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CYCLOPHOSPHAMIDE INDUCED CHANGES IN CERTAIN ENZYMOLOGICAL (GOT GPT ACP AND PARAMETERS OF ADULT MALE RATTUS NORVEGICUS.

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ABSTRACT

Cyclophosphamide (CP), [2-(bis-/2-chloro-ethyl-amino)-tetrahydro-2H-1, 2, 3 oxazaphosphorine-2-oxide], is an alky chemotherapeutic agent, which is commonly used against malignancies, such as leukemia, lymphoma, breast, lung, prostrat ovarian cancer. The aim of this study is to evaluate the side effect of CP on male albino rat in response to certain hepatic and enzymological parameters. In this investigation, total 20 albino rats were divided in to two groups of ten each. Group first ser control received i.p. injection of 0.9 physiological saline fed with standard food and water ad libitum. While, Group second rece single dose (0.2ml/100g b/w) through i.p. injection of cyclophosphamide once in a week for a period of one and five weel enzymological parameters i.e. glutamate oxyaloacetate transaminase (GOT), glutamate pyruvate transaminase (GPT), acid phospl (ACP) and. alkaline phosphatase (ALP) levels in liver and kidney were quantified after 7 and 35 days of the treatment enzymological parameters i.e. GOT, GPT, ACP and. ALP levels in liver and kidneys were significantly increased after 7 and 3! treatment of cyclophosphamide. The increased of these values was more prominent in later part of the experiment. Elevat enzymological parameters after cyclophosphamide exposure may impaires the functional activities of liver and kidney of male norvegicus.

KEYWORDS: Cyclophosphamide, Enzyme activities, GOT, GPT, ACP and ALP Rattus norvegicus.

1. INTRODUCTION

Chemotherapy of cancer has opened new possibilities and chances for improving the quality and life span. Despite this successful trend, treatment with some of the most effective anticancer drugs caused many of toxic symptoms to normal cells. Cyclophosphamide (CP) is an alkylating chemotherapeutic agent that is commonly used as an anticancer and an immunosuppressant drug. Although, CP is a known mutagenic and a pro-oxidant [1, 2, 3, 4] often used for the treatment of preparation of bone marrow transplantation, B-cell chronic lymphomas and breast cancers alone or in combination with other cytotoxic drugs. [5, 6, 7] Apart from this, CP is used clinically to treat a wide range of cancers including malignant lymphomas, myeloma, leukemia, mycosis fungoides, neuroblastoma, adenocarcinoma, retinoblastoma and breast carcinoma. [8, 9] Other clinical uses for CP include immunosuppressive therapy follows organ transplants or as a treatment for autoimmune disorders such as rheumatoid arthritis, wegener's granulomatosis and nephritic syndrome in children. [10] An ample literature implicate that elevated therapeutic dose of CP, caused liver disorders by the development of sinusoidal obstruction syndrome (veno-occlusive disease) and total serum bilirubin levels. [11, 12, 13] It has been also reported that the chemotherapeutic drugs killed dividing cells rapidly in the body, including cancer cells and normal cells. [14] Considering the above, the current study was undertaken to examine the effects of cyclophospamide on certain enzymological parameters i.e. glutamate oxyaloacetate transaminase (GOT), glutamate pyruvate transaminase (GPT), acid phosphatase (ACP) and alkaline phosphatase (ALP) levels in liver and kidney of male Rattus norvegicus.

2. MATERIAL AND METHODS

In this investigation, 20 disease free albino rats weighing 120 ± 5 gm were acclimatized and maintained at 23 ± 2^0C temperature with a 12 hours light-dark cycle in the Animal House, Laboratory of Endocrinology, Bioscience Department, Barkatullah University, Bhopal. The animals were fed with standard rat feed and water ad libitum.

2.1) Dose, preparation of drug, route and duration of administration:

Cyclophosphamide (CP) brand name-LEDOXAN known mutagenic and a pro-oxidant agent was purchased from local pharmacy market, Bhopal. CP dose (0.4mg/100g b.wt) was prepared 200mg CP was dissolved in 10 ml distilled water. Received a single dose i.e. 0.4mg/100g b wt of CP once in a week through intraperitoneal (i.p.) injection for a period of 7 and 35 days (one and five weeks).

