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## **Protective Role of Vitamin C Zinc against Toxicity Induced by Cadmium in White Rats**

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### **Abstract**

The present study was conducted to determine the protective effects of zinc and vitamin C, individually or in combination with Cd, in order to monitor their ability to improve against cadmium-induced oxidative damage in albino rats. Through research, it was found that cadmium is a toxic element that harms liver tissue. Vitamin C and zinc have been shown to have an important protective role against the toxic effects of cadmium. The study was conducted on 40 male rats and it was divided into two periods for each period of five groups, each group containing four mice. The duration of the first period was 3 weeks, and the second period was 6 weeks. This study was conducted by biochemical criteria and blood analysis in affected albino rats administering a dose of cadmium with drinking water 10 mg/L individually for (3 weeks, 6 weeks) and evaluate the role of preventive measures. The results showed a significant decrease  $p < 0.05$  in RBC, Hb and pcv in the cadmium-treated group when compared with the control group and a significant increase in WBC in the cadmium-treated group when compared to the control group for two weeks (3,6) For pcv, Hb, there were no significant differences  $p > 0.05$  for the (cd + vt.c, cd + zinc, cd + vt.c + zinc) groups.

When compared with the control between the two periods (3,6) weeks, while WBC and RBC there were significant differences  $p < 0.05$  for the treated groups (cd+vt.c, cd+zinc, cd+vt.c+zinc) when compared with control, and when

comparing between the two periods, the results showed that there were significant differences  $p < 0.05$  for the cadmium group compared with the treated groups (cd+vt.c, cd+zinc, cd+vt.c+zinc) for all blood parameters. Biochemical tests the levels of urea and creatinine in the rats treated with cadmium increased significantly ( $p < 0.05$ ) when compared to the control group and both periods, according to the hematological parameters. As for urea, there were significant differences,  $p < 0.05$ , for the treatment groups (cd+vt.c, cd+zinc, cd+vt.c+zinc) compared with control and both periods. In terms of creatinine, there were significant differences between the treatment groups (cd+vt.c, cd+zinc, and cd+vt.c+zinc) and control group ( $p > 0.05$ ) for both times. For all biochemical parameters, the cadmium group's results showed statistically significant differences ( $p < 0.05$ ). Vitamin C is known as an element essential food for all kinds of animals. In other words, these vitamins have been shown to have a protective effect against toxicity caused by metals. In conclusion, this study showed that oral exposure to cadmium caused a decrease in biochemical and hematological activities in rats, and vitamin C had a reinforcing effect against toxicity that Mineral induced A natural antioxidant that prevents increased free radical production resulting from oxidative damage to lipids and lipoproteins in many tissues.

**Keywords:** Cadmium, Zinc, Vitamin C, Amelioration Potential

### **Introduction**

Environmental Pollutions one of the primary issues plaguing the planet due to its destructive activities<sup>[1]</sup>. Pollution is defined as "a state of disequilibrium from a state of equilibrium in any system" and refers to all types of pollution. According to the following, smoking is one of the most prevalent causes of illness and death that goes unrecognized because it is an internal source of pollution, causes the greatest exposure to gases, chemicals, and heavy metals to which humans are exposed, and causes their entry into the systemic circulation to have negative health effects<sup>[3]</sup>. Metallic elements are integral elements of the environment which are difficult to remove completely. With increasing use of a wide variety of metals in industry and in our daily lives, serious problems resulting from toxic metal pollution of the environment have been reported<sup>[4, 5]</sup>. In recent years, there have been increasing ecological and global public health problems connected with environmental contamination by these metals. Dependably, human exposure has risen significantly as a result of an exponential increase of their use in several industrial, agricultural, domestic and technological applications<sup>[5]</sup>. Heavy metals can be accumulated by organisms through a

