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ORIGINAL ARTICLE

Physiological study, the protective role of curcumin extract against Ameliorating Cyclophosphamide-induced oxidative stress in Male Albino Rat model

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ABSTRACT
The yellow color of the curry spice turmeric is due to a polyphenol called curcumin recent scientific studies have established its anti-inflammatory, antioxidant, and anti-carcinogenic properties cyclophosphamide-induced oxidative stress in rats' serum. Lipid peroxidation, reduced glutathione (GSH), Malondialdehyde (MDA) and catalase the protective role of curcumin extract which improvement the levels of antioxidant enzyme in rats' serum.
Key words: - curcumin, Cyclophosphamide, oxidative stress, Rat

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INTRODUCTION

Cyclophosphamide is an alkylating agent of the nitrogen mustard class, which is an oxazaphosphorine class with immunosuppressive and potent cytotoxic effects; it was first synthesized by Arnold and colleagues in 1958 [1-5]. Cyclophosphamide is a biologically inactive prodrug that needs cytochrome-P450 mediated activation [6-9]. Cyclophosphamide-induced oxidative stress and DNA damage. Reactive oxygen species (ROS) generation and repair capacity are in equilibrated, which causes oxidative stress. Under physiological circumstances, endogenous antioxidants or antioxidant enzymes maintain cells in a reducing environment. A deterioration of mitochondrial function and cell death are the end results of elevated levels of oxidative stress, which also cause alterations to lipids, proteins, and nucleic acids [10-11]. Curcumin can help with metabolic and oxidative disorders such as type 2 diabetes, Crohn's disease and thrombosis [2]. Curcumin's chemical makeup, which includes hydroxyl and methoxy groups, is thought to be responsible for a number of qualities, including antioxidant, antibacterial, anti-inflammatory, anti-angiogenic, and antimutagenic ones [12].

MATERIALS AND METHODS

Ethics Statement

Rats were handled and euthanized according to Ethics and Guidelines for the Care and Use of Laboratory Animals; procedures were approved by the University of kufa.

Animals: -

There were 30 Adalat albino strain rats(4 months old) used in this experiment, weighing between 200 and 250 g. the animals were kept in the Biology Department's and Faculty of Science's Animal House at the University of Kufa, where the environment included a moderate temperature and a 12-hour cycle of darkness and light.

Calculating the doses of cyclophosphamide: -

It was determined that 50 mg/kg of body weight single dose was the optimum dose of cyclophosphamide to develop its therapeutic efficacy and adverse effects according to [10].

The distribution of the experimental rats: -

There were30 healthy male albino rats used. Six groups were formed from the animals (10 rats for each group). 50 animals were separated into 5 groups, with 10 animals serving as the control group.

1. G1 (control group) contained ten male rats in good health. Throughout the duration of the experiment, the animals received 0.5 ml of normal saline solution (0.9%) orally each day.\
2. G2 (cyclophosphamide group): included 10 rats for the duration of the experiment, the animals received intraperitoneal injections of cyclophosphamide at a dose of 50 mg/kg (single dose) \
3. G3 (the protection group) included 10 male rats. The five animals in this group were dosed with a solution of curcumin extract (1 ml/animal) at a concentration of 200 mg/kg body weight per day for seven days and then injected with cyclophosphamide at a dose of 50 mg/kg (a single dose).

The other five animals are from the same group and were dosed with a solution of curcumin extract per day for fourteen days and then injected with cyclophosphamide.

Animals were dosed with a solution of curcumin extract per day for fourteen, the rats were anesthetized with ketamine and xylazine and then sacrificed at the end of the experiment. After inserting a 5 ml needle into the heart to puncture it, 3–4 ml of blood was drawn for the blood sample. After being placed in gel-lined plastic tubes for 5 minutes, the blood underwent a 15-minute centrifugation procedure at 3000 rpm. For use in the investigation of various blood parameters, a serum in the quantity of 0.4 to 0.5 ml was obtained, put in a 1.5 ml Eppendorf tube, and stored in the freezer. The oral dosage for curcumin extract was 200 mg/kg per day for 14 days depended on [11].

Measuring of serum biomarker

CAT (Catalase)

The activity of CAT was assessed in the serum of experimental animals. Uses spectrophotometer and ELK Biotechnology kit

GSH (Glutathione S transferase) uses spectrophotometer and ELK Biotechnology kit.

The glutathione level was determined using spectrophotometer and ELK Biotechnology kit.

MDA (Malondialdehyde)

Malondialdehyde was measured using spectrophotometer and ELK Biotechnology kit.

