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RESEARCH ARTICLE

The Effect of Calcium Oxide Nanoparticles on Liver Enzymes in White Rats

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Abstract

Calcium is the five elements that found on the crust of the earth. It`s the essential mineral for all organisms because the important role in the functions of the life cells , and its essential mineral for binding the bone and teeth by producing calcium phosphate. The decreasing of calcium element in the body leads to weakness, dental caries, and muscle cramps. Present study was designed to evaluate the effect of calcium oxide- nanoparticles on the serum levels for liver enzymes of white rats. Two group of White rats (3 rats each group) received calcium oxide- nanoparticles which were orally administered with 50 and 100 (mg/kg) from of body weight of calcium oxide- nanoparticles suspension daily for 10 day. Serum levels of alanine aminotransferase (ALT), aspartate aminotransferase (AST), and alkaline phosphatase (ALP) were measured at start and at the end of the study. Changes of the specific parameters indicated that liver were significantly affected in both experimental groups , the changes between the levels of total alanine aminotransferase , aspartate aminotransferase and alkaline phosphatase indicate that calcium oxide- nanoparticles induces liver damage and may be lead to hepatic toxicity in the experimental rats.

Key words: *CaO-Nanoparticles, (ALT, AST and ALP), White rats.*

Introduction

Nanotechnology has revolutionized the application of commercial for products in the fields of (medicine, engineering, manufacturing, information and environmental) technology [1]. Calcium oxide is used in many applications such as cement industry, biodiesel production, biosensors, tissue engineering, petroleum industry, electric lighting and power production [2].Various physical methods were used to prepare Calcium oxide -nanoparticles [3]. Basically Calcium oxide -nanoparticles available in two ways: The commercial calcium oxide and the conventionally prepared calcium oxide [2].

Calcium oxide -nanoparticles are non-hazardous materials [4].Calcium oxide-nanoparticles showed antimicrobial effect towards test organisms *Staphylococcus epidermidis, Pseudomonas aeruginosa* and *Candida tropicalis* [5]. Calcium oxide-nanoparticles cannot be used directly with antibodies; they need to be modified with appropriate hydrophilic coating [6]. Aim of this study was to measure the oral toxicity of nanoparticle calcium oxide in white rats and the effects of these particles on biochemical parameters in the blood.

Materials and Methods

Preparation of CaO-NPs

One and a half gram of$CaCl_2.2H_2O$ (BDH Chemicals Ltd Pool England) was dissolved in (50 mL) of redistilled water. Solution was added into a round flask with stirring. About (15 ml) of NaOH (1M) was rapidly added to the mixture, and a nano-powder suspension was formed.

Suspension was kept at (75°C) for 1h. After cooling to room temperature, the particles were separated by centrifugation, and were washed with distilled water (D.W.) to remove any contaminations. The particles were then, dried in an oven at (80 °C). The CaO- NPs with a desired amount (7.5mg) was placed in

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30 ml of redistilled water and was sonicated for 5h to separate CaO -NPs and overcome the agglomeration and make the colloidal contain individual nanoparticles. After that, drop casting method used to deposited CaO layer on glass substrate to investigated the characteristics of nanoparticles by meaning of X-ray diffractometer (XRD-6000, Shimadzu, X-ray, diffractometer) with- Cukα radiation at λ = 0.154056 nm. The optical absorption of colloidal CaO-NPs was measured using a spectrophotometer (Cary, 100 Conc plus, UV-Vis-NIR, Split- beam Optics, Dual detectors). In order to observe the topography and surface roughness of deposited layer, atomic force microscopy (AFM) micrographs were taken with digital instruments, Inc. nanoscope III and dimension 3100.

Experimental Design and Procedure

Fifteen male adult of Wister rats aged 1-2 months, weighting (150- 300) g were purchased from Scientific faculty in kufa university, Iraq. Animals were housed (five rats) per plastic cage, and permitted to acclimatize under standard conditions 13 h light /11dark cycles for ten days. Then animals were divided into three sets of five animals each. Sets one to two received a dose of (50, 100 mg/kg) calcium oxide nanoparticles (CaO-NPs) body weight. The rats were orally administrated daily for (ten) days. Control group was orally administrated with distilled water (D.W.). At the finale of the study all of the rats were scarified, and the samples of the blood were collected from both (healthy control and Calcium oxide nanoparticles treated sets) via cardiac puncture, using (23 G 1¼″) needles. The samples of the blood were left to clot at (37C°) and then centrifuged at (1000 g for 10 min), and the serum were split and investigated for (ALT), (AST), and (ALP).

The procedure of experimental was approved at the Faculty of Dentistry at Kufa University and the research use the laboratory of advanced animal researches in Science Faculty at Kufa University.

