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**NATURAL DRUGS**

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**HEMATOLOGICAL AND IMMUNOBIOCHEMICAL STUDY OF  
GREEN TEA AND GINGER EXTRACTS IN EXPERIMENTALLY INDUCED  
DIABETIC RABBITS**

AHMED ELKIRDASY<sup>1,3\*</sup>, SAAD SHOUSHA<sup>2</sup>, ABDULMOHSEN H. ALROHAIMI<sup>3</sup>  
and M. FAIZ ARSHAD<sup>4</sup>

<sup>1</sup>Department of Biochemistry, Faculty of Veterinary Medicine, Menoufia University (Sadat branch), Egypt

<sup>2</sup>Department of Physiology, Faculty of Veterinary Medicine, Benha University, 13736 Moshtohor, Egypt

<sup>3</sup>Department of Biomedical Science, <sup>4</sup>Department of Pharmaceutical Science, College of Pharmacy,  
Shaqra University, Al Dawadimi, Saudi Arabia

**Abstract:** The present study was designed to investigate the effects of the extract of green tea and/or ginger on some hematological and immunobiochemical profiles in alloxan-induced diabetic rabbits. The results revealed that treatment of diabetic animals with extract of green tea and/or ginger elevated the decreased HDL-c and LDL-c but significantly decreased triglycerides, the elevated glucose and GOT concentrations. The result also displayed a non-significant increase in the levels of CRP and fibrinogen. The experiment also revealed that the elevated MDA and GSH level fell down to the normal control group. The result also showed that after green tea and/or ginger extract treatment, the lowered RBC, WBC counts, PCV, percentage of neutrophils were increased and the elevated MCV, MCH, and MCHC of diabetic rabbits were decreased to normal levels. Thus, the overall results may indicate that green tea and/or ginger extracts have a significant hypoglycemic effect in diabetic rabbits. In addition, the extracts may be capable of improving hyperlipidemia, the impaired kidney function and hemogram in alloxan-induced diabetic rabbits.

**Keywords:** alloxan, ginger, green tea, hematology, immunobiochemical

**Abbreviations:** Hb - hemoglobin, PCV - packed cell volume, RBC - red blood cell, WBC - white blood cell, MCV - mean corpuscular volume, MCH - mean corpuscular hemoglobin, MCHC - mean corpuscular hemoglobin concentration

The World Health Organisation (WHO) reported that 300 million people would suffer diabetes mellitus (DM) by the year 2025 (1). The disease is considered as important public health problem, especially in developing and 3rd world countries due to inadequate treatment (2).

Although herbal medicines have long been used effectively in treating many diseases throughout the world, the mechanisms of most of the herbals used have not been defined. Many traditional plant treatments for diabetes are also used, but most of the evidence for their beneficial effects is anecdotal (3). Ginger (*Zingiber officinale*) has been used as a spice for over 2000 years. Its root and the obtained extracts contain polyphenol compounds: 6-gingerol and its derivatives, which have high antioxidant activity (4). Ginger's high antioxidant value has

proved highly effective with its ability to scavenge a number of free radicals and protect cell membrane lipids from oxidation in a dose dependent manner (5). As in traditional medicine, rhizomes of ginger plants are consumed by women during ailment, illness and confinement. Ginger oil has been found to be an inhibitor of cyclooxygenase and lipoxygenase activities (6).

The medicinal properties of ginger were attributed to many active compounds in ginger. The major constituents in *Zingiber officinale* are the pungent vanilloids, gingerol, paradol, shogaols and zingerone (7). The antioxidant, antitumor, and anti-inflammatory pharmacologic effects of ginger were mainly attributed to these constituents (7).

Ginger acts as a hypolipidemic agent in cholesterol-fed rabbits (8). Also, it was reported (9) that

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\* Corresponding author: e-mail: afkirdasy@hotmail.com

ginger treatment significantly decreased both serum cholesterol and triglycerides. In addition, ginger decreased low density lipoprotein-cholesterol (LDL-cholesterol), very low density lipoprotein-cholesterol (VLDL-cholesterol) and triglycerides levels in apolipoprotein-E deficient mice (10). Furthermore, ethanolic extract of ginger prevented hypercholesterolemia and development of atherosclerosis in cholesterol-fed rabbits (11). It was found (12) that the ethanolic extract of ginger significantly reduced serum total cholesterol and triglycerides and increased the high density lipoprotein-cholesterol (HDL-cholesterol) levels; also, the extract can protect tissues from lipid peroxidation and exhibit a significant lipid lowering activity in diabetic rats. The study (13) revealed that serum and liver cholesterol decreased when ginger was administered to hypercholesterolemic rats.

Green tea is one of four types of tea (white, green, black, and oolong) that come from the plant *Camellia sinensis*. White tea is the least processed form of tea, while black tea leaves are fermented. Green tea leaves are steamed, not fermented and hence preserve more polyphenols (14). The beneficial effects of green tea are attributed to the polyphenols, particularly the catechins, which make up to 30% of the dry weight of green tea leaves. These catechins are present in higher quantities in green tea than in black or oolong tea, because of differences in the processing of tea leaves after harvest (15).

