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An Oral Lipid Challenge and Acute Intake of Caffeinated Coffee Additively Decrease Glucose Tolerance in Healthy Men1–3

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Abstract
Lipid-induced insulin resistance has been investigated primarily with i.v. infusions, and caffeine-induced insulin resistance, with alkaloid caffeine. The effects of orally consumed lipids and coffee have not been established and to our knowledge have never been simultaneously investigated. The goals of this study were to determine whether an oral lipid challenge and caffeinated coffee would disrupt glucose homeostasis and to characterize their respective incretin responses. It was hypothesized that oral ingestion of saturated lipids would impair glucose tolerance and that caffeinated coffee would further hinder glucose management. Ten young, healthy males participated in 5 trials in a randomized, cross-over design. At time 0 h, they underwent an oral fat tolerance test (OFTT: 1 g lipid/kg body weight) or consumed water, followed 5 h later by caffeinated (5 mg/kg) coffee, decaffeinated coffee, or water. At 6 h, volunteers underwent an oral glucose tolerance test (OGTT). Consumption of the OFTT increased glucose concentrations (P < 0.05) after a subsequent OGTT. At 7 h, caffeinated coffee produced the highest glucose concentrations (P < 0.05). Glucagon-like peptide-1 active (GLP-1a) and glucose-dependent insulinotropic polypeptide (GIP) were both increased for up to 6 h in all OFTT trials (P < 0.05). Compared to all other treatments, caffeinated and decaffeinated coffee produced higher GLP-1a response at 6.25 h (P < 0.05), whereas only caffeinated coffee increased GIP secretion (P < 0.05). These results show that oral consumption of lipids and caffeinated coffee can independently and additively decrease glucose tolerance. Incretin hormones could explain at least in part this impaired glucose homeostasis. J. Nutr. 141: 574–581, 2011.

Introduction
Acute administration of alkaloid caffeine impairs glucose homeostasis in healthy (1–4) and obese (5,6) individuals as well as in diabetic patients (6–8). Consumption of caffeine before an oral glucose tolerance test (OGTT)4 has consistently been shown to reduce the insulin sensitivity index (ISI) by 20–30% (2,5,7,9) and to decrease the glucose infusion rate during a euglycemic-hyperinsulinemic clamp by a similar extent (3,6,9–12). Caffeine’s detrimental effects on glucose management are generally attributed to adenosine receptor antagonism (9,13,14) and epinephrine action on skeletal muscle (9,11,12).

Caffeine is often consumed in beverage form, notably as caffeinated coffee. The effects of caffeine cannot be readily generalized to coffee, as caffeine only represents 2% of coffee’s compounds (15). Acutely, caffeinated coffee induces a similar impairment, albeit of lesser magnitude, on glucose management (1,2,16,17). In marked contrast, regular and long-term coffee consumption is associated with a decreased risk for type 2 diabetes in a dose-dependent fashion (18–22).

I.v. infusion of lipids is associated with an elevation in plasma FFA and a reduction in insulin-stimulated glucose uptake by 20–50% in healthy volunteers (23–31), diabetics (32), offspring of diabetic patients (33), and in both sexes (24,28). This whole-body effect is predominantly attributed to an impairment in skeletal muscle metabolism (24,25,28,30,31) and to a lesser extent to lipids’ actions on the liver (24,28–30,34). The lipid-induced impairment of glucose management may be exacerbated by SFA, which have been suggested to be more deleterious than unsaturated fatty acids both in vitro (35–39) and in vivo (40–42).

I.v. lipid infusions would exclude possible responses in intestine-derived incretin hormones, which have been demonstrated to play an important role in whole-body glucose...
tolerance. Carbohydrates (43), lipids (44–46), and coffee (16) all potentiate the release of glucose-dependent insulintropic peptide (GIP) and glucagon-like peptide-1 (GLP-1). The metabolic impact of the incretins may include both pancreatic and extrapancreatic actions (especially on skeletal muscle (47) and on gastric emptying (48)).

