Article

Acute Effect of Moderate Dose Fructose in Solid Foods on Triglyceride, Glucose and Uric Acid before and after a One-Month Moderate Sugar Feeding Period - A Randomised Controlled Trial

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Abstract: Fructose in beverages has adverse effects on lipids, glucose and insulin sensitivity after acute and chronic ingestion. There is limited data showing that chronic consumption of fructose in solid foods has harmful effects. We hypothesized that a moderate amount of fructose compared with sucrose in solid food consumed for a month would not adversely influence fasting or postprandial lipids and glucose after an acute fat and carbohydrate load. Twenty-five men and women with prediabetes and/or obesity and overweight consumed in random order two acute test meals of muffins sweetened with either fructose or sucrose, followed by 4-week chronic consumption of 42g/day of either fructose or sucrose in low fat muffins after which the 2 meal tests were repeated. Subjects were randomised to sugar type in the chronic feeding period. Sugar type had no effect on the incremental area under the curve for triglyceride or uric acid at either time point (P=0.4 and P=0.9). There was no overall difference between meal tests at baseline and after 1 month and no effect of consuming sucrose or fructose muffins for 1 month. Fasting triglyceride increased after chronic consumption of fructose by 0.31±0.37 mmol/L compared with sucrose in people with IFG/IGT only (P=0.004). Fructose at a moderate intake of <10% of energy in solid food has no different effects on postprandial triglyceride and uric acid compared with sucrose although fasting triglyceride was increased in people with IFG/IGT after 1 month of fructose muffins suggesting the need for caution.

Keywords: triglyceride; uric acid; glucose; fructose; sucrose; solid

1. Introduction

Fructose containing beverages sweetened with either sucrose or high fructose corn syrup have been associated with an increased incidence of obesity, type 2 diabetes and cardiovascular disease in longitudinal cohort studies [1-3]. Short term hypercaloric feeding studies have shown that fructose increases visceral fat, insulin resistance and hypertriglyceridemia more than a similar amount of glucose [4]. However, in a meta-analysis of isocaloric-feeding studies 7 days or longer in duration in which weight gain did not occur fructose did not have any differential effects compared to other forms of carbohydrate [5]. In 2 hypercaloric studies in this meta-analysis fructose increased triglyceride compared with other carbohydrates. Other researchers have shown that hypercaloric feeding of fructose does not increase liver or muscle fat or enhance insulin resistance in liver, muscle or adipose tissue compared with glucose [6,7]. Acute studies feeding fructose as part of a fat tolerance test [8,9] or as an addition to a mixed meal increased postprandial triglyceride levels [10-12] although some studies were negative [13,14] with no differences between fructose and glucose. Despite these potentially negative attributes of fructose, substitution for starch in type 2 diabetes lowers HbA1c and fasting glucose when the amount is <40g/day [15]. This occurs because

fructose stimulates glucokinase regulatory protein -1 [16] and doubles hepatic glucose content and enhances glycogen synthesis fourfold [17] leading to a 14% lower glucose response and a 21% lower insulin response in an OGTT with an added 7.5g fructose. Chronic fructose feeding in beverages at 25% of energy for 10 weeks increases fasting and all-day uric acid profiles in overweight and obese men and women compared with isocaloric glucose feeding [18].

It is possible that use of fructose may be beneficial in people with insulin resistance or prediabetes by reducing glucose and insulin without elevating triglyceride providing it is used in solid meals such as biscuits, cakes which are more satiating than beverages (in which all the fructose research has been conducted so far) and its intake is restricted to about 40g/day (or less than about 8% of energy). The American Diabetes Association recommended that free fructose be kept below 12% of energy to minimise triglyceride elevation [19]. Non-caloric sweeteners are widely used in beverages but little used in solid foods so there is scope for investigation of replacement of sucrose with fructose to minimize the glycemic and insulinemic response to sucrose. Optimally such foods should not be consumed by people at risk of diabetes, but the reality is that individuals continue to consume sweet foods so alternative strategies are necessary. Non-caloric sweeteners are currently under scrutiny because of their association with type 2 diabetes [1] and with enhanced glucose absorption in healthy volunteers [20]. In this study, we aimed to assess the metabolic responses to muffins containing sucrose or fructose before and after a one-month chronic consumption period in which muffins are added to the normal diet. We this aimed to see if fructose could substitute for sucrose in people with obesity and prediabetes without adverse metabolic effects. To our knowledge no study has previously used fructose solely in a solid form and compared it with sucrose and not glucose which is rarely used as a sweetener.

