Vinegar reduces postprandial hyperglycaemia in patients with type 2 diabetes when added to a high, but not to a low, glycaemic index meal.

Stavros Liatis, Sofia Grammatikou, Kalliopi-Anna Poulia, Despoina Perrea, Konstantinos Makrilakis, Evanthia Diakoumopoulou, Nl Katsilambros

TITLE PAGE

Vinegar reduces postprandial hyperglycaemia in patients with type 2 diabetes when added to a high, but not to a low, glycaemic index meal.

RUNNING TITLE

Vinegar reduces postprandial glycaemia in type 2 diabetes

LIST OF AUTHORS

Stavros Liatis[1], Sofia Grammatikou[1], Kalliopi-Anna Poulia[2], Despoina Perrea[3], Konstantinos Makrilakis[1], Evanthia Diakoumopoulou[1], Nicholas Katsilambros[1].

[1] First Department of Internal Medicine, Diabetes Center, Athens University Medical School, Laiko Hospital, Athens, Greece.

[2] Department of Nutrition, Laiko Hospital, Athens, Greece.

[3] Laboratory for Experimental Surgery and Surgical Research, Athens University Medical School, Athens, Greece

ADDRESS FOR CORRESPONDENCE

Stavros Liatis

17 Ag Thoma str.

11527 Athens

Greece

Tel: +30 210 7456261

Fax: +30 210 7791839

e-mail: s.liatis@yahoo.com

ABSTRACT

Background/Objectives

Previous studies have shown that the addition of vinegar in a carbohydrate-rich meal lowers glucose and insulin response in healthy individuals. The mechanism of how this is accomplished, however, remains unclear. The aim of the present study is to examine the effect of vinegar on glucose and insulin response in patients with type 2 diabetes (T2D) in relation to the type of carbohydrates consumed in a meal.

Patients/Methods

Sixteen patients with T2D were divided into two groups, matched for age, gender and HbA_{1c}. Patients in the first group (group A) were given a high-glycaemic index (GI) meal (mashed potatoes and low-fat milk) on two different days, with and without the addition of vinegar respectively. In the second group (group B), patients were given an isocaloric meal with the same nutrient composition but low GI (whole grain bread, lettuce, and low-fat cheese). Postprandial plasma glucose and insulin values were measured every 30 minutes for 2 hours.

Results

In group A, the incremental area under the curve of glucose ($GiAUC_{120}$) was lower after the addition of vinegar (181±78mmol·min/l vs. 311±124mmol·min/l, P=0.04). The iAUC of insulin ($IiAUC_{120}$) was also reduced but the difference was of marginal statistical significance (2368±1061μU·min/ml vs. 3545±2586μU·min/ml, P=0.056). In group B, the addition of vinegar did not affect either the $GiAUC_{120}$ (229±38mmol·min/l vs. 238±25mmol·min/l, P=0.56) or the $IiAUC_{120}$ (2996±1302 μU·min/ml vs. 3007±1255μU·min/ml, P=0.98).

Conclusions

We conclude that the addition of vinegar reduces postprandial glycaemia in patients with T2D only when it is added to a high-GI meal.

Keywords: Vinegar, acetic acid, postprandial glycaemia, glycaemic index, type 2 diabetes

INTRODUCTION

Several studies have shown that plasma glucose levels measured two hours after administering an oral glucose tolerance test (oGTT) can serve as an independent predictor of cardiovascular risk (Donahue et al, 1987; The DECODE Study Group, the European Diabetes Epidemiology Group, 1999). In addition, postprandial hyperglycemia contributes to about 30–40% of the total daytime hyperglycemia in patients with diabetes (Monnier et al, 2003). According to guidelines issued by major scientific societies, correcting postprandial hyperglycemia is part of the treatment strategy for the prevention of cardiovascular disease in patients with diabetes (American Diabetes Association, 2008; AACE Diabetes Mellitus Clinical Practice Guidelines Task Force, 2007). In addition, most authorities agree that both the amount of carbohydrates (CHO) as well as the type of CHO influence postprandial plasma glucose level, and the use of the glycaemic index (GI) and/or the glycaemic load (GL) can provide additional benefits over considering total CHO alone (Katsilambros et al, 2006).

Previous studies have shown that the addition of vinegar in a CHO-rich meal lowers glucose and insulin responses in healthy individuals (Liljeberg et al, 1998, Brighenti et al, 1995, Johnston et al, 2005, Leeman et al, 2005, Östman et al, 2005) and improves insulin sensitivity in individuals with insulin resistance and/or T2D (Johnston et al, 2004). It has also been shown that the addition of vinegar to a high-GL meal significantly reduces 60-minute postprandial glycaemia in healthy individuals (Johnston et al, 2005). The mechanism, however, remains unclear and although it has been suggested that delayed gastric emptying might be responsible for the hypoglycemic effect of vinegar, the results of relevant studies are conflicting (Brighenti et al, 1995, Liljeberg et al, 1998).