Green tea is a widely-consumed beverage and, for centuries, has been regarded to possess significant health-promoting effects (16). The health-promoting effects of green tea are mainly attributed to its polyphenol content. Green tea is a rich source of polyphenols, especially flavanols and flavonols, which represent approximately 30% dry weight of the fresh leaves (16). Catechins are the predominant flavanols and are mainly comprised of epigallocatechin gallate (EGCG), epigallocatechin (EGC), epicatechin gallate (ECG), and epicatechin (EC) (17).

Hematological and biochemical analyses after treatment with plant extracts in experimental animals are among the important methods of assessing the safety and toxicity of these plant extracts in animals and human being as well. Earlier studies have been done for investigating the effect of plant extracts on hematology and biochemistry of experimental animals but none has been done for investigating the effect of combined extracts of both green tea and ginger in rabbits. Therefore; we measured some hematological parameters for evaluation of the effect of the extract of green tea and/or ginger in diabetic-induced rabbits.

## EXPERIMENTAL

### Animals

One month before the study, the animals were treated against endo- and ecto-parasites and coccidiosis. Fifty (50) adult male rabbits of the New Zealand white strain (weighing between 1100 - 1800 g) were selected and kept for about 14 days for acclimatization. All animals were housed in stainless cages under standard laboratory conditions (temperature:  $22.5 \pm 3.58^\circ\text{C}$ ; relative humidity:  $50 \pm 20\%$ ; air ventilation: 10–15 times per hour; artificial lighting: 12 h per day), and were allowed free access to normal balanced (standard) diet purchased from Cairo company (Commercial pelleted food contained a minimum of 88% dry matter, minimum 15% crude protein, maximum 17% cellulose, maximum 10% crude ash and minimum 2300 kcal/kg metabolic energy) and to water *via* an automatic water supply system. The animals received human care according to the criteria outlined in the "Guide for the Care and Use of Laboratory Animals" prepared by the National Academy of Sciences and published by the National Institutes of Health (NIH).

### Preparation of plant extracts

#### Ginger extraction

Fresh rhizomes of ginger roots purchased from ordinary market were washed, peeled and 10 g weighed out and mashed with mortar and pestle. This was then extracted with 0.1 liter of distilled water and boiled for 5 min at  $100^\circ\text{C}$ . The extract was filtered and administered 1 mL/kg orally (30).

#### Green tea extraction

Fifteen grams of tea leaves were soaked in 500 mL of boiling water for 30 min and were then filtered. Extracts were prepared fresh daily (31). The tested plants were used in aqueous extracts at concentration of 0.2 g/mL by soaking these plants in boiling water. After 5 min, each extract was filtered and the filtrate was orally administered to rabbits at a temperature of  $37 \pm 1^\circ\text{C}$ . This filtrate was provided to rabbits as their sole source of drinking water.

### Experimental protocol

Fifty (50) adult male rabbits used in this study were divided into five experimental groups, each group consisted of 10 animals: Group 1 (G1): (control group), normal animals without treatment (standard); Group 2 (G2): diabetic group (diabetes induced by alloxan) without any treatment; Group 3 (G3): diabetic animals treated with green tea extract;

Group 4 (G4): diabetic animals treated with ginger extract; Group 5 (G5): diabetic animals treated with combined extract of both green tea and ginger.

Aqueous extracts of green tea and/or ginger were administered orally to rabbits daily using a gastric tube at dose rate of 1 mL/100 g body weight per day for 3 weeks (32). The control groups (normal and diabetic) were orally administered only with the same volume of isotonic NaCl.

Female rabbits were not used in our experiments to avoid the effect of female reproductive hormones on the measured parameters.

### *Induction of diabetes*

The most common method to induce diabetes is using phloridzin (alloxan) or streptozotocin. Considering the insensitivity of rabbits to streptozotocin, alloxan was used to induce diabetes in this study. Alloxan causes extensive  $\beta$  cell destruction in the pancreas during 18 to 24 h after injection, which leads to hyperglycemia. The reason for alloxan's selective toxicity is its structural resemblance to glucose and its mechanism of effect is producing free radicals since it can be deactivated by anti-oxidants. At the start of the experiment; the animals in the latter four groups were injected intravenously in the ear vein with 150 mg/kg of 10% alloxan (alloxan monohydrate, Sigma Chemicals Co., Egypt) dissolved in 10% solution in normal saline to induce diabetes. The control group was injected only with the same volume of isotonic NaCl as the diabetic groups received. Normal range of blood glucose in New Zealand white rabbit is ranged between 4.2-10.4 mmol/L (75-189 mg/dL with an average of 115 mg/dL) (33). Animals with blood glucose levels  $\geq$  200 mg/dL were included in the study (34). Alloxan induces diabetes through destructing Langerhans islands of the pancreas. Therefore, a large amount of insulin is released from the pancreas cells after the injection. In order to prevent hypoglycemic shock, during the first 24 h after the alloxan injection, the rabbits received 10% dextrose instead of water (35, 36). Blood glucose levels were assayed 3 days after alloxan treatment to determine which animals had become diabetic and the degree of hyperglycemia.

