agricultural and food chemistry. 2013; **61**: 7381-7386

[42] Caro JF, Triester S, Patel VK, Tapscott EB, Frazier NL, Dohm GL. Liver glucokinase: decreased activity in patients with type II diabetes. Horm Metab Res. 1995; **27**: 19-22

[43] Rizza RA. Pathogenesis of fasting and postprandial hyperglycemia in type 2 diabetes: implications for therapy. Diabetes. 2010; **59**: 2697-2707

[44] Shintani T, Yamada T, Hayashi N, *et al.* Rare Sugar Syrup Containing d-Allulose but Not High-Fructose Corn Syrup Maintains Glucose Tolerance and Insulin Sensitivity Partly via

Hepatic Glucokinase Translocation in Wistar Rats. Journal of agricultural and food chemistry. 2017; **65**: 2888-2894

[45] Shimabukuro M, Higa N, Chinen I, Yamakawa K, Takasu N. Effects of a single administration of acarbose on postprandial glucose excursion and endothelial dysfunction in type 2 diabetic patients: a randomized crossover study. J Clin Endocrinol Metab. 2006; **91**: 837-842

[46] Wachters-Hagedoorn RE, Priebe MG, Heimweg JA, *et al.* Low-dose acarbose does not delay digestion of starch but reduces its bioavailability. Diabet Med. 2007; **24**: 600-606

[47] Butler PC, Rizza RA. Contribution to postprandial hyperglycemia and effect on initial splanchnic glucose clearance of hepatic glucose cycling in glucose-intolerant or NIDDM patients. Diabetes. 1991; **40**: 73-81

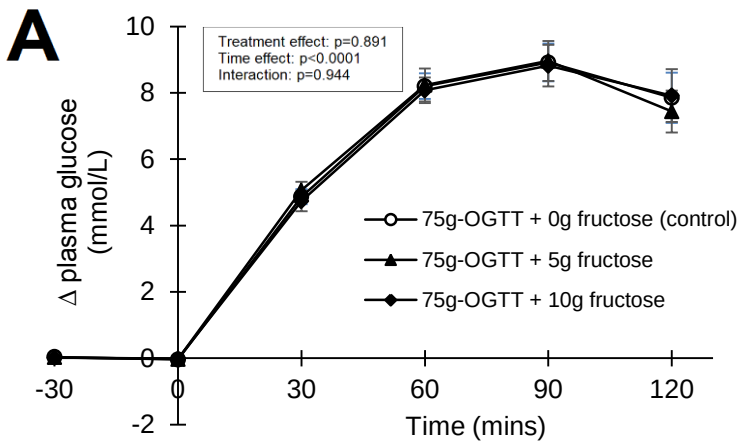
[48] Reaven GM, Chen Y-DI, M. Coulston A, *et al.* Insulin secretion and action in noninsulin-dependent diabetes mellitus: Is insulin resistance secondary to hypoinsulinemia? The American Journal of Medicine. 1983; **75**: 85-93

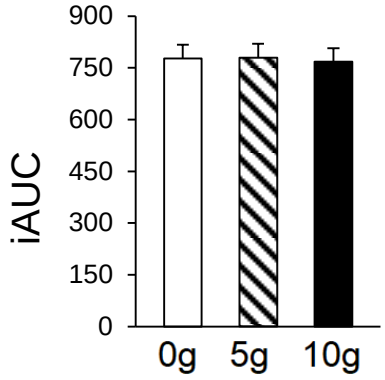
[49] Perley M, Kipnis DM. Plasma insulin responses to glucose and tolbutamide of normal weight and obese diabetic and nondiabetic subjects. Diabetes. 1966; **15**: 867-874