We hypothesized that consumption of an oral lipid load would impair glucose tolerance in a subsequent OGTT. We also postulated that caffeinated coffee would further disrupt glucose homeostasis during the postprandial lipid challenge. The incretin responses to this combination of dietary challenges were also characterized to establish if they might account for any alterations in carbohydrate homeostasis.

Materials and Methods

Participants. Eleven nonsmoking and recreationally active men were recruited from the University of Guelph community by poster and Web site advertisements. Prior to the study, volunteers were screened by a questionnaire for medical conditions and height and weight were recorded. All participants gave written informed consent to participate in the experimental procedures after the potential risks were explained to them. One participant withdrew from the study due to coffee intolerance. The study was approved by the University of Guelph Research Ethics Board.

Experimental design. Each participant came into the laboratory on 5 different occasions, separated by at least 7 d. Volunteers were instructed to withdraw from exercise, caffeine-containing products, and alcohol 48 h prior to each trial. In addition, they received a standardized meal to consume before 2000 h as their last meal before each trial (2008 kJ; 62% of energy fat, 22% of energy carbohydrate, 16% of energy fat, and 22% of energy protein). On the day of the experiment, participants reported to the laboratory at 0800 h after a 12-h fast. Upon their arrival at the laboratory, a venous catheter was placed into an antecubital vein and was kept patent by a saline solution infusion. On the first trial day, body composition was determined by bioimpedance analysis (BodyStat 1500). Throughout the day, volunteers sat quietly (read, watched movies, used a computer, etc.).

At the beginning of the experiment (time 0 h), in 4 of the 5 trials (Supplemental Table 1), the participants were given enough oral fat beverage (described below) to provide 1 g of lipid/kg body weight or the equivalent weight of water (control OGTT). After 5 h, they received either caffeinated coffee (OFTT/CAF) providing 5 mg caffeine/kg body weight, the equivalent volume of decaffeinated coffee (OFTT/DECAF), or water (control OFTT, control OGTT, OFTT/–). One hour later, at 6 h, volunteers received 75 g of dextrose in solution (OFTT/CAF, OFTT/DECAF, OFTT/–, control OGTT) (OGTT: Trutol 75, NERL Diagnostic). Except on one occasion when the 5-h water ingestion was then followed with water ingestion at 6 h (control OFTT) (Supplemental Table 1). In the one trial in which the fat beverage was not ingested, water was ingested at time 0 and 5 h, and at 6 h, 75 g of dextrose was consumed. Thus, in the control OFTT treatment, participants did not receive a coffee beverage or OGTT; in the control OGTT, they did not receive OFTT; and in the OFTT/– treatment, they received both the OFTT and OGTT but did not consume a coffee beverage. The treatments were designed to administer the OGTT 1 h after coffee consumption and at least 3 h of elevated plasma FFA, because this timing has been shown sufficient to induce caffeine-specific effects (1,2,5–7) and lipid-mediated effects (23,26) on insulin resistance. The order of treatment administration was randomized. Water was allowed ad libitum during the day.

To determine whether the consumption of the oral lipid load would disrupt glucose homeostasis, we compared the 6–8 h data from the control OGTT treatment to those of the OFTT/– treatment, because these 2 treatments differed only in the ingestion of the lipid drink at time 0 h. We included the control OFTT treatment to allow baseline measurements (without OGTT) between 6 and 8 h. To examine whether caffeinated coffee would further disrupt glucose tolerance in a context of postprandial lipid challenge, we compared the 6–8 h data from the OFTT/–, OFTT/CAF, and OFTT/DECAF treatments. OFTT/– and OFTT/CAF treatments differed only in the consumption of caffeinated coffee at time 5 h. The inclusion of the OFTT/DECAF treatment allowed the differentiation between the effects of caffeine and those of other coffee compounds on glucose tolerance. Finally, the comparison of the OFTT/– and OFTT/CAF treatments also allowed for the investigation of potential additive effects of lipids and caffeinated coffee on glucose tolerance.

