Attenuated Response of the Serum Triglyceride Concentration to Ingestion of a Chocolate Containing Polydextrose and Lactitol in Place of Sugar

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We examined the effects of ingesting a non-sugar chocolate containing polydextrose and lactitol in place of sucrose and lactose on the concentrations of plasma glucose and serum insulin and triglyceride in humans. A regular chocolate was used as the control. A crossover study was employed, and the subjects each ingested 46 g of the control or non-sugar chocolate in the experiments. Alterations in the blood components were monitored for a period of 150 min after ingestion. The control chocolate elevated the concentrations of plasma glucose and serum insulin, with the peak occurring 30 min after ingestion, but the non-sugar chocolate had a very minor effect. The serum triglyceride concentration gradually increased after ingesting the control chocolate, but was only slightly elevated 150 min after ingesting the non-sugar chocolate. An animal study also showed an attenuated response of serum triglyceride to the administration of a fat emulsion containing polydextrose and lactitol, suggesting that the triglyceride transit through the gut was promoted by these compounds. These results suggest that, compared to regular chocolate, fat absorption in the gut was less after ingesting the non-sugar chocolate, presumably resulting in less effect on body fat deposition.

Key words: non-sugar chocolate; plasma glucose; serum insulin; serum triglyceride; gut transit of fat

Chocolate is a palatable food consumed worldwide. Recent attention has been paid to the potential beneficial effects of chocolate on cardiovascular health,1) because cocoa and chocolate can be rich sources of flavonoids which have a variety of beneficial actions including as an antioxidant and modulation of vascular homeostasis.2) However, ingesting chocolate increases body fat, since the fat content of chocolate is relatively high and dietary fat is often blamed as the main dietary cause of obesity.3) A large number of people in developed countries suffer from lifestyle-related diseases, obesity and diabetes being typical examples.4) Such people have to control their body weight and elevation of blood glucose and insulin after ingesting food. Many foods have been developed for these people who need to control their body weight and blood glucose, and sugar substitutes are commonly used in these foods.

Since regular chocolate has a relatively high content of fat and sugar, non-sugar chocolate has recently been developed for those people who need to control their body weight.5) This chocolate contains polydextrose and lactitol as substitutes for sugar, and is enriched with calcium.5) It has been reported that the blood glucose level is little affected by the intake of polydextrose and lactitol6,7) and that calcium supplemented in the chocolate promotes the excretion of fatty acids into the feces in humans.5) Furthermore, it has been demonstrated in an animal study8) that polydextrose inhibits the absorption of fat by the small intestine. These findings suggest that the fate of fat in the non-sugar chocolate after its ingestion may be different from that in regular chocolate.

We examined in the present study the effect of ingesting non-sugar chocolate on the concentrations of plasma glucose and serum insulin and lipids in comparison to the effect from regular chocolate. We report here that the ingestion of non-sugar chocolate had a significantly smaller effect on the serum triglyceride (TG) concentration than that from regular chocolate. The

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Abbreviations: TG, triglyceride; FFA, free fatty acids; HDL, high-density lipoprotein; LDL, low-density lipoprotein; AUC, area under curve
mechanism responsible for this reduced response of serum TG was initially examined in rats.

Materials and Methods

Human study.

Subjects. Five healthy, non-diabetic volunteers participated in this study. All of the subjects were male students (21–23 years old) of the Nagoya Institute of Technology. Informed consent was obtained from each subject, and this study protocol conformed to the ethical guidelines of the 1975 Declaration of Helsinki as reflected in a priori approval by the human research review committee of the Nagoya University School of Medicine.

Compositions of the non-sugar and control chocolates. The non-sugar and control chocolates used in this study were prepared by Lotte Co., Ltd. The compositions of these preparations are given in Table 1. Both chocolates contained the same amount of fat, but the non-sugar chocolate contained polydextrose and lactitol in place of sucrose and lactose, and was supplemented with calcium derived from eggshell.

