How the degree of unsaturation of dietary fatty acids influences the glucose and insulin responses to different carbohydrates in mixed meals

Jean-Luc Joannic, Silvy Auboiron, Jocelyne Raison, Arnaud Basdevant, Francis Bornet, and Bernard Guy-Grand

ABSTRACT The association of fats with carbohydrates results in a lower glucose response but the influence of the nature of the dietary fatty acids has not been investigated clearly. We examined the effect of the degree of dietary fatty acid unsaturation on the postprandial glucose and insulin responses to a mixed meal. Eight young normolipidemic men consumed four different meals in random order. The meals differed in the nature of their oils and in the type of their main carbohydrates. The meals contained either a high ratio of monounsaturated to polyunsaturated n-6 fatty acids (MUFA) or a low ratio (PUFA) and either potatoes or parboiled rice. Proteins and saturated and polyunsaturated n-3 fatty acid contents were not different among meals. Blood samples were collected every 30 min for 3 h after the test meal. The glucose response was significantly lower 30 min after the parboiled rice-MUFA meal than after parboiled rice-MUFA or potato-MUFA (P < 0.05) meals. The insulin response was lower after parboiled rice-MUFA than after potato-MUFA (P < 0.05) meals. Similarly, an effect of fat appeared after 30 min. Glucose responses (F = 1.4, P < 0.01) and insulin responses (F = 5.3, P < 0.05) to both carbohydrates were significantly lower with dietary PUFA compared with dietary MUFA. In conclusion, the degree of dietary fatty acid unsaturation (18:1 compared with 18:2) may influence the glucose and insulin responses to mixed meals. Am J Clin Nutr 1997;65:1427–33.

KEY WORDS Humans, monounsaturated fat, polyunsaturated fat, carbohydrate, glucose response, insulin response, mixed meals

INTRODUCTION

The glycemic index (GI) is widely used for comparing blood glucose responses with different types of carbohydrates. The relative increase in blood glucose after the acute administration of a carbohydrate-rich food is expressed as a percentage of the response to the same amount of a standard glucose challenge (1–3). However, using the GI to predict the glucose and insulin responses to carbohydrates in a mixed meal is questionable because of the presence of other macronutrients that can modify these metabolic responses. Several studies showed that the association of protein (4), fat (5, 6), or fiber (7) with carbohydrate can reduce the postprandial glucose response. Although the influence of fat on the glucose and insulin responses to carbohydrates is well established in normal subjects (8, 9) and in subjects with non-insulin-dependent diabetes mellitus (NIDDM) (10, 11), the influence of the degree of unsaturation of the dietary fats has not been clearly documented. Only limited information on the effect of different types of fats on postprandial glucose and insulin concentrations in normal humans is available. The insulin response to mixed meals was increased by both polyunsaturated fatty acids (PUFAs) derived from fish and vegetable oils compared with the same amount of saturated fatty acids (SFAs), whereas the glucose response was increased only by unsaturated fatty acids from fish (12). However, Gatti et al (13) found no effect on the postprandial blood glucose response when adding saturated fats to a white bread meal, whereas the ingestion of olive oil or corn oil reduced the glucose response in normal subjects by 70% without altering the insulin responses. We investigated the effects of the degree of dietary fatty acid unsaturation on glucose and insulin responses to mixed meals of similar macronutrient composition containing carbohydrates with a known different GI.

SUBJECTS AND METHODS

Subjects

Eight healthy young male volunteers (aged 24 ± 1 y) with stable body weight [body mass index (BMI; in kg/m²) 21.6 ± 0.8] participated in the study after giving written informed consent to a protocol approved by the Medical Ethics Committee (Hôtel-Dieu, Paris, CCPRPB no. 93070, France). No subjects had any medical history of digestive, endocrine, or metabolic diseases. They had not been taking any medication that would interfere with glucose metabolism for the previous 6 mo. Blood pressure was normal in all subjects. Fasting blood variables were in the normal range: insulin 58.4 ± 3.9 pmol/L, 

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2 Supported by a grant from Eridania Béghin-Say, Paris.