Lipid test beverage preparation. The lipid test beverage was prepared as previously described (49). Briefly, blends of palm stearine and soybean oil (generously provided by Bunge Canada) were chemically interesterified to achieve a random distribution of fatty acids and a ratio of PUFA:SFA of 0.2. The final product was stored at −20°C until use. The FFA composition of the final product was analyzed by GC (Shimadzu, Supplemental Table 2). On the day of the experiment, the interesterified product was warmed in a microwave oven (General Electric Canada) to reach a liquid state. Warm water (73%) and 2 emulsifiers (Acetasol, Myverol (2%), and Tween 80 (0.15%), were added to the liquid fat. To ensure palatability, the drink was artificially flavored with Aspartame, a non-nutritive sweetener, and a commercially available coffee flavoring. The mixture was homogenized with an electric latte whip and served warm (−60°C).

Coffee preparation. To ensure uniformity, all the coffee used in the study was prepared by mixing a large quantity of Maxwell House Dark Roast coffee together. Caffeinated coffee was prepared according to a standard method in our laboratory (2,50) to yield a coffee that contained 621 mg caffeine/L coffee (50). The amount of coffee ingested for each participant was calculated based on this data to achieve a ratio of 5 mg caffeine/kg body weight. DECAF was prepared following the same procedure using Maxwell House decaffeinated coffee. The participants did not know whether they consumed caffeinated or decaffeinated coffee.

Circulating glucose, FFA, and lipid concentrations. Whole blood was collected in sodium heparin tubes (BD Vacutainer) for determination of glucose (YSI 2300 Stat Plus Glucose Analyzer) and for TG, total cholesterol, and HDL-cholesterol (all with Cholesterol LDX (R) lipid cassettes, Cholestech). Direct subtraction of HDL-cholesterol values from total cholesterol provided values for LDL-cholesterol. Blood for FFA analysis was collected in another sodium-heparinized tube (BD Vacutainer) and was immediately centrifuged (Beckman Allegra X-12R) at 3416 × g for 5 min. Subsequently, the supernatant was treated with NaCl, as described elsewhere (51,52), to inhibit lipoprotein lipase. This heparinized plasma was frozen at −20°C for analysis by colorimetric assay (NEFA kit, Wako Chemical) at a later date.

Serum C-peptide and TG concentrations. Blood was collected in nonheparin tubes (BD Vacutainer) previously treated with aprotinin (Sigma Aldrich). Blood was allowed to clot at room temperature and was then centrifuged (13416 × g, 10 min). The supernatant was collected and frozen at −20°C for later determination of C-peptide (Human C-peptide RIA kit, Millipore) or at −80°C for TG determination by colorimetric assay (Triglyceride GPO kit, Synchron System, Beckman Coulter) at the Guelph General Hospital.

Plasma GLP-1 active, GIP, and insulin concentrations. Blood was collected in K3-EDTA tubes (BD Vacutainer) previously treated with dipeptidyl peptidase-4 inhibitor (Millipore). Immediately after blood collection, 90 µL of aprotinin (Sigma-Aldrich) was added to the tubes. Tubes were centrifuged (1000 × g, 15 min) and the supernatant was collected and frozen at −80°C for later determination of GLP-1a, GIP, and insulin by immunoassay (Human Gut Hormone Panel, Bioplex 2200, Bio-Rad).

Dietary analysis. Volunteers were required to record food and fluid intake for 3 d prior to each trial. One participant did not keep the record for 1 d on one trial, so that the total of food record days analyzed was
Control OGTT. In addition, OFTT/DECAF and OFTT/

blood glucose in the OFTT/CAF trial reached 10.7
reflecting that the test beverage did not contain carbohydrates

Currently, TG were increased in the OFTT/

the OGTT, and were elevated (P < 0.05; Table 2). Concurrently, TG were increased in the OFTT/− and control OFTT treatments between 3 and 5 h compared to the water control (P < 0.05; Fig. 1B). In addition, all 4 treatments involving an OFTT increased the TG AUC above that of the control OGTT (P < 0.05; Table 2).

Circulating glucose, insulin, and C-peptide concentrations.

