Biochemical Assessment of Possible Protective Role of Kombucha Tea against Stressful Effect Induced by High Sucrose Dose

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Abstract

Kombucha tea is highly fermented beverage popularly consumed in many countries. The aim was to evaluate the possible protective effects of the usage of Kombucha as natural agent against the stressful effects resulted from administration of high sucrose diet to male rabbits through determination of some biochemical parameters. The results demonstrated that pretreatment with Kombucha tea in high sucrose stressed rabbit significantly improve lipid profile and antioxidant system meanwhile significant reduction of glucose, urea, creatinine, copper and non significant change in testosterone and copper levels. In conclusion, Kombucha tea was able to ameliorate serum biochemical parameters in high sucrose stressed rabbits mediated by antioxidant and lipotropic properties.

Keywords: Antioxidant, Anti-Atherosclerotic Effect, Hypoglycemic Effect, Rabbit

1. Introduction

The history of Kombucha referred to a Korean physician called Kombu, who was the first to introduce this beverage to Japanese importer (as a drink with healing properties). It is prepared by fermenting black tea with a special culture of yeasts and bacteria known as Kombucha mushroom. It is not really a mushroom, but that’s how people call it because of its shape and color when it starts forming on top of the tea after the fermentation process, but there are lots of different preparation methods and different culture that can be used. Once the beverage is finally prepared, it contains a certain amount of alcohol, acetic acid and ethyl acetate¹.

Kombucha has shown many beneficial health effects like improved energy levels, weight loss, detoxifier, antioxidants as well as antimicrobial activity against different types of bacteria like Escherichia coli, Staphylococcus aureus, Helicobacter pylori and Agrobacterium tumefaciens³.

Kombucha tea is rich with nutritive properties, beneficial bacteria, multivitamins, enzymes and essential organic acids such as acetic acid, lactic acid, folic acid, gluconic acid, glucuronic acid, usnic acid, ascorbic acid and oxalic acid which helps liver in removing toxic substances⁶.

Sucrose is a non-reducing disaccharide made up of 50% glucose and 50% fructose and has a moderately high glycemic index of 80¹⁰. Sucrose is broken down into its constituent monosaccharides, glucose and fructose by sucrase and/or isomaltase enzymes, which are located in the membrane of the microvilli lining the duodenum¹⁴.

High sucrose diet are contributed to a number of abnormalities, which include cardiovascular and renal disease such as hypertension, hypertriglyceridemia, increased collagen deposition in the heart and kidneys associated with increased oxidant concentrations and decreased antioxidant defenses as well as glucose intolerance in addition to hyper-insulinemia¹³.

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The present study was planned to show the possible effects of the usage of Kombucha as natural protective agent against the adverse stressful effects resulted from administration of high sucrose diet to male rabbits on some biochemical constituents.

2. Materials and Methods

2.1 Preparation of Kombucha Tea

Preparation of Kombucha tea was done according to 1, 3.

- Five gram of green tea leaves was soaked in one liter of freshly-boiled water for 15 minutes. The tea leaves were strained through a sieve (as green tea contains less caffeine than black tea).
- About 70-100 g of white sugar per liter of water was dissolved into the filtered infusion before it cooled.
- Then the tea was cooled to 20-25 °C. The solution was poured into a glass container. The shape of the container is unimportant but larger diameter containers work better because that allows more oxygen to get to the tea.
- Kombucha culture was added and the fermentation container was covered with a tightly, multiple layers of cheese cloth or a paper towel and was secured with a rubber band (the cloth should be porous enough to allow air to circulate so the culture can breathe, but not so porous that contamination can occur).
- The fermentation process was allowed to take place for 7-12 days (depending on the faster the fermentation the shorter period will need to be). The Kombucha culture should not be moved during this period. The temperature of the tea should fall between 20°C and 30°C.
- When the tea has attained the right degree of acidity (pH 2.7-3.2), it is ready for use.

