An extract of black, green, and mulberry teas causes malabsorption of carbohydrate but not of triacylglycerol in healthy volunteers

Litao Zhong, Julie K Furne, and Michael D Levitt

ABSTRACT

Background: In vitro studies suggest that extracts of black, green, and mulberry teas could interfere with carbohydrate and triacylglycerol absorption via their ability to inhibit α-amylase, α-glucosidase, sodium-glucose transporters, and pancreatic lipase.

Objective: We measured breath hydrogen and 13CO2 to investigate the ability of an extract of black, green, and mulberry tea leaves to induce malabsorption of carbohydrate and triacylglycerol in healthy volunteers.

Design: In a crossover design, healthy adult volunteers randomly ingested test meals with a placebo beverage or a preparation containing an extract of black (0.1 g), green (0.1 g), and mulberry (1.0 g) teas. One test meal contained 50 g carbohydrate as white rice, 10 g butter, and 0.2 g [13C]triolein, and the beverages contained 10 g sucrose. The calorie content of the second test meal consisted entirely of lipid (30 g olive oil and 0.2 g [13C]triolein), and the beverages contained 10 g sucrose. The calorie content of the second test meal consisted entirely of lipid (30 g olive oil and 0.2 g [13C]triolein). Breath-hydrogen and 13CO2 concentrations were assessed hourly for 8 h, and symptoms were rated on a linear scale.

Results: With the carbohydrate-containing meal, the tea extract resulted in a highly significant increase in breath-hydrogen concentrations, which indicated appreciable carbohydrate malabsorption. A comparison of hydrogen excretion after the carbohydrate-containing meal with that after the nonabsorbable disaccharide lactulose suggested that the tea extract induced malabsorption of 25% of the carbohydrate. The tea extract did not cause triacylglycerol malabsorption or any significant increase in symptoms.

Conclusion: This study provides the basis for additional experiments to determine whether the tea extract has clinical utility for the treatment of obesity or diabetes.


KEY WORDS Malabsorption, carbohydrate, triacylglycerol, tea extract, breath-hydrogen test, 13CO2-breath test

INTRODUCTION

It is widely believed that teas contain substances that are beneficial to health. (A search of the key words “tea health benefits” brings up >5 million entries on Google.) Although most of the alleged benefits of tea are not supported by solid scientific evidence, teas contain a variety of biologically active compounds that might influence metabolic reactions. Most of the commonly ingested teas are derived from the leaf of the Camellia sinensis plant, and various types of tea are created via manipulations (eg, drying, fermentation) of this leaf. As green tea is fermented to oolong and then to black tea, polyphenol compounds (catechins) in green tea are dimerized to form a variety of theaflavins (1); thus, these teas may have different biological activities.

A putative beneficial effect of tea is its ability to induce weight loss. Support for this contention includes a controlled human trial that showed weight loss when tea was added to a dietary regimen (2) and a mouse study that showed that administration of a tea extract with a high-fat diet eliminated the weight gain observed in the absence of tea (3). Several mechanisms have been postulated to account for this weight control. Modest increases in energy expenditure have been reported with the ingestion of oolong and green teas (4–6). In addition, tea could inhibit the absorption of carbohydrate or fat. In vitro experiments have shown that constituents of tea inhibit the activities of α-amylase (7–10) and α-glucosidase (11–16) and of intestinal sodium-dependent glucose transporters (17–21). The in vitro inhibition of pancreatic lipase (22–24) by tea extracts suggests that tea might interfere with triacylglycerol absorption. However, no in vivo studies in humans or animals have shown that tea preparations cause malabsorption of carbohydrate or fat. In the present report, we measured breath hydrogen and 13CO2 to investigate the ability of an extract of black, green, and mulberry tea leaves to induce malabsorption of carbohydrate and triacylglycerol in healthy volunteers.

SUBJECTS AND METHODS

The study was approved by the Human Studies Subcommittee of the Minneapolis Veterans Affairs Medical Center, and informed consent was obtained from all subjects.

Study A: carbohydrate- and lipid-containing test meal

Twenty healthy volunteers (10 women and 10 men) aged 23–60 y fasted after their usual dinner until the following morning (~0800), when the experiments were performed at the Minneapolis Veterans Affairs Medical Center. After collection of baseline breath samples for hydrogen and 13CO2 analysis, the subjects ingested a test meal consisting of white rice and butter.

