

RESEARCH ARTICLE

Physiological and Histological Study of Preventive effect of Antioxidants, Selenium and Vitamin C with Garlic in Rats treated with Sodium Fluoride

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ABSTRACT:

The study included two aspects: study the effect of sodium fluoride on the level of liver enzymes, the oxidation system of Glutathione (GSH), on the Malondialdehyde (MDA), concentrations of urea and creatinine. In addition to its effect on the tissues of liver and kidneys in male white rats. And the study of the protective effect of antioxidants, selenium, vitamin C in garlic on sodium fluoride toxicity, 30 adult male rats were used and randomly divided into three equal groups. The control group was given water only, and the treatment group T1 was given sodium fluoride at a concentration of (20 mg / Kg) of body weight and treatment group (T2) were injected with sodium fluoride (20 mg / kg) and water garlic extract with a concentration of (125 mg / kg) body weight. The results showed that the effect of sodium fluoride was significant. And that the antioxidants, selenium and vitamin C contained in garlic gave a significant effect in inhibiting the toxicity of sodium fluoride used.

KEYWORDS: Sodium fluoride, Garlic, Malondialdehyde, Glutathione.

1. INTRODUCTION:

Sodium fluoride is the most toxic element in the halogen family, the second after arsenic in the toxicity scale (1). Fluorides are naturally found in the earth's crust with rocks, coal, clay and soil. Fluorides are released into the air by wind-blown waste. Fluoride is released into the air from fluoride-containing materials, including minerals, coal and clay, when heated at high temperatures, phosphates, aluminum smelters, glass, bricks, corks, the largest natural source of fluorides released into the air is volcanic eruptions (2).

Fluoride poisoning is the result of exposure to large quantities of fluoride and its compounds for a long period of time, which may occur by taking water containing high levels of fluoride or accidental ingestion of some pesticides, rodenticides or chronic inhalation of industrial gases and dust (3).

Sodium fluoride is absorbed in the small intestine, causing gastrointestinal inflammation which is an ectopic effect of the lining of the gastrointestinal mucosa; this increase in fluorosis leads to fluorosis, the fluoridosis is characterized by three types: dental fluorosis, which causes tooth decay and is stained with white spots (4). Sodium fluoride has effects on liver enzymes, and the increase in levels of hepatic enzymes ALT, AST, ALP is evidence of malfunction in liver function (5), a sign of cellular breakdown in liver tissue. Several studies have indicated that excessive exposure to sodium fluoride leads to various metabolic and metabolic changes in the effectiveness of hepatic enzymes (6). Garlic attracted the attention of modern medicine because of its widespread medical use around the world, the belief that garlic helps to give more strength and maintain good health free of disease, that garlic works (reduces the risk factor for heart disease, cancer, stimulate immune functions, promotes the elimination of toxicity Antibacterial, anti-bacterial effect, anti-oxidant effect (Allicin (allyl 2-propenethiosulfinate or diallyl thiosulfinate) is the active compound plus selenium and vitamin C found in fresh garlic and garlic extract (7).

MATERIALS AND METHODS:

In this study, used 30 adult white males, weighing between 250-180 kg, were divided into three equal groups of ten animals per group, as follows:

Control group (C):

Water was given only.

Treatment group (T1):

This group of rats was daily injected with sodium fluoride with a dose (20 mg / kg) of body weight.

Treatment group (T2):

This group of rats was daily injected with sodium fluoride with a dose of (20 mg / kg) of body weight and a water garlic extract with a dose of 125 mg / kg body weight.

Prepare the garlic extract in accordance with (8, 9) by taking (20 g) of garlic powder and extract the material from it in sequence with the Soxhlet extractor in 200 ml of distilled water for 24 hours. After the experiment, which lasted for one month, the animals were anesthetized using chloroform and blood was withdrawn directly from the heart using a 5 ml syringe. The blood

was placed in a clean glass test tube from the anticoagulation pipette. The serum was isolated and kept at -20°C Tests for measuring the concentration of glutathione and monaldehyde and calculating the level of liver enzymes (10, 11, 12) and calculating the levels of urea and creatinine by (13,14). The liver and kidneys were then removed and histological sections were prepared and dyed in the form of Hematocellin - Eosin (15).

