Protective Effect of Green or Black Tea on D-Galactosamine Induced Liver Injury in Male Wistar Rats

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Abstract: Tea is the second most common drink in the world and could have a large impact on public health. In this study, the protection effect of green or black tea on D-Galactosamine (GalN) induced liver injury was determined. Hepatotoxic parameters (AST, ALT and GGT) were subsequently examined. Therefore, 90 male Wistar rats were divided into three groups, drank water, 2% green tea or 2% black tea. After 4 weeks, each group was divided into two subgroups, one injected with GalN intraperitoneally while the other group was injected with saline. Blood analysis indicated that rats treated with GalN had higher activity of AST, ALT and GGT (79.64 %, 200.43 % and 38.46 % respectively) which mean that GalN administration at a dose of 350 mg/kg body weight of rat was able to induce hepatic injury. Consumption of either green or black tea in liver injury-induced rats participates in suppressing the elevation of the serum AST and ALT activities, as indicated by a non-significant change in these enzymes activities after GalN injection. Green tea was effective in diminishing hepatotoxic indices, such as the significant reduction of serum AST and ALT activities by 59.08% and 70.4% respectively, while GGT activity reduced non-significantly by 1.74% as compared with the group of treated rats drinking water. The same reduction in the activities of the previous enzymes was observed in treated rats drinking black tea as 38.53%, 73.59% and 9.38% for AST, ALT and GGT respectively. Accordingly, it was clear that both types of tea can significantly suppress the enhancement of activities of the transferase enzymes which were induced by GalN. Moreover, green tea was more effective in hepatotoxicity protection than black tea. Furthermore, even in untreated rats, green tea was able to decrease AST activity significantly by 27.96% as compared to the control group.

Key words: Male Wistar rats, Hepatotoxic parameters, D-Galactosamine, black tea, green tea, liver.

1. Introduction:

Botanical medicines have been used traditionally by herbalists and indigenous healers worldwide for the prevention and treatment of liver disease (Zhang et al., 2013). Clinical research, in this century, has confirmed the efficacy of several plants in the treatment of liver disease, while basic scientific research has uncovered the mechanisms by which some plants provide their therapeutic effects.

Treatment options for common liver diseases such as cirrhosis, fatty liver, and chronic hepatitis are problematic. The effectiveness of treatments such as interferon, colchicine, penicillamine, and corticosteroids are inconsistent at best, and the incidence of side-effects profound. In recent years, researchers have examined the effects of plants used traditionally by indigenous healers and herbalists to support liver function and treat diseases of the liver (Zhang et al., 2013). The influence of tea consumption on lipid profile was detected and there is an inverse correlation between tea consumption and concentration of serum total cholesterol (Imai and Nakachi, 1995). The antioxidant Flavonoids and polyphenols in tea are several times more potent than Vitamin C or E (Vinson, 1998, Harborne, 1994) and may prevent blood platelets from clumping and blocking arteries. Moreover, Green and Harari (1992) demonstrated a decrease in LDL-C and an increase in HDL-C by green tea consumption.

Consumption of tea has been associated with many health benefits including the prevention of cancer due to polyphenols (Caderni et al., 2000). Many theories are focusing on the antioxidant and antiproliferative effects of polyphenolic compounds. It is also thought that these polyphenols may inhibit carcinogenesis by blocking the indogenous formation of N-nitroso compounds (Yang and Wang, 1993, Komori et al., 1993). Green tea has effects on body weight (Kao et al., 2000) and energy expenditure (Dullo et al., 1999). The weight-loss effect of EGCG in rats may have been due to a reduction in food intake and Male Sprague-Dawley rats, given EGCG orally, consumed 15% less food than did the control rats and lost 5% of their initial body weight (Kao et al., 2000).

The role of catechins in promoting weight loss was previously investigated in studies by Dulloo et al., (1999) who found that the mice receiving the green tea in their diets had a significant suppression of food intake, body weight gain and fat tissue accumulation. Leptin levels in serum showed a decrease with green
tea treatments- indicating that green tea may have a direct effect on the regulation of body weight. Tea is the oldest known medicine used for its stimulating and detoxifying properties in the elimination of alcohol and toxins, to improve blood and urine flow, to relieve joint pains, and to improve resistance to diseases (Balentine et al., 1997).