1 From NatureGen Inc, San Diego, CA (LZ), and VAMC (Research Service/151), Minneapolis, MN (JKF and MDL).
2 Supported by NatureGen Inc, San Diego, CA, and VAMC (Research Service/151), Minneapolis, MN.
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The rice was boiled for 20 min, and then individual portions (176 g containing 50 g carbohydrate) were frozen with 10 g butter. Immediately before being ingested, the meals were warmed in a microwave oven, and 0.2 g \(^{13}\)C\(_2\)stearate (Cambridge Isotope Laboratories, Andover, MA) was thoroughly mixed into the meal. Five hundred milliliters warm water and 10 g sucrose were added to the tea extract beverage or placebo preparation, which were well stirred. The subjects were randomly assigned to drink either the tea extract or the placebo beverage concurrently with the meal. Breath samples were then collected at hourly intervals for 8 h. At the end of each test period, the subjects were asked to rate a variety of symptoms—including nausea, bloating, abdominal discomfort, rectal gas, and obfuscating symptoms—on a previously described linear scale that ranged from 0 (none) to 4 (severe) (25). In addition, loose bowel movements were noted. One week later the test was repeated after the subjects ingested the opposite preparation from that ingested in the initial study.

**Study B: lipid-containing, carbohydrate-free test meal**

Ten of the subjects took part in a second study that followed the same format as study A; however, the caloric content of the meal consisted entirely of lipid (30 g olive oil plus 0.2 g \(^{13}\)C\(_2\)triolein). The tea leaf extract or placebo was similar to that described in the previous study; however, sucrose was replaced with 1 g of the noncaloric sweetener sucralose (Splenda McNeil Nutritional, Fort Washington, PA). Breath samples were obtained for \(^{13}\)CO\(_2\) measurements as described in study A.

**Test products**

The active preparation, a proprietary product, consisted of a mixture of extracts of green (0.1 g), black (0.1 g), and mulberry (1.0 g) tea leaves. The approximate quantities of the potential antiabsorptive components per dose of our tea extract beverage (measured by HPLC) were as follows: 5 mg deoxynojirimycin-type compounds, 100 mg epicatechin gallate, 300 mg epigallocatechin gallate, and 100 mg theaflavin. The control beverage consisted entirely of lipid (30 g olive oil plus 0.2 g \(^{13}\)C\(_2\)triolein). The taste of the 2 test materials differed, and subjects were aware of the preparation they received.

**Breath collections**

Expired air was sampled for hydrogen concentration as described previously (26). Breath samples for \(^{13}\)CO\(_2\) analysis were collected by having the subject expire through a straw into a glass tube (Labco Exetainer; Labco International Inc, Houston, TX), which was sealed immediately after withdrawal of the straw.

**Analyses**

Each breath collection for hydrogen measurement was analyzed for carbon dioxide (Capstar 100; CWE Inc, Ardmore, PA) to ensure that an adequate alveolar sample had been collected. The hydrogen concentration of the rare sample that contained <4.5% CO\(_2\) (5 of 360 samples) was normalized to 5% CO\(_2\) (observed hydrogen concentration \(\times\) 5%/observed carbon dioxide concentration). The hydrogen concentration was measured by gas chromatography with a molecular sieve column, nitrogen as the carrier gas, and a reduction detector (Trace Analytic, Menlo Park, CA). The atom percent (atom%) excess of \(^{13}\)CO\(_2\) in each breath sample relative to that of the baseline sample was determined by mass spectroscopy, which was performed by a commercial laboratory (Metabolic Solutions Inc, Nashua, NH).