Table 1. Participant Characteristics

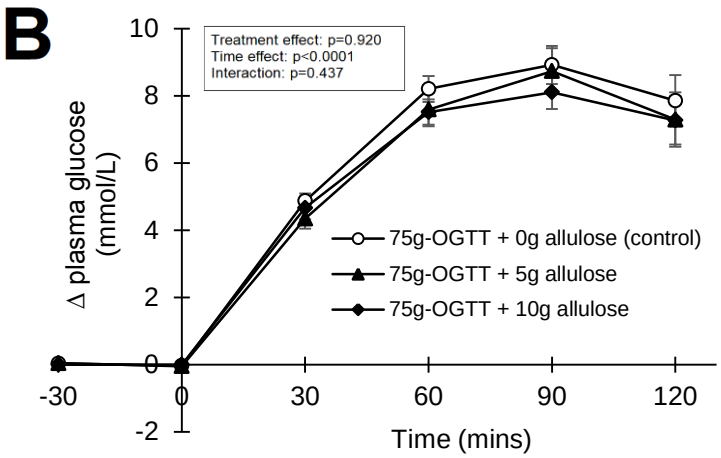
Characteristics	**Type 2 Diabetes**
Sex, M/F	12/12
Age, years	66 ± 1.2
Weight, kg	76.2 ± 3.7
BMI, kg/m^2	27.0 ± 0.9
Diabetes duration, years	11.3 ± 1.7
HbA_{1c}, %	6.7 ± 0.1
HbA_{1c}, mmol/mol	50.0 ± 1.3
Fasting blood glucose, mmol/L	6.9 ± 0.2
Diabetes therapy	
Diet alone	5
Metformin only	8
Metformin + DPP-4 inhibitor	6
Metformin + sulfonylurea	3
Metformin + thiazolidinedione	1
Metformin + SGLT-2 inhibitor	1

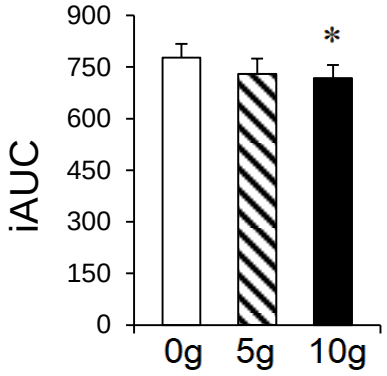
Data reported as mean ± SEM





	iAUC PG, mol·min/L
75g-OGTT + 0g fructose (control)	777.5 ± 39.9
75g-OGTT + 5g fructose	779.4 ± 40.7
75g-OGTT + 10g fructose	767.9 ± 39.3
0g vs. 5g, contrast (SE)	2.0 (25.9)
0g vs. 10g, contrast (SE)	-9.5 (25.9)
0g vs. 5g, p-value	0.940
0g vs. 10g, p-value	0.713

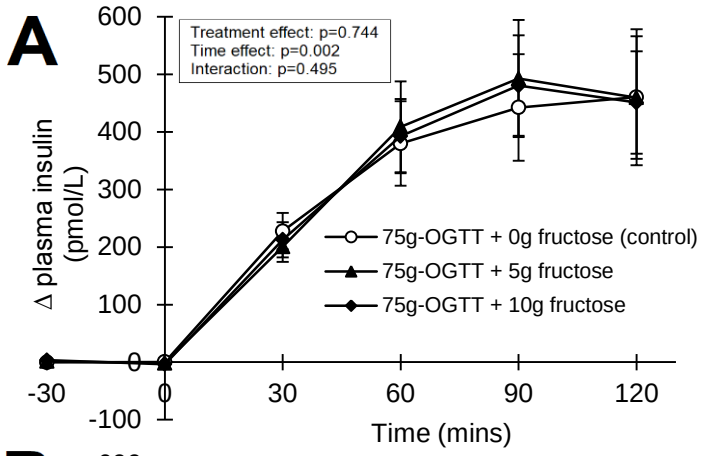


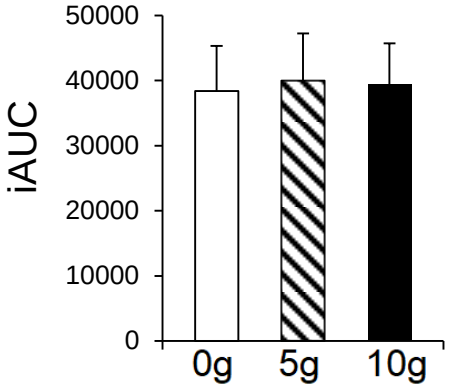


	iAUC PG, mol·min/L
75g-OGTT + 0g allulose (control)	777.5 ± 39.9
75g-OGTT + 5g allulose	729.3 ± 44.9
75g-OGTT + 10g allulose	717.4 ± 38.3
0g vs. 5g, contrast (SE)	-48.1 (24.7)
0g vs. 10g, contrast (SE)	-60.1 (24.7)
0g vs. 5g, p-value	0.051
0g vs. 10g, p-value	0.015