2.2 Laboratory Animals

Eighty male rabbits at age of 4 weeks old after winning and weighting about 500-600 gm were used in the experimental investigations of this study. Rabbits were obtained from “The Laboratory Animals Research Center”, Benha University and housed in separate wire mesh cages, exposed to good ventilation, humidity and to a 12-hr light/dark cycle.

Animals were left for 15 days for acclimatization prior to the beginning of the experiment to ensure normal growth and behavior and kept at constant environmental and nutritional conditions on a basal ration of standard pellet diet, fresh and clean drinking water were supplied ad-libitum.

3. Experimental Design

Acclimatized rabbits were randomly allocated into 4 groups as following:

- First Group (Control): Twenty rabbits, served as control, kept on basal ration only.
- Second Group (Sucrose Stressed): Twenty rabbits reared on the 25% sucrose concentration ration only.
- Third Group (Kombucha): Twenty rabbits were fed on normal diet and Kombucha solution (150 ml).
- Fourth Group (Kombucha protected): Twenty rabbits kept on equal amounts of 25% sucrose and Kombucha solution (150 ml).

3.1 Collection of Blood Samples

Blood samples were collected from all animals under experiment monthly for 4 month by ear veins in first and third sample then by slaughtering of half of rabbits in each group (Ten rabbits) in second and last sample.

Blood samples were collected in tubes without anticoagulant in clean, dry Wassermann tubes and left in slope position to clot at room temperature. The tubes were centrifuged at 3000 rpm for 5 minutes and the non hemolyzed serum was carefully separated and transferred into clean dry Eppendorf tubes which were kept frozen at -20°C until used for biochemical analysis.

All serum samples were analyzed for the determination of lipid profile (Total lipids, TAG, Total Cholesterol, HDL-C LDL-C, VLDL-C and Atherogenic Indices). As well as urea, creatinine, glucose, testosterone, zinc and copper were also determined.
3.2 Collection of Rabbit’s Liver, Heart and Kidney

After collection of the 2nd and 4th blood samples, 10 rabbits were sacrificed after two months from beginning of the experiment and the rest of 10 rabbits were sacrificed after 4 months. The liver, heart and kidney were removed and washed by ice-cold saline buffer to remove the blood and then blotted in filter papers and finally kept frozen at -20°C for biochemical analysis.

All tissues samples were analyzed for the determination of reduced Glutathione (GSH) and antioxidant enzymes (Catalase and GST), L-malondialdehyde (L-MDA) and H_2O_2.

4. Results and Discussion

Kombucha is a highly fermented tea beverage popularly consumed as a self prescribed folk remedy for numerous ailments. Kombucha is claimed to enhance cognition, aid weight loss, and prolonged life⁹.

Fructose is a major ingredient of many processed foods and has been proposed to contribute to the development of obesity and dyslipidemia. Diets rich in fructose induce hepatic steatosis and plasma hyperlipidemia². Our data was in agreement with earlier studies⁵, a short term consumption of a high-sucrose diet increases the triglyceride levels in liver and plasma. The increment in total lipids may attribute to fructose which has been shown to increase plasma concentrations of triacylglycerols and cholesterol.

Table 1. The effect of Kombucha supplementation on serum lipid profile in normal and sucrose stressed male rabbits