The objective of this study is to show, if the tea consumption is acting as a protectant against liver injury induced by GalN injection. In addition, comparing the effect of green and black in acting as liver protectant against injuries was determined. Therefore, several aspects of hepatotoxicity were measured in blood including AST, ALT, and GGT activities.

2. Material and Methods

D-galactosamine hydrochloride (C6H13NO5.HCl) was obtained from Sigma Chemical Co. Lipton green and black tea bags were obtained from the market in Jeddah. Biochemical kits for the determination of blood chemistry and enzymology were obtained from Crescent Diagnostics, KSA.

Animals

This study was carried out using a total number of 90 male Wister rats aged 8 weeks, supplied by King Fahd Medical Research Center, with a mean initial body weight of 189.57 ±1.12 g. They were randomly divided into three different groups; each group consisted of 30. They were untreated group or control group (C), green tea group (GT) which received 2% solution of green tea, Black tea group (BT) which received 2% solution of black tea. After 4 weeks, rats of each group were divided into two subgroups Control subgroup which was injected with a saline solution and treated subgroup which was injected intraperitoneally with GalN at a dose of 350 mg/kg body weight (Sugiyama et al., 1999). Rats of each subgroup were randomly distributed into cages with 5 rats per cage at a controlled temperature (24 ± 1°C), 70% relative humidity, and air flow conditions with fixed 12 hour light- dark cycles. Rats were acclimatized to the basic diet for one week before the experiment started. All the six subgroups of rats received the same basic diet in pellet form Grain Silos and Flour Mills organization, Jeddah, Saudi Arabia. This diet contained 8.44% moisture, 5.59% ash, 19.91% protein, 3.09% fat, 59.55% soluble carbohydrates and 4.08 fibers.

Tea solutions were prepared by first boiling 100 ml of water for 5 minutes to eliminate dissolved gases, including oxygen. Then, 2g of tea was added, and the mixtures maintained near the boiling point for exactly 5 minutes. The solutions were filtered and placed in dark glass water feeding bottles. Rats were maintained in this experiment for 4 weeks.

Blood samples collection

Blood samples were individually obtained from the 90 rats after 22 hours of the injection (Sugiyama et al., 1999). Blood samples were collected into dry clean centrifuge tubes by cardiac puncture under diethyl ether anesthesia. The tubes were kept at room temperature for approximately half an hour to allow blood clotting. The tubes were then centrifuged at 3000 rpm for 20 min. Clear serum was carefully separated using Pasteur pipettes, divided into aliquots and subsequently stored at -30 °C until analyses.

Biochemical Analyses

Reagents for the biochemical analysis were obtained from Crescent Diagnostics, KSA. The biochemical analyses were measured by Pharmacia Biotech Ultrospec 2000 spectrophotometer.

a. Determination of serum aspartate aminotransferase activity

Aspartate aminotransferase activity (AST) is utilized in the diagnosis of myocardial and hepatic disease. The reaction rate is monitored by observing the decrease in absorbance at 340 nm with time. A large excess of malate dehydrogenase in the reaction mixture assures that the measurement of the oxaloacetic acid is rate limiting and thus reflects the activity of AST (Tietz, 1995)

b. Determination of serum alanine aminotransferase activity

Alanine aminotransferase activity (ALT) is used primarily as a marker of hepatocellular damage and it was measured as described by Bablok (1988).

c. Determination of serum γ-glutamyltransferase

γ-Glutamyltransferase (GGT) is responsible for amino acid transport in the kidney, pancreas, liver, spleen, lungs, brain, intestine and heart. The normal serum GGT originates in the liver. Most elevations of GGT are due to liver disease. GGT catalyzes the transfer of the γ-Glutamyl group from the substrate γ-Glutamyl-3-carboxy-4-nitroanilide to glycylglycine yielding 5-amino-2-nitrobenzoate which absorbs at 405-410 nm. The change in absorbance at 405-410 nm due to the formation of the 5-amino-2-nitrobenzoate is directly proportional to the GGT activity in the sample (Tietz, 1995).

Statistical analysis

Statistical analyses of the data were performed and graphs were created on the IBM compatible computer using Statgraphics computer program package, SPSS. All data are expressed as mean with their standard deviations. One-way analysis of variance (ANOVA) was used to test for differences between experimental groups. When the ANOVA test was significant, it was followed by the least significance difference (LSD) test. The Independent-Samples T Test for paired data was employed to compare pairs of mean
values. Differences were considered significant at P< 0.05.