Statistical analysis/Used program SPSS (2006), Statistical Package for Social Science, was used to analyze the experimental data to study the effect of the different parameters in the studied traits. The differences between the averages were compared with LES (Less Significant Difference).

RESULTS:

The results of the study, as shown in Table (1), showed a significant increase in the level of probability (P <0.05) in the levels of AST, ALP, ALT in the first treatment T1 compared with control group C. While the study showed improved enzymatic parameters with significant decrease in T2 compared with T1.

Table (4-1) The effect of sodium fluoride and the water extract of garlic plant on some of the enzymatic parameters of male white male rats in nesting (n = 10)

Standards / Groups	AST (V/L) Rate ± standard error	ALT (V/L) Rate ± standard error	ALP (V/L) Rate ± standard error
Control C	15.38±0.11a	10.37±0.07a	80.58±0.12a
T1	29.9±0.24b	21.41±0.11b	111.53±0.33b
T2	17.73±0.17a	12.54±0.14a	91.5±0.61a
L.S.D	0.554	0.347	1.224

*Figures represent the mean ± represents the standard error

*Different letters in one column indicate significant differences (P <0.05) between the totals.

*C represents the control group * T1 representing the animal group of sodium fluoride

*T2 represents the animal group of sodium fluoride with the water extract of garlic.

The study showed a significant increase in the levels of liver enzymes (AST, ALT, ALP) at the first treatment of T1 with sodium fluoride. While T2 treatment with the water extract of garlic led to the return of enzymatic standards to normal levels. The results were shown in Table (2) indicate a significant decrease in GSH level with significant increase in MDA lipid levels when

treated with T1 group of sodium fluoride compared to control group C. We note the return of the normal levels of GSH levels with significant decrease in the level of MDA in the treatment of the second group T2 with sodium fluoride with the water extract of garlic compared to the T1 group.

Table (2): Effect of sodium fluoride and water extract of garlic plant at the level of glutathione and lipid oxidation in the two interventions (n = 10)

Standards / Groups	Glutathione GSH (U/ mL)	MalondialdehydeMDA (µmol/L)
Control C	2.43±0.08 a	1.67±0.02 a
T1	1.22±0.03 b	2.43±0.05 b
T2	1.96±0.02 a	1.79±0.01 a
L.S.D	0.162	0.098

* Figures represent the mean ± represents the standard error

* Different letters in one column indicate significant differences (P <0.05) between the totals.

* C represents the control group * T1 representing the animal group of sodium fluoride

* T2 represents the animal group of sodium fluoride with the water extract of garlic.

Treatment of T1 group with garlic extract and sodium fluoride resulted in the return of the levels of MDA and GSH to their normal levels compared to the T1 group treated with sodium fluoride. The study also showed a significant increase in the levels of urea and creatinine at T1 treatment with sodium fluoride. While the standards improved with T2 treated by the water extract of garlic (Table 3)

Table (3): The effect of sodium fluoride and the water extract of garlic plant at the serum level of urea and creatinine for males of white rats in the interventions (n = 10)

Standards / Groups	Urea (mg/dl)	Creatinine (mg/dl)
Control C	21.54±0.25 a	0.62±0.02 a
T1	39.89±0.24 b	1.59±0.04 b
T2	25.66±0.39 a	0.82±0.02 a
L.S.D	0.920	0.102

* Figures represent the mean ± represents the standard error
 * Different letters in one column indicate significant differences (P <0.05) between the totals.
 * C represents the control group * T1 representing the animal group of sodium fluoride
 * T2 represents the animal group of sodium fluoride with the water extract of garlic.

As for the histological study, we observe the disappearance of the radial order of the hepatic cells around the central vein, which appears to be congested with enlargement of the hepatic corpuscles, with bubbles in the cytoplasm. 2 There is a clear bleeding inside the sinuses and infiltration of the inflammatory cells within the liver tissue, with obvious congestion and hyperplasia. Bile duct.

At the treatment of T2 sodium fluoride and the water extract of garlic we observe the return of the radiological arrangement of hepatic cells around the central vein and show hepatic cells natural and hexagonal faces that appear naturally. And a simple expansion in hepatocellular fibrosis as pictured (3).