The aim of the present study is to examine the effect of vinegar on postprandial glycaemic and insulin response in patients with type 2 diabetes in relation to the GI of their meal.

Patients-Methods

Sixteen patients with T2D, treated either with diet alone or with metformin monotherapy at a maximum dose of 850 mg daily, accepted to participate in the study. They were selected from a pool of patients with T2D attending the outpatient Diabetes Clinic of Laiko University Hospital in Athens, Greece. Participants were divided into two groups, matched for age, sex, body mass index (BMI), duration of diabetes and HbA1c. To be included in the study, patients needed to have an HbA1c < 7.5%, a fasting plasma glucose < 8.9 mmol/l, and no history of major complications related to diabetes. All participants were given explanations about the purpose and the procedures of the study, which was approved by the ethics committee of the Laiko Hospital, and then signed an informed consent. The study required that patients visit the laboratory of the Diabetes Clinic for both their visits in the morning, after a 12-hour fasting period.

Patients in the first group (group A) were given a high-GI mixed meal (meal A) on two different days, separated by one week (± 2 days), with and without the addition of 20g of wine vinegar (6% acetic acid, AB Vassilopoulos S.A) respectively, corresponding to 1.2g of acetic acid. In the second group (group B), patients were given an isocaloric meal with the same nutrient composition but a low GI (meal B), using the same protocol (tested on two different days, with and without vinegar, respectively). Meal A was composed of pureed potatoes (50g of mashed potato instant, GIOTIS S.A) and low-fat (1%) milk (250 ml, FAGE S.A.), while meal B was composed of whole grain bread (100g whole grain bread from wheat and rye, German

Type, Katselis, NUTRIART S.A.), lettuce (55g), and low-fat cheese (13% fat, 20g, La Vache qui rit light, BEL S.A.). The macronutrient composition, GI and GL of the test meals are shown in Table 1. The GI and GL were calculated on the basis of validated international tables (Foster-Powell K et al , 2002). Water (200 ml) was served with each meal. The meals were served in random order on the two occasions and were consumed steadily within 10 minutes.

Measurement of body weight, height, waist circumference and seated blood pressure (BP) were obtained during the two scheduled visits, pre-prandially. Vein blood samples were drawn prior to each meal and every 30 minutes for a total period of 2 hours for analysis of plasma glucose and insulin levels. Plasma glucose was measured by the hexokinase method. Plasma insulin was measured in duplicate using BioSource INS-Irma immunoradiometric assay kits.

Statistical analysis

Analysis of the data was performed using the SPSS statistical package (SPSS 15.0, IL, USA). The incremental 2-hours area under the curve (iAUC) for postprandial glucose and insulin plasma values were calculated for each patient-test by using the trapezoidal rule and compared by using the paired samples, Student's t-test. In addition, the effect of vinegar on plasma glucose and insulin concentrations (at baseline, and 30′, 60′, 90′ and 120′ after the meal consumption) was assessed, in both groups, by using the one-way repeated-measures ANOVA. Between groups comparisons, regarding demographic and clinical parameters have been performed by the independent samples Student's t-test. P values < 0.05 were considered statistically significant.

Results

Table 2 shows that the two groups of participants were similar in terms of gender, age, body mass index (BMI), waist circumference, diabetes duration, HbA_1c, and proportion of patients treated with metformin. Fasting plasma glucose and plasma insulin values at baseline (time 0′) were also not statistically different between the two groups and between the two days of the experiment (within each group).

In group A (high-GI meal), the mean $GiAUC_{120}$ was significantly lower when vinegar was added than when the meal was consumed alone (181±78 mmol·min/l vs. 311±124 mmol·min/l, P=0.04). Six out of eight participants of that group (A), showed a lower $GiAUC_{120}$ when vinegar was added in the meal. Regarding plasma insulin, the mean $IiAUC_{120}$ was lower when vinegar was added than when the meal was consumed alone. However, this difference was of marginal statistical significance (2368±1061 μU·min/ml vs. 3545±2585 μU·min/ml, P=0.056). Five out of eight participants in group A showed a lower $iAUC_{120}$ after vinegar was added in the meal.

In group B, the addition of vinegar in the low-GI meal did not significantly affect either glucose or insulin response [$GiAUC_{120}$: 229±38mmol·min/l (with vinegar) vs. 238±25mmol·min/l (without vinegar), P=0.56; $IiAUC_{120}$: 2996±1302μU·min/ml (with vinegar) vs. 3007±1255μU·min/ml (without vinegar), P=0.98].

One-way repeated measures ANOVA analysis (Figure 1) showed a significant lowering effect of vinegar addition on glucose response in group A (F=7.3, P=0.03), but not in group B (F=3.0, P=0.13). There was also a significant interaction effect between vinegar addition and glucose concentration over time (0-120 min, F=4.9, P=0.004) in group A but not in group B (F=0.43, P=0.79). The interaction effect (in group A) was significant at times 60′ (P=0.034), 90′ (P=0.03) and 120′ (P=0.02), (Figure 1). The same type of analysis, when applied for insulin response (Figure 2),

showed a marginal but non-significant lowering of plasma insulin concentration after vinegar addition in group A (F=5.2, P=0.054). In group B, vinegar addition didn't affect insulin response (F=0.01, P=0.95). The interaction between vinegar addition in the meal and insulin concentration over time (0-120 min) was statistically significant in group A (F=3.7, P=0.03) but not in group B (F=0.84, P=0.46). The interaction effect (in group A) was significant at times 90′ (P=0.04) and 120′ (P=0.03), (Figure 2).