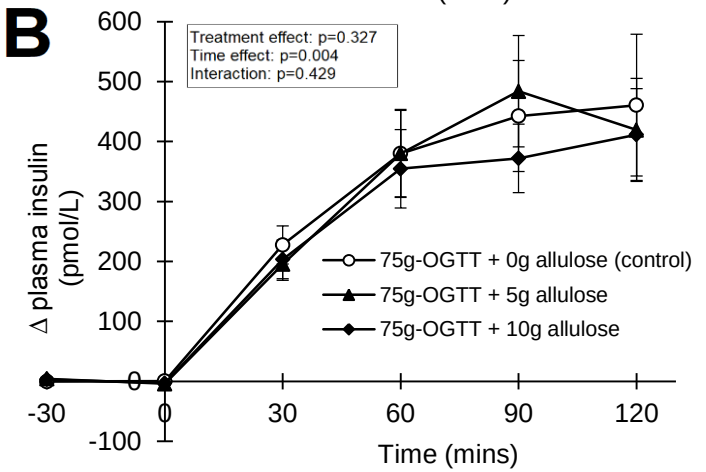
Figure 1. A) Effect of small doses of fructose on incremental change and incremental area under the curve (iAUC) for plasma glucose (PG) following consumption of 75g-OGTT + 0g fructose (control), 75g-OGTT + 5g fructose and 75g-OGTT + 10g fructose in 24 participants with type 2 diabetes. Mean fasting glucose concentrations were similar prior to consumption of 75g-OGTT (control), 75g-OGTT + 5g fructose and 75g-OGTT + 10g fructose at 7.64±0.25, 7.61±0.27 and 7.49±0.26 mmol/L, respectively ($p>0.05$, linear mixed-effects models). B) Effect of small doses of allulose on incremental change and incremental area under the curve (iAUC) for PG following consumption of 75g-OGTT + 0g allulose (control), 75g-OGTT + 5g allulose and 75g-OGTT + 10g allulose in 24

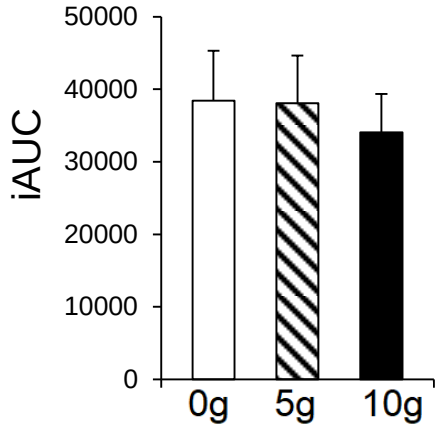
participants with type 2 diabetes. Mean fasting glucose concentrations were similar prior to consumption of 75g-OGTT (control), 75g-OGTT + 5g allulose and 75g-OGTT + 10g allulose at 7.64±0.25, 7.39±0.27 and 7.42±0.29 mmol/L, respectively ($p>0.05$, linear mixed-effects models). *represents a statistically significant difference ($p<0.05$, linear mixed-effects models) compared with control (0g). Data reported as mean ± SEM.





	iAUC PI, pmol·min/L
75g-OGTT + 0g fructose (control)	38405 ± 6917
75g-OGTT + 5g fructose	39963 ± 7285
75g-OGTT + 10g fructose	39345 ± 6353
0g vs. 5g, contrast (SE)	1557.7 (1921.8)
0g vs. 10g, contrast (SE)	940.2 (1921.8)
0g vs. 5g, p-value	0.542
0g vs. 10g, p-value	0.195





	iAUC PI, pmol·min/L
75g-OGTT + 0g allulose (control)	38405 ± 6917
75g-OGTT + 5g allulose	38054 ± 6599
75g-OGTT + 10g allulose	34065 ± 5292
0g vs. 5g, contrast (SE)	-350.9 (2256.7)
0g vs. 10g, contrast (SE)	-4340.2 (2256.7)
0g vs. 5g, p-value	0.962
0g vs. 10g, p-value	0.151

Figure 2. A) Effect of small doses of fructose on incremental change and incremental area under the curve (iAUC) for plasma insulin (PI) following consumption of 75g-OGTT + 0g fructose (control), 75g-OGTT + 5g fructose and 75g-OGTT + 10g fructose in 24 participants with type 2 diabetes. Mean fasting insulin concentrations were similar prior to consumption of 75g-OGTT + 0g fructose (control), 75g-OGTT + 5g fructose and 75g-OGTT + 10g fructose at 79.4±12.1, 82.7±12.9 and 81.4±10.0 pmol/L, respectively ($p>0.05$, linear mixed-effects models). **B)** Effect of small doses of allulose on incremental change and incremental area under the curve (iAUC) for PI following consumption of 75g-OGTT + 0g allulose (control), 75g-OGTT + 5g allulose and 75g-OGTT + 10g allulose in 24 participants with type 2 diabetes. Mean fasting insulin concentrations were similar prior to consumption of 75g-OGTT + 0g allulose (control), 75g-OGTT + 5g allulose and 75g-OGTT + 10g allulose at 79.4±12.1, 80.6±11.9 and 74.1±8.7, respectively ($p>0.05$, linear mixed-effects models). *represents a statistically significant difference ($p<0.0125$, linear mixed-effects models) compared with control (0g). p-values correspond to log-transformed data due to non-normal distribution of residuals. Data reported as mean ± SEM.

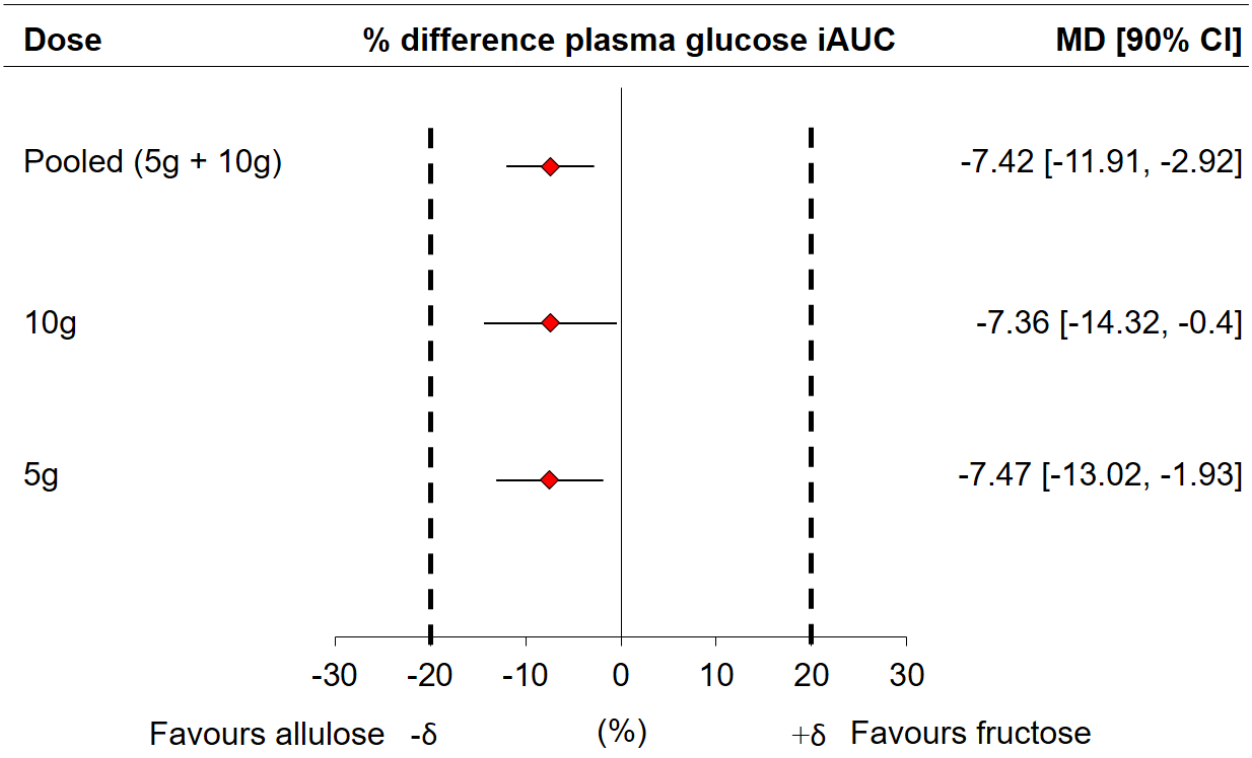


Figure 3. Equivalence assessment comparing the effect of allulose with fructose on plasma glucose incremental area under the curve (iAUC). % difference plasma glucose iAUC = $[(\text{allulose}_{\text{iAUCglucose}}/\text{control}_{\text{iAUCglucose}}) - (\text{fructose}_{\text{iAUCglucose}}/\text{control}_{\text{iAUCglucose}})]$ x 100%. Equivalence margins (+δ, - δ) were set at -20%, +20%. If the 90% CIs completely fell within the equivalence margins, then allulose was considered equivalent to fructose. If either the upper or lower bound of the 90% CI fell outside the equivalence margins, then the assessment was considered inconclusive. If the 90% CIs fell either completely above or completely below the equivalence margins, then allulose was considered inferior or superior to fructose, respectively.