2. Materials and Methods

Participants

Selection criteria.

Participants were either overweight or obese (BMI >25). People with impaired fasting glucose (IFG glucose>5.5 mmol/L) or impaired glucose tolerance (IGT 2-hour glucose 8-11.1 mmol/L) after 75g oral glucose tolerance testing with finger prick glucose values were recruited from previous acute studies at the University of South Australia over the last 3 years. Participants with frank diabetes were excluded. Subjects on any medication were excluded.

Study plan

Participants attended the Sansom clinic at the University of South Australia from February to June 2016 for baseline visits on 2 mornings one week apart and were randomised to consume after an 8hr fast, 2 muffins over a period of 15 minutes (muffin S - sucrose one week, followed by muffin F - fructose the following week or vice versa). Subjects were asked to avoid alcohol and strenuous exercise in the 24h preceding the tests. Following the baseline tests participants were asked to consume two low fat muffins/day for 4 weeks before repeating the acute studies (again in random order) on 2 mornings one week apart. They were randomly allocated to consume either sucrose or fructose muffins for this month using a computerised randomisation program by a technician not involved in measurement or analysis of results. The research personnel enrolling the participants and providing the muffins were unaware of the muffin code as were the participants.

At each of the visits, before and following the consumption of the test muffins, we took venous blood samples at regular interval (every 30mins for 180 minutes) and measured glucose, triglyceride and uric acid levels on a Konelab using standard commercial kits.

Composition of muffins

Muffins were cooked in the research kitchens at the University of South Australia. The muffins for the acute studies (before and after the chronic phase) each contained 21g sucrose or fructose and 21g of polyunsaturated fat while the muffins for the chronic phase contained the same amount of sugar but had a lower amount of fat at 11g per muffin to minimise the energy load. The two muffins/day for the chronic phase represented a total of 17-25% of daily energy intake depending on gender with the sugar at 6-8% of energy. No instructions were given about replacement of foods with muffins and it is possible energy intake could have increased by 17-25% for one month and induced measurable weight gain. No measurement of energy or sugar intake was made before or during the intervention to endure the intervention was as free-living as possible.

Ethics.