Discussion

The main finding of the present study is that in patients with T2D, postprandial hyperglycaemia is reduced when vinegar is added to a high-GI meal. However, postprandial hyperglycaemia is not affected by the addition of the same amount of vinegar to an isocaloric, of equal CHO-content, low-GI meal.

Previous studies, in healthy adults, have shown that several organic acids (including propionic acid, lactic acid and acetic acid) exhibit a postprandial plasma glucose and insulin lowering effect. The mechanism by which these substances produce this effect is not fully elucidated. In the case of acetic acid, some evidence suggests that the delay in gastric emptying (estimated by paracetamol measurements in the blood) may explain the reduced glycaemic and insulinaemic responses (Hunt JN et al., 1969, Liljeberg H et al., 1998). Acidity and fatty acids are known to affect stomach motility through duodenal receptor-mediated mechanisms (Hunt et al., 1972, Lin HC et al., 1990). In another relevant study (Brighenti F et al, 1995), however, gastric emptying as measured by ultrasonography was not affected by the addition of vinegar in a mixed-meal. In addition to delayed gastric emptying, other possible implicated mechanisms include a lower rate of starch hydrolysis in the upper small intestine

(Brighenti F et al., 1995), suppression of disacharidase activity (Ogawa et al., 2000) and earlier induction of satiety (Östman et al., 2005).

The effect of vinegar on postprandial glucose response in T2D has been examined previously only in one study with 10 participating patients (Johnston C et al., 2004). It was shown that the consumption of 20g apple cider vinegar immediately before a CHO-rich mixed meal (87g total CHO) improved (non-significantly) postprandial insulin and glucose fluxes and insulin sensitivity (estimated using a composite score). Postprandial increase in glucose after vinegar addition to a CHO-rich mixed meal has also been examined in type 1 diabetes (Personal communication with Mitrou P et al., data accepted for publication), using artificial pancreas. It was shown that vinegar, compared to placebo, reduced glucose $AUC_{0\text{-}240}$ by almost 20%. However, caution should be paid in patients with type 1 diabetes and gastroparesis, since it has been shown that in such patients, vinegar addition to food reduces the gastric emptying rate even further, and this might be a disadvantage regarding to their glycaemic control (Hlebowicz J et al., 2007).

In our study, the consumption of similar amount of vinegar together with a CHO-rich (51g), high-GI (86) meal significantly reduced postprandial plasma glucose by 42% as indicated by $GiAUC_{120}$. We were not able to compare our results with those of the study by Johnston et al. (2004), since they only reported incremental glucose values at 30 and 60 minutes. As it can be extrapolated from their published figure, however, at 60 minutes after glucose ingestion, in the vinegar group, plasma glucose seems to have decreased by 1 mmol/l, while in our study, it decreased by 2 mmol/l. Different characteristics of the two diabetic populations might be responsible for this difference (these are not provided in Johnston et al,, 2004).

Previous reports on healthy individuals have shown that when 20g of vinegar were added to CHO-rich, high-GI mixed meals, glucose response decreased by 11% - 54% (Johnston C et al., 2005: 54%, Liljeberg et al., 1998: 35%, Sugiyama et al., 2003: 25-35%, Brighenti et al., 1995: 30%, Östman et al., 2005: 11%,). The higher decrease observed in our study might be attributed to the fact that patients with diabetes exhibit (by definition) higher postprandial glucose excursions than healthy persons, which leads to a higher proportional glucose-lowering effect of any anti-hyperglycamic intervention.

The novel finding of the present study is that when the vinegar was added to an isocaloric mixed meal, with the same total-CHO content but having a remarkably lower GI, the glucose-lowering effect of vinegar disappeared (Figure 1). Interestingly, a previous study in healthy individuals (Johnston C et al., 2005) showed that adding vinegar (in the form of apple cider vinegar, 20g, 5% acetic acid) to a high-GI/high GL mixed meal (GI: 96, GL: 81) reduced the 60-min postprandial glycaemia significantly, by about 55%. The addition of the same amount of vinegar, however, on a mixed meal with similar GI (=91) but lower GL (=48) still reduced the 60-min glucose response, although not in a statistically significant proportion. The difference with the present study is that while their meal exhibited a low-GL but still a high-GI value (the difference in GL was due to a lower CHO content), our study had *both* low-GL and low GI values.