The Analyzing of the Enzymes

The serum of animal was analyzed for (ALT), (AST), and (ALP) using standard diagnostic Fuji film Kit (Japan).

Statistical Analysis

Data were stated as (mean ± standard deviation). The statistical analyses of the experimental values were compared to their control. The analysis of ANOVA variance in (SPSS) software (Version 19.0) was used to show the difference of significant between (experimental and control sets). The difference significant was studied to be $p < 0.05$ or fewer.

Results

Structural Properties of CaO-NPs

The different Three of crystal phases CaO, $CaCO_3$ and Ca $(OH)_2$ are shown in Fig.1. The presence of these three crystal phases in the inactivated (CaO) starting material is related to it's the chemical composition; present the impurities and the potential surface interactions with humidity in ambient conditions. The phase of CaO is the principal one, while $CaCO_3$ and especially the phases of Ca $(OH)_2$ are only detectable having a volume fraction much the lower when compared to the phase of CaO. X-ray diffraction studies shows that the microwave assisted synthesized materials were pure monophasic cubic CaO-NPs and crystal structures agree well with corresponding reported (JCPDS) data (JCPDS) powder diffraction data card no. 77-23.

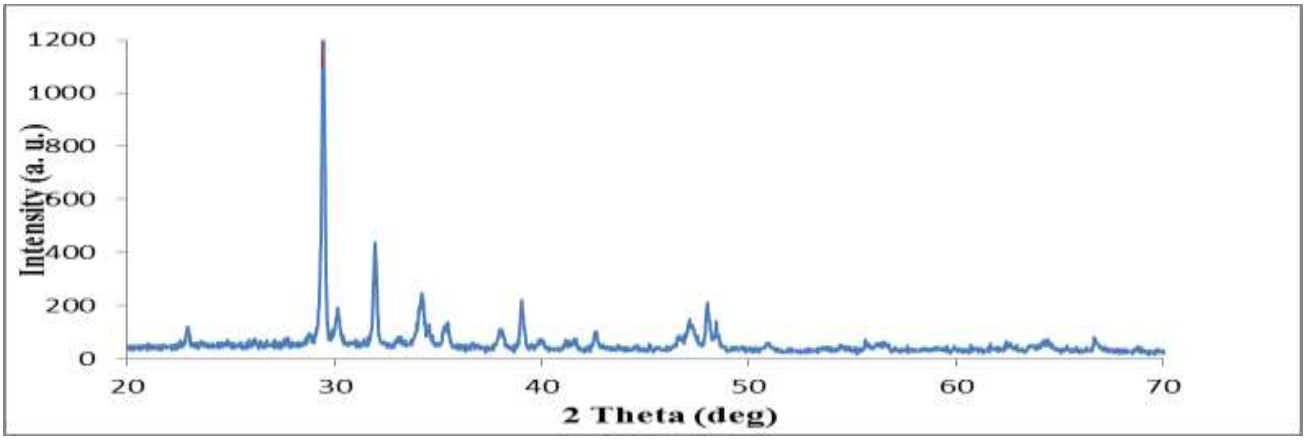

Fig.1: XRD pattern of synthesized CaO-NPs

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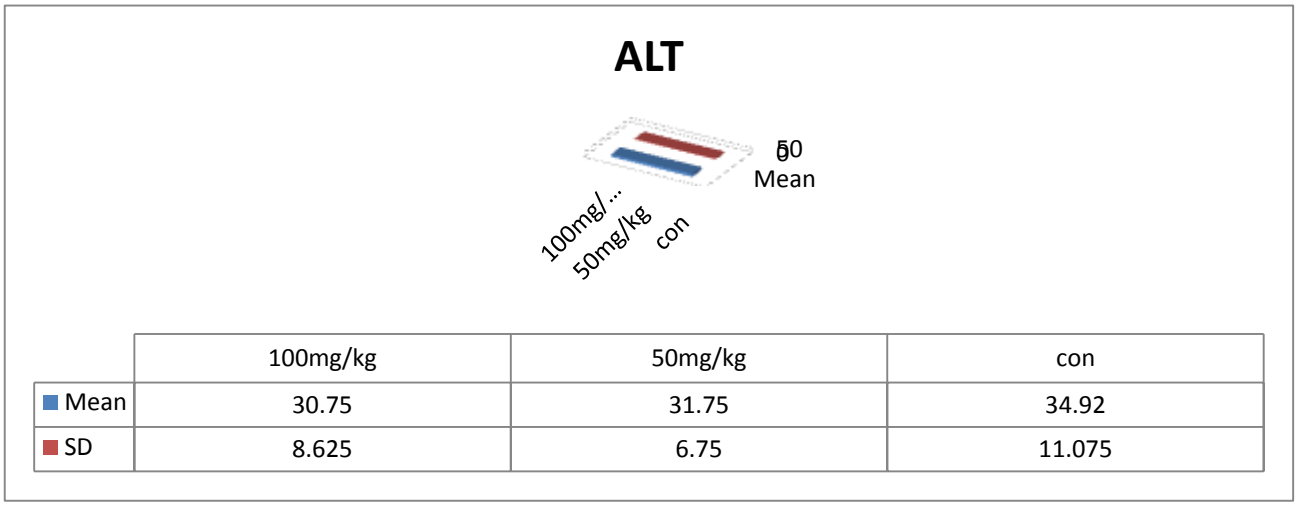
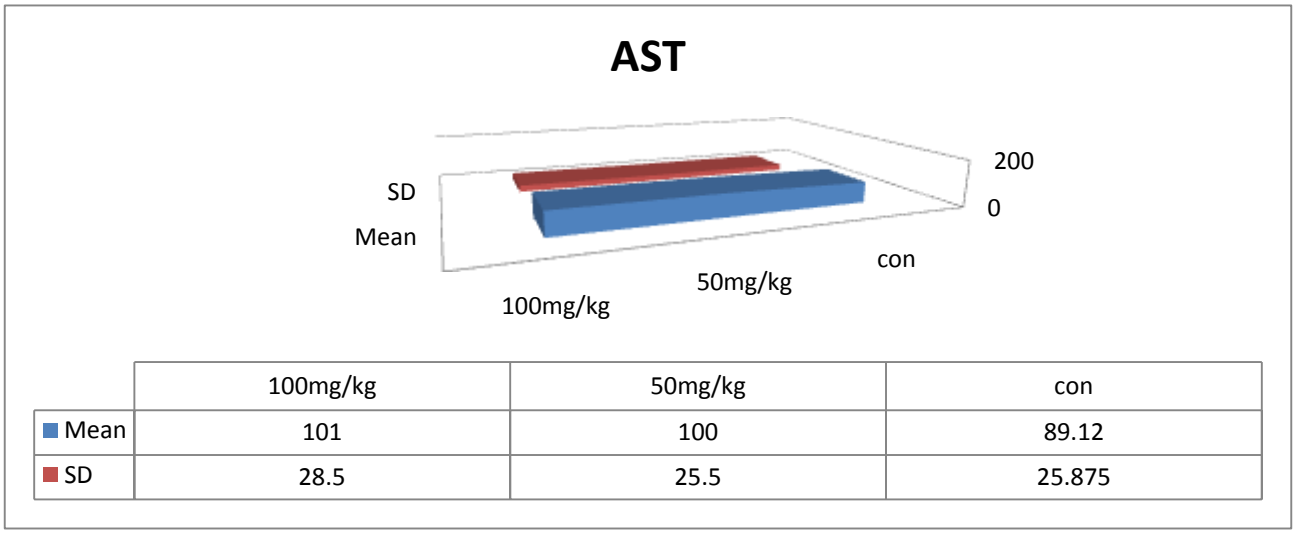
Effect of CaO-NPs on Liver Enzymes

The increasing effect and decreasing concentration of calcium oxide nanoparticles on the enzymes of liver, are presented in Figure two (2), three (3), and four (4). As shown, the enzymes (AST), and (ALP) levels varied in the serum of both calcium oxide nanoparticles treated groups. A remarkable decrease of ALP activity was observed from both groups. AST levels were significantly ($P< 0.05$) increased in 50 and 100 (mg/ kg) of calcium oxide nanoparticles treated group. There were no changes of (ALT) levels in both the experimental groups.