variety of pathways, including absorption, respiration, and ingestion, with high toxicity to many organs of both animals and humans [6, 7]. Cadmium is one of the most toxic heavy metals. It is used in the manufacture of alloys, dyes, electronic compounds, stabilizers, and, in particular, in rechargeable nickel-cadmium batteries. Hence, its presence in the environment has increased with the industrial development [8, 9]. Reactive oxygen species (ROS) generation and the onset of oxidative stress are both induced by toxicity in many organs [10]. Cd exposure also promotes tissue damage brought on by lipid peroxidation [11]. Both enzymatic and non-enzymatic antioxidant mechanisms are present in live cells to protect them from oxidative damage. Due to modifications in the systems controlling gene expression, cadmium causes oxidative stress by impairing the antioxidant enzyme system [12]. When the cellular oxidoreduction balance is upset, it can cause significant harm to tissues and organs, which can impede their ability to operate [8]. The antioxidant state under cadmium poisoning in many systems is a topic that researchers are currently focusing on. Endogenous antioxidants are well known to be crucial in Antioxidant defense mechanisms against oxidative stress that show the protective character of particular biological processes [10, 13]. Cigarette smoke, food, and water all contain it. The toxic effects of cadmium at the tissue level are still poorly known in biological systems, though, and neither are the processes and pathways that cause them. Several research have demonstrated this. Vitamin C, sometimes referred to as ascorbic acid and ascorbate, is a nutrient that is included in many foods and is also available as a dietary supplement. Scurvy can be prevented and treated with it. For the enzymatic digestion of certain neurotransmitters as well as for tissue healing, vitamin C is a crucial nutrient. This is crucial for the proper operation of numerous enzymes as well as the immune system. As an antioxidant, it also functions [14]. Vitamin C was first isolated in 1928, classified as a naturally active vitamin in 1933, and discovered in 1912. [15] According to the World Health Organization's List of Essential Medicines, it is one of the safest and most effective medicines required in a health service. There's a commonly available, budget-friendly over-the-counter vitamin C supplement [16]. Vitamin C is found in foods include strawberries, broccoli, kiwi, bell peppers, and citrus fruits. Heat or extended storage could cause the amount of vitamin C of processed foods to decrease [17].

## Materials and Method

### Chemicals

Cadmium as cadmium chloride (CdCl<sub>2</sub>) and vitamin C (ascorbic acid) zinc as zinc chloride (ZnCl<sub>2</sub>) were obtained from India's Cd-Fine chemicals and Loba Chemicals. coming from Sigma Chemical Co. The maximum purity was used for all the substances used in this study.

### Experimental design

In this study, 40 male rats were used, and they were randomly distributed into five groups of equal number, as each group included 8 males, 4 of which were used in each of the two experimental periods 3 of and 6 weeks for each group, and the five groups were treated as follows:

1-The first group: control

2- The second group: 10 mg/L of CdCl<sub>2</sub> was dissolved in drinking water

3-The third group: 10 mg/L of CdCl<sub>2</sub> was dissolved in drinking water

+ vitamin C (500Mg/L of drinking water)

4-fourth group: 1: 10 mg/L of CdCl<sub>2</sub> was dissolved in drinking water

+ zinc (50mg/L of drinking water

5-fifth group: 10 mg/L of CdCl<sub>2</sub> was dissolved in drinking water

+ vitamin C (500mg/L of drinking water) + zinc (50mg/L of drinking water)

### Blood Sample Collection

Animal sacrifice and blood draw Animal sacrifice Animal sacrifice after the end of the two experimental periods [6, 3] weeks, after anesthesia with chloroform by inhalation. A large amount of blood is drawn directly from the heart through a puncture in the heart. Physiological blood analyses, while a portion of another substance in air (3 ml) was placed in a tube containing an anticoagulant, the blood was then separated from the centrifuge (3000 rpm for 15 min) The final loop was opened after it up to 20 degrees Celsius until the checks. Liver, liver), then keeping the organs in formalin solution (10%) until performing histological resection.

These tests included blood complete count (CBC) including (WBC, Hb, PCV, WBC), and measuring Kidney function Parameters Values Serum urea and Creatinine

### Laboratory tests

1-Hematological analysis

To use an automated auto-analyzer xp-300 (Symex) to analyze EDTA blood, hematological parameters were performed conducted. Blood complete count (CBC) measures included (WBC, Hb, PCV, WBC).