In group B, the low GI meal produced (as expected) a lower glycaemic and insulin response than the high-GI meal (without vinegar). It must be emphasized that these figures cannot be directly compared, since the two meals were tested in two different groups of diabetic patients (matched, however, for age, gender, BMI, duration of diabetes and level of glycaemic control). The low GI of the meal was mainly due to

its high fiber content. Nevertheless, it was clearly shown that the addition of vinegar in the low-GI meal did not decrease postprandial glucose and insulin response (Figures 1 & 2).

To our knowledge, the effect of vinegar on high-fiber/low-GI meals has not been examined in the past. In the study by Johnston C et al. (2005), the GL of the low-GL mixed meal was 48, which is still quite higher than the one presented in our study (19.8, see Table 1), while the GI was 96 compared to only 38 in the present study. The low GI/GL meal in our study still produced a substantial increase in postprandial glucose and insulin levels, which wasn't affected by the addition of vinegar.

As mentioned above, the mechanism by which vinegar reduces postprandial glycaemia/insulinemia could be attributed to the decrease in the rate of gastric emptying, which in turn leads to slower CHO absorption. It can be hypothesized that the slow CHO absorption caused by a high-fiber/low-GI meal is not further affected by the effect of vinegar on gastric emptying. In other words, slow CHO absorption caused by fiber, is not further delayed by vinegar. However, we believe that more studies are needed to elucidate the extent that other physiological mechanisms affect glucose absorption in the presence of acetic acid.

A limitation of the present study is that the four different test meals (high/low GI, with/without vinegar) were tested in two *different* groups of patients with T2D, matched, however, for main demographic and clinical characteristics (Table 2). This approach was chosen in order to avoid the need to test every patient at four different days and confine the necessary experimental days to two per patient, since no patient was willing to participate having to be tested at four different occasions.

In conclusion, the addition of vinegar on high-GI meals might serve as a protective measure to avoid excessive postprandial rise in plasma glucose, in patients with

"early" T2D. Postprandial hyperglycaemia is generally recognized as an important factor affecting overall glycaemic control, while, according to several reports, is also believed to consist an independent predictor of cardiovascular events (The DECODE Study Group, the European Diabetes Epidemiology Group, 1999). According to the results of the present study, however, when the consumed meal had a low GI value, vinegar addition did not further suppress postprandial glucose response.

Acknowledgements

The authors would like to thank Mr Elias Kourpas for his assistance to write the article in proper English.

Conflict of interest

Nothing to declare

References

1. AACE Diabetes Mellitus Clinical Practice Guidelines Task Force (2007): American Association of Clinical Endocrinologists medical guidelines for clinical practice for the management of diabetes mellitus. *Endocr Pract* **13 (Suppl 1)**, 1-68.
2. American Diabetes Association (2008): Standards of Medical Care in Diabetes. *Diabetes Care* **31 (Suppl 1)**, S12-S54
3. Brighenti F, Castellani G, Benini L, Casiraghi MC, Leopardi E, Crovetti R, et al (1995) Effect of neutralized and native vinegar on blood glucose and acetate responses to a mixed meal in healthy subjects. *Eur J Clin Nutr* **49,** 242-247
4. Donahue RP, Abbott RD, Reed DM, Yano K (1987): Postchallenge glucose concentration and coronary heart disease in men of Japanese ancestry: Honolulu Heart Program. *Diabetes* **36**, 689-692.
5. Ebihara K, Nakajima A. (1988) Effect of acetic acid and vinegar on blood glucose and insulin responses to orally administered sucrose and starch. Agric Biol Chem 52:1311-1312
6. Hlebowicz J, Darwiche G, Björgell O, Almér LO. (2007) Effect of apple cider vinegar on delayed gastric emptying in patients with type 1 diabetes mellitus: a pilot study. *BMC Gastroenterol* **7**, 46.
7. Hunt JN, Knox MT (1969). The slowing of gastric emptying by nine acids. *J Physiol* **201,** 161-179
8. Hunt JN, Knox MT (1972) The slowing of gastric acid by four strong acids and three weak acids. *J Physiol* **222,** 187-208

9. Johnston CS, Buller AJ (2005) Vinegar and peanut products as complementary foods to reduce postprandial glycemia. *J Am Diet Assoc* **105,** 1939-1342.

10. Johnston CS, Kim CM, Buller AJ. (2004) Vinegar improves insulin sensitivity to a high-carbohydrate meal in subjects with insulin resistance or type 2 diabetes. *Diabetes Care* **27,** 281-282

11. Katsilambros N, Liatis S, Makrilakis K (2006): Critical Review of the International Guidelines: What Is Agreed upon - What Is Not? *Nestle Nutr Workshop Ser Clin Perform Programme* **11,** 207-218.

