



## Effect of Nano-Piperine and Vitamin C in Antioxidant Status in Dexamethasone Treated Male Rats

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### Abstract

The study was designed to investigate whether nano-piperine and vitamin C (Vit. C) have antioxidant activity and/or it act through induction of the endogenous antioxidants. The study involved 48 male Wister rats (90 days old, 150±10g weight) randomly assigned to 6 groups (8 per group) and administered orally for 30 days. T1 was given 15 mg/kg B.W. nano-piperine, T2 was given 25 mg/kg B.W. vitamin C, T3 was given 2mg/kg B.W. dexamethasone, T4 was given both at the same time, and T5 was given both at the same time. Malondialdehyde (MDA) concentration and activities of superoxide dismutase (SOD), catalase (CAT) and glutathione (GSH). were measured in blood serum. The study found a substantial rise ( $p<0.05$ ) in MDA levels in the T3 group given dexamethasone compared to the control group. Significant reduction ( $p<0.05$ ) in MDA levels was seen in T4 and T5 groups compared to T3. Additionally, MDA levels increased significantly ( $p<0.05$ ) in T1 and reduced in T2 compared to the control group. The T1 and T2 groups showed a substantial increase ( $p<0.05$ ) in SOD, CAT, and GSH compared to the control group. T3 had the lowest SOD, CAT, and GSH and other experimental groups. Additionally, T4 and T5 showed a substantial increase ( $p<0.05$ ) in these parameters compared to T3. It can be concluded that nano-piperine and Vit. C has antioxidant activity and plays a great role in reducing free radicals and increasing endogenous antioxidants.

**Key word: Dexamethasone, Antioxidants, Nano-Piperine, Vit.C**

### Introduction

Damage to cells and tissues can be caused by oxidative stress, an imbalance between the body's antioxidants and free radicals. Oxidative stress arises when the oxidant/antioxidant balance is disrupted, leaving the body vulnerable to a steady increase in the production of reactive oxygen species (ROS) and reactive nitrogen species (RNS). Reactive oxygen species (ROS) refers to a group of molecules and free radicals produced when molecular oxygen reacts with certain molecules (1). Free radicals naturally produced by the body as a

byproduct of normal metabolism, they are highly reactive and short-lived molecules. Toxic chemical exposure can also cause the body to produce free radicals (2). Antioxidants are man-made or naturally occurring substances that have the potential to prevent or slow down certain types of cellular damage. Enzymatic and non-enzymatic antioxidants are the two main categories used to describe antioxidants. Antioxidant enzymes include the catalases, peroxiredoxins (PRXs), superoxide dismutase (SODs), glutathione



peroxidases (GPXs), and the four enzymes of the ascorbate–glutathione pathway (3). Increase in the levels of anti-oxidants in the tissue can protect the tissue from damage caused by free radicals. piperine possess antioxidant property. Apart from anti-oxidant property it can maintain the levels of superoxide dismutase, catalase, glutathione peroxidase, glutathione-s-transferase and glutathione in addition to reduce the thiobarbituric acid reactive substances(4). Vitamin C has been referred to as L-ascorbic acid since it was discovered as a scurvy

## Materials and methods

### Experimental design

48 Male rats had been divided to 6 equal groups ( 8 male rats of each group) and treated as follow:

1. Control (C):- Drenched Orally with 0.5 ml distilled water daily for 30 days.
2. T1 group :- Drenched orally with Nano-piperine (15mg/ kg B.W) Suspended in 0.5 ml distilled water daily for 30 days.(7)
3. T2 group:- Drenched orally with Vit. C (25 mg/ kg B.W) Suspended in 0.5 ml distilled water daily for 30 days.(8).
4. T3 group :- Drenched orally with dexamethason(2mg/ kg B.W) Suspended in 0.5 ml distilled water daily for 30 days (9)
5. T4 group :- Drenched orally with nano piperine(15mg/kg B.W) and dexamethasone (2 mg/ kg b.w) suspended in distilled water daily for 30 days.
6. T5 group:-Drenched orally with Vit. C(25mg/kg B.W) and dexamethasone (2 mg/ kg B.W) suspended in distilled water daily for 30 days.