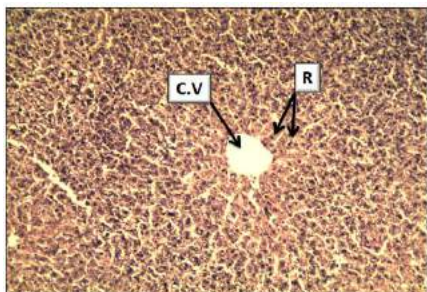


Fig.(1) liver rat. Control group C. Note the normal tissue of the liver where we note that the normal structure of the hepatocytes (R) which occur radially around the central vein (C.V) and the bile duct appear normal within the hepatic tissue (BD) 10X H and E.

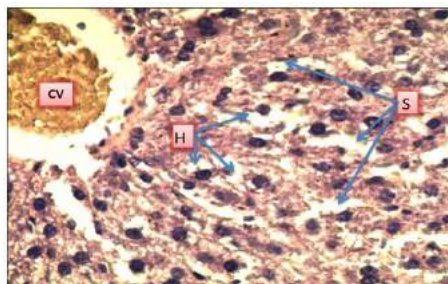


Fig.(2) T1 rat liver. The disappearance of the hepatic arrangement of hepatic cells around the central vein, which appears to be congested (CV), with the expansion of hepatic corpuscles (S) with swelling of hepatic cells where we observe the presence of bubbles within the cytoplasm 40X H&E

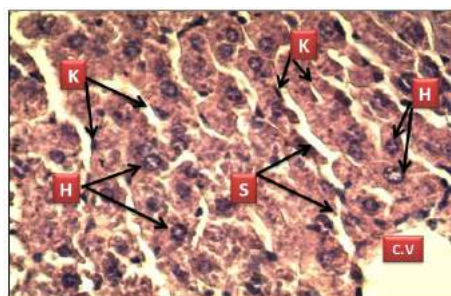


Fig.(3) T2 rat liver. Note that there is a radiographic arrangement of hepatic cells around the central vein (C.V) and hepatic cells showing normal and six-faceted (H) and simple expansion in hepatic gibbons (S) with proliferation of K-40X and E coffer cells.

The current study, as has shown in Figure (4,5) showed the tissue effects of the adult tissues of the T1 group. We notice severe hemorrhage in the renal tissue with an enlarged renal tubule lining and a clear atrophy of some renal glomeruli and a clear degeneration of the lining of the tubules.

The tissue histories of the adult tissues of the T2 group showed the presence of normal enlarged round glomeruli with narrow-lining tubes lined with normal vertical cells except for the degeneration of some of the lining cells of these tubules.

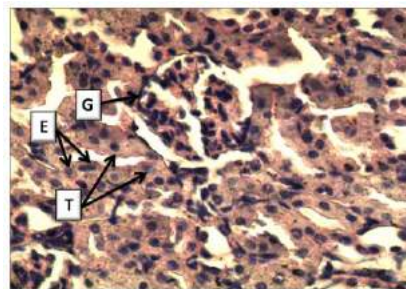


Fig.(4) College rat. Control group C. Note that there are normal and round natural glomeruli (G) with the presence of natural (T) nuclei, which appear in the form of vertical cells or a natural cube (E) 40X H and E

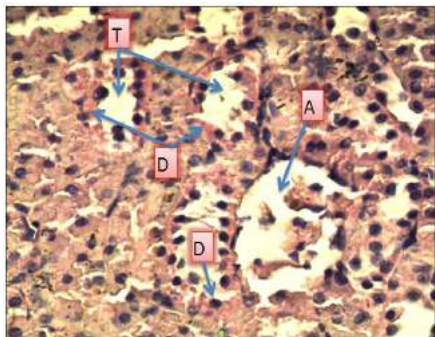


Fig.(5) College T1 rat. Acute renal glomerular atrophy (A) with enlarged renal tubular tubules (T), clear degeneration of dendritic cells (D). 40X H and E.

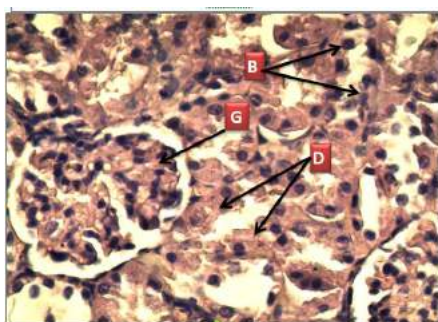


Fig.(6) College T2 rat. We observe the presence of normal enlarged round glands (G) and some degeneration of the endothelial cells of these tubules (D) and the presence of TUBULAR BASOPHILIA of the B-cells. 40XH and E.