3. Results

In this study, we first examined if tea drinking was effective in preventing liver injury induced by GalN injection as assessed by AST, ALT and GGT activities. Rats were given free access to green and black tea with or without GalN injection (treated and untreated rats). The injection was done at the end of the experimental period (4 weeks). Weights and food consumption of animals were determined weekly. There was no mortality in animals at all, during the experimental period. Table 1 and figures 1, 2 and 3 showed the effects of green tea and black tea consumption on serum AST, ALT and GGT activities in untreated and GalN-treated rats.

Table 1: Effect of green tea and black tea consumption on serum aspartate aminotransferase, alanine aminotransferase and γ-glutamyl transferase activities in untreated and galactosamine treated rats.

<table>
<thead>
<tr>
<th></th>
<th>Groups</th>
<th>Untreated rats</th>
<th>Treated rats</th>
<th>Significance</th>
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<tbody>
<tr>
<td>AST [U/l]</td>
<td>C</td>
<td>52.07 ± 4.88</td>
<td>93.54 ± 9.00</td>
<td>*</td>
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<tr>
<td></td>
<td>GT</td>
<td>37.51 ± 1.50</td>
<td>38.28 ± 1.25</td>
<td>NS</td>
</tr>
<tr>
<td></td>
<td>BT</td>
<td>51.79 ± 2.95</td>
<td>57.50 ± 1.36</td>
<td>NS</td>
</tr>
<tr>
<td>ALT [U/l]</td>
<td>C</td>
<td>25.46 ± 15.62</td>
<td>76.49 ± 10.90</td>
<td>*</td>
</tr>
<tr>
<td></td>
<td>GT</td>
<td>22.43 ± 13.90</td>
<td>22.64 ± 14.60</td>
<td>NS</td>
</tr>
<tr>
<td></td>
<td>BT</td>
<td>18.08 ± 8.22</td>
<td>20.20 ± 6.90</td>
<td>NS</td>
</tr>
<tr>
<td>GGT [U/l]</td>
<td>C</td>
<td>2.08 ± 1.7</td>
<td>2.88 ± 2.14</td>
<td>*</td>
</tr>
<tr>
<td></td>
<td>GT</td>
<td>2.25 ± 1.03</td>
<td>2.83 ± 1.19</td>
<td>NS</td>
</tr>
<tr>
<td></td>
<td>BT</td>
<td>2.32 ± 1.80</td>
<td>2.61 ± 1.07</td>
<td>NS</td>
</tr>
</tbody>
</table>

Values are presented as means with their standard deviations, n=5, NS: Non-significant; *: significant at p< 0.05; AST: Aspartate aminotransferase; ALT: Alanine aminotransferase; GGT: γ-glutamyl transferase, C: Rats drinking water; GT: Rats drinking green tea; BT: Rats drinking black tea.

Figure 1: Effect of green tea and black tea consumption on serum aspartate aminotransferase activities in untreated and galactosamine treated rats.
C: Rats drinking water; GT: Rats drinking green tea; BT: Rats drinking black tea. C+ d: Treated rats drinking water; GT+ d: Treated rats drinking green tea; BT+ d: Treated rats drinking black tea.

Figure 2: Effect of green tea and black tea consumption on serum alanine aminotransferase activities in untreated and galactosamine treated rats.
C: Rats drinking water; GT: Rats drinking green tea; BT: Rats drinking black tea, C+ d: Treated rats drinking water; GT+ d: Treated rats drinking green tea; BT+ d: Treated rats drinking black tea.
Figure 3: Effect of green tea and black tea consumption on serum γ-glutamyl aminotransferase activities in untreated and galactosamine treated rats.
C: Rats drinking water; GT: Rats drinking green tea; BT: Rats drinking black tea, C+ d: Treated rats drinking water; GT+ d: Treated rats drinking green tea; BT+ d: Treated rats drinking black tea.

Injection of GalN caused a significant increase in the AST, ALT and GGT activities compared to control groups. On the other hand, GalN treatment failed to induce any significant change in the aminotransferase enzymes and GGT enzyme of the green and black tea group. The three experimental groups differ significantly in the activity of AST. Furthermore, the GalN-treated rats drinking green tea recorded the lowest activity of AST (38.28 ± 1.25 U/l), while the highest activity was observed in GalN-treated rats drinking water (93.54± 9.00 U/l). No statistical differences were apparent in ALT activity between the green tea and the black tea groups. But they were significantly lower than the control group. The results did not note any statistical difference between all the groups of the GalN-treated rats in respect to GGT activity.