| parameter       | Groups               | 1st month       | 2nd month       | 3rd month       | 4th month       |
|-----------------|----------------------|-----------------|-----------------|-----------------|-----------------|
| Total lipids (mg/dl) | Control             | 241.05 ± 5.75   | 248.44 ± 4.83   | 287.36 ± 4.01   | 314.58 ± 4.37   |
|                 | Sucrose Stressed     | 378.30 ± 8.85   | 396.18 ± 7.29   | 414.46 ± 6.56   | 451.75 ± 7.14   |
|                 | Kombucha only        | 269.61 ± 6.41   | 282.50 ± 5.51   | 323.30 ± 8.96   | 360.23 ± 9.76   |
|                 | Kombucha + Sucrose   | 322.53 ± 8.26   | 339.27 ± 8.53   | 36.61 ± 4.81    | 397.72 ± 5.24   |
|                 | Control              | 36.68 ± 1.86    | 52.00 ± 1.94    | 93.50 ± 1.12    | 101.91 ± 1.20   |
| Triacylglycerols (mg/dl) | Sucrose Stressed     | 161.25 ± 5.44   | 236.05 ± 8.51   | 223.18 ± 2.57   | 274.78 ± 2.79   |
|                 | Kombucha only        | 56.08 ± 5.06    | 104.24 ± 4.84   | 80.67 ± 1.94    | 131.13 ± 2.11   |
|                 | Kombucha + Sucrose   | 105.39 ± 3.89   | 166.57 ± 4.25   | 126.79 ± 4.01   | 161.30 ± 4.36   |
|                 | Control              | 49.50 ± 3.49    | 58.32 ± 4.17    | 70.63 ± 2.97    | 76.98 ± 3.22    |
| Total cholesterol (mg/dl) | Sucrose Stressed     | 63.86 ± 5.09    | 70.11 ± 3.49    | 75.64 ± 1.83    | 82.44 ± 1.99    |
|                 | Kombucha only        | 38.75 ± 1.39    | 53.46 ± 2.92    | 62.52 ± 2.09    | 68.14 ± 2.27    |
|                 | Kombucha + Sucrose   | 57.71 ± 2.23    | 58.87 ± 3.05    | 68.09 ± 1.79    | 75.39 ± 1.94    |
|                 | Control              | 28.77 ± 0.98    | 26.26 ± 24.10   | 20.41 ± 1.78    | 22.24 ± 1.93    |
| HDL-C (mg/dl)    | Sucrose Stressed     | 12.24 ± 1.99    | 10.87 ± 0.97    | 9.33 ± 1.04     | 8.54 ± 1.13     |
|                 | Kombucha only        | 15.16 ± 1.92    | 22.74 ± 1.60    | 16.91 ± 1.31    | 17.06 ± 1.32    |
|                 | Kombucha + Sucrose   | 22.74 ± 1.60    | 24.42 ± 1.18    | 20.56 ± 1.46    | 20.75 ± 1.48    |
|                 | Control              | 14.19 ± 3.22    | 14.90 ± 2.53    | 15.31 ± 1.95    | 15.88 ± 2.12    |
| LDL-C (mg/dl)    | Sucrose Stressed     | 15.55 ± 0.86    | 23.13 ± 2.34    | 16.63 ± 1.95    | 19.10 ± 1.79    |
|                 | Kombucha only        | 11.16 ± 1.97    | 12.39 ± 1.36    | 13.80 ± 1.31    | 15.04 ± 1.42    |
|                 | Kombucha + Sucrose   | 13.30 ± 16.32   | 19.67 ± 1.06    | 13.50 ± 0.97    | 14.70 ± 1.05    |
|                 | Control              | 7.33 ± 0.37     | 10.40 ± 0.38    | 18.69 ± 1.08    | 20.36 ± 1.17    |
| VLDL-C (mg/dl)   | Sucrose Stressed     | 32.24 ± 3.88    | 47.20 ± 3.00    | 42.47 ± 0.50    | 46.28 ± 0.55    |
|                 | Kombucha only        | 21.22 ± 3.88    | 23.40 ± 0.84    | 27.97 ± 0.37    | 26.33 ± 0.40    |
|                 | Kombucha + Sucrose   | 22.52 ± 0.77    | 30.07 ± 0.85    | 35.07 ± 0.79    | 36.57 ± 0.86    |
Table 2. The effect of Kombucha supplementation on atherogenic indices in normal and sucrose stressed male rabbits