**Statistics and calculations**

The significance of differences between means observed with the 2 treatments was determined by 2-tailed paired \(t\) test. The quantity of carbohydrate malabsorption induced by the tea extract preparation was estimated by first determining the difference between the sum of breath-hydrogen concentrations observed over hours 1–8 when subjects ingested tea extract or placebo. The grams of carbohydrate represented by this difference in hydrogen were estimated by comparison with the previously observed difference in the sum of hydrogen concentrations over hours 1–8 when 55 healthy subjects ingested 10 g lactulose or a noncaloric beverage (27). The excess sum of breath-hydrogen concentrations observed with 10 g lactulose averaged 6.2 \(\mu\)mol/L; thus, carbohydrate malabsorption induced by tea extract in the present study was estimated from the formula

\[
\text{Malabsorption (g)} = \frac{(\sum [H_2] \text{ hours } 1–8_{\text{tea extract}} - \sum [H_2] \text{ hours } 1–8_{\text{placebo}})}{10 \text{ g} / 6.2 \text{ \(\mu\)mol/L}}
\]

**RESULTS**

**Breath-hydrogen concentration**

The mean (±SEM) hourly breath-hydrogen concentrations observed after ingestion of the rice and butter meal with each of the 2 treatments are shown in **Figure 1**. The hydrogen concentration at baseline was not significantly different from that at 1 h. However, the curves significantly diverged by 2 h, with the breath-hydrogen concentration being significantly greater in the group receiving the tea extract beverage at each hourly time point from 2 to 8 h. The sum of the breath-hydrogen concentrations for hours 1–8 (a value that closely approximated the area under the

![Figure 1](image-url)
carbohydrate as rice, 10 g butter, and 0.2 g [13C]triolein with the tea extract. Study A: results for 20 healthy subjects who ingested a meal consisting of 50 g of carbohydrate in the meal over the 8-h test period. The significance of the differences was determined with a 2-tailed paired t test. The values were significantly greater in the tea extract group than in the placebo group (P = 0.014 at 5 h and P < 0.001 at 6–8 h). Study B: results for 10 subjects who ingested a carbohydrate-free meal consisting of 30 g olive oil and 0.2 g [13C]triolein with the tea extract or placebo solution, both of which contained 1 g sucralose (noncaloric sweetener). No significant differences were observed at any time point (P > 0.2).

Breath-[13C]CO2 measurements

The mean (±SEM) hourly [13C]atom% excesses (hourly values minus baseline value) for the 2 treatments when subjects ingested the rice and butter meal (study A) are shown in Figure 2. Although the measurements at hours 1–4 were not significantly different between the 2 treatments, the values were significantly higher for tea extract than for placebo at hours 5–8. The sum of the values for hours 1–8 averaged 0.0256 ± 0.0017 and 0.0213 ± 0.0019 atom% excess for the tea extract and the placebo, respectively (P = 0.014). The [13C]atom% excess values after ingestion of the lipid-containing (30 g olive oil plus 0.2 g [13C]triolein) carbohydrate-free meal (study B) are shown in Figure 2. In contrast with the results in study A, the sum of the values for hours 1–8 for the tea extract (0.012 ± 0.0025 atom% excess) was virtually identical to that with the placebo (0.012 ± 0.0023 atom% excess) (P = 0.95), and none of the hourly measurements showed significant differences (P > 0.2) between the 2 treatments.

**FIGURE 2.** Mean (±SEM) atom% excess of [13C] in expired air after ingestion of 2 different test meals: a tea extract (□) or a placebo (○). The significance of the differences was determined with a 2-tailed paired t test. Study A: results for 20 healthy subjects who ingested a meal consisting of 50 g carbohydrate as rice, 10 g butter, and 0.2 g [13C]triolein with the tea extract or placebo solution, both of which contained 10 g sucrose. Values at 0–4 h were not significantly different between the 2 treatment groups. The values were significantly greater in the tea extract group than in the placebo group at 5–8 h (P = 0.014 at 5 h and P < 0.001 at 6–8 h). Study B: results for 10 subjects who ingested a carbohydrate-free meal consisting of 30 g olive oil and 0.2 g [13C]triolein with the tea extract or placebo solution, both of which contained 1 g sucralose (noncaloric sweetener). No significant differences were observed at any time point (P > 0.2).