DISCUSSION:

Moral elevation in liver enzymes AST, ALT, ALP is attributed to the formation of free radicals that attack the plasma membranes of the liver cells leading to the leakage of these enzymes (16). That the state of oxidative stress resulting from the increase of effective oxygen aggregates is the result of the breakdown of DNA, proteins and fats in liver cells, which leads to the degeneration of these cells and then crashing, and their contents to the bloodstream, including the enzymes above (17).

He explained that garlic extract has the effectiveness of protecting the liver from the toxicity of fluoride by reducing the process of oxidation by reducing levels (urinary F2-isoprostane, 8-hydroxyguanosine) and serum or through increased activity of the serum (SOD) superoxide dismutase and prevent the enzymes of the liver to rise and protect the cell membrane from Free radical attack (18).

The reduction in the level of GSH in T1 was due to the occurrence of oxidative stress due to the continuous treatment of sodium fluoride. As a result of the active GSH involvement in the prevention of oxidation in oxidative stress, either through the direct removal of free radicals or by enzymes that have a core substance such

as GSH peroxidase leading to an increase in the consumption of GSH and its transformation into an ineffective form of diphtheriaGSH (19). The significant increase in the level of MDA is due to the effect of sodium fluoride on the stimulation of the enzyme Fatty-acetyl-coA and the direct oxidation of fatty acids, which leads to increased production of H₂O₂ of internal origin and thus contributes to the production of fat oxidation (20).

The treatment of garlic extract resulted in a significant increase in the level of GSH and a decrease in morbidity in MDA due to the active role of garlic in the activation of the enzymatic and non-enzymatic antioxidant system within the body, as well as containing natural antioxidant foods such as vitamin C and selenium which protect LDL-C from Oxidation process (21). They confirmed (22) the presence of allicin, selenium, germanium, vitamin C and flavonoids found in garlic because they have the ability to remove toxic effects of heavy metals by binding and destroying them (23).

That the increase in levels of urea and creatinine in the treatment of T1 is attributable to fluoride poisoning, which leads to a lack of kidney function as the low rate of urea secretion leads to an increase in the level of urea in the blood (24). The high level of creatinine occurred due to the high level of protein and the proportion of plasma glucose in the plasma and to the oxidative stress associated with increasing the generation of free radicals that have the ability to work in the MDA in the kidney and the low level of antioxidants and antioxidant enzymes (25).

The treatment of T2 with the extract of garlic water to the return of levels of creatinine and urea to normal levels compared with the group T1, because of the inclusion of garlic on the compound (S-allyl cysteine; S-acssluphoxide), which contributes to reduce the effectiveness of Alkaline phosphate in the liver and garlic has a significant impact in the contribution reduction Concentration of this compound in the serum (26).

The effect of sodium fluoride on liver tissue is that fluoride has the ability to form free radicals such as OH hydroxyl and oxygen root, which increase the concentration of the liver, which leads to fat oxidation in hepatic cells and inhibits the antioxidant enzymes in the liver, Superoxide oxidase (SOD), and the effectiveness of (GUT) and catalytic catheter (27).

The changes in the kidney tissue are due to fluoride poisoning with T1, resulting in soft tissue functions by penetrating the cellular membranes into soft tissues, especially kidney tissues, promoting lipid peroxidation

and reducing the effectiveness of the antioxidant enzymes, which cause apoptosis of cells with decomposition renal tubule cells (28).

And that the positive effects of garlic in the treatment T1, T2 liver and kidney tissues may be due to its active role in the activation of the system of enzymatic and non-enzymatic antioxidants within the body as well as contain the natural food antioxidants such as vitamin C and selenium(29). Vitamin C is an antioxidant because it is a source of energy and activates many enzymes and metabolic processes. It activates the enzyme adenylcyclase and inhibits the phosphate esterase enzyme, which leads to increased levels of adenosine monophosphate (cAMP). Metabolism in tissues requires this, And differentiation of cells (30). Some research has indicated that selenium has positive effects in tissue lesions in cases of fluoride poisoning (31).

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