Effect of Statroltea on lipid metabolism in rats fed on high-fat diet

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Abstract: The present study was undertaken to evaluate the anti-obesity potential of a phenolic-rich herbal tea beverage produced in northern regions of Cameroon from the leaves of Stathmostelma sp. (Statroltea). The effect of Statroltea (5 mg/Kg/day) on lipid metabolism was assayed in male rats fed on either standard or high-fat diet for 60 days. Rats of both diet fed Statroltea presented significant (P < 0.05) reduced body weight than their controls with a rate of reduction varying from 5 to 15%. The consumption of Statroltea for 60 days significantly lowered abdominal fat index, blood total and LDL cholesterol, triglycerides and hepatic lipids (P < 0.05). Fecal lipids were found to be more excreted in all Statroltea fed rats. This is the first report on the effect of Stathmostelma sp. leaves on lipid metabolism and the study demonstrates that Statroltea, an herbal tea from Stathmostelma sp. leaves has a potential as anti-obesity functional food beverage.

Keywords: Obesity, Lipid Metabolism, Statroltea, Herbal Tea, Functional Food Beverage

1. Introduction

Obesity is a medical condition characterized by an excessive accumulation of fat in the body [1]. About 1.2 billion people worldwide are overweight and at least 300 million of them are obese with World Health Organization projecting that more than 700 million adults worldwide will be obese by 2015 [2]. Obesity is strongly associated with metabolic syndrome which is characterized by the presence of insulin resistance, hypertension, and hyperlipidemia [3]. Therefore, prevention and treatment of obesity are important for achieving a healthy life [1]. Owing to the adverse side effects associated with many antiobesity drugs, more recent trials have focused on screening herbal sources that have been reported to reduce body weight with minimal side effects [3].

Many teas (green tea, mate tea, etc.) have been widely used all over the world for weight reduction purpose [4, 5]. Their putative effects have been attributed to phenolic compounds which are reported to inhibit in vitro activity of pancreatic lipase, a key enzyme involved in the digestion of dietary lipids [6].

A phenolic-rich herbal tea beverage obtained from the leaves of Stathmostelma sp. (Statroltea) is produced in the northern regions of Cameroon. Feumba et al. [7] reported that Statroltea inhibits in vitro pancreatic lipase activity. However, no work has been reported on its in vivo activities. It is against this background that the study aims to investigate the in vivo anti-obesity potential of Statroltea.

2. Material and Methods

2.1. Production of Statroltea

The fresh leaves of Stathmostelma sp. were harvested in Ngaoundere, Cameroon and identified at the Cameroon National Herbarium (Voucher N° 59014). Leaves were roasted at 144°C for 20min and cooled to room temperature. Roasted leaves were ground and bagged in 3 grams. Bags containing 3g of roasted leaves were brewed for 23min in 300mL of preheated water at 60°C to give an infusion called in this study Statroltea. Statroltea was dried at 40°C to obtain a dry residue that was used in the study. The composition of Statroltea expressed in percentage of total soluble solids (TSS) is recorded in Table 1.
2.2. Effect of Statroltea on Lipid Profile of Rats

2.2.1. Experimental Animals

Male Wistar rats, aged 3–4 months, weighing 250–350g, were purchased from the animal house of the Laboratory of Animal Physiology, Faculty of Sciences of the University of Ngaoundere, Cameroon. Animals were maintained in standard laboratory conditions (23 ± 2°C, 12h photoperiod) having free access to tap water and food. Procedures used in the study were approved by the Animal Ethics Committee of the Ngaoundere University. Rats were weighed and examined for physical abnormalities a day before initiation of test.

2.2.2. Diet Administration

The experimentation was conducted during 60 days. Animals were fed either with high-fat diet (10 rats) or with standard diet (10 rats). The standard and the high-fat diets were formulated as reported in Table 2. For each diet, rats housed individually received distilled water (5 control rats) while treated groups (5 animals) received Statroltea at the dose of 5 mg/kg.

2.2.3. Statroltea Administration

Statroltea was administrated daily for 60 days. A mass of Statroltea (1.25-1.75mg) corresponding to the dose of 5mg/Kg of body weight was dissolved in 5mL of distilled water. The tea solution was mixed with 3g of appropriate meal and the tea treated-meal was given to each animal.