RESULTS

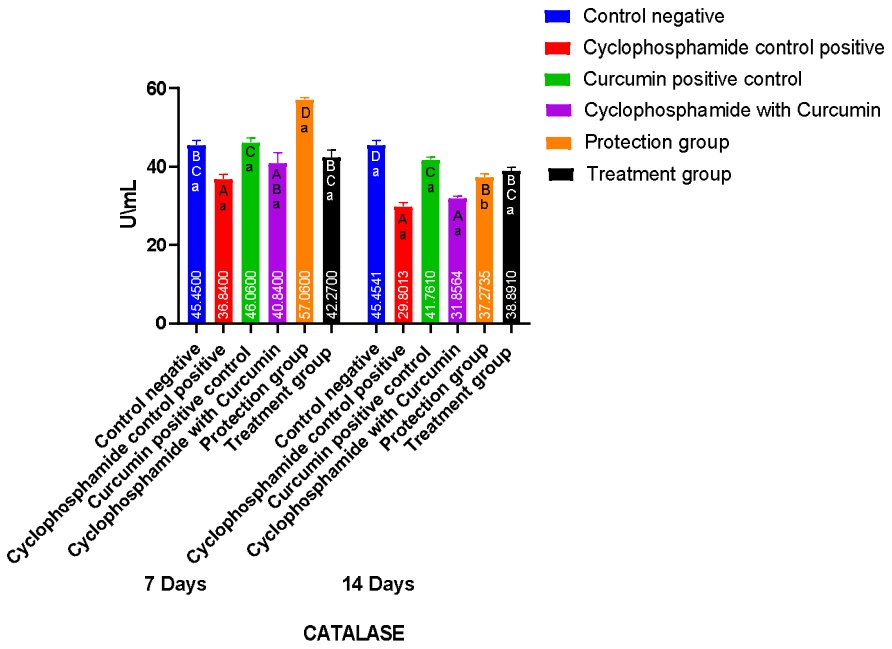
The groups treated with cyclophosphamide and curcumin chemicals and sample collection on 7th and 14th days after the experimental time were different from the control group considerably (P 0.05), according to the antioxidant biomarker data. The groups are statistically diverse, as indicated by various capital letters.The different small letters indicate the statistical difference (P< 0.05) in the same group at different times (comparison between the 7th day and the 14th day.

CAT (Catalase)

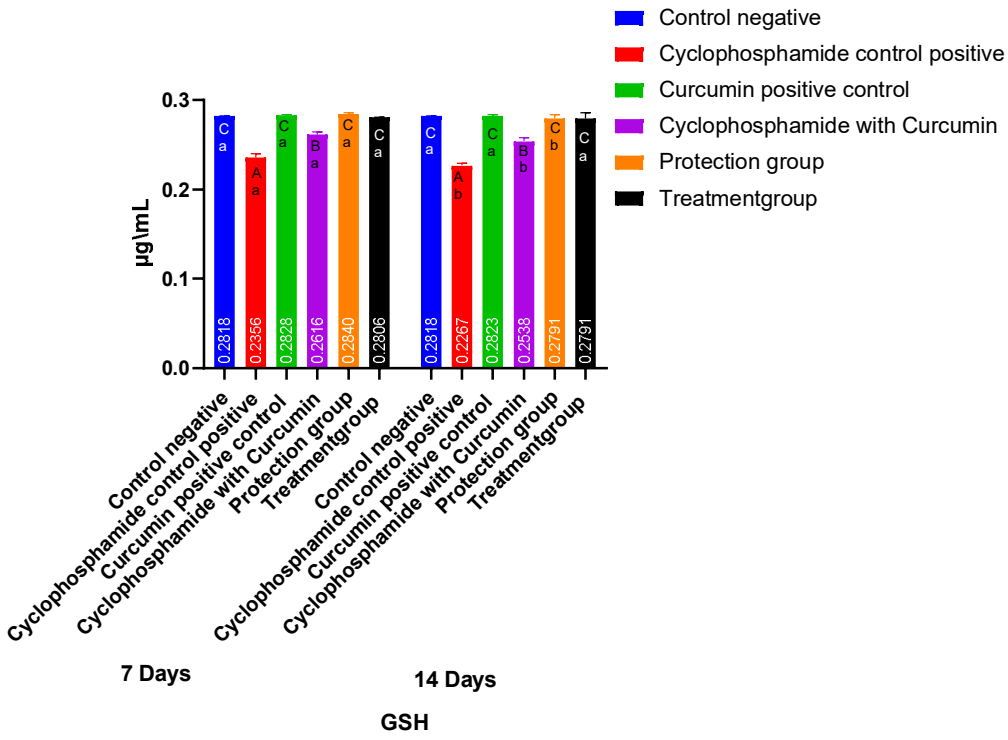
Antioxidant biomarkers Comparing the Cyclophosphamide-treated group (Cyclophosphamide control positive group) of rats to the control group, there was a significant (P< 0.05) decline in the catalase enzyme activity. Inversely, Cyclophosphamide group protected by curcumin extract (protection group) displayed a significant (P <0.05) increase when compared to the cyclophosphamide and control groups, The protection group showed a significant (P< 0.05) increase on the 7th day when compared with the same group on the 14th day.