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Received July 10, 1996.

Accepted for publication December 15, 1996.
glucose 4.68 ± 0.07 mmol/L, triacylglycerol 0.81 ± 0.1 mmol/L, total cholesterol 4.05 ± 0.12 mmol/L.

Subjects were instructed to not deviate from their regular activities and dietary habits during the study. However, they were asked to avoid eating hazelnuts, walnuts, or salmon, and to limit fish consumption to no more than twice per week to control their n-3 PUFA intake. They were monitored by a 3-d food record during the 4 consecutive wk of the experiment. Their usual energy and macronutrient intakes were evaluated according to nutrient tables (14, 15). The subjects consumed a typical Western diet with a usual energy consumption (mean: 12 406 ± 531 kJ/d). Proteins, carbohydrates, and fats accounted for 14.6 ± 0.5, 45.2 ± 1.3, and 37.5 ± 1.1% of energy, respectively. The ratio of PUFAs to SFAs was 0.25 ± 0.02, and the ratio of monounsaturated fatty acids (MUFAs) to PUFAs was 4.95 ± 0.65. Alcohol intake was 6.2 ± 0.9 g/d.

Test meals

After an overnight fast of 12 h, the subjects arrived at the research hospital unit at 0800. At 0830, they were served a carbohydro-carbohydrate-rich standardized breakfast (1254 kJ, 12% protein, 70% carbohydrate, and 18% fat). The subjects stayed in a semirecumbent position during the experiment. At 1130, the test meal was consumed steadily for 20 min. The four experimental meals were given in random order separated by an interval of 7 d according to a Latin-square design. The meals differed only in the nature of unsaturated fats (MUFA or PUFA) and in the type of carbohydrates (rice or mashed potatoes) and consisted of freshly prepared commercially available foods. The rice was parboiled, long-grain white rice (Japanese type and Ariette variety). The GI of instant mashed potatoes is 83 ± 1, and the GI of the parboiled long-grain white rice is 47 ± 3, with the glucose as a reference of 100 (16). The total energy content was 5265 kJ for the meals containing instant mashed potatoes and 5200 kJ for the rice meals. Protein, carbohydrate, and fat accounted for 14%, 38%, and 47% of the energy provided by the meals. Composition of the meals is given in Table 1. Two kinds of fat were used: a mixture of high-oleic sunflower (70%) and rapeseed (30%) oils with a high ratio of MUFA to PUFA (4.3) or a mixture of sunflower (60%) and soybean (40%) oils with a MUFA-PUFA ratio (0.4). Those were combined with two types of carbohydrates as follows: potato-MUFA, rice-MUFA, potato-PUFA, or rice-PUFA. The oils were provided by Lesieur (Neuilly/Seine, France), and their fatty acid composition was analyzed by the Institut Scientifique d’Hygiène Alimentaire (Longjumeau, France) as shown in Table 2. The amounts of SFAs and n-3 PUFAs were kept constant among test meals. The rice was cooked for 20 min in 1000 mL water with 7 g salt/L. The instant mashed potatoes were restored with 500 mL boiled water that contained 7 g salt/L.

**Measurements**

When the subjects arrived at the hospital unit an intravenous cannula equipped with disposable obturators (Jelco-Critikon, Chatenay-Malabry, France) was inserted into an antecubital vein. The fasting blood variables were measured before breakfast. Postprandial blood samples were collected just before the test meal (baseline) and every 30 min for 3 h.