| parameter                        | Groups             | Sample interval       |
|----------------------------------|--------------------|-----------------------|
|                                  |                    | 1st month | 2nd month | 3rd month | 4th month |
| Cardiac risk ratio (CRR)         | Control            | 1.53 ± 0.14 | 2.40 ± 0.31 | 3.46 ± 0.45 | 3.76 ± 0.48 |
|                                  | Sucrose Stressed   | 5.06 ± 0.87 | 4.71 ± 0.32 | 8.26 ± 95.25 | 8.99 ± 103.82 |
|                                  | Kombucha only      | 1.74 ± 1.08 | 2.46 ± 0.29 | 4.07 ± 0.38 | 4.42 ± 0.42 |
|                                  | Kombucha + Sucrose | 2.54 ± 0.25 | 1.93 ± 0.11 | 3.40 ± 0.29 | 3.70 ± 0.31 |
|                                  | Control            | 0.10 ± 0.03 | 0.32 ± 0.10 | 0.64 ± 0.02 | 0.70 ± 0.02 |
|                                  | Sucrose Stressed   | 1.03 ± 0.09 | 1.17 ± 0.02 | 1.32 ± 0.03 | 1.44 ± 0.03 |
|                                  | Kombucha only      | 0.86 ± 0.02 | 0.70 ± 0.02 | 0.68 ± 0.03 | 0.73 ± 0.03 |
|                                  | Kombucha + Sucrose | 0.79 ± 0.03 | 0.72 ± 0.02 | 0.58 ± 0.04 | 0.62 ± 0.04 |
|                                  | Control            | 0.82 ± 0.14 | 1.43 ± 0.31 | 2.49 ± 0.46 | 2.71 ± 0.49 |
| Atherogenic index (AI) ratio     | Sucrose Stressed   | 4.09 ± 0.87 | 3.74 ± 0.32 | 7.29 ± 0.95 | 7.93 ± 1.02 |
|                                  | Kombucha only      | 0.77 ± 0.09 | 1.92 ± 0.29 | 3.20 ± 0.38 | 3.58 ± 0.42 |
|                                  | Kombucha + Sucrose | 1.76 ± 0.25 | 0.96 ± 0.11 | 2.43 ± 0.29 | 2.64 ± 0.31 |

Table 3. The effect of Kombucha supplementation on serum urea, creatinine and glucose in normal and sucrose stressed male rabbits

| parameter            | Groups                | Sample interval       |
|----------------------|-----------------------|-----------------------|
|                      |                       | 1st month | 2nd month | 3rd month | 4th month |
| Urea (mg/dl)         | Control               | 24.30 ± 1.72 | 29.16 ± 1.04 | 31.10 ± 0.75 | 31.65 ± 1.13 |
|                      | Sucrose Stressed      | 24.30 ± 0.62 | 33.04 ± 1.42 | 32.07 ± 1.42 | 34.95 ± 1.52 |
|                      | Kombucha only         | 19.87 ± 1.00 | 25.27 ± 1.38 | 26.24 ± 1.81 | 28.59 ± 1.97 |
|                      | Kombucha + Sucrose    | 18.70 ± 1.59 | 2.10 ± 1.73 | 25.27 ± 1.53 | 27.54 ± 1.26 |
|                      | Control               | 0.63 ± 0.03 | 0.58 ± 0.03 | 0.74 ± 0.07 | 0.81 ± 0.07 |
|                      | Sucrose Stressed      | 0.68 ± 0.03 | 0.65 ± 0.05 | 0.77 ± 0.03 | 0.84 ± 0.03 |
|                      | Kombucha only         | 0.60 ± 0.02 | 0.55 ± 0.05 | 0.70 ± 0.01 | 0.75 ± 0.01 |
|                      | Kombucha + Sucrose    | 0.41 ± 0.02 | 0.48 ± 0.03 | 0.68 ± 0.03 | 0.73 ± 0.03 |
|                      | Control               | 84.31 ± 2.32 | 91.41 ± 2.94 | 101.08 ± 6.02 | 110.18 ± 3.28 |
| Creatinine level (mg/dl) | Sucrose Stressed      | 98.56 ± 2.27 | 129.76 ± 2.01 | 174.96 ± 3.27 | 95.35 ± 3.56 |
|                      | Kombucha only         | 79.21 ± 1.62 | 85.05 ± 17.75 | 87.48 ± 3.27 | 95.35 ± 3.56 |
|                      | Kombucha + Sucrose    | 81.11 ± 2.87 | 105.81 ± 2.35 | 119.14 ± 3.07 | 137.74 ± 3.34 |
| Glucose level (mg/dl) |                       |                       |           |           |           |
|                      |                       |                       |           |           |           |
|                      |                       |                       |           |           |           |