GSH (Glutathione S transferase)

The cyclophosphamide-treated group shows there was a significant (P< 0.05) decline in the glutathione-S transferase enzyme activity when compared to the control group. The protection groups showed a significant (P<0.05) increase when compared with the cyclophosphamide-treated group. The cyclophosphamide-treated group displayed a substantial (P<0.05) low in glutathione S transferase enzyme levels on the 7th day when compared with the same group on the 14th day. On the other hand, rest groups showed no significant changes (P > 0.05) when compared between different times.



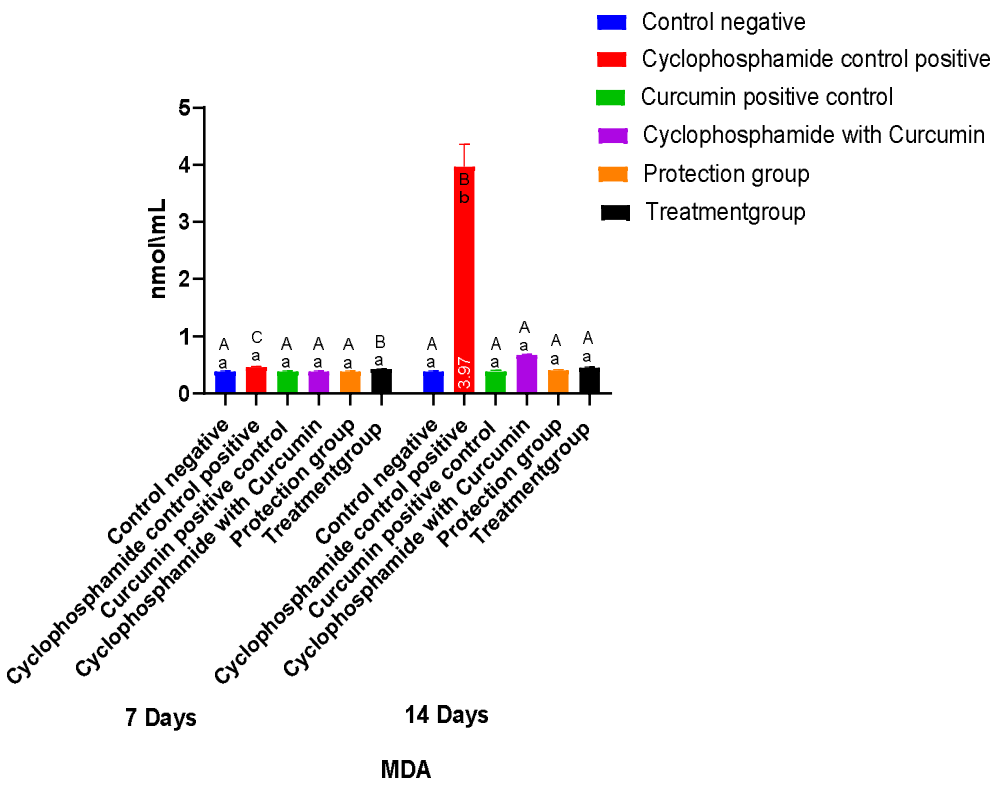
(Figure_1) Level of catalase (UI\ml) in the serum of rat animal groups



(Figure -2) Level of GSH (µg\ml) in the serum of rat animal groups.

MDA (Malondialdehyde)

The cyclophosphamide-treated group shows there was a significant (P< 0.05) increase in the malondialdehyde when compared with the control group, the protection group shows a significant (P< 0.05) decline. When compared with the cyclophosphamide-treated group. In contrast, the results showed that there was a significant (P < 0.05) increase in the level of malondialdehyde in the cyclophosphamide-treated group on the 14th day when compared with the same enzyme levels on the 7th day.



Figurer_3: Level of MDA (nmol\mL) in the serum of rat animal groups.

DISCUSSION

Antioxidant enzyme tests showed that cyclophosphamide treatment in animals resulted in a diversity of changes in the activity of these enzymes, which include a significant reduction in the levels of GSH and CAT, which indicate a decline in the efficiency of the endogenous antioxidant factors, and a raise in the amount of MDA, which confirms lipid peroxidation. According to studies agree with our study. Curcumin's effects on oxidative stress markers are linked to its abilities to reduce reactive oxygen and nitrogen, interact with metals, and control a wide range of enzymes. Thus, the antioxidant properties of curcumin have been proven [12].

CONCLUSIONS

This study found that curcumin extract is an effective anti-inflammatory, anti-oxidative, Thus, cyclophosphamide's adverse effects as an oxidative agent were lessened by curcumin extract.

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