Rats consuming green tea were significantly lower in AST activity (37.51 ± 1.50 U/l) than rats consuming either water (52.07 ± 4.88 U/l) or black tea (51.79± 2.95U/l). On the other hand, there were no statistical differences recorded between rats drinking water, green tea or black tea in either serum ALT or GGT activities.

Table 2 and figure 4 showed the effect of green tea and black tea consumption on body weight of untreated rats. At the beginning of the experimental period the average body weights of rats were approximately similar in all groups (189.57 ±1.12 g). Our data showed that there were significant differences in body weight of rats between the three groups during the first week. While during the last three weeks, of the experimental period, the green tea and black tea groups did not differ significantly in body weight and they were significantly lower than the control group.

Table 2: Effect of green and black tea consumption on body weight (g) of untreated rats

<table>
<thead>
<tr>
<th>Groups</th>
<th>Weeks</th>
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<tbody>
<tr>
<td></td>
<td>0</td>
</tr>
<tr>
<td>C</td>
<td>189.37 ± 16.54</td>
</tr>
<tr>
<td>GT</td>
<td>190.60 ± 14.08</td>
</tr>
<tr>
<td>BT</td>
<td>188.37 ± 12.09</td>
</tr>
</tbody>
</table>

Values are presented as means with their standard deviations, n=30, Mean values in the same column with different superscript letters were significantly different (p<0.05; one-way ANOVA and LSD-tests). C: Rats drinking water; GT: Rats drinking green tea; BT: Rats drinking black tea.

Table 3 and figure 5 showed the effects of green tea and black tea consumption on feed consumption of untreated rats. Feed consumption was lower significantly in green tea group than in either control or black tea groups during the first week. Statistical Differences were observed in feed consumption between the three groups of animals during the last three weeks of the experimental period. Rats drinking water consumed a significantly higher amount of food than did those drinking either green tea or black tea. However, rats drinking green tea consumed significantly less food than those drinking black tea.

Figure 4: Effect of green tea and black tea consumption on body weight of untreated rats.
Control: Rats drinking water; Green Tea: Rats drinking green tea; Black Tea: Rats drinking black tea.
Table 3: Effect of green tea and black tea consumption on feed consumption [g] of untreated rats

<table>
<thead>
<tr>
<th>Groups</th>
<th>Weeks</th>
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<tbody>
<tr>
<td></td>
<td>0-1</td>
</tr>
<tr>
<td>C</td>
<td>212.58 ± 23.64</td>
</tr>
<tr>
<td>GT</td>
<td>177.78 ± 32.28</td>
</tr>
<tr>
<td>BT</td>
<td>202.47 ± 19.05</td>
</tr>
</tbody>
</table>

Values are presented as means with their standard deviations, Number of rats for each group is 30, Mean values in the same column with different superscript letters were significantly different (p<0.05; one-way ANOVA and LSD-tests), C: Rats drinking water; GT: Rats drinking green tea; BT: Rats drinking black tea.

Table 4 and figure 6 showed the effects of green tea and black tea consumption on dietary efficiency ratio of untreated rats.

For the entire experimental period, the highest value of dietary efficiency ratio was obtained with rats of control group (consuming water). For the first week, there were significant differences between all the experimental groups of rats in dietary efficiency ratio. However, consumption of black tea or green tea had no significant difference on dietary efficiency ratio values during the last three weeks, but they were significantly lower than that of the control group.

Table 4: Effect of green tea and black tea consumption on dietary efficiency ratio [g body weight gain/g feed consumption] of untreated rats.

<table>
<thead>
<tr>
<th>Groups</th>
<th>Weeks</th>
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<tbody>
<tr>
<td></td>
<td>0-1</td>
</tr>
<tr>
<td>C</td>
<td>0.105 ± 0.034</td>
</tr>
<tr>
<td>GT</td>
<td>0.001 ± 0.060</td>
</tr>
<tr>
<td>BT</td>
<td>-0.060 ± 0.064</td>
</tr>
</tbody>
</table>

Values are presented as means with their standard deviations, Number of rats for each group is 30, Mean values in the same column with different superscript letters were significantly different (p<0.05; one-way ANOVA and LSD-tests), C: Rats drinking water; GT: Rats drinking green tea; BT: Rats drinking black tea.