Fasting whole-blood glucose concentrations were normal and comparable for each treatment (−4.4 mmol/L). In all 5 treatments, glucose remained at fasting levels during the first 6 h, reflecting that the test beverage did not contain carbohydrates (Supplemental Table 3). In all treatments except the control OFTT, glucose levels increased rapidly at 6 h, upon initiation of the OGTT, and were elevated (P < 0.05) compared to the control OFTT treatment between 6.5 and 8 h (Fig. 2A). At 7 h, blood glucose in the OFTT/CAF trial reached 10.7 ± 0.4 mmol/L, which was higher (P < 0.05) than OFTT/DECAF, OFTT/−, and control OGTT. In addition, OFTT/DECAF and OFTT/− both

induced intermediate glucose levels that were higher (P < 0.05) than control OGTT (Fig. 2A). AUC analysis during the OGTT period (6–8 h) showed that all 4 treatments involving the OGTT increased blood glucose AUC above that of control OFTT. In addition, compared to the control OGTT treatment, glucose AUC was elevated in OFTT/− (32%; P = 0.097), OFTT/DECAF (46%; P < 0.05), and OFTT/CAF (65%; P < 0.05) (Table 3).

Plasma insulin responses mirrored the blood glucose responses (Fig. 2B) and remained at fasting level up to 6 h (Supplemental Table 4). Between 6.25 and 8 h, the consumption of the OGTT increased insulin levels beyond the OFTT control (P < 0.05). Moreover, the OFTT/CAF treatment resulted in elevated insulin concentrations (P < 0.05 vs. OGTT between 6.5 and 7.5 h; Fig. 2B). In addition, the OFTT/CAF insulin concentration was also elevated compared to OFTT/− at 7 h and OFTT/DECAF at 7.5 h (P < 0.05; Fig. 2B). Between 6 and 8 h, insulin AUC for the OGTT treatments were greater than that of the OFTT control (P < 0.05). In addition, OFTT/CAF produced higher insulin AUC than the OGTT control (55%; P < 0.05) (Table 3).

In agreement with the insulin and glucose data, serum C-peptide concentrations for all 5 treatments remained at baseline levels (−0.2 nmol/L) during the first 6 h of the experiment, with no differences between treatments (data not shown). C-peptide concentrations for OFTT control did not differ from fasting

Results

Lipids. The participants’ baseline characteristics were within the normal range (Table 1). Consumption of the OFTT elevated plasma FFA between 5 and 7 h above the water control (P < 0.05; Fig. 1A). Similarly, all OFTT treatments, except for OFTT/DECAF, elevated FFA 8 h AUC (P < 0.05; Table 2). Concurrently, TG were increased in the OFTT/− and control OFTT treatments between 3 and 5 h compared to the water control (P < 0.05; Fig. 1B). In addition, all 4 treatments involving an OFTT increased the TG AUC above that of the control OGTT (P < 0.05; Table 2).

**TABLE 1** Baseline characteristics of and fasting whole-blood measurements for 10 healthy men

<table>
<thead>
<tr>
<th>Variables</th>
<th>Mean values</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age, y</td>
<td>22.9 ± 0.4</td>
</tr>
<tr>
<td>Height, m</td>
<td>1.8 ± 0.03</td>
</tr>
<tr>
<td>Weight, kg</td>
<td>78.9 ± 4.8</td>
</tr>
<tr>
<td>BMI, kg/m²</td>
<td>24.7 ± 0.8</td>
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<tr>
<td>Body fat, %</td>
<td>16.5 ± 1.4</td>
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<tr>
<td>TC, mmol/L</td>
<td>3.8 ± 0.2</td>
</tr>
<tr>
<td>HDL-C, mmol/L</td>
<td>1.2 ± 0.1</td>
</tr>
<tr>
<td>TG, mmol/L</td>
<td>1.1 ± 0.1</td>
</tr>
<tr>
<td>LDL-cholesterol, mmol/L</td>
<td>2.2 ± 0.2</td>
</tr>
<tr>
<td>TC:HDL-C</td>
<td>3.4 ± 0.3</td>
</tr>
<tr>
<td>Exercise, activities/wk</td>
<td>3.2 ± 0.5</td>
</tr>
</tbody>
</table>

1. Values are presented as means ± SEM, n = 10.
2. TC, total cholesterol; HDL-C, HDL cholesterol.
3. Activities: one session of moderate aerobic or resistance exercise.