**Analytic methods**

Samples were centrifuged immediately (4 °C for 15 min at 785 × g) and frozen at −20 °C for later assay. Plasma glucose was assayed with a glucose-oxidase method (Galway, Ireland; intraassay reproducibility 1%) on a Beckman Autoanalyzer II (Fullerton, CA). Plasma insulin was measured by radioimmunoassay (ERIA Diagnostics Pasteur, Marnes la Coquette, France; intraassay reproducibility 4%). Plasma triacylglycerols (BioMérieux, Marcy-l’Étoile, France; intraassay reproducibility 2%) and fatty acids (Wako, Neuss, Germany; intraassay reproducibility 2%) were determined by enzymatic methods.

**Statistical analysis**

This study was constructed in a Latin-square design with four treatments (k² = 16). A sample of eight subjects was taken to respond to the conditions of 2k². Three kinds of statistical analysis were performed on the SUPERANOVA microcomputer program (Abacus Concepts Inc, Berkeley, CA) used on a Macintosh personal computer (Cupertino, CA). First, the Latin-square design consisted of a three-way analysis of variance (ANOVA) and tested the effect of these factors: days, subjects, and meals. Multiple comparisons were made by using the method of Newman and Keuls (17) at P < 0.05. Second, data

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**Table 1**

Composition and portion size of test meals

<table>
<thead>
<tr>
<th>Food item</th>
<th>Chemical composition</th>
<th>Portion size</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Available Carbohydrates</td>
<td>Protein</td>
</tr>
<tr>
<td>Mashed potatoes</td>
<td>81</td>
<td>8</td>
</tr>
<tr>
<td>Rice</td>
<td>78</td>
<td>8</td>
</tr>
<tr>
<td>White bread</td>
<td>23</td>
<td>3</td>
</tr>
<tr>
<td>Apple</td>
<td>14</td>
<td>0</td>
</tr>
<tr>
<td>Water</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Steak</td>
<td>0</td>
<td>28</td>
</tr>
<tr>
<td>Cheese</td>
<td>1</td>
<td>5</td>
</tr>
<tr>
<td>Oils</td>
<td>0</td>
<td>0</td>
</tr>
</tbody>
</table>