The protocol was approved by the Human Ethics Committee of Australia and all volunteers gave written informed consent. The trial was registered by the Australian New Zealand Clinical Trials Registry (ANZCTR www.anzctr.org.au/) ACTRN12618000125224
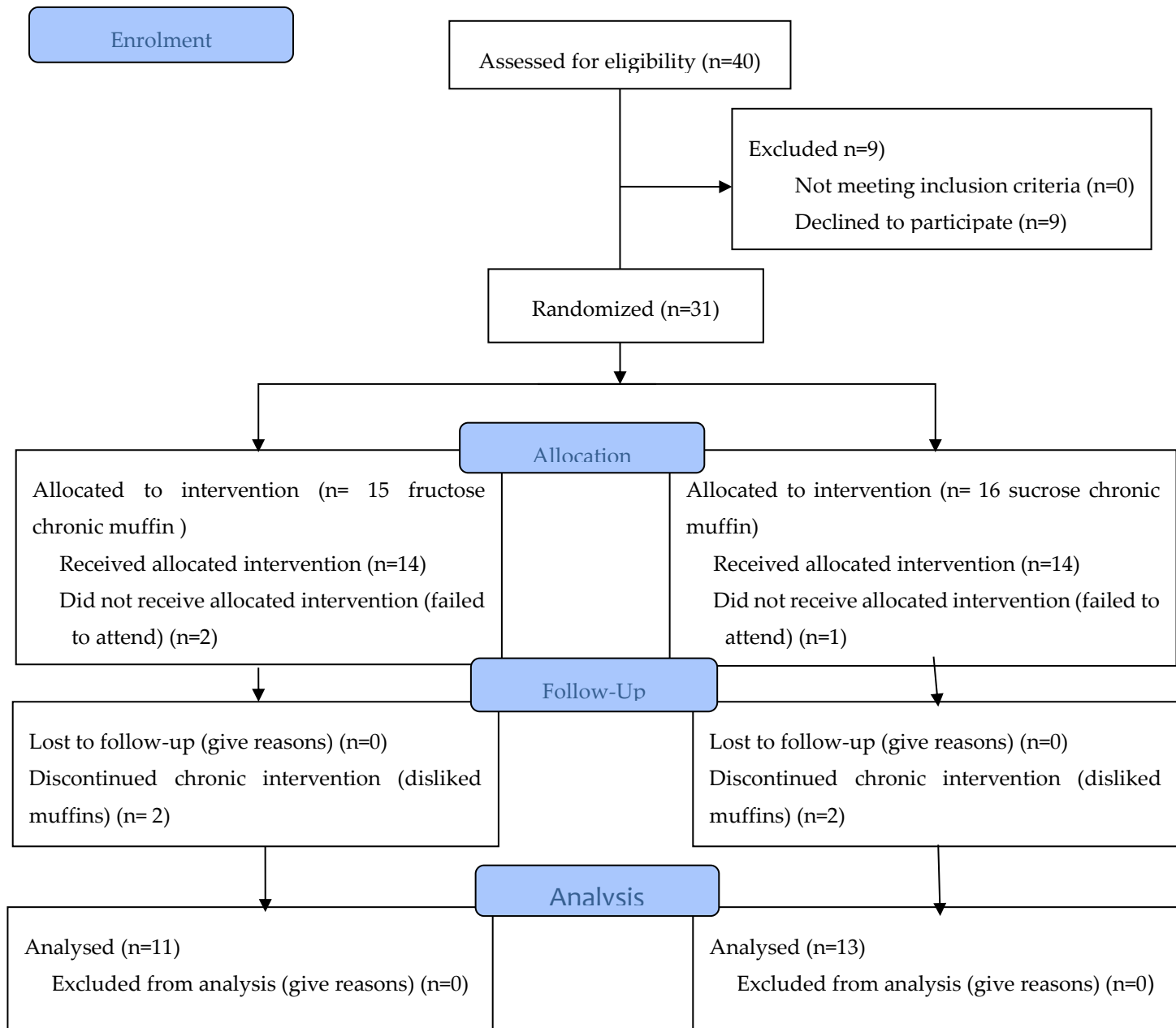
Statistics

All data shown are mean plus standard deviation (SD). Data was analysed by repeated measures ANOVA of iAUC (incremental area under curve) of triglyceride and uric acid with type of chronic sugar as a between subject factor and weight as a covariate in a secondary analysis.   Statistics were obtained for acute sugar type separately at baseline and at 1 month, and month by acute sugar in a final model which also examined the between subject factor of chronic sugar type. Data were significant if P was <0.05. No adjustment was made for multiple tests. Post-hoc analysis was performed contrasting people with IFG/IGT and those without and weight was included as a covariate in secondary analysis. The data analyst was blinded to the code for acute and chronic sugar until analysis was completed. Twenty-four subjects were required to complete the study to detect a difference in triglyceride iAUC of 30% (80% power, P<0.05, SD of difference 1.2 mmol/L.3h). The primary endpoints were the contrasts between sugars for incremental triglyceride AUC and uric acid AUC at baseline and 1 month with secondary endpoints of fasting triglyceride levels at 1 month as well as sugar, sugar by time and sugar by time by month for each individual time point value for triglyceride, glucose and uric acid. For the secondary endpoint the study had enough power to detect a 35% difference in fasting triglyceride between sugars with a SD of 30%. There was no statistical difference in characteristics between the 40 eligible for the study and the 24 who completed and whose data was analysed.

Results

Thirty-one volunteers were enrolled and 3 failed to commence. The CONSORT Flow Diagram is presented in Figure 1.