**FIGURE 1** Time course for plasma FFA (A) and serum TG (B) concentrations in men during the 8-h protocol for OFTT control, OFTT/CAF, OFTT/DECAF, OFTT/−, and OGTT control. Data are means ± SEM, n = 10. Means at a time without a common letter differ, P < 0.05.
values at any point during the protocol. In contrast, compared to the OFTT control, C-peptide was elevated ($P < 0.05$) between 6.5 and 8 h for OFTT/CAF, between 7 and 8 h for OFTT/–, and between 7.5 h and 8 h for OFTT/DECAF and control OGTT. C-peptide concentrations were also greater in OFTT/DECAF and OFTT/– compared to the OGTT control at 8 h ($P < 0.05$; data not shown). From 6 to 8 h, the C-peptide AUC data for OFTT/CAF, OFTT/DECAF, and OFTT/– were all greater than those of OFTT control ($P < 0.05$; Table 3).

The ISI for the OGTT period was 5.9, 6.2, 7.6, and 7.5 for OFTT/CAF, OFTT/DECAF, OFTT/–, and OGTT control, respectively. Although OFTT/CAF ISI was 22% lower than the value for the OGTT control, the treatments did not differ.

**Incretins: plasma GLP-1a and GIP**. Consumption of the lipid test beverage elevated ($P < 0.05$) plasma GLP-1a (2–6 h; Fig. 3A) and GIP (30 min–6 h; Fig. 4A) above the water control. At 6 h, consumption of 75 g of dextrose transiently ($P < 0.05$) increased GLP-1a above baseline levels at 6.25 h in the OGTT treatment, whereas in the OFTT/– treatment, the GLP-1a concentration did not differ between 6 and 6.25 h ($P = 0.83$). Consequently, there were no differences in GLP-1a concentrations between OFTT/– and the OGTT control at 6.25 h. In contrast, both OFTT/CAF and OFTT/DECAF treatments considerably increased ($P < 0.05$) GLP-1a, compared to the OFTT control (6.25–6.5 h), OFTT/– (6.25 h), and OGTT control (6.25–6.5 h) (Fig. 3B).

The consumption of the OGTT alone also increased GIP levels above baseline values ($P < 0.05$) and this increase was sustained throughout the last 2 h of the protocol. In contrast to GLP-1a responses, only the OFTT/CAF treatment elevated the GIP concentration compared to all other treatments at 6.25 h ($P < 0.05$). At 6.5 h, OFTT/CAF GIP levels were still higher than both OGTT and OFTT controls ($P < 0.05$) and tended to be higher than OFTT/DECAF ($P = 0.06$) (Fig. 4B).

The analysis of the whole protocol (0–8 h) compared to the 6- to 8-h period led to very different findings regarding the incretins. The AUC analysis for the entire experimental period (0–8 h) revealed that OFTT control, OFTT/CAF, OFTT/DECAF, and OFTT/– produced larger ($P < 0.05$) GLP-1a AUC than the OGTT control (Table 2). In contrast, the AUC analysis for the OGTT period (6–8 h) showed that the OGTT control treatment triggered the largest GLP-1a response, which was greater than that of OFTT/CAF ($P < 0.05$) and OFTT/DECAF ($P < 0.05$) and also elevated 9.5- and 11-fold compared to the OFTT control ($P = 0.12$) and OFTT/– ($P = 0.10$), respectively (Table 3). The GIP AUC computed between 0 and 8 h was elevated 4-fold in OFTT control, OFTT/CAF, OFTT/DECAF, and OFTT/– treatments compared to OGTT control ($P < 0.05$; Table 2). On the other hand, the GIP AUC analysis during the 2-h OGTT revealed that OFTT/CAF, OFTT/DECAF, and OGTT control produced higher responses than the OFTT control ($P < 0.05$; Table 3).