12. Leeman M, Ostman E, Björck I (2005) Vinegar dressing and cold storage of potatoes lowers postprandial glycaemic and insulinaemic responses in healthy subjects. Eur J Clin Nutr **59**,1266-1271

13. Liljeberg H, Björck I. (1998) Delayed gastric emptying rate may explain improved glycaemia in healthy subjects to a starchy meal with added vinegar. *Eur J Clin Nutr* **52**, 368-371

14. Lin HC, Doty JE, Reedy TJ, Meyer JH (1990). Inhibition of gastric emptying by acids depends on pH, titratable acidity, and length of intestine exposed to acid. *Am J Physiol* **259,** 1025-1030

15. Mitrou P, Raptis AE, Lambadiari V, Boutati E, Petsiou E, Spanoudi F et al (2010). Vinegar decreases postprandial hyperglycemia in patients with type 1 diabetes. *Diabetes Care* **33**, e27.

16. Monnier L, Lapinski H, Colette C (2003). Contributions of fasting and postprandial plasma glucose increments to the overall diurnal hyperglycemia of type 2 diabetic patients: variations with increasing levels of HbA(1c). *Diabetes Care* **26**, 881-885.

17. Ogawa N, Satsu H, Watanabe H, Fukaya M, Tsukamoto Y, Miyamoto Y, et al. (2000) Acetic acid suppresses the increase in disaccharidase activity that occurs during culture of caco-2 cells. *J Nutr* **130**, 507-513

18. Ostman E, Granfeldt Y, Persson L, Björck I. (2005) Vinegar supplementation lowers glucose and insulin responses and increases satiety after a bread meal in healthy subjects. *Eur J Clin Nutr* **59,** 983-988.

19. Sugiyama M, Tang AC, Wakaki Y, Koyama W. (2003) Glycemic index of single and mixed meal foods among common Japanese foods with white rice as a reference food. *Eur J Clin Nutr* **57**, 743-752.

20. The DECODE Study Group, the European Diabetes Epidemiology Group (1999): Glucose tolerance and mortality: comparison of WHO and American Diabetes Association diagnostic criteria. *Lancet* **354**, 617-621.

Figure legends

Figure 1. Mean plasma glucose concentration in group A (high-GI meal) and in group B (low-GI meal) with and without the addition of vinegar.

Figure 2. Mean plasma insulin concentration in group A (high-GI meal) and in group B (low-GI meal) with and without the addition of vinegar.

Table 1: Macronutrient composition of the two test meals

Macronutrients	Meal 1	Meal 2
Energy (Kcal)	279	279
Carbohydrate (g)	51	52
Fat (g)	2.5	2.5
Protein (g)	13	12
Dietary Fiber (g)	4	8
GI	86	38
GL	44	20

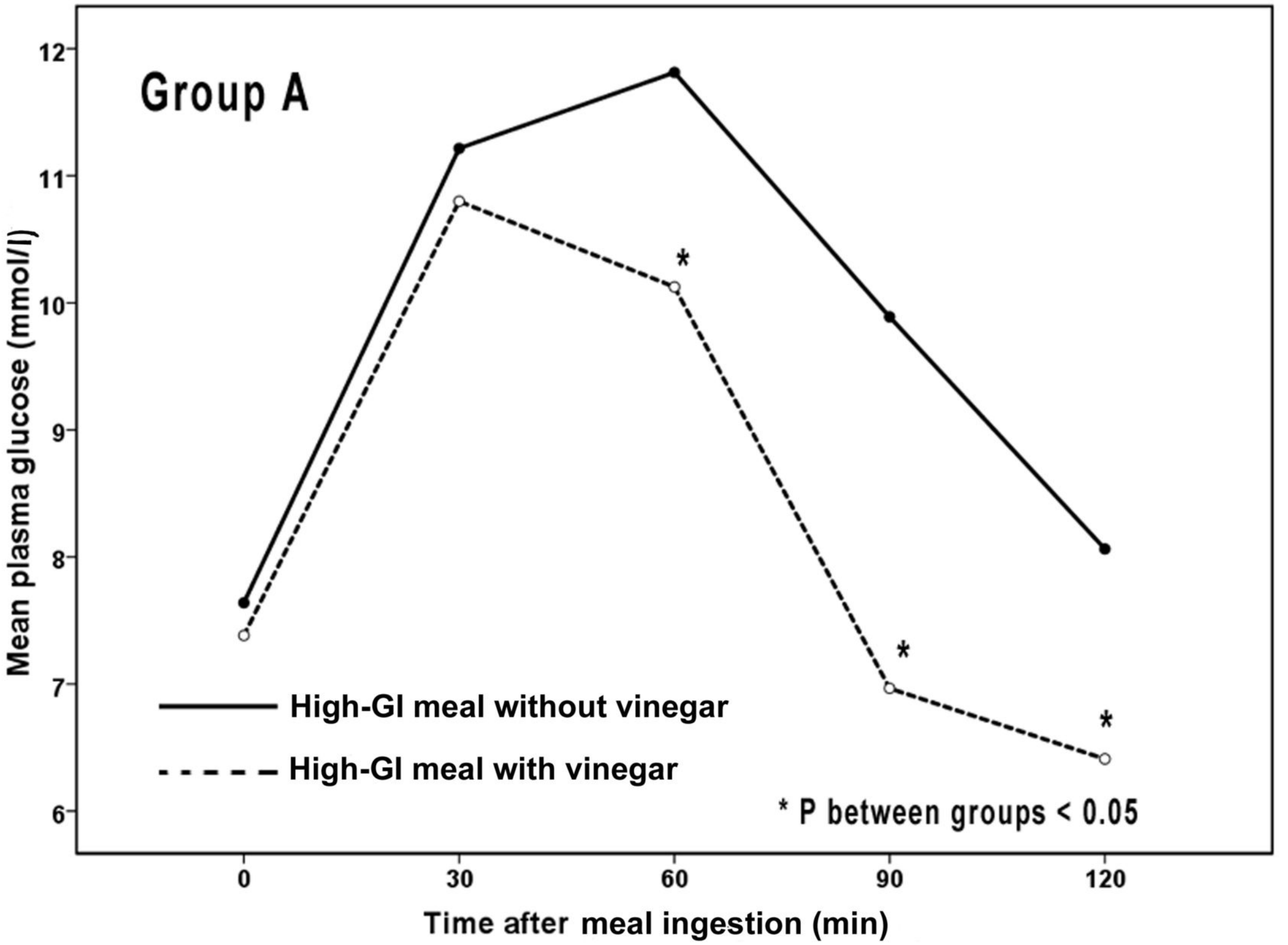
GI = Glycaemic index, GL = Glycaemic Load