Figure 1 CONSORT Flow Diagram

Enrolment

Assessed for eligibility (n=40)

Excluded n=9)
    Not meeting inclusion criteria (n=0)
    Declined to participate (n=9)

Randomized (n=31)

Allocation

Allocated to intervention (n= 15 fructose chronic muffin)
    Received allocated intervention (n=14)
    Did not receive allocated intervention (failed to attend) (n=2)

Allocated to intervention (n= 16 sucrose chronic muffin)
    Received allocated intervention (n=14)
    Did not receive allocated intervention (failed to attend) (n=1)

Follow-Up

Lost to follow-up (give reasons) (n=0)
Discontinued chronic intervention (disliked muffins) (n= 2)

Lost to follow-up (give reasons) (n=0)
Discontinued chronic intervention (disliked muffins) (n=2)

Analysis

Analysed (n=11)
    Excluded from analysis (give reasons) (n=0)

Analysed (n=13)
    Excluded from analysis (give reasons) (n=0)

Fifteen men and 13 women commenced the study (Table 1). Fifteen participants had impaired fasting glucose (>5.5 mmol/L) or impaired glucose tolerance and 3 had both; 10 had normal glucose tolerance and 17 were obese. The 28 participants who commenced had an average BMI of 32.3 kg/m2, age of 44.7 years, a fasting glucose of 5.3 (SD 0.89) mmol/L and a 2-hour glucose of 6.6 (SD 1.8) mmol/L. There was no statistical difference between those allocated to fructose muffins and those allocated to sucrose muffins. Four dropped out after the first 2 acute meal studies. Seventeen people with IFG/IGT and 7 with normal glucose tolerance completed the study. Weight gain over 1 month in the 24 (11 fructose, 13 sucrose) who completed this section was 0.53kg (P=0.2) with 6 of this group losing weight. Volunteers could not distinguish the type of sugar in the muffins.

Table 1. Baseline Characteristics of completers

	Normal Glucose Tolerance	Impaired Glucose tolerance	Chronic sugar Fructose	Chronic Sugar Sucrose
IFG/IGT		17	8	9
Normal	7		3	4
Gender	3M, 4F	10M, 7F	6M,5F	5M, 8F
Age	48.5±18.2	53.1±16.5	55±18	47±15
BMI	31.2±3.3	32.1±4.9	31.3±4.9	32.5±3.9
Fasting Glucose	5.0±0.5	6.0±0.8	5.6±0.8	5.8±0.9
2hr Glucose	6.1±1.2	7.9±1.7	7.8±2.0	7.0±1.5
Fasting TG	1.14±0.26	1.47±0.92	1.37±0.92	1.32±0.64

Triglyceride

Fasting triglyceride and weight before and after 4 weeks of muffin consumption are presented in Table 2.

Table 2. Fasting triglyceride and weight before and after 4 weeks of muffin consumption

	Fasting Triglyceride mmol/L				Weight Kg			
	Chronic Fructose		Chronic Sucrose		Chronic Fructose		Chronic Sucrose	
	Baseline	1 month	Baseline	1 month	Baseline	1 month	Baseline	1month
IFG/IGT	1.55±1.03 (n=8)	1.95±1.12 (n=8)	1.51± 1.04 (n=9)	1.59±0.97 (n=9)	85.2±13.5 (n=8)	86.2±13.7 (n=8)	97.1±14.4 (n=9)	97.8±14.3 (n=9)
Normal	1.03±0.18 (n=3)	1.09±0.26 (n=3)	1.31±0.52 (n=4)	1.11±0.34 (n=4)	86.8±14.5 (n=3)	87.3±14.5 (n=3)	95.6±8.7 (n=4)	94.2±9.3 (n=4)

Data was analyzed using repeated measures ANOVA with month (0,1), sugar (fructose or sucrose) and IFG/IGT (yes/no) as factors. There was a strong interaction between IFG/IGT and month (P=0.001) and a weak interaction between month and chronic sugar (P=0.036). Analysing only those with IFG/IGT (n=17) there was a strong effect of month (P<0.001) and an interaction between month and chronic sugar (P=0.004). Change in weight over the month was a significant covariate (p=0.014) but the effect of weight gain was seen only in those with IFG/IGT (an increase of 0.8kg) where there was no difference seen between the chronic sugars. There was no effect seen in those without IFG/IGT (n=8)

Following the acute meal tests there was no difference between fructose and sucrose -containing muffins in triglyceride iAUC with an increase in triglyceride from 1.46 mmol/L at baseline to 2.53 mmol/L at 180 minutes after fructose and an increase from 1.43 mmol/L to 2.25 mmol/L at 180 minutes after sucrose (P=0.14 for iAUC triglyceride). There was a time by sugar interaction with a slightly delayed response after fructose with a peak at 180 minutes while after sucrose it was higher from 30 minutes onwards with a peak at 150 minutes followed by a slight fall at 180 minutes (P<0.001 time by sugar) (Figure 2). Average triglyceride was 1.83 mmol/l after fructose and 1.89 mmol/l after sucrose (NS).