**Discussion**

This experiment was undertaken to examine whether an oral load of saturated lipids and/or ingestion of caffeinated coffee would disrupt glucose homeostasis in healthy men and to characterize the incretin responses to these stimuli. We hypothesized that the acute consumption of an oral load of lipids and caffeinated coffee would independently and additively impair glucose management during a subsequent OGTT. The main

<table>
<thead>
<tr>
<th>Treatment</th>
<th>FFA (nmol/L)</th>
<th>TG (nmol/L)</th>
<th>Glucose (nmol/L)</th>
<th>Insulin (pmol/L)</th>
<th>C-peptide (pmol/L)</th>
<th>GLP-1a (pmol/L)</th>
<th>GIP (pmol/L)</th>
</tr>
</thead>
<tbody>
<tr>
<td>OFTT control</td>
<td>1.7 ± 0.3</td>
<td>3.7 ± 0.9</td>
<td>-1.6 ± 0.7</td>
<td>32 ± 9</td>
<td>-0.6 ± 0.2</td>
<td>70 ± 12</td>
<td>181 ± 24</td>
</tr>
<tr>
<td>OFTT/CAF</td>
<td>1.7 ± 0.2</td>
<td>2.7 ± 0.7</td>
<td>8.4 ± 0.5</td>
<td>721 ± 118</td>
<td>1.5 ± 0.4</td>
<td>77 ± 12</td>
<td>202 ± 16</td>
</tr>
<tr>
<td>OFTT/DECAF</td>
<td>1.2 ± 0.3a</td>
<td>1.6 ± 0.8a</td>
<td>5.7 ± 1.0c</td>
<td>547 ± 81b</td>
<td>1.3 ± 0.3ab</td>
<td>86 ± 22b</td>
<td>181 ± 21b</td>
</tr>
<tr>
<td>OFTT/–</td>
<td>1.7 ± 0.2b</td>
<td>2.6 ± 0.8b</td>
<td>6.2 ± 1.1bc</td>
<td>509 ± 107bc</td>
<td>1.2 ± 0.4b</td>
<td>76 ± 14b</td>
<td>204 ± 27b</td>
</tr>
<tr>
<td>OGTT control</td>
<td>0.7 ± 0.2a</td>
<td>0.8 ± 0.4a</td>
<td>4.4 ± 0.7b</td>
<td>426 ± 88b</td>
<td>1.1 ± 0.3b</td>
<td>8 ± 5a</td>
<td>37 ± 6a</td>
</tr>
</tbody>
</table>

**TABLE 2** AUC for circulating glucose, insulin, C-peptide, GLP-1a, and GIP in men during the 8-h protocol following OFTT control, OFTT/CAF, OFTT/DECAF, OFTT/–, and OGTT control treatments.

**FIGURE 2** Time course for whole-blood glucose (A) and plasma insulin (B) concentrations in men throughout the last 3 h of the protocol for OFTT control, OFTT/CAF, OFTT/DECAF, OFTT/–, and OGTT control. Data are means ± SEM, $n = 10$. Means at a time without a common letter differ, $P < 0.05$. The ISI for the OGTT period was 5.9, 6.2, 7.6, and 7.5 for OFTT/CAF, OFTT/DECAF, OFTT/–, and OGTT control, respectively. Although OFTT/CAF ISI was 22% lower than the value for the OGTT control, the treatments did not differ.
findings were that ingestion of lipids and caffeinated coffee independently and additively increased the blood glucose concentration and that both GLP-1α and GIP responses were dampened during an OGTT if preceded by an oral lipid challenge.