2 Kidney function Parameters Values Serum urea and Creatinine.

## Results

### Hematological Parameters values

**Table 1:** Differences in Hematological Parameters values (RBC) between the treatment groups are 3 weeks (control, cd, cd+ vt.c, cd + zinc, cd+ vt.c+zinc) and the treatment groups being 6 weeks (control, cd, cd+ vt.c, cd + zinc, cd+ vt.c+zinc)

Treated groups	RBC (10X <sup>6</sup> )		Univariate Tests	Multivariate Tests
	Mean ± SD			
	3 W.	6 W.		
Control	4.93±0.15	5.27±0.47	0.0001*	LSD=0.339 p-value=0.0001*
Cd	4.23±0.12	3.18±0.05		
Cd + Vt.C	5.80±0.10	4.42±0.27		
Cd + Zinc	5.82±0.11	4.81±0.09		
Cd + Vt.C. + Zinc	5.60±0.10	6.08±0.15		
LSD	0.210	0.467		
p-value	0.0001*	0.0001*		

The results of the study showed a significant decrease  $p < 0.05$  in the rate of red blood cells in the cadmium group when compared with the control group, while the results showed a significant increase  $p < 0.05$  for the treatment groups (cd + vt.c, cd + zinc, cd + vt.c + zinc) when compared in the cadmium group for two [3, 6] weeks. Significant differences were found between the treated groups cd + vt.c, cd + zinc, cd + vt.c +zinc when compared with each other and with control for two periods [3, 6] weeks.

**Table 2:** Differences in Hematological Parameters values (Hb) between the treatment groups are 3 weeks (control, cd, cd+ vt.c, cd + zinc, cd+ vt.c+zinc) and the treatment groups being 6 weeks (control, cd, cd+ vt.c, cd + zinc, cd+ vt.c+zinc)

Treated groups	Hb (mg/dl) Mean ± SD		Univariate Tests	Multivariate Tests
	3 W.	6 W.		
Control	13.00±1.00	12.33±0.58	0.0001*	LSD=0.886 p-value= 0.327
Cd	10.00±1.00	9.67±0.29		
Cd + Vt.C	11.73±0.25	11.13±0.21		
Cd + Zinc	10.67±0.29	11.12±0.20		
Cd + Vt.C. + Zinc	11.72±0.21	11.77±0.12		
LSD	1.204	0.585		
p-value	0.002*	0.0001*		

The results of the study showed a significant decrease (p<0.05) in the size of packed cells in the cadmium group when compared with the control group and groups treated (cd + vt.c, cd + zinc, cd + vt.c + zinc). For two periods (3.6) weeks, while the results showed a significant increase p < 0.05 for the treatment groups (cd + vt.c, cd + zinc, cd + vt.c +zinc) when compared to the cadmium group for two periods [3, 6] weeks. There are significant differences (p>0.05) for the treated groups (cd + vt.c, cd + zinc, cd + vt.c + zinc) when compared with the control for a period of [3, 6] weeks.

**Table 3:** Differences in Hematological Parameters values (PCV) between the treatment groups are 3 weeks (control, cd, cd+ vt.c, cd + zinc, cd+ vt.c+zinc) and the treatment groups being 6 weeks (control, cd, cd+ vt.c, cd + zinc, cd+ vt.c+zinc)

Treated groups	PCV (%) Mean ± SD		Univariate Tests	Multivariate Tests
	3 W.	6 W.		
Control	39.00±3.00	38.67±1.53	0.0001*	LSD=2.109 p-value= 0.431
Cd	34.33±0.58	32.67±0.58		
Cd + Vt.C	37.67±0.58	36.68±0.58		
Cd + Zinc	38.00±1.02	36.10±1.00		
Cd + Vt.C. + Zinc	38.33±0.58	37.34±0.58		
LSD	2.698	1.694		
p-value	0.024*	0.0001*		

Compared the cadmium group to the control group and the groups treated (cd + vt.c, cd + zinc, and cd + vt.c + zinc), the study's results revealed a significantly lower level of PCV (p 0.05). the results for the treatment groups (cd + vt.c, cd + zinc, and cd + vt.c +zinc) when compared to the cadmium group for two periods [3,6] weeks, whereas for two periods (3.6) weeks, the results revealed a significant increase (p 0.05) for the cadmium group. Throughout a duration of [3, 6] weeks, there are significant differences (p>0.05) between both the treated groups (cd + vt.c, cd + zinc, cd + vt.c + zinc) and the control.