Table 4. The effect of Kombucha supplementation on serum testosterone, zinc and copper in normal and sucrose stressed male rabbits

| Parameter              | Groups                | Sample interval       |
|------------------------|-----------------------|-----------------------|
|                       |                       | 1st month | 2nd month | 3rd month | 4th month |
| Testosterone concentration (ng/ml) | Control | 1.32 ± 0.04 | 1.80 ± 0.18 | 2.01 ± 0.23 | 1.93 ± 0.34 |
|                        | Sucrose Stressed      | 1.56 ± 0.14 | 2.13 ± 0.27 | 3.11 ± 0.38 | 3.18 ± 0.35 |
|                        | Kombucha only         | 0.98 ± 0.03 | 1.03 ± 0.06 | 1.05 ± 0.03 | 1.37 ± 0.27 |
|                        | Kombucha + Sucrose    | 1.16 ± 0.02 | 1.19 ± 0.05 | 1.09 ± 0.05 | 1.06 ± 0.32 |
|                        | Control               | 177.62 ± 2.38 | 185.40 ± 4.37 | 205.74 ± 6.15 | 227.78 ± 6.70 |
| Zinc concentration (µg/dl) | Sucrose Stressed      | 141.50 ± 3.57 | 156.89 ± 6.12 | 165.61 ± 4.05 | 175.12 ± 4.40 |
|                        | Kombucha only         | 194.15 ± 1.99 | 200.23 ± 2.72 | 218.34 ± 5.34 | 228.57 ± 5.82 |
|                        | Kombucha + Sucrose    | 159.69 ± 3.00 | 171.03 ± 2.78 | 179.49 ± 6.75 | 196.39 ± 7.35 |
Table 5. The effect of Kombucha supplementation on liver, heart and kidney - reduced glutathione (GSH), antioxidant enzymes (Catalase and GST), L-malondialdehyde (L-MDA) and H₂O₂ in normal and sucrose stressed male rabbits