4. Discussion:

Treatment options for liver diseases are problematic and effective treatments are difficult. Physicians and patients are in need of effective therapeutic agents with a low incidence of side effects. Thus, they examined the effects of plants used traditionally by indigenous healers and herbalists to support liver function and treated diseases of the liver (Kim and Song, 2013). For many years, tea has been consumed in many countries for a very long time, and today interest is growing because scientific reports indicate that tea could bring benefits for health and may help prevent chronic diseases (Tijburg et al., 1997; Katiyar and Mukhtar, 1996). Finding about the beneficial effects of tea for health is meaningful because of the popularity of this beverage around the world. Other studies, however, are not consistent with such beneficial effects (Yang and Landau, 2000). Liver
injury is caused by different agents, such as chemicals, viruses, chemicals, alcohol, and auto-immune diseases (Sugiyama et al., 1999). GaIN administration to rats resembles viral hepatitis both biochemically and histologically (Wojcicki et al., 2001) and inhibits the energy metabolism of hepatocytes (Mangeney et al., 1982). Moreover, Sire et al., (1983) showed that GaIN injures the enzymes involved in the transport of substrates to the mitochondria and modifies the phospholipid composition of membranes. Furthermore, GaIN is thought to induce hepatotoxicity by inhibiting the synthesis of RNA and protein through a decrease in cellular UTP concentration, which finally leads to the necrosis of liver cells which can be measured by the activities of certain liver function enzymes, such as aminotransferases (Decker and Keppler, 1974). The mechanisms responsible for GaIN toxicity are characterized by inhibition of nuclear RNA synthesis accompanied by nuclear fragmentation or by inhibition of protein synthesis followed by accumulation of aggregates between the stacks of rough endoplasmic reticulum (Wojcicki et al., 2001).

Our study was designed to examine whether drinking green tea or black tea could protect rats against GaIN-induced liver injury, as assessed by various serum markers such as AST, ALT and GGT. In addition, some performance parameters such as body weight, food consumption and dietary efficiency ratio were determined during the experimental period. Liver injury is caused by different agents such as GaIN which induces inflammatory response in the liver that in some aspects resembles the reaction seen clinically in viral hepatitis (Wojcicki et al., 2001). GaIN is usually used in combination with other hepatotoxic substances, such as lipopolysaccharide in mice (Bajjat et al., 2000). In the present study, we used GaIN alone to induce liver injury because rats are more sensitive to GaIN than are mice (Sugiyama et al., 1999).

The extent of liver injury can be easily estimated by measuring the activities of certain enzymes, e.g., ALT and AST (Decker and Keppler, 1974). Results of blood analysis in this study indicated that rats treated with GaIN had higher activities of AST, ALT and GGT (79.64%, 200.43% and 38.46% respectively) than did untreated rats. These results were in accordance with the reports of Mangene et al., (1985), Wojcicki et al., (2001) and Ferencikova et al., (2003). It may be concluded from our findings that GaIN administration at a dose of 350mg/Kg body weight of rat was able to induce hepatic injury, as evidenced by the elevation of the previous enzymes. Moreover, our results confirm the previous findings that aminotransferases, especially ALT, represent highly specific index of hepatocellular injury and are much more sensitive to minimal or moderate damage of the liver than the other hepatic function tests (Wojcicki et al., 2001).

It would be interesting in this study to be able to clarify the hepatoprotective activity of tea against GaIN-induced liver injury, and also to know which type of tea is more effective than the other in protecting the liver from injury. However, in the comparison between untreated and treated groups of animals, our results found that consumption of either green or black tea in liver injury-induced rats participates in suppressing the elevation of the serum AST, ALT and GGT activities, as indicated by a non-significant change in these enzymes activities after GaIN injection. On the other hand, in respect to the comparison between the rats in the treated groups, our data demonstrated that pretreatment of rats with green tea was effective in diminishing hepatotoxic indices, such as, the significant reduction of serum AST and ALT activities by 59.08 % and 70.4 % respectively, while GGT activity reduced non-significantly by 1.74 % than did the group of treated rats drinking water. The same trend of reduction in the activities of the previous enzymes was observed in treated rats drinking black tea as 38.53 %, 73.59 % and 9.38% for AST, ALT and GGT respectively.