These acute tests were repeated after 1 month of consumption of muffins containing either sucrose or fructose and iAUC triglyceride was not different between the sugars (P=0.5).   The same pattern was seen with fructose increasing from 1.55 mmol/L to 2.71 mmol/L (mean 1.97 mmol/l) and sucrose increasing from 1.58 mmol/L to 2.49 mmol/L (mean 2.02 mmol/l).

With all data points from month 0 and month 1 were analysed together the main effect of acute sugar on iAUC triglyceride was not different (P=0.3) and there was no effect on month on iAUC (p=0.5) and no effect of   chronic sugar type (p=0.5). People consuming sucrose muffins for a month had an overall mean triglyceride of 2.0 mmol/L (across all time points) at month 0 and 1.99 at month 1. With the fructose chronic muffins overall triglyceride was 1.70 mmol/L at month 0 and 1.87 mmol/L at month 1(NS).

For the secondary endpoint of fasting triglyceride overall there was no significant effect of chronic sugar. However, when IFG/IGT was added as a factor a strong interaction with month (P=0.001) was present. Analysing only those with IFG/IGT (n=17) there was a strong effect of month (P<0.001) and an interaction between month and chronic sugar (P=0.004). Fasting triglyceride was 1.51 mmol/L and 1.59mmol/L for chronic sucrose and 1.55mmol/L and 1.95 mmol/L for chronic fructose at month 0 and month 1 respectively (Table 2). The difference in this group in the effect of chronic sugar was an increase over the month of 0.40±0.21 mmol/L for fructose and 0.09±0.17 mmol/L for sucrose with a difference between the two sugars of 0.31±0.37 mmol/L (95% CI 0.11-0.51).   Change in weight over a month was a significant covariate (P=0.014) but weight gain was seen only in those with IFG/IGT (an increase of 0.8kg) but there was no significant difference in weight gain between the chronic sugars. There was no effect on weight seen in those without IFG/IGT (n=8).

Total cholesterol (0.33 mmol/L), LDL cholesterol (0. 24 mmol/l) and HDL cholesterol (0.08 mmol/L) increased significantly over the 1 month feeding period with no differences between muffin types.

Uric acid

Uric acid decreased after each muffin acutely (P<0.001) and iAUC was not different between the two sugar types (p=0.9) at either baseline or 1 month with no differences between baseline and 1 month. (Figure 3).

Blood glucose

As expected, the glucose profiles were quite different between the two muffin types with a time by sugar P value of P<0.001 for iAUC over 180 min. (Figure 4). The mean difference in glucose was 0.41 mmol/L. Blood glucose increased from 5.5 to 5.9 mmol/L after fructose muffins and from 5.6 mmol/L to 6.8 mmol/L after sucrose muffins. On repeat testing after 1 month of muffin consumption glucose after fructose muffins increased from 5.5 mmol/L to 6.4 mmol/L and from 5.5 mmol/L to 6.6 mmol/L after sucrose muffins with an overall significance between sugars of P<0.001 (iAUC over 180 min) with no significant difference between the baseline and one-month tests (P=0.14). Fasting glucose was not different after 1 month of muffin consumption.
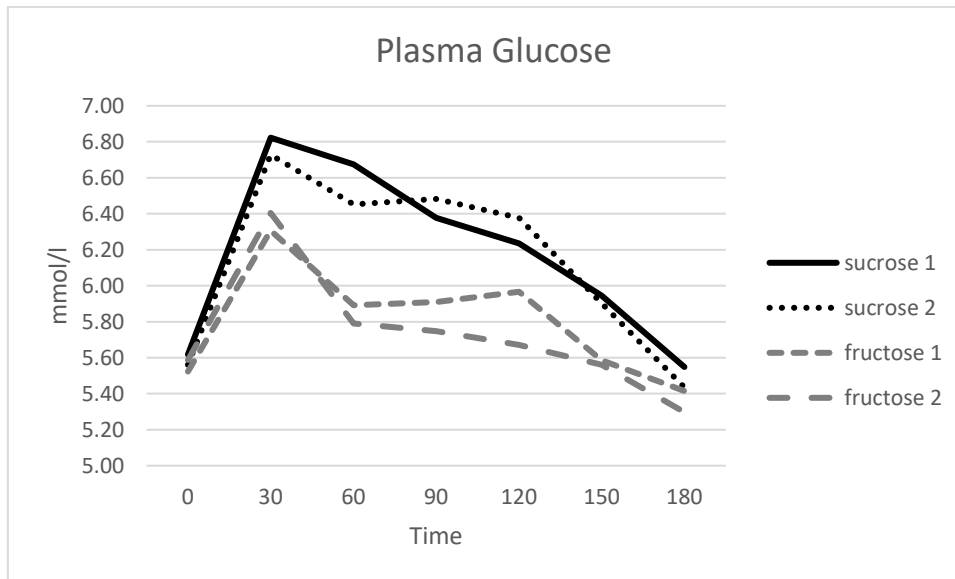
Figure 2