## Results

### Concentration of malondialdehyde MDA ( $\mu\text{mol/mL}$ ).

treatment. Vitamin C was coined to replace "fat-soluble vitamin A" and "water-soluble vitamin B." The name also encompasses L-dehydroascorbic acid, which the body rapidly converts to L-ascorbic acid (5). Piperine has been used in many conventional medications to relieve numerous infections, as well as anti-septic and diuretic, and to enhance-digestion, as well as insecticides and anti-bacterial (6). Our study aimed to investigate the antioxidant effect of nano-piperine and vitamin C in dexamethasone-treated male rats.

Male rats have been monitored throughout the experimental periods. After the termination of experiment the animals were dissected and blood samples were collected then blood serum samples were detached for assessment of MDA concentration and activities of SOD, CAT and GSH.

Dexamethasone, piperine and Vit.C had been obtained from sigma, USA.

### Laboratory measurements

MDA ,CAT,GSH and SOD were estimated according to A colorimetric method by using kit(Biosolar, China)

### Ethical approval:

The researchers obtained ethical approval from the research Ethical Approval Committee of the College of Veterinary Medicine, University of Al-Qadisiyah

### Statistical Analysis:

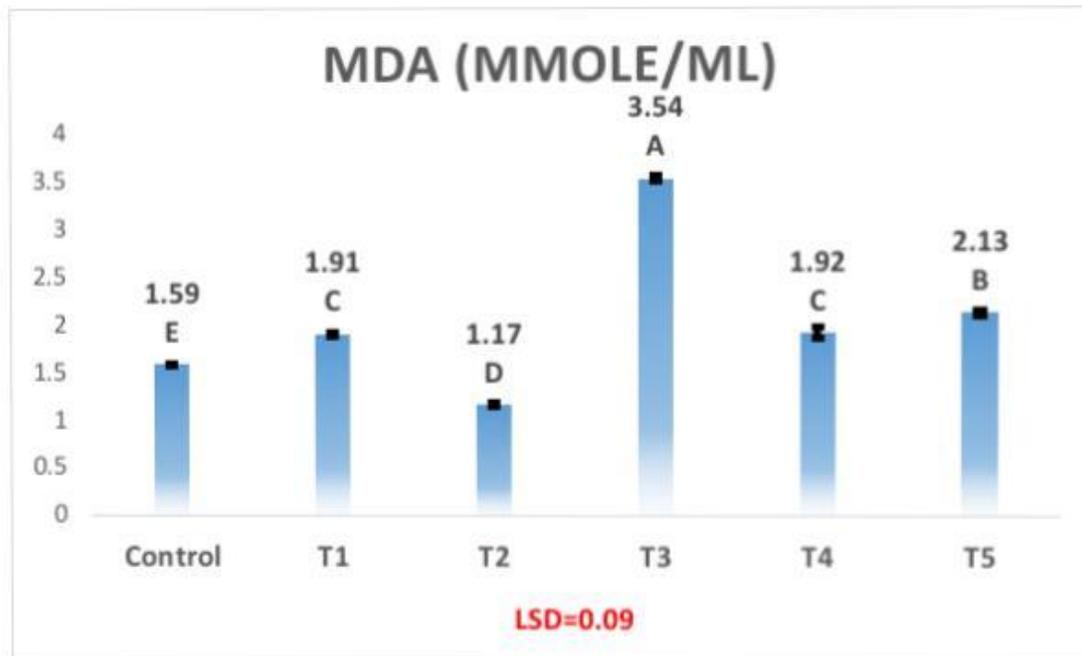
Results were detailed as mean  $\pm$  standard error of the mean (SEM). Contrasts were achieved using one way analysis of variance (ANOVA). When  $p < 0.05$ . All statistical analysis by (SPSS, version 22 ) (10).