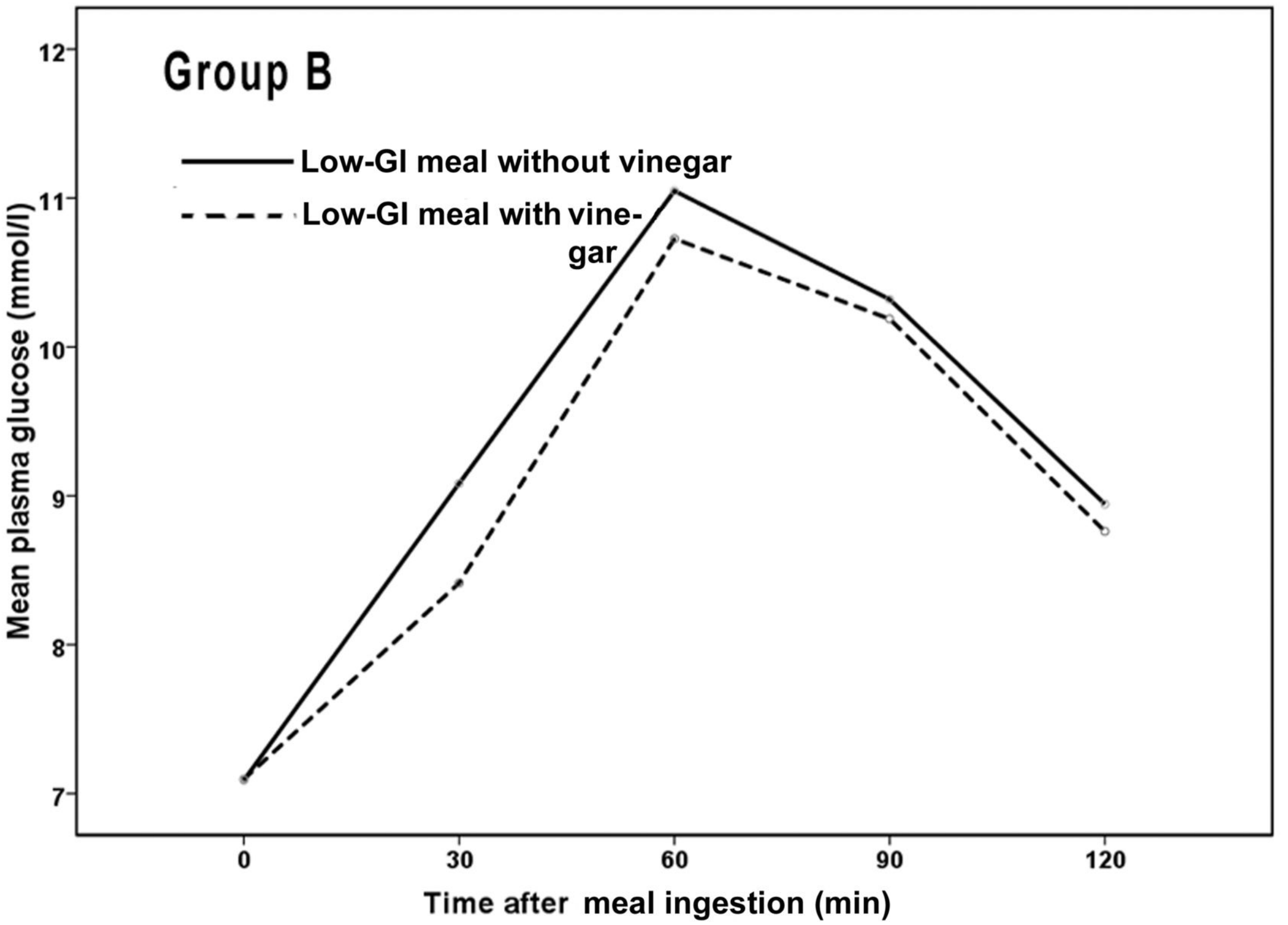
Table 2. Demographic and clinical characteristics of the study participants.