### *Collection of blood samples*

At the end of the experimental period (3 weeks), blood samples were collected from experimental animals through the ear vein into a set of labeled sterile bottles containing K3-EDTA for hematological parameters determination with careful considerations to avoid hemolysis during sampling and transfer. Another set of tubes without K3-

EDTA was used to collect blood, immediately covered, left to clot and centrifuged at  $4000 \times g$  for 15 min at room temperature for separation of serum that will be used for immunobiochemical parameters assays.

### *Immunobiochemical parameters*

The levels of serum triglycerides, HDL-cholesterol and LDL-cholesterol were estimated according to methods described in (37). Lipid peroxidation (malondialdehyde concentration, MDA) and reduced blood glutathione (GSH) concentration were determined according to the methods of (38). Glucose level was measured by the method described by (39). Aspartate aminotransferase (AST; SGOT) and alanine aminotransferase (ALT; SGPT) were measured using commercial kits.

Immunoglobulins were measured using the method of (40). C-reactive protein (CRP) was measured using the method of (41). All other chemicals used were of analytical reagent grade.

### *Hematological parameters*

The packed cell volume (PCV) was measured by the microhematocrit centrifuge (40). Hemoglobin (Hb) concentration was determined by the cyanomethemoglobin technique (42). The red blood cells (RBCs) and white blood cells (WBCs) counting methods were based on the dilution of obtained blood with diluting fluids (Hayem & Turke) in RBCs and WBCs counting pipettes (42). Individual cells were then counted in the counting chamber (hemocytometer). Giemsa's staining method was used for the differential count of WBCs.

### *Statistical analysis*

Data were statistically analyzed by one-way analysis of variance followed by Duncant's test (PC-stat computer program). Finally, least significant difference (L.S.D) was used to test the difference among treatments. Results were considered statistically significant when  $p \leq 0.05$ .

## **RESULTS**

As shown in Figures 1, 2, alloxan produced significant hyperlipidemic action, where a significant ( $p \leq 0.05$ ) increase was recorded in the levels of plasma triglycerides and LDL-c when compared with normal group, but HDL-c level was significantly ( $p \leq 0.05$ ) decreased. However, post-treatment with extract of green tea and/or ginger ameliorated the hyperlipidemic action of alloxan on lipid profile by changing either up or down the

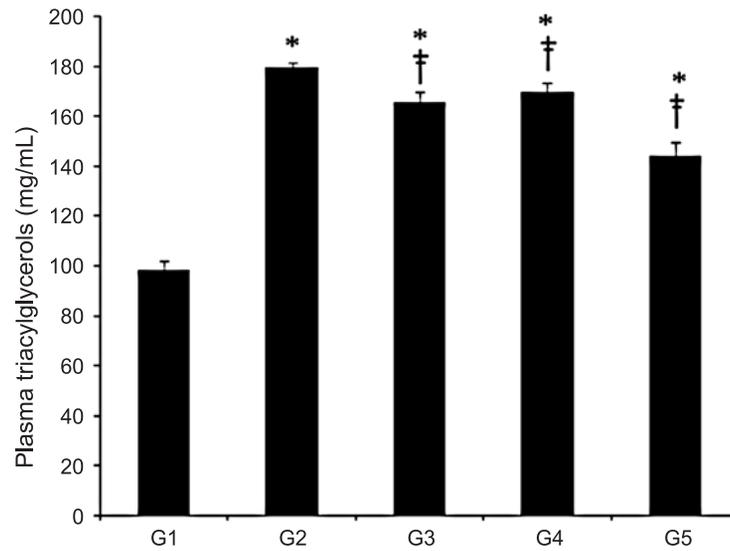


Figure 1. Plasma triacylglycerols in normal control, diabetic and diabetic treated animals. Values are the mean  $\pm$  SEM. \*  $p < 0.05$  vs. control group (G1) and ‡  $p < 0.5$  vs. diabetic group (G2)

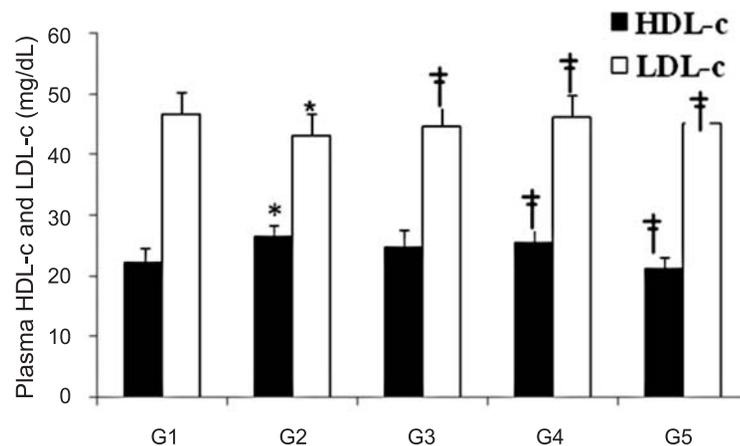


Figure 2. Plasma HDL-c and LDL-c levels in normal, diabetic and diabetic treated animals. Values are the mean  $\pm$  SEM. \*  $p < 0.05$  vs. control group (G1) and ‡  $p < 0.5$  vs. diabetic group (G2)

level to be normal or near the normal in case of HDL-c and LDL-c but decreasing significantly ( $p \leq 0.05$ ) the level of triglycerides. In case of post-treatment by oral administration of both green tea and ginger extracts, the level of triglycerides decreased significantly ( $p \leq 0.05$ ) more than the effect of post-treatment with oral administration of either green tea or ginger extract alone as shown in Figure 1.

Serum glucose concentration increased significantly ( $p \leq 0.05$ ) in diabetic rabbits but treatment of diabetic rabbits with extract of green tea and/or gin-

ger decreased the elevated glucose concentration significantly ( $p \leq 0.05$ ); however, glucose concentrations were still significantly higher ( $p \leq 0.05$ ) than those of the control group as shown in Figure 3.

Highly significant ( $p \leq 0.05$ ) increase in both GPT and GOT was recorded in diabetic group compared to normal control group. Post-treatment of diabetic animals with extract of green tea or ginger returned their level to its normal level again. However, post-treatment of diabetic animals with extract of both green tea and ginger increased GOT significantly but not GPT as shown in Figure 4.

Regarding to MDA and GSH, alloxan-induced diabetic animals showed significant increase in MDA and a significant ( $p \leq 0.05$ ) decrease in GSH levels compared to their equivalent levels in normal control animals as shown in Figure 5. However, post-treatment of diabetic animals with extract of either green tea or ginger resulted in significant ( $p \leq 0.05$ ) increase in MDA

as an indicator of lipid peroxidation process, while post-treatment of diabetic animals with extract of both green tea and ginger reduced significantly ( $p \leq 0.05$ ) the level of MDA returning it to its normal control level. In case of GSH, post-treatment of alloxan-induced diabetic animals with extract of green tea and/or ginger return GSH level to its normal control animals.

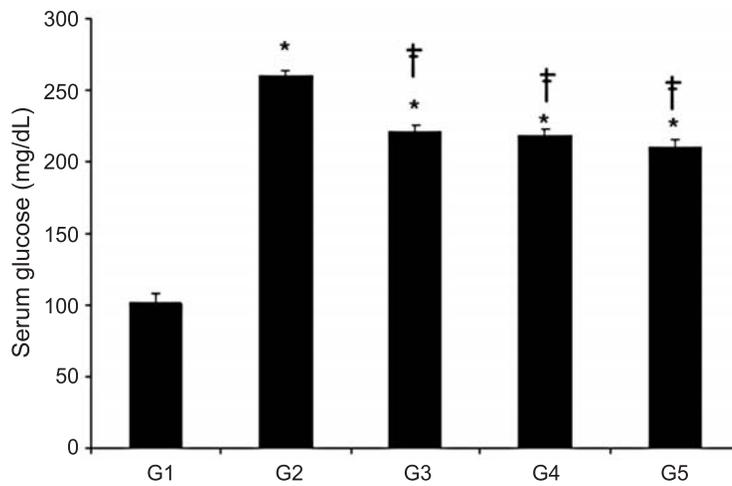


Figure 3. Serum glucose levels in normal, diabetic and diabetic treated animals. Values are the mean  $\pm$  SEM. \*  $p < 0.05$  vs. control group (G1) and ‡  $p < 0.5$  vs. diabetic group (G2)

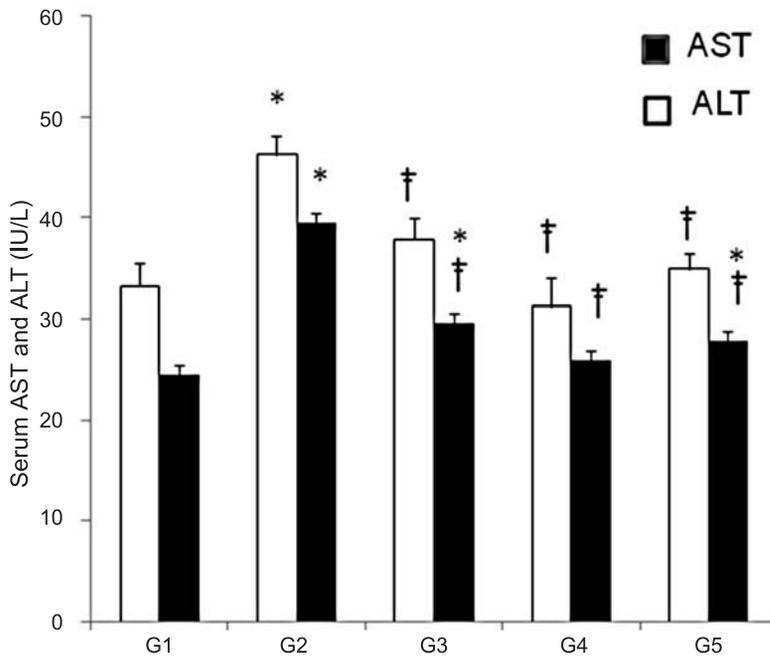


Figure 4. Serum AST and ALT levels in normal, diabetic and diabetic treated animals. Values are the mean  $\pm$  SEM. \*  $p < 0.05$  vs. control group (G1) and ‡  $p < 0.5$  vs. diabetic group (G2)

Regarding CRP and fibrinogen, alloxan-induced diabetic animals showed significant ( $p \leq 0.05$ ) increase in the levels of CRP and fibrinogen, however, post-treatment of diabetic animals with extract of green tea and/or ginger showed slight but not significant ( $p \leq 0.05$ ) increase in the levels of CRP and fibrinogen as shown in Figure 6.

Regarding immunoglobulins, alloxan decreased significantly IgG, IgM, and IgA compared to their levels in normal control group, as shown in Figure 7. Post-treatment of diabetic ani-

mals with either extract of green tea or ginger alone decreased significantly ( $p \leq 0.05$ ) the levels of immunoglobulins. However, post-treatment of diabetic animals with a mixture extract of both green tea and ginger decreased significantly ( $p \leq 0.05$ ) the immunoglobulins but in a lesser degree than groups post-treated with extract of either green tea or ginger alone, as shown in Figure 7.

As shown in Table 1, the results revealed that the RBC and WBC counts, PCV and neutrophil percentage decreased significantly ( $p \leq 0.05$ ), while

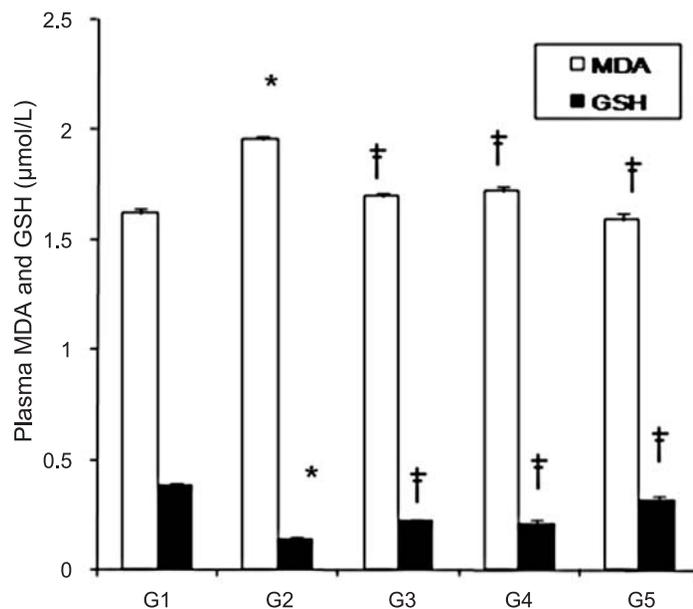


Figure 5. Plasma MDA and GSH levels in normal, diabetic and diabetic treated animals. Values are the mean  $\pm$  SEM. \*  $p < 0.05$  vs. control group (G1) and †  $p < 0.5$  vs. diabetic group (G2)

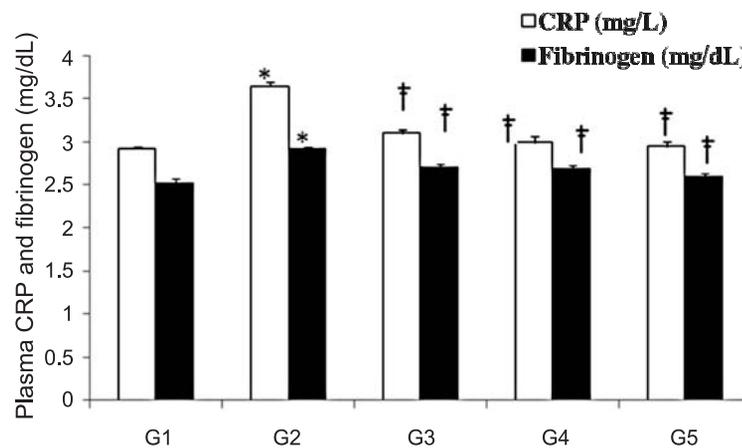


Figure 6. Plasma CRP and fibrinogen levels in normal, diabetic and diabetic treated animals. Values are the mean  $\pm$  SEM. \*  $p < 0.05$  vs. control group (G1) and †  $p < 0.5$  vs. diabetic group (G2)