Experimental design. A crossover study was employed. The subjects were instructed not to perform intense exercise and to finish dinner by 21:00 h on the day before the experiment. After dinner, the subjects were allowed to take plain water only. On the day of the experiment, the subjects came to the laboratory by 8:30 h, ingested 46 g of chocolate (non-sugar or control) experiment, the subjects came to the laboratory by

Table 1. Components and Energy in the Control and Non-Sugar Chocolate Samples

<table>
<thead>
<tr>
<th>Component</th>
<th>Control</th>
<th>Non-sugar</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>% (w/w)</td>
<td></td>
</tr>
<tr>
<td>Cacao mass</td>
<td>27.4</td>
<td>27.4</td>
</tr>
<tr>
<td>Cocoa butter</td>
<td>9.1</td>
<td>9.1</td>
</tr>
<tr>
<td>Vegetable oil</td>
<td>6.9</td>
<td>6.9</td>
</tr>
<tr>
<td>Powdered milk&lt;sup&gt;1&lt;/sup&gt;</td>
<td>14.9</td>
<td>14.9</td>
</tr>
<tr>
<td>Sucrose</td>
<td>40.9</td>
<td>—</td>
</tr>
<tr>
<td>Lactitol</td>
<td>—</td>
<td>19.4</td>
</tr>
<tr>
<td>Polydextrose&lt;sup&gt;2&lt;/sup&gt;</td>
<td>—</td>
<td>15.1</td>
</tr>
<tr>
<td>Calcium derived from eggshell</td>
<td>—</td>
<td>4.9</td>
</tr>
<tr>
<td>Whey mineral</td>
<td>—</td>
<td>0.3</td>
</tr>
<tr>
<td>Sweetener</td>
<td>—</td>
<td>1.2</td>
</tr>
<tr>
<td>Others&lt;sup&gt;3&lt;/sup&gt;</td>
<td>0.8</td>
<td>0.8</td>
</tr>
<tr>
<td>Energy (kcal/chocolate ingested)&lt;sup&gt;4&lt;/sup&gt;</td>
<td>250</td>
<td>194</td>
</tr>
</tbody>
</table>

<sup>1</sup>Regular powdered milk and powdered milk without lactose were used for preparation of the control and non-sugar chocolates, respectively. The regular powdered milk consisted of 27.1% protein, 25.5% lipid, 38.9% carbohydrate (exclusively composed of lactose), 5.9% ash, and 2.6% water; powdered milk without lactose consisted of 24.5% protein, 26.5% lipid, 44% carbohydrate (41% polydextrose and 3% other carbohydrate without lactose), 2% ash, and 3% water.

<sup>2</sup>Powdered milk for the non-sugar chocolate contained polydextrose. The total polydextrose content in the non-sugar chocolate was 21.2%.

<sup>3</sup>Others components include emulsifier and flavoring.

<sup>4</sup>46 g of chocolate was ingested by each subject.

then remained relaxed, sitting on a chair for 150 min. Blood (about 4 ml) was taken just prior to ingesting the chocolate and 15, 30, 45, 60, 90, 120, and 150 min after ingesting (a total of eight timed samples).

The same subjects participated in both experiments (non-sugar and control chocolates) which were conducted with 1-week interval.

Analyses of plasma glucose and serum insulin and lipids were conducted by SRL Inc. (Tokyo, Japan). The total areas under the curves for plasma glucose, and serum insulin and TG were calculated according to the method of Wolever et al.<sup>9</sup>

Animal study.

Animals and fat emulsions used. Male Wistar rats aged 10 weeks were used. The chocolate was replaced with a fat emulsion in this study, the control fat emulsion (corresponding to the control chocolate) containing about 27% sucrose and the non-sugar fat emulsion (corresponding to the non-sugar chocolate) containing polydextrose + lactitol in place of sucrose, as shown in Table 2. All procedures involving the animals were approved by the experimental animal care committee of the Nagoya Institute of Technology.