Fig. (1) showed a significant difference ( $p < 0.05$ ) in MDA concentration among the experimental groups. The results of the current



study showed highest significant concentration of MDA recorded in T3 ( $3.54 \pm 0.03$ ) which dosed with dexamethasone as compared with the control group. While there was a significant decrease ( $p < 0.05$ ) in concentration of MDA in T4 and T5 groups ( $1.92 \pm 0.06$  and  $2.13 \pm 0.03$ )

respectively in compare with T3. Furthermore, the result showed a significant increase ( $p \leq 0.05$ ) in concentration of MDA in the T1 group ( $1.91 \pm 0.01$ ) while lessened in T2 ( $1.17 \pm 0.01$ ) in compare with the control group ( $1.59 \pm 0.003$ ).

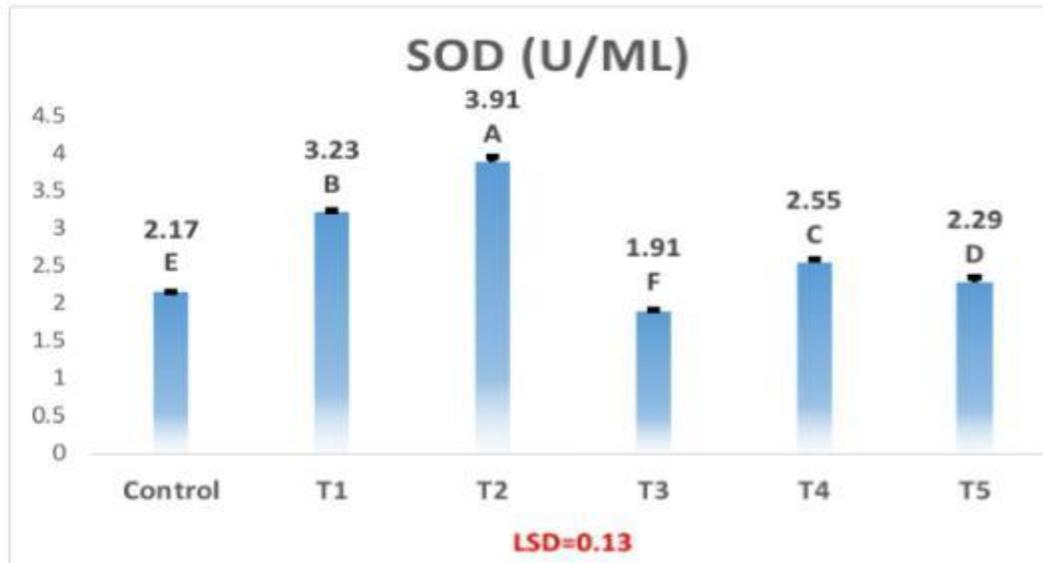


**Fig 1: Effect of nano-piperine, dexamethasone and vit. C on MDA concentration**

**Activity of Superoxide dismutase (SOD) (U/ML).**

The results explained in fig. (2) showed a significant variance ( $p < 0.05$ ) in SOD activity among the experimental groups. The results indicated a significant rise ( $p < 0.05$ ) in SOD activity in T1 and T2 groups ( $3.23 \pm 0.021$  and

$3.91 \pm 0.055$ ) respectively in compare with the control group ( $2.17 \pm 0.004$ ). On the other hand, T3 group recorded the lowest SOD activity ( $1.91 \pm 0.014$ ) among the other experimental groups. Furthermore, there was a significant rise in the T4 and T5 groups ( $2.55 \pm 0.044$  and  $2.29 \pm 0.067$ ) respectively as compared with T3 .



**Fig 2: Effect of nano- piperine, dexamethasone and vit. C on SOD activity**

#### **Activity of Glutathione (GSH) (MMOLE/ML)**

Fig. (3) revealed a significant difference ( $p < 0.05$ ) in GSH activity among the experimental groups. The results of the present study exhibited a significant rise ( $p < 0.05$ ) in GSH activity in T1 and T2 ( $3.64 \pm 0.011$  and  $3.26 \pm 0.048$ ) respectively as compared with the all of the groups. T3 group recorded the lowest GSH activity ( $1.54 \pm 0.022$ ) among the other experimental groups. Furthermore, there was a significant increase in the T4 and T5 groups ( $2.81 \pm 0.066$  and  $2.74 \pm 0.043$ ) respectively as compared with T3 group