**TABLE 1**

Comparison of symptoms reported by healthy subjects in the 8-h period after ingestion of a standard carbohydrate- and lipid-containing meal plus a tea extract or placebo

<table>
<thead>
<tr>
<th>Symptom†</th>
<th>Tea extract group</th>
<th>Placebo group</th>
<th>P2</th>
</tr>
</thead>
<tbody>
<tr>
<td>Headache</td>
<td>1.16 ± 0.27†</td>
<td>0.71 ± 0.27</td>
<td>0.11</td>
</tr>
<tr>
<td>Fullness</td>
<td>0.77 ± 0.17</td>
<td>0.59 ± 0.19</td>
<td>0.44</td>
</tr>
<tr>
<td>Itching</td>
<td>0.07 ± 0.05</td>
<td>0.02 ± 0.02</td>
<td>0.33</td>
</tr>
<tr>
<td>Incomplete evacuation</td>
<td>0.23 ± 0.14</td>
<td>0.13 ± 0.10</td>
<td>0.33</td>
</tr>
<tr>
<td>Nausea</td>
<td>0.70 ± 0.23</td>
<td>0.23 ± 0.19</td>
<td>0.06</td>
</tr>
<tr>
<td>Excessive rectal gas</td>
<td>0.61 ± 0.21</td>
<td>0.21 ± 0.12</td>
<td>0.12</td>
</tr>
<tr>
<td>Fatigue</td>
<td>1.13 ± 0.25</td>
<td>0.97 ± 0.26</td>
<td>0.56</td>
</tr>
<tr>
<td>Bloating</td>
<td>0.45 ± 0.19</td>
<td>0.26 ± 0.13</td>
<td>0.31</td>
</tr>
<tr>
<td>Abdominal pain</td>
<td>0.41 ± 0.20</td>
<td>0.13 ± 0.17</td>
<td>0.67</td>
</tr>
</tbody>
</table>

†Symptoms were rated on a linear scale of 0 (none) to 4 (severe).

DISCUSSION

We used measurements of breath-hydrogen and of breath-[13C]CO2 to determine whether ingestion of a tea extract preparation induced malabsorption of carbohydrate or fat. Carbohydrate malabsorption provides substrate for most of the hydrogen produced in humans, which can be assessed by measuring breath-hydrogen concentrations (28, 29). In contrast, fat is not fermented to carbon dioxide by the colonic bacteria, and carbon dioxide production from lipid reflects the host’s metabolism of absorbed lipid. Studies using triolein labeled with [13C] (30, 31) or [14C] (32, 33) showed that fat malabsorption documented by fecal fat measurements was associated with a reduction in labeled carbon dioxide excretion.

In the present study, the subjects ingested standard meals with a beverage containing tea extract or placebo. The initial test meal contained 60 g carbohydrate (50 g starch as white rice and 10 g sucrose in the tea extract or the placebo) and 10.2 g fat. White rice was used as the complex carbohydrate because, in contrast with most complex carbohydrates, rice starch is nearly completely absorbed by healthy subjects (34). Thus, a rice meal allows breath testing to more sensitively determine whether a manipulation significantly increases hydrogen excretion, ie, causes starch malabsorption. As shown in Figure 1, the breath-hydrogen concentration declined with the placebo, which indicated that residual fermentable colonic substrate was not replenished via malabsorption of carbohydrate in the test meal. In contrast, the ingestion of tea extract resulted in increased breath-hydrogen concentrations, which were significantly greater than the values observed with placebo for each hourly measurement between 2 and 8 h. Thus, the tea extract clearly induced malabsorption of the carbohydrate.
The carbohydrate malabsorption induced by the tea extract was estimated by comparing the difference in breath-hydrogen concentrations between the tea extract and placebo groups with those observed previously (27) in healthy volunteers who ingested 10 g lactulose (see Equation 1). This calculation suggested that ≈15 g of the 60 g of carbohydrate in the test meal was not absorbed. This may be a low estimate because nonabsorbed material in the test meal could have been fermented less rapidly than is lactulose (35).

Elucidation of the mechanisms and specific tea extracts responsible for the carbohydrate malabsorption will require additional studies. Mulberry leaf contains alkaloids of the 1-deoxyxojirimycin type that inhibit intestinal α-glucosidase (14). Green tea supplies epicatechin gallate and epigallocatechin gallate, compounds that inhibit mucosal sodium-glucose transporters (17, 18). Black tea, via its theaflavin content, is an inhibitor of α-amylase (8). The quantities of these compounds in one dose of our tea extract preparation are equivalent (depending on the compound) to that which would be contained in 5–20 cups (1.2–4.8 L) of conventional tea. The extraction process reduced the caffeine of the tea extract to 50% of that of 1 cup (0.24 L) of conventional tea.