**Table 4:** Differences in Hematological Parameters values (WBC) between the treatment groups are 3 weeks (control, cd, cd+ vt.c, cd + zinc, cd+ vt.c+zinc) and the treatment groups being 6 weeks (control, cd, cd+ vt.c, cd + zinc, cd+ vt.c+zinc)

Treated groups	WBC (10 <sup>3</sup> ) Mean ± SD		Univariate Tests	Multivariate Tests
	3 W.	6 W.		
Control	6.33±0.58	6.67±0.58	0.0001*	LSD=0.693 p-value= 0.0001*
Cd	9.53±0.68	11.33±0.15		
Cd + Vt.C	4.57±0.23	5.53±0.30		
Cd + Zinc	4.40±0.26	7.63±0.38		
Cd + Vt.C. + Zinc	5.37±0.21	5.13±0.32		
LSD	0.799	0.679		
p-value	0.0001*	0.0001*		

In comparison to the control group and the treated groups (cd + vt.c, cd + zinc, and cd + vt.c + zinc), the study's results

revealed a substantial increase (p 0.05) in WBC in the cadmium group. When compared to the cadmium group, the treated groups (cd + vt.c, cd + zinc, and cd + vt.c + zinc) showed a significant decrease over two periods of 3.6 weeks (p< 0.05). [3, 6] weeks, there are statistically significant differences (p > 0.05) between both the treated groups (cd + vt.c, cd + zinc, and cd + vt.c + zinc) and the control group.

**Kidney function Parameters Values**

**Table 5:** Differences in Creatinine The treatment groups are 3 weeks (control, cd, cd+ vt.c, cd + zinc, cd+ vt.c+zinc) and the treatment groups being 6 weeks (control, cd, cd+ vt.c, cd + zinc, cd+ vt.c+zinc)

Treated groups	Creatinine (mg/ml) Mean ± SD		Univariate Tests	Multivariate Tests
	3 W.	6 W.		
Control	0.73±0.06	0.77±0.06	0.029*	LSD=0.251 p-value=0.115
Cd	1.07±0.23	0.93±0.06		
Cd + Vt.C	0.87±0.21	0.63±0.15		
Cd + Zinc	0.70±0.00	0.80±0.20		
Cd + Vt.C. + Zinc	0.67±0.06	0.87±0.21		
LSD	0.262	0.274		
p-value	0.037*	0.228		

According to the results of one study, the creatinine concentration in the cadmium group increases significantly over two periods when compared with the control (p<0.05). [3, 6]. There were no significant differences between the groups treated with cd + vt.c, cd + zinc, cd + vt.c + zinc and the control group (p > 0.05).

It was also observed that there was a significant increase (p<0.05) for the cadmium group compared to the treated groups (cd + vt.c, cd + zinc, cd + vt.c + zinc).

**Table 6:** Differences in urea the treatment groups are 3 weeks (control, cd, cd+ vt.c, cd + zinc, cd+ vt.c+zinc) and the treatment groups being 6 weeks (control, cd, cd+ vt.c, cd + zinc, cd+ vt.c+zinc)

Treated groups	Urea (mg/ml) Mean ± SD		Univariate Tests	Multivariate Tests
	3 W.	6 W.		
Control	22.33. ±1.53	24.33 ±2.08	0.0001*	LSD=3.821 p-value=0.001*
Cd	36.33. ±1.15	38.67±0.58		
Cd + Vt.C	28.00±2.00	29.00±2.00		
Cd + Zinc	29.00±1.00	22.67±2.52		
Cd + Vt.C. + Zinc	27.67±2.52	20.33±4.51		
LSD	3.151	4.836		
p-value	0.007*	0.002*		

According to the results of a study, the urea concentration increased in the cadmium group over the control group over two periods (p 0.05). (3, 6).