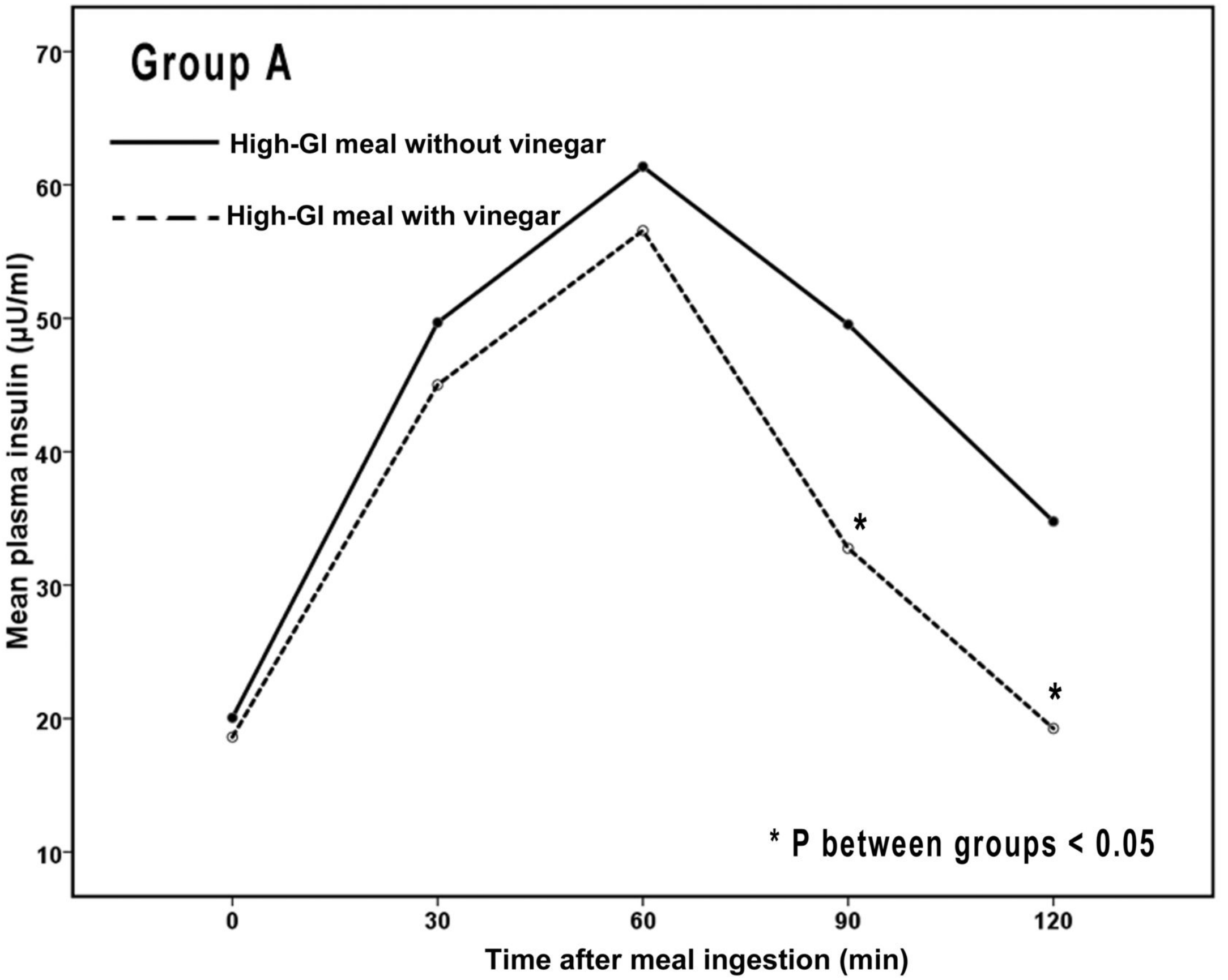
[Continuous variables are presented as mean value (SD)].