Breath-¹³CO₂ measurements did not support the concept that the tea extract inhibited triacylglycerol absorption. Rather, the ¹³CO₂ concentration was significantly greater when tea extract accompanied the standard meal (Figure 2), a finding that cannot be explained by tea extract enhancing the absorption of lipid because [¹³C]tri olein absorption should approach 100% with the placebo. One possible explanation is that the extract-induced carbohydrate malabsorption resulted in more rapid oxidation of newly absorbed, labeled lipid because of the lesser availability of glucose for energy utilization. It is also possible that the tea extract caused more rapid oxidation of absorbed [¹³C]oleic acid, independent of differences in carbohydrate absorption (4). To differentiate between these 2 possibilities, 10 subjects ingested a lipid-containing (30 g olive oil containing [¹³C]tri olein), carbohydrate-free meal plus the tea extract or placebo. No significant differences in [¹³C]CO₂ excretion were observed, which suggested that the higher ¹³CO₂ noted with the carbohydrate-containing meal reflected the influence of the extract-induced carbohydrate malabsorption on lipid metabolism.

The ability of a tea extract to inhibit carbohydrate absorption has potential clinical utility for weight control and the treatment of diabetes. Assuming that the tea extract causes malabsorption of 25% of ingested carbohydrate, striking weight loss would be expected providing that caloric intake was not commensurately increased and the caloric content of malabsorbed carbohydrate was unavailable to the host. Malabsorption of 25% of 400 g carbohydrate/d would reduce the caloric availability by ≈146 000 calories (16 kg fat) per year. Although it is commonly assumed that the host obtains no calories from materials entering the colon, the colonic absorption of carbohydrate fermentation products results in an appreciable conservation of calories (36). Thus, weight loss would be less than the predicted 16 kg/y.

For centuries, teas have been used as a treatment for diabetes mellitus in Asia. Multiple studies have shown that extracts of mulberry and other teas reduce blood glucose in type 2 diabetic persons (37–39) and in animal models of diabetes (40–43). This hypoglycemic effect generally has been attributed to alterations of the intermediary metabolism of glucose. The present study indicates that carbohydrate malabsorption induced by the tea extracts also could influence blood glucose concentrations. A similar observation has been reported with an extract of the root of _Salacia oblonga_ (44, 45). This extract reduces glucose absorption via inhibition of intestinal α-glucosidase by 2 compounds, salcinol and kotanol, that differ in structure from the α-glucosidase inhibitors in the tea preparation. It also should be noted that 2 α-glucosidase–inhibiting drugs of bacterial origin (acarbose and miglitol) are available for the treatment of diabetes. However, the use of these drugs in the United States has been limited by side effects (eg, gas and diarrhea) and by a relatively minor effect on blood glucose concentrations. The structure of α-glucosidase inhibitors in mulberry tea differs from that of acarbose, but is similar to that of miglitol. It remains to be determined whether carbohydrate malabsorption induced by our tea extract offers any benefits over those obtained with acarbose or miglitol. Although a significant increase in gaseous symptoms was not reported after ingestion of either the tea extract or the placebo (Table 1), studies in which tea extract is administered with each meal should be performed before it can be claimed that tea extract–induced carbohydrate malabsorption is associated with fewer symptoms than has been observed with α-glucosidase–inhibiting drugs. Indeed, it would be surprising if the degree of malabsorption observed with the tea extract (25% of total ingested carbohydrate) were not associated with some degree of gaseousness and loose stools.

Extracts of black, green, and mulberry teas have been consumed for many years by enormous numbers of Asians, and these products are considered safe. Green and black tea extracts also are widely used in the Western world. Although tea extracts have been shown to interact with the metabolism of other drugs (46, 47), serious complications possibly attributable to ingestion of these extracts are rare (48). Thus, although the potential for unintended serious side effects is seemingly low, rare unexpected side effects of the extract can be confidently excluded only after the product has been consumed in an environment where medical surveillance is adequate to detect the problem.

JFK helped design the protocol, recruited the subjects, and analyzed the data. MDL contributed to the design of the protocol, analyzed the data, and wrote the manuscript. LZ was involved in the design of the protocol but had no involvement in the collection or analysis of the data. LZ is president of NatureGen, the company that provided the tea extract and placebo used in this study. JFK and MDL had no financial interest in NatureGen or any other type of conflict of interest with the material presented in this article.

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