Serum Triglyceride
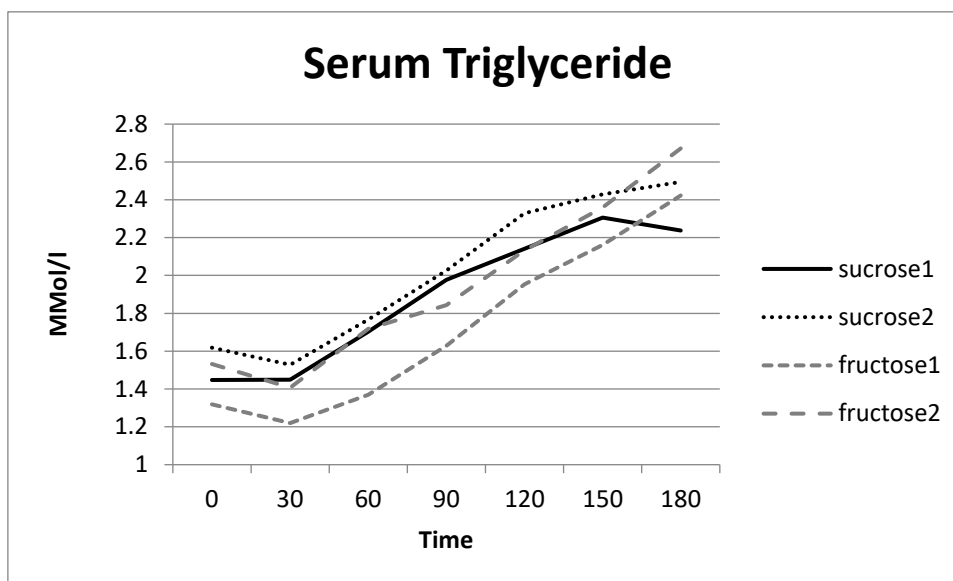
Effect of acute feeding of fructose and glucose on serum triglyceride before (1) and after (2) 4 weeks of 42g/day of sugar. Analysed by repeated measures ANOVA of iAUC with acute sugar and month as repeated measures and chronic sugar as a between subject factor.   Fructose 1 and sucrose 1: (P=0.14) . Fructose 2 and sucrose 2. (p=0.5).   Overall P=0.3 for acute sugar, P=0.2 for acute sugar by month, P=0.5 for acute sugar by month by chronic sugar. N=24 for each line