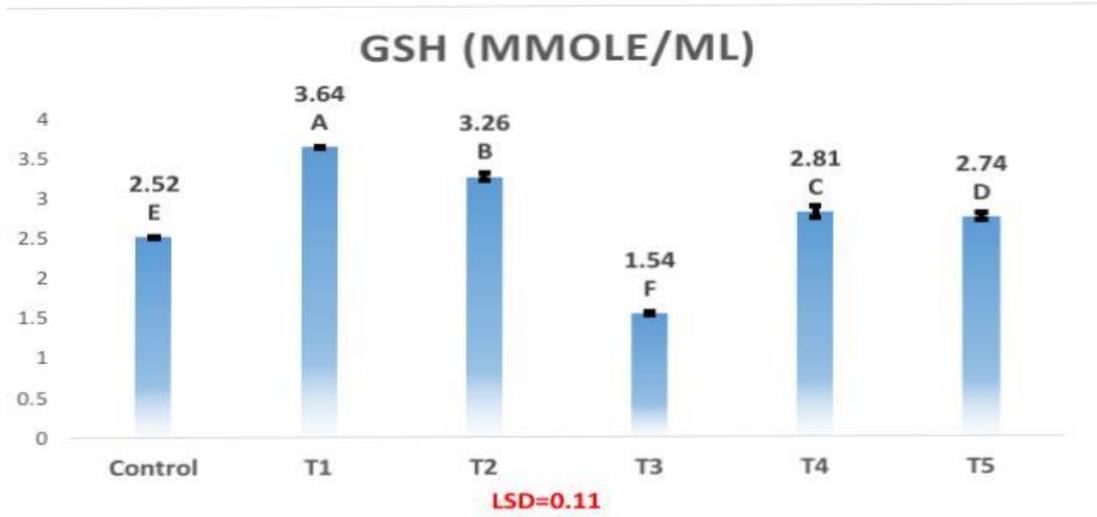


Fig 3: Effect of nano-piperine, dexamethasone and vit. C on GSH activity.

#### Activity of Catalase (CAT)(U/ML)

Fig. (4) indicated a significant difference ( $p \leq 0.05$ ) in serum CAT activity among the all experimental groups. The highest significant difference ( $p < 0.05$ ) in CAT activity was recorded in the T1 group ( $1.31 \pm 0.009$ ) and T2 ( $0.98 \pm 0.017$ ) as compared with control group ( $0.63 \pm 0.004$ ). While, T3 recorded the lowest CAT activity as compared with the all of the groups. The results showed a significant increase ( $p < 0.05$ ) in CAT activity in T4 and T5 ( $1.03 \pm 0.016$  and  $0.81 \pm 0.019$ ) as compared with T3.