| parameter            | Groups                        | Liver | Heart | Kidney |
|----------------------|-------------------------------|-------|-------|--------|
|                      |                               | After 2 months | After 4 months | After 2 months | After 4 months | After 2 months | After 4 months |
| GSH (mg/g. tissue)   | Control                       | 723.89 ± 29.63 | 666.13 ± 27.06 | 35.22 ± 1.26 | 31.28 ± 1.19 | 96.71 ± 2.14 | 88.86 ± 1.99 |
|                      | Sucrose Stressed              | 397.89 ± 20.50 | 336.26 ± 21.83 | 21.35 ± 0.82 | 17.16 ± 0.75 | 35.37 ± 2.70 | 29.41 ± 6.73 |
|                      | Kombucha only                 | 771.62 ± 20.41 | 797.23 ± 23.43 | 39.79 ± 1.27 | 33.56 ± 1.15 | 102.85 ± 4.35 | 94.61 ± 2.45 |
|                      | Kombucha + Sucrose            | 528.28 ± 28.29 | 567.69 ± 23.98 | 26.67 ± 1.42 | 22.07 ± 1.26 | 75.06 ± 4.38 | 62.15 ± 4.15 |
| Catalase (U/g. tissue)| Control                       | 316.76 ± 14.16 | 300.92 ± 13.87 | 78.55 ± 3.09 | 74.97 ± 2.93 | 272.88 ± 3.79 | 254.91 ± 10.27 |
|                      | Sucrose Stressed              | 218.60 ± 12.52 | 187.80 ± 12.32 | 37.44 ± 1.51 | 31.14 ± 1.26 | 125.51 ± 8.37 | 109.16 ± 7.77 |
|                      | Kombucha only                 | 342.19 ± 13.82 | 323.81 ± 13.54 | 84.47 ± 0.82 | 79.41 ± 0.74 | 213.03 ± 4.47 | 197.41 ± 9.46 |
|                      | Kombucha + Sucrose            | 274.61 ± 11.71 | 261.40 ± 14.73 | 64.62 ± 4.66 | 55.76 ± 4.14 | 258.55 ± 5.68 | 230.10 ± 9.37 |
| GST activity (U/g. tissue) | Control                       | 301.77 ± 4.38 | 287.75 ± 4.15 | 146.39 ± 3.56 | 139.07 ± 4.02 | 273.85 ± 9.97 | 260.16 ± 9.47 |
|                      | Sucrose Stressed              | 209.34 ± 2.40 | 186.31 ± 2.23 | 85.44 ± 4.14 | 79.63 ± 3.85 | 114.37 ± 12.26 | 93.29 ± 11.40 |
|                      | Kombucha only                 | 320.02 ± 9.93 | 300.94 ± 9.03 | 149.52 ± 3.69 | 144.99 ± 3.35 | 260.19 ± 2.07 | 247.83 ± 1.79 |
|                      | Kombucha + Sucrose            | 265.10 ± 22.93 | 242.44 ± 20.41 | 127.34 ± 3.98 | 113.33 ± 3.54 | 194.18 ± 12.15 | 214.72 ± 10.80 |
| L-MDA (nmol/g. tissue)| Control                       | 5.09 ± 2.43 | 7.48 ± 2.30 | 566.53 ± 19.80 | 583.75 ± 18.81 | 23.91 ± 2.99 | 32.28 ± 2.84 |
|                      | Sucrose Stressed              | 30.29 ± 2.45 | 35.49 ± 2.27 | 794.34 ± 6.76 | 739.16 ± 6.28 | 90.18 ± 2.39 | 98.88 ± 2.22 |
|                      | Kombucha only                 | 5.72 ± 1.04 | 7.67 ± 0.95 | 646.61 ± 14.22 | 634.50 ± 12.74 | 28.44 ± 1.97 | 35.17 ± 1.79 |
|                      | Kombucha + Sucrose            | 12.90 ± 3.28 | 16.65 ± 25.59 | 736.29 ± 8.29 | 655.29 ± 7.37 | 50.27 ± 2.04 | 42.80 ± 1.81 |
| H₂O₂ (mM/g. tissue)  | Control                       | 13.95 ± 0.77 | 16.54 ± 0.73 | 0.75 ± 0.05 | 0.82 ± 0.04 | 0.71 ± 0.09 | 0.79 ± 0.08 |
|                      | Sucrose Stressed              | 39.04 ± 1.01 | 45.85 ± 0.93 | 3.17 ± 0.05 | 4.17 ± 0.04 | 4.51 ± 0.38 | 5.34 ± 0.35 |
|                      | Kombucha only                 | 10.87 ± 2.33 | 14.11 ± 2.12 | 0.69 ± 0.04 | 0.73 ± 0.03 | 0.34 ± 0.06 | 0.38 ± 0.05 |
|                      | Kombucha + Sucrose            | 20.70 ± 0.62 | 35.41 ± 0.55 | 1.85 ± 0.05 | 1.77 ± 0.04 | 1.89 ± 0.04 | 1.66 ± 0.03 |
Moreover, hypertriglyceridemia after simple carbohydrate feeding results from the induction of de novo lipogenesis, the enhanced rate of hepatic VLDL, triglyceride synthesis and a decrease in peripheral triglyceride clearance. In addition, in subjects consuming fructose, significant increased circulating levels of remnant lipoproteins, small dense LDL and oxidized LDL were reported.

Fructose feeding can induce free radical formation by a number of mechanisms. It causes down regulation of the key enzymes of the hexose monophosphate pathway, namely glucose-6-phosphate dehydrogenase and 6-phosphogluconate dehydrogenase that generate a reduced environment in the form of NADPH and NADH. Impaired regeneration of NADPH could result in an increased oxidative state of the cell.