Experimental design. The effect of administrating the fat emulsion on the serum glucose and TG concentrations was examined with 5 rats in each group. On the day of the experiment, the fat emulsion (3 ml/200 g of body weight) was orally administered at about 13:00 to rats that had been starved for 18 h. Blood (about 0.1 ml) was obtained from the tail vein before and 30, 60, 90, 120, and 150 min without anesthesia after administration. Serum was obtained from the blood sample, and the serum glucose and TG concentrations were measured by enzymatic methods.

Since the serum TG concentration in the control group was at the highest level 120 min after the fat emulsion had been administered (see Fig. 4), the remaining amount of TG in the gut was examined at this same time using another 5 rats in each group. The rats were kept under the same conditions and submitted to the same treatment as that already described. Rats

Table 2. Composition of the Fat Emulsions

<table>
<thead>
<tr>
<th>Component</th>
<th>Fat emulsion</th>
<th>Control</th>
<th>Non-sugar</th>
</tr>
</thead>
<tbody>
<tr>
<td>Non-salt butter</td>
<td>6.1</td>
<td>6.1</td>
<td></td>
</tr>
<tr>
<td>Cocoa butter</td>
<td>31.0</td>
<td>31.0</td>
<td></td>
</tr>
<tr>
<td>Vegetable oil</td>
<td>9.1</td>
<td>9.1</td>
<td></td>
</tr>
<tr>
<td>Sucrose</td>
<td>53.7</td>
<td>—</td>
<td></td>
</tr>
<tr>
<td>Lactitol</td>
<td>—</td>
<td>25.7</td>
<td></td>
</tr>
<tr>
<td>Polydextrose</td>
<td>—</td>
<td>28.0</td>
<td></td>
</tr>
<tr>
<td>Emulsifier</td>
<td>0.1</td>
<td>0.1</td>
<td></td>
</tr>
<tr>
<td>Distilled water</td>
<td>100</td>
<td>100</td>
<td></td>
</tr>
</tbody>
</table>

The rats were fasted overnight, before the fat emulsion (3 ml/200 g of body weight) was orally administered.
were then anesthetized with sodium pentobarbital (50 mg/kg of body weight) 120 min after the administration, and the stomach, small intestine, and cecum were divided by tying. The small intestine was further divided into its proximal and distal portions with approximately equal lengths by tying. The luminal contents of each segment were collected by using 10 mM sodium taurocholate as the rinsing solution, and the amount of TG in each sample was determined as reported previously.8) The heart and perirenal adipose tissue were removed, freeze-clamped at liquid-nitrogen temperature, and stored at −80°C until need for analyses. The lipoprotein lipase activity in the tissues was measured as reported previously,10) 1 unit of activity being defined as the amount of lipoprotein lipase that catalyzed the release of 1 μmol FFA per hour at 37°C.

Statistical analysis. Data obtained were analyzed for statistical significance by the Wilcoxon signed-rank test in the human study and by repeated ANOVA and Fisher PLSD in the rat study. P < 0.05 was considered to be significantly different.

Results and Discussion

Human study

It has been reported that polydextrose and lactitol had almost no effects on plasma glucose and serum insulin.6,7) The non-sugar chocolate included these substances in place of sucrose contained in the control (regular) chocolate. As we anticipated, control chocolate ingestion significantly elevated the plasma glucose and serum insulin concentrations with a clear peak 30 min after ingestion, while the non-sugar chocolate ingestion had only a minor effect on these blood components (Fig. 1). We calculated the areas under the curves (AUC) for plasma glucose and serum insulin, with both areas being much less from the non-sugar chocolate than from the control chocolate (Table 3).

Chocolate is a high-fat food, and both chocolate samples used in this study contained about 35% (w/w) fat. The control chocolate gradually elevated the serum TG concentration between 30 min and 150 min after ingestion (Fig. 2). On the other hand, the non-sugar chocolate had almost no effect on the TG concentration up to 120 min after ingestion and only slightly raised it by 150 min after ingestion (Fig. 2). AUC for the non-sugar chocolate was less than one fourth of that for the control chocolate (Table 3), suggesting that the availability of fat in the non-sugar chocolate may have been less than that in the control chocolate. This is a new finding obtained for humans.