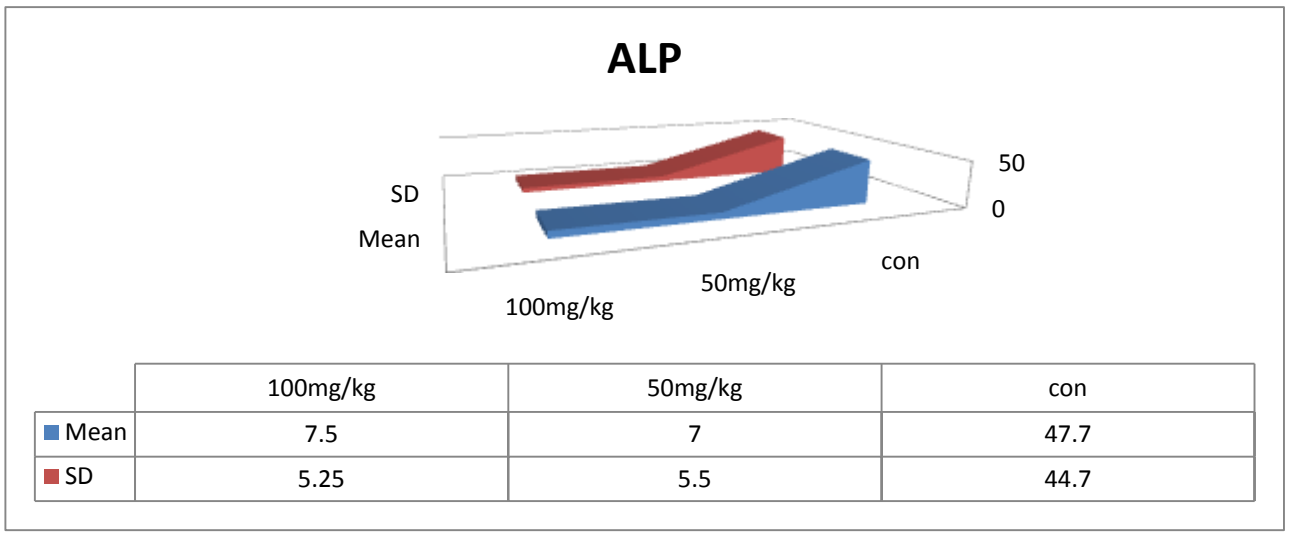
	100mg/kg	50mg/kg	con
Mean	30.75	31.75	34.92
SD	8.625	6.75	11.075

Fig.2: Alanine aminotransferase (ALT) levels in White rats for these groups



	100mg/kg	50mg/kg	con
Mean	101	100	89.12
SD	28.5	25.5	25.875

Fig.3: Aspartate aminotransferase (ALT) levels in White rats for these groups



	100mg/kg	50mg/kg	con
Mean	7.5	7	47.7
SD	5.25	5.5	44.7

Fig.4: Alkaline phosphatase (ALP) levels in White rats for these groups

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Discussions

The study has shown that Calcium oxide nanoparticles are widely used due to their antimicrobial effects. Humans can be exposed to these nanomaterials via a number of routes with the nanoparticles tending to accumulate in vital organs [5]. According to Gatti [7], nanoparticle deposition in vital organs or tissues could induce cellular damage. Currently, the application of industrial calcium oxide nanoparticles is increasing, calcium oxide is widely used in biodiesel production [8], and calcium oxide nanoparticles can be used as potential drug delivery agent for biomedical applications, antimicrobial activity [4,6].

The liver is the major place for the changes of biological and it protect the body from the foreign substances and the xenobiotic chemicals, identifying the causes for hepatic toxicity. The liver excretes the substances into the bile; consequently biliary organism is also exposed to (NPs). The previous study have shown that the diverse toxins with the different mechanisms, including: Activation of (alcohol degeneration, the peroxidation of lipid membrane), inhibition the synthesis of protein, the disruption of calcium homeostasis, and activation the receptor of enzymes that cause the damage to the cells of liver [9].

The enzymes such as (ALT and AST), are metabolic enzymes in the liver, which are dysfunctional enzymes in the serum and the plasma. Level of these enzymes in cytoplasm cells of the liver is number of times more than the extracellular fluid. When the cells of hepatic and the membrane are scratched or died [10], the quantity of these enzymes elevation in the blood stream and this quantity of altitude is an indication of the damage to the liver [11]. In the present study, rats were exposed to different doses of calcium oxide nanoparticles; resulted in significant difference between the levels of AST and ALT enzymes within the groups. Although the levels of serum AST were increased in the CaO-NPs treated rats when compared to that of controls, this change was not significant in among the experimental groups. These results may be lead to CaO-NPs induce acute hepatic damage, value of the serum (ALT) as an indicator of the necrosis in hepatocellular). ALP is nothing but a cholestatic enzyme liver. The cholestasis is a condition that causes incomplete or obstruction of the bile ducts.

The bile duct gives bile from liver into gall bladder, and then into intestines. The bile is the fluid discharged from the cells of the liver and helps the body to cleave the fat, process the cholesterol and catch free of the toxins. If bile duct is damaged, ALP can get reversed up and leak out from liver into the stream of the blood [12-14]. In this study, reduced ALP levels were observed in the serum of calcium oxide nanoparticles treated rats. Therefore, in this study the significantly decreased in the ALP level enzymes and its reason, is not clear but may be associated to the growth of rats.

Conclusion

Toxicity of calcium oxide nano particles on the adult of male rats treated by orally of the diverse dilutions of calcium oxide nanoparticles was assessed by measuring some enzymes of the liver, the results showed significantly increase and decrease in the enzymes of liver starting from CaO nanoparticles dilutions of 50 and 100 (mg/ kg) animal from the body weight. Conversely, additional investigation is needed to impact the study of CaO nanoparticles on the health and the safety of human.

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