1 Data given by the manufacturer.
2 According to the nutrient table in reference 15.
3 Analyzed by the Institut Scientifique d’Hygiène Alimentaire (Longjumeau, France).
TABLE 2
Fatty acid composition of mixtures of oils and foods
d| Fatty acid | High-oleic sunflower and rapeseed oil | Sunflower and soybean oil | Steak | Cheese |
<table>
<thead>
<tr>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>12:1n−9</td>
<td>0.0</td>
<td>0.0</td>
<td>0.0</td>
<td>0.1</td>
</tr>
<tr>
<td>14:1n−9</td>
<td>0.0</td>
<td>0.0</td>
<td>1.0</td>
<td>1.5</td>
</tr>
<tr>
<td>16:1n−9</td>
<td>0.2</td>
<td>0.1</td>
<td>4.2</td>
<td>1.9</td>
</tr>
<tr>
<td>17:1n−9</td>
<td>0.0</td>
<td>0.0</td>
<td>0.0</td>
<td>0.4</td>
</tr>
<tr>
<td>18:1n−9</td>
<td>70.0</td>
<td>24.5</td>
<td>39.0</td>
<td>22.5</td>
</tr>
<tr>
<td>Σ MUFA</td>
<td>70.2</td>
<td>24.6</td>
<td>44.2</td>
<td>26.4</td>
</tr>
<tr>
<td>18:2n−6</td>
<td>16.2</td>
<td>56.7</td>
<td>3.3</td>
<td>2.1</td>
</tr>
<tr>
<td>Σ n−6 PUFA</td>
<td>16.2</td>
<td>56.7</td>
<td>3.3</td>
<td>2.1</td>
</tr>
<tr>
<td>18:3n−3</td>
<td>2.3</td>
<td>3.2</td>
<td>0.5</td>
<td>0.7</td>
</tr>
<tr>
<td>18:4n−3</td>
<td>0.0</td>
<td>0.2</td>
<td>0.4</td>
<td>0.3</td>
</tr>
<tr>
<td>20:4n−3</td>
<td>0.9</td>
<td>0.0</td>
<td>0.1</td>
<td>0.0</td>
</tr>
<tr>
<td>Σ n−3 PUFA</td>
<td>3.2</td>
<td>3.4</td>
<td>1.0</td>
<td>1.0</td>
</tr>
<tr>
<td>4:0</td>
<td>0.0</td>
<td>0.0</td>
<td>0.0</td>
<td>3.4</td>
</tr>
<tr>
<td>6:0</td>
<td>0.0</td>
<td>0.1</td>
<td>0.1</td>
<td>2.8</td>
</tr>
<tr>
<td>8:0</td>
<td>0.0</td>
<td>0.0</td>
<td>0.0</td>
<td>1.8</td>
</tr>
<tr>
<td>10:0</td>
<td>0.0</td>
<td>0.1</td>
<td>0.1</td>
<td>4.1</td>
</tr>
<tr>
<td>12:0</td>
<td>0.0</td>
<td>0.0</td>
<td>0.1</td>
<td>4.4</td>
</tr>
<tr>
<td>13:0</td>
<td>0.0</td>
<td>0.0</td>
<td>0.0</td>
<td>0.1</td>
</tr>
<tr>
<td>14:0</td>
<td>0.1</td>
<td>0.2</td>
<td>3.1</td>
<td>11.9</td>
</tr>
<tr>
<td>15:0</td>
<td>0.0</td>
<td>0.4</td>
<td>0.4</td>
<td>1.2</td>
</tr>
<tr>
<td>16:0</td>
<td>5.3</td>
<td>9.5</td>
<td>28.2</td>
<td>27.5</td>
</tr>
<tr>
<td>18:0</td>
<td>3.7</td>
<td>4.3</td>
<td>14.5</td>
<td>8.2</td>
</tr>
<tr>
<td>20:0</td>
<td>0.5</td>
<td>0.4</td>
<td>0.5</td>
<td>0.8</td>
</tr>
<tr>
<td>22:0</td>
<td>0.0</td>
<td>0.0</td>
<td>0.4</td>
<td>0.0</td>
</tr>
<tr>
<td>Σ SFA</td>
<td>9.6</td>
<td>14.6</td>
<td>47.3</td>
<td>66.2</td>
</tr>
<tr>
<td>Σ n−9/n−6</td>
<td>4.3</td>
<td>0.4</td>
<td>13.4</td>
<td>12.6</td>
</tr>
</tbody>
</table>

1 Results are expressed as the percentage of total fatty acid methyl esters.
2 MUFA, monounsaturated fatty acids; PUFA, polyunsaturated fatty acids; SFA, saturated fatty acids.

were subjected to an ANOVA according to a two factorial arrangement, by using the general linear model (GLM) procedure, to determine the main fat or carbohydrate effects. These effects are expressed as variance ratio (F) and probability. The observed statistical differences were also found by using single paired t tests. Third, data were subjected to an ANOVA with repeated-measures at P < 0.05 to determine the time-course variations of plasma glucose and insulin responses for each meal. The total area under the 0- to 180-min curve was calculated by the trapezoidal method (18). Results are given as mean ± SEM.

RESULTS

The baseline data did not differ among subjects on each experimental day. The ANOVA revealed no order effect. No difference was found for the time course of plasma triacylglycerols and fatty acids, regardless of the dietary conditions (data not shown).

Effect of fat

Plasma glucose responses

The time course of plasma glucose responses and the corresponding total area under the curves are shown in Figure 1 (top). At 30 min, the glucose response to rice-PUFA was significantly lower (P < 0.05) than to rice-MUFA. No significant difference was found between the two potato meals, but the glucose response after potato-MUFA tended to be higher. The glucose response to rice-PUFA was significantly lower (P < 0.05) than to potato-MUFA. The glucose response to rice-MUFA was not significantly different from the potato meals. No other differences among meals were found up to 180 min. The total area under the plasma glucose curve was not significantly different among the four meals.