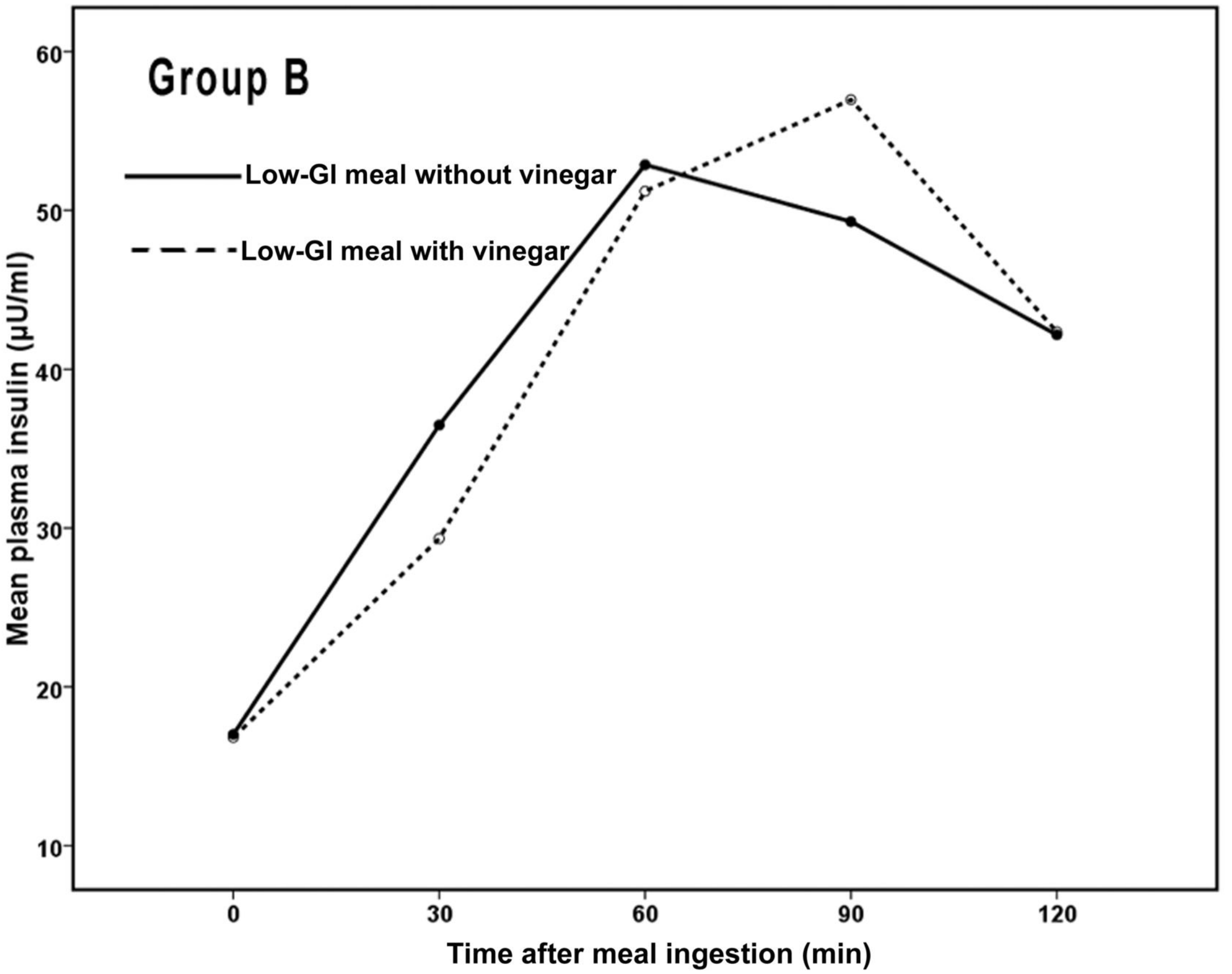
		Group A			**Group B**		**p**
Gender (males/females)	3/5			4/4		0.31	
Age (years)	57.4 (8.0)			61.4 (8.4)		0.37	
BMI (Kg/m^2)	29.8 (1.9)			30.1 (3.3)		0.84	
Waist circumference (cm)	103 (7)			107 (9)		0.82	
Duration of diabetes (years)	3.6 (4.0)			4.8 (3.5)		0.33	
Metformin therapy (yes/no)	3/5			3/5		1.0	
HbA1c (%)	6.3 (1.0)			6.0 (0.5)		0.56	
	Day 1	**Day 2**	**p**	**Day 1**	**Day 2**		
Fasting plasma glucose (mmol/l)	7.6 (0.8)	7.4 (0.6)	0.33	7.1 (0.5)	7.1 (0.4)	0.95	
Fasting plasma insulin (μU/mL)	20.1 (6.1)	18.6 (5.2)	0.29	17.0 (6.6)	16.8 (8.7)	0.88	

BMI: Body Mass Index


Group A
Mean plasma glucose (mmol/l)
12
11
10
9
8
7
6
High-GI meal without vinegar
High-GI meal with vinegar
*
*
*
* P between groups < 0.05
0
30
60
90
120
Time after meal ingestion (min)


Group B
Low-GI meal without vinegar
Low-GI meal with vinegar
Mean plasma glucose (mmol/l)
12
11
10
9
8
7
0
30
60
90
120
Time after meal ingestion (min)


Group A
High-GI meal without vinegar
High-GI meal with vinegar
Mean plasma insulin (µU/ml)
70
60
50
40
30
20
10
*
*
* P between groups < 0.05
0
30
60
90
120
Time after meal ingestion (min)


Group B
Low-GI meal without vinegar
Low-GI meal with vinegar
Mean plasma insulin (µU/ml)
60
50
40
30
20
10
0
30
60
90
120
Time after meal ingestion (min)