Actually, the finding in this study that tea normalized or tended to normalize the increased activities of these enzymes (AST, ALT and GGT), in response to GaIN administration, supports the idea that tea could protect against hepatotoxicity. Lin et al (2009) found that D-GaIN induced acute liver injury via hypoxia/hypoperfusion-enhanced mitochondrial apoptosis and contributing to oxidative stress and inflammation in the liver. Green tea can counteract the D-GaIN-induced acute liver injury via the attenuation of apoptotic and proinflammatory signaling by the upregulation of anti-apoptotic mechanism.

In addition, when we compare between green and black tea effects on the elevated enzymes activities in the treated rats, it was clear that both type of tea can significantly suppress the enhancement of activities of the transferase enzymes which induced by GaIN. Moreover, green tea was more effective in hepatotoxicity protection than black tea in term of AST (59.08% vs. 38.53% in green tea and black tea respectively). We may conclude from our results that green tea had a significant preventive effect on liver injury more than black tea in respect to AST activity. These results were in agreement with the studies of Sai et al., (1998) and He et al., (2001). Furthermore, even in untreated rats, green tea was able to decrease AST activity significantly by 27.96 % as compared to the control group. Supporting our results, in an animal model of viral hepatitis, pretreatment with green tea extract significantly prevented increases in hepatic transaminases levels in a dose-related manner (Hayashi et al., 1992). The beneficial effect of drinking green tea over black tea may be attributed to its higher
content of epicatechins, since they remain relatively unchanged compared with the fresh tea leaves, whereas in black tea, they are mostly oxidized during the fermentation process (Graham, 1992).

Since catechins have been considered as the major effective component of green tea, many beneficial effects of green tea are attributed to them. They are powerful antioxidants, which are thought to be at least in part responsible for green tea’s hepatoprotective activity (Miyagawa et al., 1997). But this effect of green tea is not dependent on its direct antioxidation effects alone. Green tea catechins have been shown to maintain intracellular protein thiol levels. Protein thiol he maintain the intracellular reduction-oxidation (redox) balance, protein tertiary configuration and therefore cellular function (Miyagawa et al., 1997). Moreover, Hikino et al., (1985) showed that, when catechins were added to a medium of primary cultured rat hepatocytes, they had a preventive effect on GalN-induced liver injury.

In contrast to these studies, Sugiyama et al., (1999) indicated that the major part of the effect of green tea cannot be ascribed to tea catechins which was in line with the study of Wada et al., (1999) who reported that the protective effect of green tea against GalN-induced liver injury was ascribed mainly to flavonoid glycosides, theanine and soluble dietary fibers, whereas the effect of tea catechins was relatively weak. On the other hand, He et al., (2001) suggested that the protective effect of green tea against liver injury might be attributed mainly to caffeine. Such contrasting results, concerning the major effective component of green tea, may be due to a number of confounding factors including diet, tea concentration, tea preparation, animal species and the experimental period.

With this information, one might surmise that green tea could be used as part of liver injury treatment protocol; although more human research is needed in this area before a solid recommendation can be made.

In general, the present study indicated that food intake and body weight of rats drinking tea (green and black) was significantly lower than that of rats drinking water during the experimental period. Moreover, the green tea group consumed less food than the black tea group. In accordance with our results, previous report of Naismith et al., (1969) showed that growth and food intake of rats were suppressed by dietary caffeine containing beverages. Furthermore, He et al., (2001) suggested that the effect of green tea on growth and food intake could be ascribed to caffeine, while Yang and Landau, (2000) reported that decreased nutrient absorption and increased energy expenditure may both contribute to the suppression of growth and food intake in response to tea consumption.

On the other hand consumption of black tea did not affect daily food intake nor body weight gain (Greger and Lyle, 1988, De Vos and De Schrijver, 2003). Tea concentration used may be not sufficient to induce a decrease in food intake and body weight gain. Controversially, Miura et al., (2001) found that the body weight in the tea group were greater than those in the control group.

However, the data of the present study is not sufficient to evaluate the practical benefit of tea since this experiment employed a single intraperitoneal administration of a lethal dose of GalN, which is different from the actual exposure route and frequency of intake for humans. To clarify the benefits of tea, it is important, in practical terms, to further examine the complete effectiveness of tea after repeated administrations of this toxicant, to assess the influence of different tea fractions and different tea concentrations. In addition, it’s also important to evaluate undesirable health-related consequences that may arise from ingestion of large amounts of tea.

References:


