

THE EFFECT OF SOME BIOCHEMICAL PARAMETERS AND SOME MINERAL SUBSTANCES IN RATS WITH DIFFERENT DOSES OF ELLAGIC ACID

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ABSTRACT

Background: The present study was conducted for the purpose of determining the dose of Ellagic Acid with which it may show its effects on some biochemical parameters and in some mineral substances. **Materials and Methods:** For this purpose, 40 male rats (Wistar-albino) that weighted 200-250 g were used in the study. A total of 5 groups of rats were formed and there were 8 rats in each group; the Control Group, Group 1 (EA 3 mg/kg), Group 2 (EA 10 mg/kg), Group 3 (EA 12 mg/kg), and Group 4 (EA 20 mg/kg). After 8 weeks of oral Ellagic Acid (EA) administration every other day, the measurements of the biochemical parameters in blood samples were

made with a modular auto-analyzer device by using a commercial kit. **Results:** When the mineral substance levels were compared with the Control Group, it was determined that the Glukoz, T.bil., İ.bil., albumin, Mg, Cl, UIBC levels increased in all groups to which EA was administered, and the Ca, Fe, urea, BUN levels decreased. **Conclusion:** As a result, based on the data obtained in the present study, it may be claimed that Ellagic Acid decreases the absorption of some mineral substances.

KEYWORDS: Ellagic Acid, Rat, Minerals, biochemical parameters.

INTRODUCTION

Ellagic Acid is a phenolic compound that exists in some fruits, and has antioxidant, anticarcinogenic and antifibrosis features. When Ellagic Acid is $\text{Ph} > 7$, it yields chelates with

bivalent cations like Zn^{2+} , Ca^{2+} , Fe^{2+} , Cd^{2+} , Cu^{2+} . Ellagic Acid is produced when ellagitannins undergo some chemical treatments.^[1] Ellagic Acid is a substance, which has antioxidant, anti-inflammatory, immunomodulatory, antimycotic, anti-carcinogenic^[2,3] and antihyperlipidemic^[4] activities. It also has an anti-aging effect.^[5]

Trace elements are the inorganic substances that participate in the catalytic, enzymatic and structural activities in the organism, and must be taken from outside with nutrients and water. The trace elements that are taken into the organism bind to various blood proteins and spread to all tissues. The trace element amounts are closely related with various factors like nutrition, age, disease, and the environment. The elements are necessary for proper functioning of cell functions. When they lack in the organism, metabolic abnormalities appear, and when they exist in excessive amounts, increases are observed in toxicity. The minerals that build up the human and animal organism are necessary to take in through diets at adequate levels since they exist in many physiological functions like growth, and reproductive and immune system.^[6] It is already known that trace elements play important roles in biological processes that cover the activation or inhibition of enzymatic reactions. For these reasons, the relations between trace elements and oxidative stress and cancer diseases were reported in previous studies.^[7]

The present study was planned to examine the effects of Ellagic Acid, which is used in the treatment and prevention of various diseases, on some biochemical parameters and mineral substance levels by administering it at different doses.

MATERIALS AND METHODS

Animals - In the present study, 40 male rats (Wistar-albino) that weighted 200-250 g, which were obtained from Experimental Animals Unit of Yüzüncü Yıl University, were used. The rats were kept in cages where continuous feed and fresh water were provided at 12 hours dark/light cycle; and the temperature was set as $22 \pm 2^{\circ}C$. The rats were selected randomly, and were divided into 5 groups.

Design of experiment groups - There were 8 rats in each group: The Control Group, Group 1 (EA 3 mg/kg), Group 2 (EA 10 mg/kg), Group 3 (EA 12 mg/kg), and Group 4 (EA 20 mg/kg). Throughout the 8-week study period, the Ellagic Acid (EA), which was dissolved in corn extract oil, was administered through oral gavage at the doses that are given above every other day. The Control Group was given corn extract oil through oral gavage.

Samples Collection - After the Study Period, 75 mg/kg Ketamine HCl was injected through intraperitoneal way, and the blood samples were taken through ponction from the left ventricle of the heart of the animals. These blood samples were centrifuged at 3000 rpm at +4°C for 10 min to obtain their serums.

Biochemical analysis—T The biochemical parameters were measured by using Cobas-brand kit with a Modular PP (Roche) Auto Analyzer. Following the necessary procedures on the blood samples, the samples were analyzed in ICP-OES (Inductively-Coupled Plasma-Optic Emission Spectroscopy) Device to determine the levels of mineral substances.

Statistical analysis - T One way analysis of variance (One-way ANOVA) and Duncan's multiple comparison tests were applied to the data, and the difference between the data was determined ($P < 0.05$).

RESULTS

The levels of elements in rat serum are shown in fig.1. The levels of Calcium (Ca), magnesium (Mg), sodium (Na), potassium (K), Chlorine (Cl), Iron (Fe), Unsaturated Iron Binding Capacity (UIBC) were assessed. The values of some biochemical parameters (Glukoz, Total bilirubin, Indirect bilirubin, albumin, urea, blood urea nitrogen (BUN)) of the study are shown in fig 2.

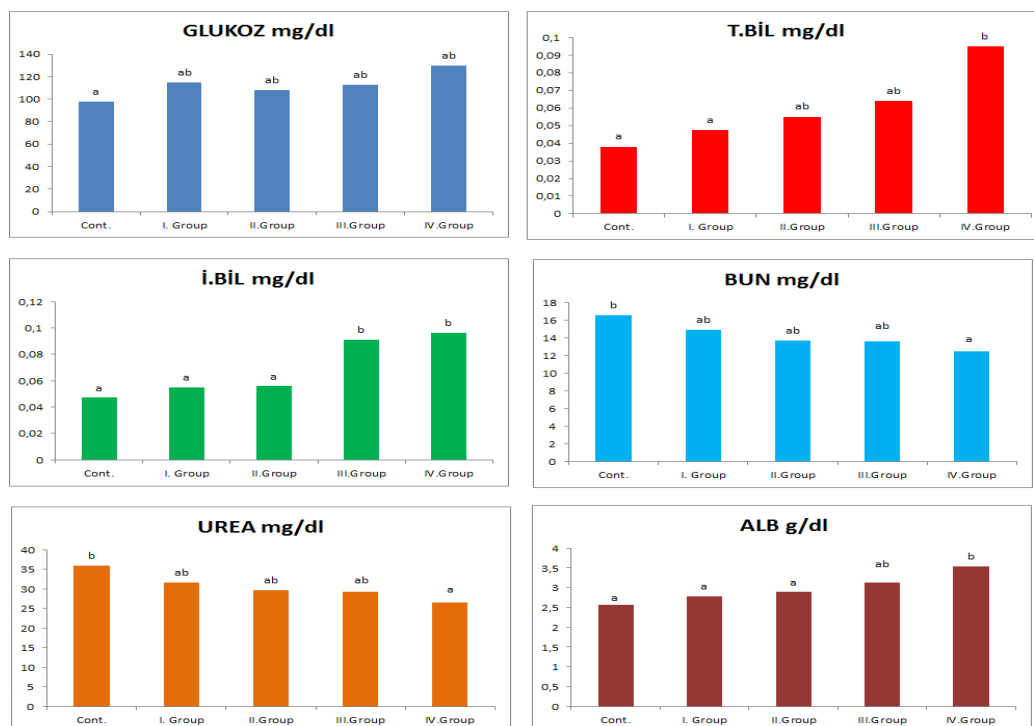


Figure 1: Glukoz, Total bilirubin, Indirect bilirubin, albumin, urea, BUN levels.

a, b: The difference among the group averages bearing different letters in the same column is statistically significant. $p < 0.05$.

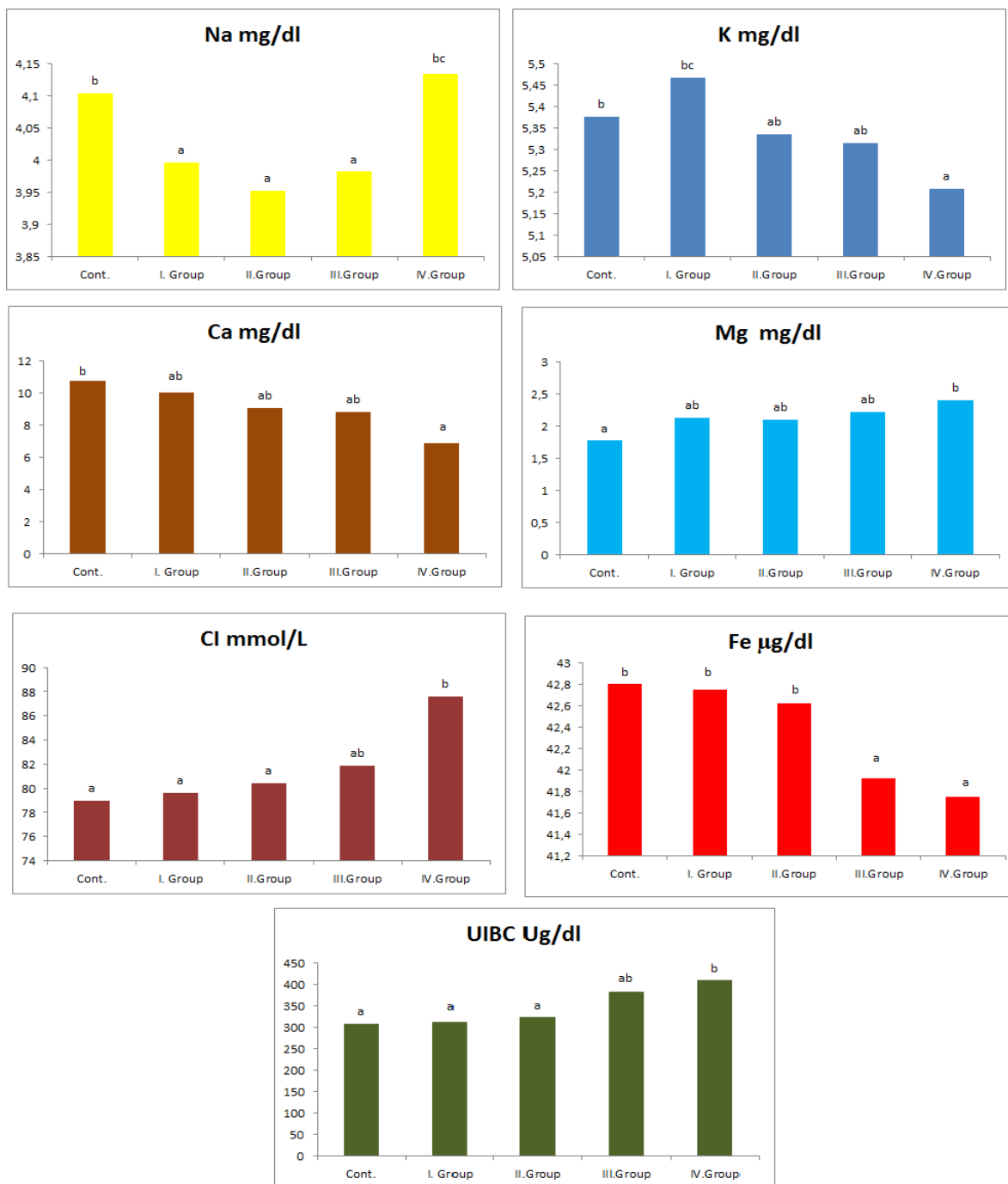


Figure 2: Calcium(Ca), magnesium(Mg), sodium(Na), potassium(K), Chlorine (Cl), Iron (Fe), Unsaturated Iron Binding Capacity (UIBC) levels.

a, b,: The difference among the group averages bearing different letters in the same column is statistically significant. $p < 0.05$.

DISCUSSION

The other components in diet might decrease or increase the absorption on minerals as well as the antagonistic interactions among them. The mineral distribution, their chemical forms and the interaction among minerals are important factors in the plants taken in with diets.^[6]

Phenolic Phytochemicals like Ellagic Acid are good chelators. For this reason, these phytochemicals may chelate ions in an effective way. They modify the Ca ions in the cellular matrix or in the cytosol.^[8] In a previous study, it was reported that Ellagic Acid increases Ca ions in the liver tissue that was exposed to Aluminum and had a healing effect.^[9] The Ca ions in cells are important regulators in various cellular processes, which include muscle contraction, secretion and cell division. Calcium has significant functions especially in bones, blood, the circulation, muscle system and in nerve conduction.^[10] Similarly, like calcium, magnesium is a structural component for bones, muscles and nerves.^[11] In our study, we determined that the Ca levels decreased.

Magnesium plays a role in the activation of many enzymes. In addition, it is also important in protein, nucleic acid, fat synthesis and muscle contraction.^[12] It was determined in our study that the Mg level increased as the dose increased.

Ellagic Acid also makes chelates with Fe ions. The increase in free Fe²⁺ in the cells results in lipid peroxidation, and therefore, in membrane damage.^[13] In a previous study, significant increases were shown in Fe level in groups to which pomegranate juice, which is known to have intense Ellagic Acid, was given.^[14] It was determined that iron-binding agents were highly effective in the fight against bacterial, parasitic and viral infections.^[15] In the present study, Fe levels decreased in high doses (3st and 4nd Groups) were administered. U_{1bc} is the unsaturated iron-binding capacity in the blood. Basically, it is considered as the measurement of all proteins that can bind to iron. It is employed to diagnose the anemia stemming from iron deficiency. U_{1bc} level increased in the third and fourth groups.

Cl, exists in biological systems as the main anion, which ensures that the extracellular volume is protected. Ionic balance plays a role in the osmotic pressure, water balance and regulation of acid-base balance by passing through the cell membrane. It is important in controlling the blood pH, the ready transfer of the chlorine in exchange for bicarbonate in the renal collection tubule that exist between the erythrocytes and the plasma and between the gastrointestinal tract. The major negative effect of increasing chlorine intake (i.e. sodium

chloride) is the increase in the blood pressure levels. Higher blood pressure is the risk factor for ischemic heart diseases, stroke and kidney diseases, which are the main causes of morbidity and mortality in Europe. Depending on the increasing Cl consumption in the form of sodium chloride, blood pressure increases depending on the dose. Cl is not a carcinogenic substance; however, high amounts of sodium chloride may increase the sensitivity to carcinogenic effects of carcinogens like nitrosamines, and gastric infection with *H. pylori*.^[16] In our study, Cl level increased in the third and fourth groups.

The function of the potassium is closely related to sodium in the body. As sodium consumption increases, the increase in potassium consumption may be more beneficial, because in addition to other benefits, it might also decrease the negative effects of high sodium consumption on blood pressure. Some studies reported that two nutritional rates were important factors in cardiovascular diseases and mortality. In addition, in Randomized Controlled Trials (RCTs), evidence was reported showing that a combination of increasing potassium and decreasing sodium intake might be influential in reducing blood pressure and cardiovascular mortality. Among the other benefits of increased potassium intake, there are increases in the glucose control, glucose intolerance, and insulin resistance. WHO reported that increased potassium showed differences according to the intervention types (supplementary foods or enriched foods), potassium supplementation types (potassium citrate, potassium chloride or others), and gender. It shows that the increase in potassium intake obtained from supplements or nutrients has a beneficial effect on blood pressure.^[17]

Sodium is a vital component in the human diet for cellular activity regulation and nervous system functioning, and is the main cation in the extracellular fluid in the body. It is also required for the maintenance of the plasma volume, acid-base balance, and transmission; and must be taken with nutrients. Sodium is required in products that are specifically formulated for blood regulation, and its deficiency might cause serious disorders in the bodily functions. It regulates the body fluids as an electrolyte, and conveys the electrical impulses in the body, helps to the regulation of the nerves and muscle contractions. In a previous study, it was reported that flavonoids were effective antioxidants with their chelate and antiradical mechanisms.^[18] Studies were conducted which showed that Ellagic Acid is effective in colon, esophagus and skin cancers.^[19,20] In cholestatic rats, it was reported that Ellagic Acid had modulator effects in serum and liver in Cu and Zn levels.^[21] Na and K are elements that are related closely to each other. Na exist in the cartilage and skin at the highest level, and K

exist in the liver, spleen and kidney at the highest level. Na plays an important role in osmotic pressure balance.^[17] Na and K increased in the first group, and decreased in the second and third groups. In the present study of ours, although decrease were detected in Na level in the first, second and third dose of Ellagic Acid, a increases was detected when the dose was increased. K levels were the opposite of the changes in Na level.

Urea and BUN are among important indicators that inform the doctor about renal functions. Sunil *et al.*,^[22] Conducted a study and did not report any differences in the glucose, total bilirubin, and albumin levels when the group to which Ellagic Acid was administered was compared with the Control Group. However, in our study, the glucose amount increased depending on the increase in the dose. In a study, which was conducted with pomegranate oil, which contained plenty of Ellagic Acid, the authors detected the total protein and albumin were increased, and the bilirubin and BUN levels did not change.^[23] In the present study of ours, the albumin amount increased at the highest dose administered. BUN and urea levels decreased in the fourth group. In another study, no changes were reported in the bilirubin level in the group to which Ellagic Acid was administered. However, when it was given together with cyclosporin, it reduced the increase in the group to which only cyclosporine was administered.^[24] In the present study, the total bilirubin and indirect bilirubin levels increased parallel to the increase in the dose.

CONCLUSIONS

The biochemical parameters of Ellagic Acid, its regulatory effects on mineral substances were shown in our study. As the Ellagic Acid dose increased, it showed its effect better. It may be argued that Ellagic Acid makes changes in the absorption of minerals. When Ellagic Acid dose is adjusted, this should be considered. It should not be ignored when it has different effects on different minerals.

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