Table 1. Basic proximate and phenolic composition of Statroltea

<table>
<thead>
<tr>
<th>Nutrients</th>
<th>(mg% TSS)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total free sugars</td>
<td>349.68 ± 6.09</td>
</tr>
<tr>
<td>Reducing free sugars</td>
<td>330.57 ± 5.56</td>
</tr>
<tr>
<td>Crude proteins</td>
<td>666.87 ± 21.09</td>
</tr>
<tr>
<td>Phenolic compounds</td>
<td>(mg% TSS)</td>
</tr>
<tr>
<td>Total Phenolics</td>
<td>97.64 ± 1.35</td>
</tr>
<tr>
<td>Total flavonoids</td>
<td>68.10 ± 3.59</td>
</tr>
<tr>
<td>Total tannins</td>
<td>16.17 ± 0.41</td>
</tr>
</tbody>
</table>

Values represent mean ± SD of 3 replicates; Total phenolics were expressed as mg gallic acid equivalents % TSS; total flavonoids were expressed as mg quercetin equivalents % TSS; Total tannins were expressed as tannic acid equivalents % TSS; Total free sugars and reducing free sugars were expressed as fructose equivalents % TSS; Crude proteins were expressed as N x 6.25.

Table 2. Composition of standard and high-fat diets

<table>
<thead>
<tr>
<th>Ingredients (%)</th>
<th>Standard diet</th>
<th>High-fat diet</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cassava starch</td>
<td>69.5</td>
<td>39.5</td>
</tr>
<tr>
<td>Casein</td>
<td>21.0</td>
<td>21.0</td>
</tr>
<tr>
<td>Soy bean oil</td>
<td>5.0</td>
<td>5.0</td>
</tr>
<tr>
<td>Palm oil</td>
<td>0.0</td>
<td>30.0</td>
</tr>
<tr>
<td>Vitamin mix</td>
<td>1.0</td>
<td>1.0</td>
</tr>
<tr>
<td>Mineral mix</td>
<td>3.5</td>
<td>3.5</td>
</tr>
</tbody>
</table>

Rats were given ad libitum portion of untreated standard or high-fat meal after they have completely eaten the tea-treated meal.

2.2.4. Measurement of Food Intake and Variation in Body Weight

The food intake of all animals was assessed daily and the body weights of rats were recorded every ten days after which variations in body weights were calculated.

2.2.5. Measurement of Blood Lipids

At the end of the treatment, animals were fasted overnight and anesthetized with chloroform and blood collection from the jugular vein. Blood samples were collected into heparinized centrifuge tubes. The level of total cholesterol, triglycerides, and HDL-cholesterol were measured using commercial enzymatic kits (Spinreact, Spain) and the LDL-cholesterol concentration in the plasma was calculated using the equation described by Friedewald et al. [8].

2.2.6. Measurement of Abdominal Fat Index, Hepatic and Fecal Lipids

At the end of the experimental period, abdominal fat and liver were excised, washed in NaCl (0.9%) and weighed. The abdominal fat index was represented by the ratio abdominal fat/body weight. Feces collected every ten days during the course of the experiment were dried in an oven at 50°C, ground and homogenised. To determine hepatic or faecal lipid content, a sample of ground liver or feces was extracted in 5mL of the mixture of chloroform: methanol (2:1 v/v), using the method described by Folch et al. [9].

2.3. Statistical Analysis

Statistical analysis was performed for all parameters. One-way analysis of variance (ANOVA) was used to determine statistical difference between control and treated groups with P values less than 0.05 considered as significant.

3. Results and Discussion

3.1. Effect of Statroltea on Body Weight

The ingestion of high-fat diet resulted in gradual increase in body weight with time whereas there was no significant change in body weight in those fed on the standard diet (Fig. 1). This result is in accordance with that of Amin and Nagy [10] who reported that feeding rats on high fat diet significantly increased their body weight. The consumption of Statroltea has brought the variation in body weight from a positive to a negative rate with a percentage of reduction situated between 5 to 15%. According to Grundy et al. [11], it has been recommended that weight reduction programs should focus on achieving a modest weight loss of 7–10% of the initial weight. Statroltea can therefore be used in weight reduction programs. This reductive effect in body weight of rats by Statroltea is irrespective of the type of diet because the ingestion of Statroltea did produced weight losses in all the Statroltea-treated groups irrespective of the diet consumed with these weight losses occurring from the
beginning of the ingestion of Statroltea. Similarly, Sogawa et al. [12] found that low energy diet containing 3% of green tea or 3% tokushima tea produced significant weight loss in male rats. In the same light, Hala et al. [13] reported that 5mL of green tea extract prepared freshly and given three times daily to a high fat fed male rats produced a significant decrease in their body weight gain. Kao et al. [14] also reported that purified EGCG (50 – 100mg/kg), a green tea catechin, significantly reduced or prevented an increase in the body weight of lean and obese Zucker rats with the weight losses attributed to food intake reduction.