When pooling data from both PUFA or MUFA meals (Figure 2, top), a significant effect of the type of fat appeared at 30 min (F = 1.40, P < 0.01), ie, the glucose response was significantly lower with PUFA than with MUFA meals, regardless of the type of carbohydrate. No significant effect of fat was found on the total area under the curve of mean glucose responses.

Plasma insulin responses

The time course of plasma insulin responses and the corresponding total area under the curves are shown in Figure 1 (bottom). At 30 min, no difference was found between the two rice meals or between the two potato meals. At this time, however, the insulin response to rice-PUFA was significantly lower (P < 0.05) than to potato-MUFA. The total area under the curve of the plasma insulin response showed no difference between the two rice meals or between the two potato meals. However, the overall insulin response to both rice meals was significantly lower (P < 0.05) than to the potato-MUFA meal.

When pooling data from both PUFA and MUFA meals (Figure 2, bottom), an effect of fat (F = 5.30, P < 0.05) was
observed only at 30 min, i.e., the insulin response was significantly lower with PUFA than with MUFA meals, regardless of the type of carbohydrate. The overall insulin response tended to be lower with PUFA meals than with MUFA meals ($P = 0.16$).

**Effect of carbohydrate**

*Plasma glucose responses*

When pooling data from rice or potato meals (Figure 2, top), a significant effect of carbohydrate was found at 180 min ($F = 8.46, P < 0.02$), i.e., the rice meals induced a higher glucose response than the potato meals, independent of the type of dietary fat. Between 90 and 180 min, glucose concentration increased significantly after the rice meals ($P < 0.05$) but did not differ after the potato meals. No significant effect of carbohydrate was found on the total area under the curve of the mean glucose responses.

*Plasma insulin responses*

A difference among the four meals (Figure 1, bottom) was observed at 90 min, when the insulin response to rice-PUFA was significantly lower ($P < 0.05$) than to potato-PUFA or to potato-MUFA, and the insulin response to rice-MUFA was significantly lower ($P < 0.05$) than to potato-PUFA.

When pooling data from rice or potato meals (Figure 2, bottom), an effect of carbohydrate was found at 30 min ($F = 6.64, P < 0.05$) and at 90 min ($F = 1.4, P < 0.01$) on the insulin response, i.e., the rice meals induced a significantly lower insulin response than the potato meals, independent of the nature of the fat. The overall mean insulin response to the rice meals was significantly lower than to the potato meals ($F = 3.40, P < 0.001$), regardless of the type of fat. Between 30 and 180 min, however, the insulin concentration decreased after potato meals containing any of the two oils ($P < 0.05$) but did not significantly decrease after the rice meals.

**DISCUSSION**

We compared the glucose and insulin responses to various mixed meals that differed in the nature of their fatty acids (PUFA compared with MUFA) and in the GI of their prevalent carbohydrate (potato with a high GI compared with parboiled rice with a low GI). The major finding was that the degree of unsaturation of the dietary fat influenced the early postprandial glucose and insulin responses. Plasma glucose and insulin responses reached a peak at 30 min. At this time, the increase in plasma glucose and insulin concentrations was significantly lower after PUFA meals than after MUFA meals, regardless of
the main carbohydrate in the meal. Despite the different GI of the main carbohydrate, both MUFA meals produced similar glucose responses. The MUFA blunted the differences in plasma glucose response usually found between rice and potatoes when ingested alone (19, 20), whereas the PUFA did not. Thus, the meal containing carbohydrates with low GI (rice) and MUFA resulted in the same glucose response as the meal containing carbohydrates with the high GI (potatoes) and PUFA. This effect is likely to be related to the degree of fatty acid unsaturation because MUFA and PUFA oils do not differ in their carbon chain length (93% and 90% 18-carbon fatty acids, respectively).