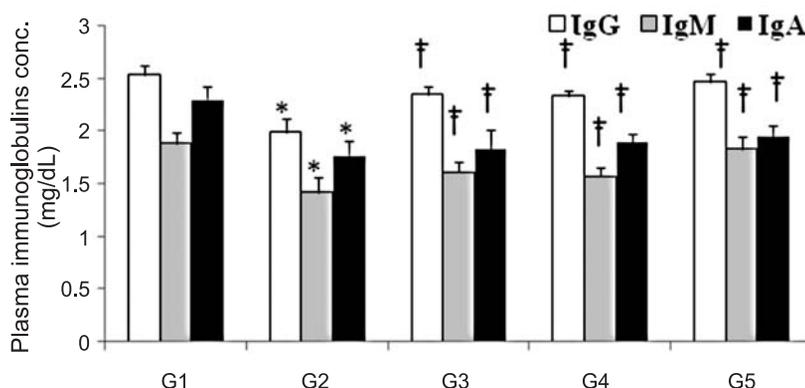


Figure 7. Plasma immunoglobulins concentrations in normal, diabetic and diabetic treated animals. Values are the mean  $\pm$  SEM. \*  $p < 0.05$  vs. control group (G1) and ‡  $p < 0.5$  vs. diabetic group (G2)

Table 1. Comparative hematological values of normal control (G1), diabetic (G2) and diabetic treated animals (G3, G4, G5). Values are expressed as the mean  $\pm$  SD. Means in the same row with different superscripts significantly differ ( $p < 0.05$ ). Number of rabbits in each group = 10.

Parameter	G1	G2	G3	G4	G5
PCV (%)	37.00 $\pm$ 2.9 <sup>a</sup>	29 $\pm$ 2.5 <sup>b</sup>	33 $\pm$ 2.8 <sup>c</sup>	32 $\pm$ 2.9 <sup>c</sup>	36 $\pm$ 3.2 <sup>a</sup>
Hb (g/dL)	12.45 $\pm$ 0.9 <sup>a</sup>	12 $\pm$ 1.1 <sup>b</sup>	12.2 $\pm$ 1.2 <sup>b</sup>	12.4 $\pm$ 1.1 <sup>a</sup>	12.47 $\pm$ 0.9 <sup>a</sup>
RBCs ( $\times 10^6/\text{mm}^3$ )	5.72 $\pm$ 0.4 <sup>a</sup>	4.1 $\pm$ 0.3 <sup>b</sup>	5.2 $\pm$ 0.1 <sup>c</sup>	5.4 $\pm$ 0.2 <sup>a</sup>	5.6 $\pm$ 0.2 <sup>a</sup>
MCV ( $\mu^3$ )	64.46 $\pm$ 5.4 <sup>a</sup>	70.73 $\pm$ 4.9 <sup>b</sup>	63.46 $\pm$ 5.1 <sup>a</sup>	59.26 $\pm$ 5.8 <sup>c</sup>	64.29 $\pm$ 5.2 <sup>a</sup>
MCH (pg)	21.6 $\pm$ 0.7 <sup>a</sup>	29.27 $\pm$ 1.2 <sup>b</sup>	23.46 $\pm$ 1.5 <sup>a</sup>	23 $\pm$ 1.2 <sup>a</sup>	22.26 $\pm$ 0.9 <sup>a</sup>
MCHC (%)	33.65 $\pm$ 1.8 <sup>a</sup>	41.37 $\pm$ 1.5 <sup>b</sup>	37 $\pm$ 1.8 <sup>c</sup>	38.75 $\pm$ 1.3 <sup>c</sup>	34.64 $\pm$ 0.8 <sup>a</sup>
WBCs ( $\times 10^3/\text{mm}^3$ )	5.4 $\pm$ 0.5 <sup>a</sup>	3.1 $\pm$ 0.9 <sup>b</sup>	4.4 $\pm$ 0.6 <sup>c</sup>	4.8 $\pm$ 0.8 <sup>d</sup>	5.3 $\pm$ 0.6 <sup>a</sup>
Neutrophils (%)	30 $\pm$ 1.3 <sup>a</sup>	25 $\pm$ 1.5 <sup>b</sup>	29 $\pm$ 1.3 <sup>a</sup>	28 $\pm$ 1.2 <sup>a</sup>	33 $\pm$ 1.3 <sup>c</sup>
Lymphocytes (%)	61 $\pm$ 2.3 <sup>a</sup>	63 $\pm$ 2.1 <sup>a</sup>	61 $\pm$ 1.8 <sup>a</sup>	62 $\pm$ 1.7 <sup>a</sup>	60 $\pm$ 1.4 <sup>a</sup>
Monocytes (%)	4 $\pm$ 0.3 <sup>a</sup>	5 $\pm$ 0.4 <sup>a</sup>	4 $\pm$ 0.6 <sup>a</sup>	3 $\pm$ 0.5 <sup>a</sup>	4 $\pm$ 0.3 <sup>a</sup>
Eosinophils (%)	3 $\pm$ 0.2	4 $\pm$ 0.1	3 $\pm$ 0.1	4 $\pm$ 0.1	2 $\pm$ 0.2
Basophils	2 $\pm$ 0.1	3 $\pm$ 0.1	3 $\pm$ 0.1	3 $\pm$ 0.1	1 $\pm$ 0.1

PCV = packed cell volume; Hb = hemoglobin; RBC = red blood cell count; WBC = white blood cell count; MCV = mean corpuscular volume, MCH = mean corpuscular hemoglobin, MCHC = mean corpuscular hemoglobin concentration.