Figure 1. Variation in the body weight of male rats fed standard or high-fat diet with or without Statroltea

3.2. Effect of Statroltea on Food Intake and Abdominal Fat Index

Food intake is regulated by a variety of endocrine parameters (leptin, neuropeptides, glucagon, etc.) which are influenced by peripheral factors and by central neuroendocrine systems [15]. From Table 3, it is noticeable that Statroltea produced an increase in the food consumption of rats fed with the standard diet or the high-fat diet. Statroltea may influence any components involved in food intake. On the other hand, Bajerska et al. [16] found that the administration of 1.1% and 2.2% of green tea aqueous extract to high-fat diet fed rats produced no change in their food intake. In normal fed diet rats, Nakamura et al. [17] reported no change in food intake when green tea polyphenol (0.01, 0.05, 0.1, 0.2, 0.5 and 1.0 g/kg) are consumed. The body fat index (Table 3) depicts that final parametrial adipose tissue weight was significantly increased by feeding high-fat diet to male rats compared to their controls (P < 0.05). Statroltea produced a relative reduction of 15.88% of this abdominal fat accumulation in male rats. This reduction of body fat index produced by Statroltea is concomitant with several reports on effects of traditional Camellia sinensis tea components on body fat accumulation.

Table 3. Effect of Statroltea on food consumption and abdominal fat index of rats

<table>
<thead>
<tr>
<th>Diet</th>
<th>Groups</th>
<th>Food consumption (mg/g BW/day)</th>
<th>Abdominal index (mg/ g BW)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Standard</td>
<td>Control</td>
<td>186.79 ± 15.55</td>
<td>19.46 ± 1.27</td>
</tr>
<tr>
<td>diet</td>
<td>Statroltea</td>
<td>290.08 ± 50.00^b</td>
<td>20.54 ± 1.30</td>
</tr>
<tr>
<td>High-fat</td>
<td>Control</td>
<td>174.82 ± 3.66</td>
<td>43.46 ± 3.63^a</td>
</tr>
<tr>
<td>diet</td>
<td>Statroltea</td>
<td>284.10 ± 5.82^b</td>
<td>35.69 ± 2.75^b</td>
</tr>
</tbody>
</table>

a. Value significantly different from the control of the standard diet group (P < 0.05)
b. Value significantly different from the control within the same diet group (P < 0.05)

Table 4. Effect of Statroltea on blood lipids of male rats

<table>
<thead>
<tr>
<th>Blood lipids</th>
<th>Standard diet</th>
<th>Statroltea</th>
<th>High-fat diet</th>
<th>Statroltea</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Control</td>
<td></td>
<td>Control</td>
<td></td>
</tr>
<tr>
<td>Total cholesterol (mg/ dL)</td>
<td>76.10 ± 1.13</td>
<td>58.48 ± 1.88^b (21.87%)</td>
<td>83.04 ± 3.40^a</td>
<td>45.93 ± 7.71^b (37.30%)</td>
</tr>
<tr>
<td>Triglycerides (mg/ dL)</td>
<td>135.12 ± 2.50</td>
<td>123.47 ± 4.16^b (5.71%)</td>
<td>140.54 ± 10.09</td>
<td>114.12 ± 3.33^b (18.80%)</td>
</tr>
<tr>
<td>HDL- cholesterol (mg/ dL)</td>
<td>27.09 ± 2.37</td>
<td>23.66 ± 2.16</td>
<td>24.12 ± 3.28</td>
<td>22.90 ± 7.02</td>
</tr>
<tr>
<td>LDL- cholesterol (mg/ dL)</td>
<td>16.93 ± 1.22</td>
<td>9.52 ± 1.10^b (43.77%)</td>
<td>33.81 ± 7.76^c</td>
<td>12.84 ± 3.17^b (55.18%)</td>
</tr>
<tr>
<td>Atherogenic Index (1)</td>
<td>2.93 ± 0.48</td>
<td>2.28 ± 0.10^b (19.06%)</td>
<td>4.05 ± 0.89^a</td>
<td>2.01 ± 0.11^a (49.97%)</td>
</tr>
<tr>
<td>Atherogenic Index (2)</td>
<td>0.76 ± 0.13</td>
<td>0.41 ± 0.08^b (45.86%)</td>
<td>1.56 ± 0.21^c</td>
<td>0.60 ± 0.17^b (59.53%)</td>
</tr>
</tbody>
</table>