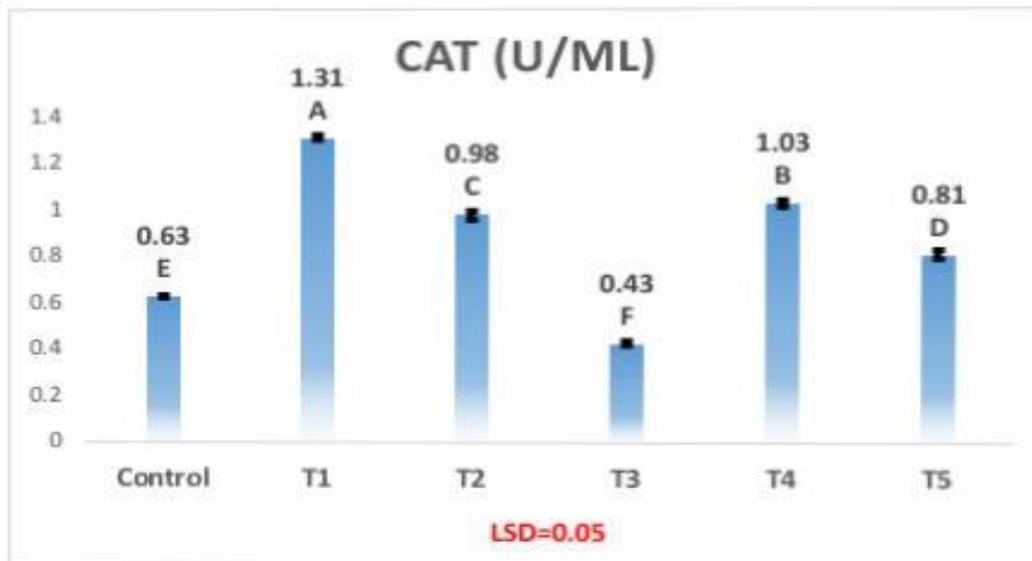


Fig 4: Effect of nano-piperine, dexamethasone and vit. C on CAT activity.



## Discussion

According to the result of our study and compared to dexamethasone treated group, the MDA levels in other groups significantly decreased. MDA is known as an indicator of terminal lipid peroxidation. Plasma levels of malondialdehyde were raised in dexamethasone groups, which may be described by the imbalance between augmented production of free oxygen radicals and impaired antioxidative defence. This imbalance could be viewed as the primary cause of chronic inflammation and the component responsible for its maintenance. Dexamethasone causes increase in MDA concentration and decreased in GSH and CAT (11). Reduced SOD and CAT activity will accumulate these highly reactive free radicals and the loss of cell membrane integrity and function (12). According to earlier studies on the effect of dexamethasone on oxidative stress in the small intestine and kidneys, the mean serum MDA level significantly increase in the dexamethasone-treated group. Similarly, to MDA, lipid peroxidation was enhanced in patients treated by corticosteroid therapy (13). According to the data, the MDA level in T2, T4 group and T5 group was significantly lower than in T3 group, which could explain the preventive impact of piperine and vit.C which act as antioxidants agents in reducing the MDA concentration. Recent research has shown that prenatal vitamin C supplementation significantly reduces lipid peroxidation and MDA generation (14). Salihu *et al.* (2022) (15) Whom showed that evaluated plasma MDA levels in patients with moderate asthma and found no significant changes between inhaled steroid users and non-users. Within cells, protein disulfide isomerase and glutaredoxins catalyse the reaction that maintains glutathione's reduced state. Ascorbic acid can neutralise hydrogen peroxide and other reactive

oxygen species (16). Glutathione, a peptide containing cysteine, is prevalent in aerobic organisms. It can be synthesized in the body from other amino acids; therefore, its consumption in food is not required. Glutathione is an antioxidant because its cysteine component contains a thiol group, which may be oxidized and reduced (17). Glutathione reductase is responsible for maintaining the reduced form of glutathione in cells, allowing it to reduce oxidants and other compounds. This category includes ascorbate in the glutathione peroxidases, glutathione-ascorbate cycle, and glutaredoxins. Because of its great concentration and significant role in maintaining the cell's redox state, glutathione is one of the greatest essential antioxidants in cells (18). Superoxide dismutase (SOD) is the major antioxidant and first detoxifying enzyme within cells. This enzyme is a vital component of the body's first line of defence against reactive oxygen species. It is naturally produced and secreted by the liver. It catalyses the transformation of two molecules of the hazardous superoxide anion into hydrogen peroxide and molecular oxygen. As SOD is a metalloenzyme, its function needs a metal cofactor (18). Nano-piperine which possesses protective effects, protects against oxidative stress damage by blocking or quenching free radicals, ROS, and lipid peroxidation and has been reported to function as a low-concentration hydroxyl radical scavenger (19). GSH, a major endogenous defence, is believed to protect cell membranes from peroxidative damage. Glutathione reduces the concentrations of both lipid peroxides and activated oxygen species. GSH concentrations in tissues are proportional to detoxification capacity. Detoxifying the body of superoxide ions and hydroxyl radicals is likely one of the greatest



effective defence mechanisms against numerous diseases (20).

## Conclusion

from the results obtained from this study, one can conclude that nano-piperine and vitamin c

play an important role in improving oxidants-antioxidants status.

**Conflict of Interest:** there is no conflict of interest.

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