The mechanism responsible for the attenuated serum TG response to the ingestion of non-sugar chocolate seems to involve the physiological action of polydextrose, lactitol, and calcium in the digestive tract. It has been reported that polydextrose retarded the absorption of TG in the small intestine of rats,5) and that lactitol and polydextrose promoted bowel movement.7,11,12) Furthermore, it has been suggested that calcium supplemented in non-sugar chocolate inhibited the absorption of saturated fatty acids by the digestive tract.5) These findings strongly suggest that polydextrose, lactitol and calcium supplemented in the non-sugar chocolate sample were responsible for the small rise in the serum TG concentration after ingestion. Meanwhile, none of the subjects had diarrhea after consuming the non-sugar chocolate in the present study, presumably because the consumption of lactitol and polydextrose was less than the reported amounts that can cause diarrhea.13,14)

We examined the effects of the control and non-sugar chocolate samples on the concentrations of serum free fatty acids (FFA) and on the total, high-density lipoprotein (HDL) and low-density lipoprotein (LDL) cholesterol. The ingestion of both chocolate samples
gradually decreased the serum FFA concentration, although there was no significant difference between them (data not shown). Concentrations of total, HDL and LDL cholesterol were not affected by the ingestion of either chocolate sample (data not shown).

Animal study
The rat study was conducted to elucidate the physiological mechanism responsible for the attenuated serum TG response to non-sugar chocolate ingestion by humans. It was confirmed that the administration of the control fat emulsion containing sucrose (corresponding to the control chocolate) raised the serum glucose and TG concentrations (Fig. 3). The serum TG concentration showed an apparent peak 120 min after administration. On the other hand, administration of the non-sugar fat emulsion containing polydextrose and lactitol induced less increase in the serum glucose than that of the control fat emulsion, and almost no effect on the serum TG concentration, similar to the effect observed in humans. The serum glucose concentration increased during the period from 0 to 30 min after administrating even the non-sugar fat emulsion, so this phenomenon may be attributable to handling the rats for blood sampling.

The remaining amount of TG in the luminal content of each segment of the digestive tract was measured 120 min after administrating the emulsion. In the control fat emulsion group, the greatest amount of TG was found in the distal small intestine, and the minimal amount in the cecum (Fig. 4). On the other hand, in the non-sugar fat emulsion group, the amount of TG was similar between the distal small intestine and cecum. The TG content in the distal small intestine was significantly less and that in the cecum was much greater in the non-sugar group than in the control group, and it was noted that some fat was excreted from the anus at the time of sacrifice by the non-sugar fat emulsion group. These findings suggest that the transit of fat through the gut was promoted by polydextrose and
lactitol. This effect of the sugar substitutes may have been involved in the mechanism responsible for the low response of serum TG to administration of the non-sugar fat emulsion.

The lipoprotein lipase activity plays an important role in the TG uptake into tissues, so the enzyme activities in the rat heart and perirenal adipose tissue were measured. The enzyme activities in the heart and adipose tissue were little different between the control and non-sugar groups (5.46 ± 0.70 vs. 5.72 ± 0.93 units/g of tissue for the heart and 1.26 ± 0.77 vs. 1.86 ± 0.60 for the adipose tissue, respectively). Therefore, the lipoprotein lipase activities in these tissues might not have contributed to the small response of serum TG to administration of the non-sugar fat emulsion.

In conclusion, we found attenuated responses to the ingestion of the non-sugar chocolate for not only plasma glucose and serum insulin, but also for the serum TG concentration when compared to responses to regular chocolate ingestion. The results of the rat study suggested that polydextrose and lactitol in the non-sugar chocolate may have promoted fat movement through the gut. These findings imply that the non-sugar chocolate may have less effect on body fat deposition than regular chocolate does.

References