a. Value significantly different from the control of the standard diet group (P < 0.05)
b. Value significantly different from the control within the same diet group (P < 0.05)

d. Value significantly different from the control of the high-fat diet group (P < 0.05)

Han et al. [4] found that the parametrial adipose weight significantly decreased from 2.08 to 1.04 after adding 0.5% Thea sinensis saponins to the high-fat diet for 11 weeks. Ikeda et al. [18] observed a relative decrease of 21.85% in adipose tissue weight or visceral fat deposition after administration of 1% tea catechins for 23 days to rats.
According to Tokimitsu [19], long term ingestion of tea catechins stopped accumulation of body fat in mice with high-fat diet induced obesity. However this effect was also found in non obese rats and was probably due to the activation of hepatic lipid metabolism [20].

3.3. Effect of Statroltea on Blood Lipids

The effects of Statroltea on blood lipids profile are recorded in Table 4.

It can be observed that consuming a high fat diet resulted in an increase in total cholesterol and LDL cholesterol in rats compared to rats fed on standard diet. This observation is in agreement with Woo et al. [21] who stated that in animal models of high fat diets, dramatic increases in serum total cholesterol and LDL-cholesterol but relatively slight changes in HDL-cholesterol were observed. The role of Statroltea in countering lipemic aberrations accompanying diet-induced hyperlipidemia was investigated and recorded in Table 4. The consumption of Statroltea for 60 days significantly lowered both total and LDL cholesterol (P < 0.05) and the relative reduction rates in all the Statroltea treated groups were respectively above 20 and 40%. Similarly, Zdunczyk et al. [22] stated that an extract of catechin from green tea was found to decrease the total cholesterol and LDL-fraction. By reducing total cholesterol and LDL cholesterol, Statroltea would reduce the incidence of coronary events [23].

Atherogenic index indicates the deposition of foam cells or plaque or fatty infiltration or lipids in heart, coronaries, aorta, liver and kidneys. It was observed that, the higher the atherogenic index was, the higher the risk of oxidative damage of the above organs. Table 4 shows that the consumption of a high-fat diet significantly increased atherogenic indexes exposing high-fat fed animals to more elevated risk of metabolic diseases. Statroltea ingestion significantly reduced atherogenic indexes for both diet (P < 0.05). Ramadan et al. [24] reported that black tea and green tea extracts given at concentrations of 50 and 100mg/Kg to male rats significantly reduced in dose dependant manner atherogenic indexes in obese groups but brought no significant change in standard diet fed rats. It is worthy to mention that the reduction of atherogenic indexes by Statroltea was relatively more marked in high-fat fed groups than in standard fed groups.

On the other hand, the concentration of HDL cholesterol in all the groups was not affected by Statroltea. This effect may be due to regularity in the activity of lecithin:cholesterol acyl transferase (LCAT), which may contribute to the regulation of blood lipids. LCAT plays a key role in incorporating free cholesterol into HDL and transferring back to VLDL or IDL, which is taken back by the liver cells [25].

Table 4 also revealed significant reduction in triglyceride levels in all the Statroltea fed rats. This effect may be related to the inhibition of enzymes responsible of fat metabolism (pancreatic lipase, lipoprotein lipase, and glycerophosphate dehydrogenase) since polyphenols from herbal sources have been reported to inhibit these enzymes [26, 27]. These observations were consistent with Uchimaya et al. [28] who found that total plasma cholesterol and triglyceride levels in mice fed a standard diet containing 5% black tea polyphenol extracts were respectively reduced to 79% and 67.5%. Controversially, Murase et al. [29] reported that high-fat diet supplemented for 11 months with green tea catechins up to 0.5% produced a significant decrease in total cholesterol but did not alter plasma triglyceride in mice.