MCV, MCH, and MCHC increased significantly ( $p \leq 0.05$ ) in alloxan-induced diabetic rabbits compared to normal control animals. Green tea and/or ginger extracts treatment increased ( $p \leq 0.05$ ) the lowered RBC and WBC counts, PCV and neutrophil percentage in diabetic rabbits. However, green tea and/or ginger extracts treatment decreased the elevated MCV, MCH, and MCHC of diabetic rabbits to normal level. The other parameters as percentages of lymphocytes, monocytes, eosinophils and basophils showed non significant changes among normal and treated groups.

## DISCUSSION

In the present study, statistical analysis revealed that alloxan-induced diabetic rabbits showed significant reduction as shown in the levels of serum triglycerides and LDL-c, but serum HDL-c level statistically increased (Figs. 1 and 2). However, post-treatment of diabetic animals with green tea or ginger extract ameliorated the lipid lowering action of alloxan, but in case of post-treatment with combination of green tea and ginger extracts, the level of triglycerides decreased significantly ( $p \leq$

0.05) more than the effect of post-treatment with green tea or ginger extract alone. These findings are in agreement with earlier studies including (9, 12) and others. It was concluded that the hypocholesterolemic effect of ginger or green tea could have possibly resulted from the inhibition of cellular cholesterol biosynthesis after the consumption of the extract (10). Furthermore, it was reported (18) that the reduction of cellular cholesterol biosynthesis is associated with increased activity of the LDL receptor, which in turn leads to enhanced removal of LDL from blood, resulting in reduced serum or plasma cholesterol concentration. In our study, we can attribute this reduction in most lipid parameters to the severe lipolysis caused by both extracts to overcome the effect of insulin deficiency.

Referring to Figure 3, the alloxan induced diabetic treated groups (G3, G4 and G5) showed significant decrease ( $p \leq 0.05$ ) in the level of glucose in comparison with diabetic untreated group (G2). In the present investigation, ginger and green tea extracts caused significant hypoglycemic effect in diabetic rabbits. Such a phenomenon of hypoglycemic response with green tea extract has already been reported (19). Figure 3 showed a significant increase in blood glucose levels in diabetic rabbits because of the destruction of pancreatic  $\beta$  cells by alloxan. The hypoglycemic effect of ginger increased gradually and was observed to be maximal at the end in group G5 when we used the two extracts together. The decrease in blood glucose levels was due to the antidiabetic compounds of ginger and different types of catechins present in green tea.

Regarding the effect of green tea and/or ginger extracts in alloxan-induced diabetic rabbits on the oxidant status and liver function enzymes; the present investigation showed that there were significant decreases ( $p \leq 0.05$ ) in lipid peroxidation and elevated plasma total antioxidant capacity and blood reduced glutathione concentration in the treated groups 3, 4 and 5 in comparison with the diabetic group 2 (Figs. 4 and 5). This finding strongly confirms the antioxidant properties of ginger reported in previous investigations. Ginger has been reported to have a lowering effect on lipid peroxidation by influencing the enzymatic blood level of superoxide dismutase, catalase, and glutathione peroxidase (20). It has been also shown that ginger reduces cellular oxidation, scavenge superoxide anion and hydroxyl radicals. Ginger free phenolic and ginger hydrolyzed phenolic fractions exhibit free radical scavenging activity. Depletion of tissue GSH levels enhances cellular damage caused by oxidative stress (21). Significant depletion of GSH ( $p < 0.05$ ) in dia-

betic rats suggests its increased utilization against reactive oxygen species (22). However, green tea and ginger treatment in diabetic rabbits reversed the GSH to normal levels, what shows that ginger has an antioxidant property. The antioxidative activity of green tea and ginger was attributed to scavenging superoxide anion and hydroxyl radicals by some phenols of ginger and catechins in green tea. Theoretically, our results may be due to the presence of many antioxidant compounds, phenolic ketone derivatives, catechins and volatile oils present in ginger and green tea.

Regarding the liver function enzymes, as shown in Figure 4, there were significant decreases ( $p \leq 0.05$ ) in the activity of mean values of GOT and GPT activity in treated groups 3, 4 and 5 in comparison to group 2. These results go with those showed earlier (23) and attributed this disturbance in enzyme activity to lipid peroxidation produced as a result of diabetes. In the present work, the normalization of enzyme activity may be due to the effect that comes from the usage of the green tea and ginger extracts which leads to a lowering effect of free radicals on the cell membrane of liver cells. These results go with those given previously from GSH and MDA (38).