In comparison to the treated groups (cd+vt.c, cd+ zinc, and cd+vt.c+zinc), a significant increase (p0.05) was reported for the cadmium group. For both times [3, 6]. However, the results revealed that the treated groups (cd+vt.c, cd+zinc, and cd+vt.c+zinc) had substantial differences from one another. When contrasted with the control group

**Discussion**

blood tests for Red blood cells are the basic building blocks of blood, and one of its most crucial functions is the transfer of gases from the lungs to the tissues and vice versa. Blood is a measure to determine the changes that occur in the body, whether they are pathological or physiological. The

results of blood tests for rats treated with cadmium chloride in this study showed the occurrence of anemia, as evidenced by a significant decrease ( $p < 0.05$ ) in the values of the blood parameters represented in the number of red blood cells, hemoglobin concentration, and the volume of packed blood cells compared with the results recorded. Anemia is one of the symptoms associated with cadmium poisoning. Rum says this because the results of blood tests for rats treated with cadmium chloride [18, 19, 20] that the explanation for the occurrence of anemia includes a number of possibilities, including what is indicated that the competition of cadmium with iron during its absorption in the gastrointestinal tract leads to a decrease in its level in the blood. However, the methods of administration, the amount of the dose given, and the duration of exposure differed (Graber, and Krantz 1989). Accordingly, abnormalities in erythropoietin production result in a decrease in the production of blood cells [21, 22] and the cause of this may be attributed to cadmium's interference with the lack of oxygen supply to tissue cells, which inhibits the work of energy houses for their energy production. Additionally, cadmium poisoning increases the production of free radicals, which are known to negatively affect body functions, including the function of red blood cells. This is because free radicals attack cellular components, such as fats, particularly those that contain unsaturated fatty acids found in the membranes of red blood cells, leading to cellular fragility. The authors are Stohse and others [23] Together with the previously listed factors, osteoporosis brought on by cadmium exposure may also be to blame for the drop in hemoglobin levels and packed blood cell volume [24]. The outcomes of the current study revealed that the cadmium chloride treatment had caused a steady, considerable increase in the number of white blood cells. liver and kidneys [25] addition to that in cases of inflammation, the bone marrow will release immature cells, immature cells, due to the ability to form new cells, which leads to an increase in the total number of white blood cells, [26] Cadmium is a highly toxic heavy metal and a major environmental pollutant that is harmful to many animal and human tissues [27, 28]. In human plasma, zinc and vitamin C is a potent water-soluble antioxidant. It removes reactive oxygen and nitrogen species such as super anions, hydroxyl radicals, and peroxy radicals. In the present investigation, during the experimental period, the body weight of mice fed cadmium decreased significantly and continuously over time. It has been observed that the body weight of albino rats decreases significantly by [29, 30]. The decrease in body weight in the Cd-treated group of mice is related to the work of other authors [31, 32]. In the current study, analysis of animal samples treated with vitamin C and zinc showed an increase in body weight compared to the group of mice treated with cadmium. These findings were consistent with a separate study that found that ascorbic acid inhibits free radicals that damage cell membranes through oxidative stress. Animals exposed to cadmium gained more body weight after receiving vitamin C treatment. [33] This result is consistent with results Compared with the cadmium-treated mice, the WBC and RBC of the cadmium-treated mice were significantly lower than those in the control group, according to the results of the current study. The number of erythrocytes and leukocytes were significantly decreased in albino mice given CdCl [34, 29], according to reports. Compared to the cadmium-treated mice, the vitamin C-treated group showed increased blood values. According to

other research, dietary intake of vitamin C [32] significantly raised blood parameters. On the other hand, vitamin C pretreatment showed a protective effect in cadmium poisoning on the hematological value [35]. Serum concentrations of creatinine and urea The measurement of creatinine in the blood is the optimal measure for testing kidney function, and it is anhydrous creatine produced from creatine phosphate, Phosphocreatine, after the loss of the phosphate group, then it passes through the blood to the kidneys to be excreted with the urine [36] The urea test is also one of the tests The task is combined with creatinine [37], thus the urea level test is used as an indicator to guarantee the effectiveness of the functional performance of both the liver and kidneys [38]. The cadmium group and over the duration of the experiment compared with its concentration in the control group, as these results were comparable with the work of [39], where a significant increase in the blood creatinine level was reported in male rats exposed to cadmium chloride. It was also similar to the study of [40], as he noticed a significant increase in the level of creatinine in rats treated with cadmium, and this increase may be due to the effect of cadmium chloride on the level of kidney filtration as well as on the processes of excretion and reabsorption in the urinary tubules. On the other hand, the study that he completed suggested. Vitamin C and zinc are strong antioxidants against oxidative stress. Vitamin C and zinc with cadmium significantly improve urea and creatinine concentrations if they return to levels close to those of the control group.

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