In the same line, De Vos and De Schrijver [30] found that the consumption of black tea by rats significantly lowered esterified and total cholesterol concentrations in plasma, while contents of free cholesterol and triglycerides were not influenced. The difference observed between the effect of Statroltea and that of green or black tea may be due to the amount of tea ingested, their content and composition in polyphenols. It can also be due to their content and composition in others bioactive compounds beside polyphenols which may act synergistically to influence lipid metabolism.

3.4. Effect of Statroltea on Liver Lipids

The behaviour of liver lipids after ingestion of Statroltea is displayed in Table 5. A high-fat diet may induce hepatic triglycerides accumulation, owing to the import of excess amounts of fatty acids into the liver. The excess fatty acids are then esterified and stored as triglycerides [31]. It can be observed that the deposition of lipids in the liver is reduced by the consumption of Statroltea administrated at 5 mg/Kg irrespectively of the type of diet (P > 0.05). This is consistent with Uchiyama et al. [28] who reported that giving normal or high-fat diet supplemented with black tea polyphenols to rats did not decrease liver total lipids until the supplemented concentration reached 5%. In the same light, Nakamura et al. [17] stated that feeding animals with green tea polyphenols from 0.01 to 0.2g/Kg produced no significant changes in hepatic lipids parameters (total cholesterol, triglycerides, phospholipids), but higher concentrations (0.5 to 1.0g/Kg) provoked a decrease in hepatic triglycerides. This decrease in hepatic triglyceride is possibly due according to Kobayashi-Hattori et al. [32] to an increased hepatic-β oxidation activity.

3.5. Effect of Statroltea on Fecal Lipids

Fig. 2 displays the lipids excretion during the study. It can be observed that the pattern of fecal lipid excretion did not differ with the diet under study. For both diet, Statroltea ingestion increased significantly (P < 0.05) the amount of excreted fecal lipids. This is concomitant of Uchiyama et al. [28] who reported that fecal excretion in female mice fed a high fat diet containing 5% black tea polyphenols extract increased 1.7-fold compared to mice only fed a high fat diet. In the same light, Hsu et al. [33] found that lipid excretion into feces was significantly higher in the polyphenol-enriched oolong tea period (12.9 - 19.3g/ 3
days) than in the placebo period (7.3 - 9.4g/3 days).

Table 5. Effect of Statroltea on liver lipids of rats

<table>
<thead>
<tr>
<th>Liver lipids</th>
<th>Standard diet</th>
<th>Control</th>
<th>High- fat diet</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Control</td>
<td>Statroltea</td>
<td>Control</td>
</tr>
<tr>
<td>Total lipids (mg/g of tissue)</td>
<td>74.16 ± 3.20</td>
<td>61.74 ± 0.09(a)</td>
<td>82.53 ± 5.28</td>
</tr>
<tr>
<td>Triglycerides (mg/g of tissue)</td>
<td>1.29 ± 0.06</td>
<td>1.08 ± 0.10(a)</td>
<td>1.83 ± 0.06(a)</td>
</tr>
<tr>
<td>Total cholesterol (mg/g of tissue)</td>
<td>0.89 ± 0.06</td>
<td>0.75 ± 0.03(b)</td>
<td>1.57 ± 0.06(a)</td>
</tr>
</tbody>
</table>

\(a\) Value significantly different from the control of the standard diet group (P < 0.05)

\(b\) Value significantly different from the control within the same diet group (P < 0.05)

4. Conclusion

The present study demonstrates that the herbal tea beverage from *Stathmostelma sp.* regulates lipid metabolism and has a potential in the prevention of high-fat diet-induced obesity. The related effects are significant reduction of body weight, abdominal fat index, blood total cholesterol, LDL-cholesterol, triglycerides, hepatic lipids and significant increase in fecal lipids. This is the first report on the in vivo effect of Statroltea and the study demonstrates that Statroltea has a potentiality as anti-obesity herbal tea. Further studies are necessary to determine the mechanism of action of Statroltea on all enzymes implicated in the lipid metabolism and to ascertain its safety.

References