Statistically, as showed in Figure 6, fibrinogen and CRP both showed significant decrease ( $p \leq 0.05$ ) in the treated groups 3, 4 and 5 when compared with diabetic group 2. The mean values of both fibrinogen and CRP showed significant increase ( $p \leq 0.05$ ) in comparison with the control one. These findings come in line with that stated in (24) and (25). The obtained data clarify platelet inhibition (PI) found in the diabetic patient by revealing a significant interaction in elevated plasma fibrinogen in the presence of DM. In this investigation, the effect of green tea and ginger extract administration lead to decrease of inflammatory response caused by DM on endothelial cells. This was very clear in Figure 6, especially in group 5, due to the usage of both green tea and ginger extracts. This may be attributed to antiplatelet effect of different phenols and catechins present in both green tea and ginger.

Immunological status of the animals under experiment is shown in Figure 7. The statistical analysis of the obtained data revealed that there was a significant decrease ( $p \leq 0.05$ ) in the mean values of immunoglobulins in diabetic group 2 in comparison with the control group 1. Administration of green tea and ginger extracts led to significant increase ( $p \leq 0.05$ ) in treated groups 3, 4 and 5. The obtained results come in accordance with that stated

in (26). Theoretically, the obtained data can be attributed to fact that the ginger phenols and tea catechins may possess immunostimulant effects and this point needs further investigation, which may be our future research point.

Referring to Table 1 in the present study, which indicated that treatment with extracts of green tea and/or ginger might ameliorate some disturbed hematological parameters of diabetic rabbits, it has been suggested that anemia occurrence in DM is due to the increased non-enzymatic glycosylation of RBC membrane proteins, which correlates with hyperglycemia (27). Oxidation of these glycosylated membrane proteins and hyperglycemia in DM caused an increase in the production of lipid peroxides causing hemolysis of RBC.

Hematological indices are indicators and reflection of the effects of dietary treatments on animals in terms of the quality of feed ingested and nutrients available to the animal to meet its physiological requirements. The values of hemoglobin (Hb), an iron-containing conjugated protein that performs the physiological function of transporting oxygen and carbon dioxide, which didn't show any significant changes in diabetic or diabetic treated groups compared with those on the control diet group, suggest that the animals didn't suffered depressed respiratory capability at any group indicating that the oxygen-carrying capacity of the blood of the animals are not affected either in diabetic or diabetic treated groups. Thus, increased RBC count of green tea and/or ginger treated diabetic rabbits could be due to the lowered lipid peroxide level in RBC membrane leading to a decreased susceptibility of RBC to hemolysis. Since non-enzymatic glycosylations of membrane proteins correlate with hyperglycemia (27), it might be said that green tea and ginger extract produced their effect by decreasing the elevated glucose concentration in green tea and ginger extract treated diabetic rabbits. However, more studies by measuring the RBC fragility, and serum folic acid, iron, cobalt, vitamin B<sub>12</sub> and calcium levels are needed to demonstrate the exact mechanism of action of green tea and ginger extracts on increased RBC count of diabetic rabbits. Therefore, our results suggest that the extracts of green tea and ginger stimulate the synthesis (erythropoiesis) and concentration of erythrocytes till normalizing RBC in anemic diabetic rabbits. The corresponding statistical decrease in the PCV (measure of the volume of blood consisting of solid cells) of the diabetic animals and its normalizing with green tea or ginger extract treatment suggest their role in erythropoiesis. Taken together, the

results of RBC, Hb and PCV suggest that the extracts of green tea and ginger possess antioxidant properties and help in RBC membrane stabilization by binding to proteins and carbohydrates which are components of RBC membrane and therefore may prevent breakdown of RBC membrane and antagonize the anemic effect of alloxan.

Neutrophils ingest and kill bacteria and have been called the body's first line of defense against bacterial infections (28). It has been suggested that the body's defense mechanism against infections was disturbed due to the disturbed neutrophil function in diabetes (29). In this experiment, we demonstrated that treatment of diabetic rabbits with green tea and ginger extract increased the lowered neutrophil percentage of WBC to the control level. This result indicated that green tea and/or ginger treatment might also increase the defense mechanism of the body against infections in diabetic rabbits. As mentioned above, it was found that RBC count increased to control level in green tea and/or ginger extract treated diabetic rabbits. Therefore, return of blood indices (MCV, MCH, MCHC) in diabetic treated rabbits could be due to a normalized RBC count in these rabbits. From the hematological results, it is apparent that oral administration of green tea and/or ginger extract might decrease the diabetes-induced disturbances of hematological parameters in alloxan-induced diabetic rabbits.

Data of the present study revealed that daily treatment with green tea and ginger extracts markedly improves hematological, immunobiochemical statuses of rabbits with alloxan-induced diabetes. In conclusion, our study suggests that green tea and ginger extracts if taken together may have more beneficial effects in diabetes as they may have synergistic action, thus holding the hope of a new generation of anti-diabetogenic drugs. However, comprehensive chemical and pharmacological research is required to find out the exact mechanism of green tea and ginger for their antidiabetogenic effect and to identify the active constituent(s) responsible for their effect.

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