**Consumption of oral lipids disrupts glucose homeostasis.** The present findings suggest that the consumption of an oral lipid load decreased glucose tolerance in healthy men. The lipid drink provided ~80 g of lipids, which increased plasma FFA to 0.72–0.84 mmol/L after 6 h, a level comparable to the postprandial lipemia in diabetic patients (24,54) and similar to postprandial lipemia in diabetic patients (24). This lipid increase was sufficient to elicit exaggerated blood glucose responses during an OGTT in all OFTT treatments (OFTT/−, OFTT/CAF, and OFTT/DECAF) compared to the OGTT control (Table 3; Fig. 2A). These elevated glucose concentrations were present despite similar insulin responses among OFTT/−, OFTT/DECAF, and OGTT control treatments, suggesting that insulin action was disrupted following lipid ingestion (Fig. 2B).

Disrupted glucose management in the present study is also illustrated by the fact that the glucose AUC reported here are far greater than what was previously reported from our laboratory for comparable healthy volunteers. In fact, compared to the mean of values we have reported for healthy men [159 mmol/L–120 min (1,2,55)], glucose AUC for OFTT/− and OGTT control were elevated 2.7- and 2.1-fold, respectively. In addition, in the present investigation, the ISI in both the OGTT control and OFTT/− trials were reduced by 31% compared to similar studies in healthy males (1,2,9). There is no reason to think that differences in participants’ fitness or adiposity can explain these glucose AUC discrepancies (Table 1). Rather, the increases in FFA and/or TG through prolonged fasting (OGTT control) or oral lipid consumption (OFTT/−) probably explain the higher glucose AUC and decreased ISI in the current investigation.

Notably, in the OGTT control treatment, dextrose was ingested 6 h after the start of the protocol, i.e. after at least 18 h of fasting, in order to match the treatments chronologically. However, this prolonged fasting resulted in increases in FFA (Fig. 1A) that likely negatively affected glucose tolerance. It is impressive that despite this apparent blunted glucose tolerance in the OGTT control treatment, there was still evidence that consumption of the OFTT further disrupted glucose homeostasis. Thus, the current protocol probably portrays a conservative image of the glucose intolerance induced by the oral lipid beverage.

**Consumption of caffeinated coffee impairs glucose management in healthy men.** It has been extensively reported that alkaloid caffeine (3,9–12,55,56) and caffeinated coffee (2,16,17) induce insulin resistance. This action is usually attributed to the antagonism of the adenosine A1 receptor, but elevated FFA have also been considered as a complementary mechanism. The present study is the first to our knowledge to characterize this effect in the context of postprandial lipid challenge. The consumption of caffeinated coffee 5 h after an oral load of lipid increased glucose and insulin levels beyond those of lipids only or lipids and decaffeinated coffee (Fig. 2A,B). The concurrent increases in insulin and glucose responses suggest that insulin action, and not insulin secretion, is impaired with the combined action of lipids and caffeinated coffee, a situation that is further supported by the C-peptide data. The glucose AUC for the OFTT/CAF treatment was elevated by 65% compared to the OGTT control, whereas OFTT/− and OFTT/DECAF only increased AUC by 32 and 46%, respectively. Taken together, these data support the interpretation that caffeinated coffee and lipids disrupt glucose tolerance in an additive manner. At the level of skeletal muscle, lipid-induced insulin resistance is thought to be mediated by the disruption of phosphoinositide

![FIGURE 3](image338x108to530x376)
3-kinase-dependent insulin signaling (24–26). In contrast, caffeine-induced insulin resistance is presumably mediated by adenosine A1 receptor antagonism through a phosphoinositide 3-kinase-independent pathway (56). It is then possible that these 2 independent pathways converge to mediate this additive effect of lipids and caffeinated coffee.

The magnitude of insulin resistance induced by the combination of oral lipids and caffeinated coffee was important. At 7 h, the OFTT/CAF treatment produced a very high glucose concentration [10.7 ± 0.4 mmol/L (P < 0.05 vs. OFTT/ – and OFTT/DECAF)], which remained as high as 8.6 ± 0.7 mmol/L at the end of the 2-h OGTT. Although the participants were lean, active, and healthy, this latter result is within the range of impaired glucose tolerance [plasma glucose between 7.8 and 11.1 mmol/L after a 2-h OGTT (57)]. Moreover, ISI in the OFTT/ – treatment never differed from those of the OFTT or OGTT controls. Data are means ± SEM, n = 10. Means at a time without a common letter differ, P < 0.05.