Our study is in accordance with who reported that fructose enriched diet, resulted in increased lipid peroxidation and impaired antioxidant status. Moreover, reported that, increases in the levels of the Thiobarbituric Acid reactive Substances (TBARS) and hydroperoxides were observed in the liver of fructose fed rats.

 reported that the increased rate of \( \text{H}_2\text{O}_2 \) generation in high sucrose diet rat could result from decreased levels of reduced CoQ10, vitamin E and reduced glutathione and elevated activity of Cu/Zn-SOD in the mitochondrial intermembrane space. The excess \( \text{H}_2\text{O}_2 \) in the inter membrane space may cross the external mitochondrial membrane and affect the redox state of the entire liver by decreasing the concentration of reduced GSH. Additionally, the reduced activity of catalase observed in sucrose fed rats liver homogenate may contribute to increased levels of lipid peroxidation and protein carbonylation in whole liver cells.

Kombucha prevents central body fat accumulation and decreases postprandial adiponectin expression induced by a carbohydrate rich diet in insulin resistant subjects. Our results nearly similar to that a decrease in serum triacylglycerol, VLDL concentration after administration of Kombucha tea (Table 1) due to low activity of hydroxyl methyl glutaryle Co A responsible for cholesterol synthesis. Also, observed that, hypcholesteremic effect of Kombucha tea (Table 2) refer to present gluconoacetobacteria sp4 that attributed to modified Kombucha tea has powerful effect.

Kombucha tea exhibit hypoglycemic effect, reduced the insulin requirements and improved insulin resistance (Table 3) so, it increased the release of TG from the liver and decreased the flux of free fatty acids from peripheral adipose tissue back to the liver. The administration of Kombucha resulted in reductions of total cholesterol concentration and transaminases activity in serum. Moreover, Kombucha been shown to be less susceptible to oxidation. This binding is directly related to an increase of the LDL resistance to oxidation.

In addition, the anti-atherosclerotic effect (Table 2) of Kombucha demonstrated in rabbits on a high-lipid diet related to suppression of inflammation and decreasing the concentration of TG and LDL-C and improved the level of HDL-C that has been linked to a lower risk of coronary heart disease.

The obtained data belonging to the effect of Kombucha on testosterone concentration (Table 4) nearly agree with that, Kombucha significantly decreased serum testosterone as well as sperm count, sperm motility, the weight of prostate, testis, epididymis, seminal vesicle, weights of the testicle and seminal vesicle. Moreover, Kombucha is known as a phytoestrogen compound since it contains phenolic compounds. Also reported that, the Kombucha contains phytoestrogen which may has an inhibitory effect on the enzyme 17- \( \text{B} \)-hydroxy steroid 11-hydrogenase, therefore, the synthesis of testosterone in adrenal cortex is reduced.

The significant decrease of testosterone level after administration of Kombucha tea may attribute to hypocholesterolemic effect which may be decrease the testosterone testicular synthesis. The inhibition of de novo synthesis pathway of cholesterol biosynthesis negatively affects testosterone level in addition to cholesterol concentration in the tissues, body weight gain and ALT with no successful compensatory mechanism as related with testosterone level.
biosynthesis pathway may result in a decline in plasma testosterone concentration which may lead to a marked decrease in the fertility index and sperm cell count\(^\text{23}\).

Kombucha improved oxidative stress and repair any damage which generates free radicals and alters antioxidants scavenging enzyme (Table 5)\(^\text{28}\). The results was in accordance with\(^\text{21}\) that, the administration of purified Kombucha is able to reduce the oxidative damage and suppresses oxidative stress as monitored by the elevation activity of the main anti-oxidative enzyme, catalase and decreases lipid peroxidation products in liver.

In addition, the positive impact of treatment with Kombucha on the antioxidant enzymes GPx, GRx, CAT and SOD could be explained with two possible mechanisms. First, the antioxidant effect of Kombucha may prevent further glycosylation and peroxidation of proteins by interacting with free radicals and hence minimizing their noxious effects. Second, Kombucha may induce protein synthesis of these enzymes as reported by\(^\text{28}\).

5. Conclusion

Our results conclude that, the administration of high sucrose diet to male rabbits accompany by significance disturbance of lipid profile and antioxidant system as well as increase levels of glucose, urea, creatinine and Kombucha tea was able to ameliorate serum biochemical parameters.

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