Figure 3
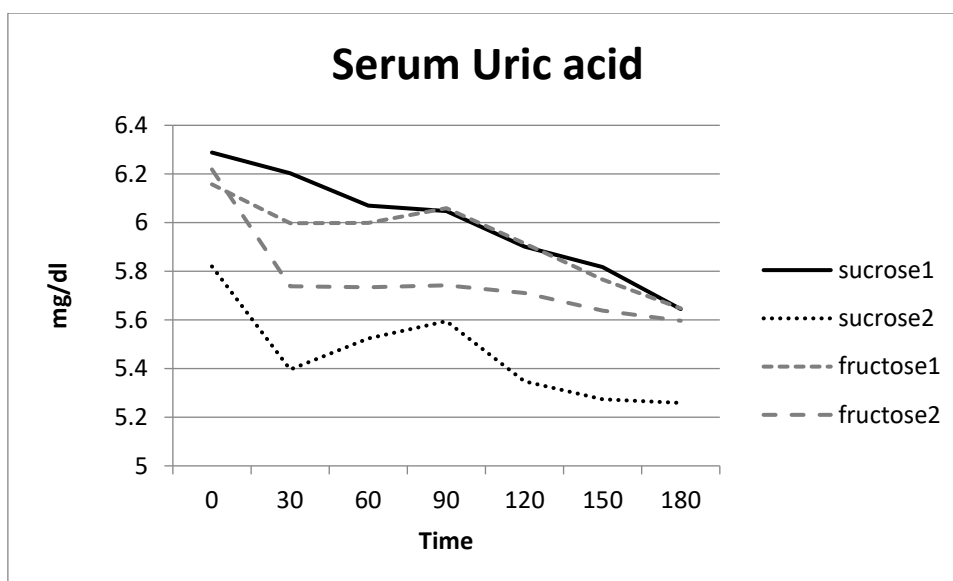
Serum Uric acid

Effect of acute feeding of fructose and glucose on serum uric acid before and after 4 weeks of 42g/day of sugar. Analysed by repeated measures ANOVA of iAUC with acute sugar and month as repeated measures and chronic sugar as a between subject factor. No effect of   acute sugar or acute sugar by month (p=0.6 to 0.9). Interaction between acute sugar, month and   chronic sugar feeding (p=0.09).n=24 for each line

Figure 4

Effect of acute feeding of fructose and glucose on plasma glucose before and after 4 weeks of 42g/day of sugar. Analysed by repeated measures ANOVA with acute sugar, time and month as repeated measures and chronic sugar as a between subject factor. Fructose 1 and sucrose 1. $P<0.001$ for main effect of sugar. $P<0.001$ for time by sugar. Fructose 2 and sucrose 2. Main effect of sugar $P=0.001$, Time by sugar $P=0.007$. No effect of chronic sugar feeding ($P=0.5$). Overall time by sugar $P<0.001$. N=24 for each line

Discussion

In this group of overweight/obese individuals 42g of fructose as an intrinsic part of a high fat, solid meal containing starch and protein had no effect on plasma triglyceride over 3 hours compared with sucrose.   Fructose led to a lower glucose level compared with the sucrose-sweetened muffin meal. This may have advantages in this population at risk of progression to diabetes by minimising the demand for insulin without adverse effects on lipids. These results contrast with other acute studies of fructose using liquid meals containing sugar and fat only [8,9]. These studies have focused on determining whether fructose is worse than glucose at exacerbating metabolic abnormalities, but these studies have no practical outcomes as glucose is rarely used as a caloric sweetener whereas fructose has been used in the past in people with type 2 diabetes as an alternative to sucrose. One study [9] with lean healthy subjects showed that fructose at a dose of 0.75g/kg combined with fat at a dose of 0.5g/kg in beverage form but with no starch or protein increased postprandial triglyceride from 120-360 minutes after the meal with a maximum difference of 0.8 mmol/L at 300 and 360 minutes compared with the glucose meal. Our fat load was much lower at an average of 0.22 g/kg (varying from 0.18 to 0.28) but this represents a normal large food fat load.   Singleton et al [13] found that both glucose and fructose augmented the triglyceride response to a fat load compared to no added sugar, but no differences were seen between the two sugars.

When a sugar containing beverage is consumed with a solid meal fructose augments plasma triglyceride compared with glucose [10,11]. Teff et al [11] compared the effects of a high fructose or a high glucose beverage added to meals over 24 h in 12 normal weight women. Although the total nutrient intake was normal at 55% carbohydrate, 30% fat and 15% protein, 30% of energy came from the beverages with an average intake of free sugars over the day of over 130g. With this large amount of fructose serum triglyceride increased by 30-60% compared with glucose at the same time point with a 35% increase in fasting triglyceride the next day. However maximum triglyceride levels achieved were still low at 1.2 mmol/L as they were normal weight women without disturbed glucose homeostasis.   Similar but greater effects were seen in overweight men and women [12].