However, the integrated insulin and glucose responses during the 180-min postmeal period (area under the curve) were not significantly different, although a trend to a lower area was seen after the PUFA meals. The metabolic consequences of increased early glucose and insulin responses remain to be evaluated. Given that insulin is involved in the secretion of triacylglycerol-rich lipoproteins (21, 22) and in lipoprotein lipase activity (23), it is possible that a 23% increase in early postprandial insulin might influence the early postprandial lipid metabolism. The fat content of the present test meals (47% was close to the fat consumption of many Western populations (42%) (24, 25). Whether the effect of the degree of unsaturation is present when the fat content of the meal is lower remains speculative.

Several factors may account for the influence of the dietary fat on the glucose and insulin responses. Differences in gastric emptying may play a role. It is well-known that the glucose response to a mixed meal is strongly correlated with the rate of gastric emptying, which is faster after the ingestion of potato than rice (26). It also has been shown that the gastric emptying of a protein and carbohydrate meal is delayed either by adding fat (27) or by infusing lipids into either the ileum or duodenum (28). The effects of nutrients could be related to a direct influence of carbohydrate (29) or fat (30, 31) on the small intestinal nutrient receptors that control gastric emptying, although the existence of specific intestinal receptors for fats has not been established in humans (32). Whether the differences in the unsaturation of fat can influence the gastric emptying remains unknown.

Dietary fatty acids may also interact with food digestion by modulating digestive enzyme activities. A study in 1995 (33) showed that a high-fat diet compared with a low-fat diet had a
tendency for higher output of gastric lipase and significantly increased the gastric lipase activity in healthy humans. However, the effect of fat on starch hydrolysis by \( \alpha \)-amylase is less known. It has been shown that the rate of hydrolysis of starch (potatoes or legumes) in vitro by \( \alpha \)-amylase was not modified when fat was added in the reaction mixture (34). Whether the nature of dietary fatty acids might affect the enzyme activity of either gastric lipase or \( \alpha \)-amylase in vivo is unknown.

In this experiment, differences in the degree of fatty acid unsaturation influenced the insulin responses to mixed meals. At 30 min, the differences in insulin responses paralleled those in plasma glucose responses. Most of the insulin differences reflect those obtained with the glucose responses, as evidenced by the time course of the insulin to glucose ratio that paralleled the insulin responses (data not shown). However, at 90 min, no difference in insulin responses was found between meals with MUFA oils, whereas a significant difference between rice and potato meals was found with PUFA oils. No significant difference was found in glucose concentration 90 min after the meal. These results raise the question of the influence of PUFAs on insulin secretion and sensitivity. In vitro studies performed in 1979 and 1994 on perfused mouse islets of Langerhans or isolated perfused rat pancreas showed that the insulin release induced by a physiologic glucose concentration (close to 5 mmol/L) was increased by unsaturated fatty acids (35, 36) but was not affected by saturated fatty acids (36, 37). Insulin secretion was enhanced as the degree of fatty acid unsaturation increased (36).

Our data show that regardless of the nature of the oil the late glucose responses differed between rice and potato meals. At 180 min, a significant effect of carbohydrates was observed: the rice meal induced a second glucose peak. This response has not been described previously. Whether this effect is specific to rice or is attributable to the high fat content of the meals remains to be determined.

This study showed that the degree of unsaturation of fat influenced the glucose and insulin responses to mixed meals. Thirty minutes after both rice or potato meals, glucose and insulin responses were lower with PUFA than with MUFA meals. Moreover, at this time, after the MUFA meals, the glucose responses were similar regardless of the GI of the major carbohydrate. Thus, in healthy subjects, the GI of the carbohydrate contained in mixed meals cannot predict accurately the glucose and insulin responses because the degree of unsaturation of dietary fatty acids also influences these metabolic responses.

REFERENCES