FIGURE 4 Time course for plasma GIP concentrations in men throughout the 8-h protocol (A) and during the 2-h OGTT (B) for OFTT control, OFTT/CAF, OFTT/DECAF, OFTT/–, and OGTT control. Data are means ± SEM, n = 10. Means at a time without a common letter differ, P < 0.05.

Lipids, coffee, and caffeine mediate incretin release from the intestine. The ingestion of lipid was sufficient to induce the secretion of GLP-1a (3-fold) and GIP (6-fold) for up to 6 h. While GLP-1a has previously been shown to respond to lipids as well as carbohydrates (44), the current study expands these findings to GIP, showing that this hormone is also responsive to lipids. Moreover, to our knowledge, the present study is the first to document that consumption of oral lipids decreases incretin responses to a subsequent glucose challenge. As expected, the OGTT control produced large and rapid increases in both GLP-1a and GIP at 6.25 h upon consumption of dextrose. In contrast, after consumption of the lipid load, glucose did not induce incretin secretion (Figs. 3B, 4B; Table 3).

In fact, between 6.25 and 8 h, GLP-1a and GIP concentrations in the OFTT/ – treatment never differed from those of the OFTT or OGTT controls. The fact that 75 g of glucose failed to increase incretin secretion after a lipid challenge is a novel finding and is consistent with the knowledge that GLP-1 secretion is blunted in type 2 diabetic patients (59). The lack of incretin responses in the present study could partly mediate the blunted glucose tolerance described after lipid ingestion. Therefore, a high-fat diet and/or elevated circulating FFA, often characteristic of obesity and diabetic states, could lead to metabolic issues based on interference with the regulation incretin secretion and/or action.

In addition, the present findings suggest that coffee can independently modulate incretin responses. During the OGTT, the ingestion of both caffeinated and decaffeinated coffee increased the GLP-1a concentration above that of OFTT/ –, a treatment that differed only by the consumption of water instead of coffee. Moreover, only caffeinated, but not decaffeinated coffee, elevated the GIP concentration at 6.25 and 6.5 h. These results are comparable with those of Johnson et al. (16), who showed that GIP, but not GLP-1a, was higher after ingestion of caffeinated compared to decaffeinated coffee (0.1 < P > 0.05). Taken together, these findings suggest that caffeine itself may be a potent stimulus for GIP secretion, whereas GLP-1a could be responsive to other coffee compounds. Chlorogenic compounds (60,61) and quinides (62) are 2 classes of substances present in coffee that were associated with improved glucose tolerance. It is possible that action of these bioactive compounds in the gastrointestinal tract mediate GLP-1a release. Furthermore, the association among coffee, caffeine, and incretin secretion may explain at least in part the negative correlation between long-term coffee consumption and type 2 diabetes risks (18,19).

Admittedly, several other gastrointestinal factors may have responded to coffee and/or lipids in the present study. However, we are confident that this study examined 2 of the most bioactive gastrointestinal molecules, GLP-1a and GIP, and that they played an important role in the present results. In addition, our results are consistent with previous reports, suggesting that the insulin resistance described here is a direct effect of coffee and lipids and not secondary to the modulation of gastric emptying, intestinal transit time, or other gastrointestinal factors.

In conclusion, the present study documents that the ingestion of an oral lipid load and caffeinated coffee can, independently and additively, blunt glucose tolerance in healthy men. We not only confirmed that caffeine has a negative effect on glucose homeostasis, but also demonstrated for the first time to our knowledge that this impairment is still evident in a postprandial...
lipid context. In addition, the modulation of incretins by lipids and coffee could explain at least in part their respective effects on glucose metabolism during the OGTT. That the combination of saturated lipids and caffeinated coffee, 2 common items in Western diets, has such a profound effect on glucose tolerance and insulin action even in young, lean, active adults is highly relevant for the prevention and control of insulin resistance and type 2 diabetes.

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Literature Cited