Previously we performed a study in healthy lean subjects examining 55g of fructose in solid muffins containing protein and fat and found no difference between fructose and sucrose or sucralose in triglyceride levels over 5 hours [21]. Both glucose and insulin levels were lower with fructose. In this study despite the presence of pre-diabetes or obesity there was still no difference between fructose and sucrose muffins on postprandial triglyceride levels up to 3 hours and glucose was lower as expected with the fructose muffin.   This data is very similar to a recent meta-analysis [22]. It is possible greater effects could be seen on triglyceride levels after 5 hours.

We asked volunteers to consume 2 muffins/day containing 42g of fructose or sucrose for one month and then repeated the acute tests. Fasting triglyceride and glucose overall were not altered by a month of muffin consumption and the repeat acute response was the same as the first one regardless of chronic muffin type. However, in people with IFG/IGT fasting triglyceride at 1 month was increased with a moderate difference between sucrose muffins and fructose muffins of 0.36 mmol/L but numbers were small in each group.   Body weight overall increased non-significantly overall by 0.5 kg which is similar to the Stanhope study [4] in which a weight gain of 1.8kg for the glucose group and 1.4kg for the fructose group was seen after 10 weeks with a larger daily sugar load. However, in the IFG/IGT group body weight increased significantly by 0.8kg with no differences between chronic sugar types.   The postprandial TG peak in the Stanhope study [4] was 1,5-2 times greater with fructose than with glucose as was fasting apoB and fasting LDL. However fasting TG increased only after glucose and not after fructose which contrasts with our study. Fasting glucose and insulin increased 4-fold from baseline with fructose compared with glucose while AUC glucose and insulin in the OGTT doubled, consistent with increased insulin resistance. In our study we saw no changes in fasting glucose, although a meta-analysis suggested small doses of fructose could lower fasting glucose [19]; a more recent meta-analysis of 11 chronic studies (2-10 weeks) with 14 treatment arms found no differences in fasting glucose overall or with fructose substituted for sucrose [23]. Our results in people with IFG/IGT who experienced weight gain with chronic muffin feeding partially agree with the systematic review and meta-analysis of Chiavaroli et al [5] who found fructose only had adverse effects on lipids when fed at an energy level of 21-35% with energy in excess of requirements. Isocaloric substitution of glucose with fructose in chronic studies has no effect on fasting triglyceride while substitution for sucrose lowers fasting triglyceride but there were only 3 studies in this latter group. Overall triglyceride was lowered by 0.08 mmol/L [22]while in isocaloric acute studies fructose was not different to other carbohydrates in its effect on postprandial triglyceride [22] although in studies that follow TG levels over 24 hours the level tends to increase with fructose later in the day.

Teff et al [12] found no effect of either glucose or fructose containing drinks with food on uric acid over 23 hr in obese men and women while Cai et al [24] found that 75g of fructose alone increased uric acid over 3 hours compared with glucose.   Le et al [25] demonstrated a similar effect with 70g of high fructose corn syrup over 6 hours as did Stanhope et al [26] over 24 h with beverages contributing 17.5% and 25% of energy as fructose fed for 2 weeks.   In this study beverages contributing only 10% of energy elevated postprandial triglyceride while the two higher doses increased fasting LDL cholesterol, apoB, non-HDL cholesterol. Fasting uric acid was increased only if fed in a 35% of energy excess [27] although this paper has been criticised [28].

Conclusions.

Fructose at a moderate intake of about 6-8% of energy as part of a solid meal has no adverse effects nor any beneficial effects on postprandial triglyceride and uric acid over 3 hours at baseline or after a 4-week feeding period in which no overall weight gain occurred. People with IFG/IGT have a moderate increase in fasting triglyceride after 1 month of eating fructose muffins and gaining weight but this needs confirmation with larger numbers, but caution in using this amount of fructose chronically is required in this particular group.

Limitations

Limitations of this study is that we did not extend our acute studies to 5-6 hours as differences may have appeared later and we did not compare fructose in solid foods to fructose in liquid form, nor did we perform acute meal tests without sugars. We did not assess background dietary intake which may have been differentially altered by the two muffin types. The conclusions of this study are limited to a moderate intake of fructose and may not apply to much larger amounts for longer periods of time.

Acknowledgements

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