2.3) Experimental Design:

Total 20 albino rat were divided into two groups of ten each. **Group first** received normal physiological 0.9 NaCl saline solutions through intraperitoneal (i.p.) with standard food and water ad libitum served as control group. While, group second received a single dose i.e 0.2 ml/100g b.wt. of CP once in a week through i.p. injection for a period of one and five weeks. The five animals from each group were sacrificed on day 8^{th} and 36^{th} by cervical dislocation and liver and kidney were quickly removed, cleaned, blotted, weighed, homogenated in 0.25M sucrose solution and processed for GOT, GPT, ACP and ALP estimations. GOT and GPT were quantified by using Reitman and Frankel (1957) [15] methodology and ACP and ALP were estimated by Bergmeyer and Bernt (1963) [16] methodology. The results were statistically analyzed by using Student 't'test.

3. RESULTS AND DISCUSSION

3.1 Results

The animals treated with CP (0.2 ml/100g b.wt/animal/week) for 7 and 35 days altered hepatic and renal GOT, GPT, ACP and ALP levels in Rattus norvegicus (Figure-1, 2, 3 & 4).The GOT and GPT levels in liver and kidney were significantly increased after 7 and 35 days of the CP treatments in compression to control groups (Figure-1, 2). These changes were more pronounced in later part of the experiment. In connection to this, the hepatic ACP and ALP were also elevated significantly in both the durations (Figure-3, 4).But the renal ACP and ALP were insignificantly elevated after 7 day of CP treatment (Figure-3, 4). While, it significantly elevated in later part of the experiment.

3.2 Discussion

Chemotherapy is the non-specific cytotoxic action of both tumour cells and normal healthy cells. [17] Cyclophosphamide is a widely prescribed non-cell-cycle-specific antineoplastic drug which is known to cause toxic effects including hepatotoxicity. [18] There is pharmacologic evidence that the breakdown of CP into biologically active alkylating compounds takes place principally in the liver. [19] Therefore, the changes observed in epithelial cells of the proximal segment of uriniferous tubules were presumably the result of the cytotoxic effect of metabolites of CP. This cytotoxicity resulted in necrosis of tubular epithelial cells within the first few hours after the alkylating agent was administered, in a manner similar to that observed in the urinary bladder. [20] Abraham et. al., 2007 [21] reported that CP may induce nephrotoxicity secondary to decrease in the activities of lysosomal protein digestive enzymes with consequent accumulation of abnormal amounts of protein in the kidney. Earlier and recent studies have demonstrated that increased generation of both reactive oxygen and nitrogen species by CP in kidney tissues plays a critical role in the pathogenesis of CP-induced kidney damage. [22, 23, 24] Evidences suggest that oxidative stress play a predominant aetiological role in CP induced. [25, 26, 27] Several studies indicate that CP has a pro-oxidant character, and generation of oxidative stress after CP administration leads to decrease in the activities of antioxidant enzymes and increase in lipid per-oxidation in liver, lung and serum of mice and rats. [28, 29, 30, 31, 32, 33]

Cyclophosphamide altered liver and kidney functions by modulating all liver enzymes. [34, 21] Enzymes are biological molecules that catalyze (i.e., increase the rates of) chemical reactions. [35, 36] Enzymes are specific proteins that increase the rate of chemical reactions in human body. Enzymes occur naturally in the human body and without them, we couldn't survive. These enzymes continually regenerate while maintaining the body's systems and protecting us from disease. They keep the body functioning properly and ward off deadly diseases. The liver and kidney is the richest source of both GOT and GPT enzymes. And these are marker enzymes for assessment of liver and kidney functions. Any damage to the liver and kidney cells will result in the increase in both of these enzymes. [37] Increased tissue ACP and ALP are symptoms of chemical induced tissue injury along with hepatocellular necrosis. [38] The elevation of ACP and ALP that our present findings are consistent with. [39] Acid phosphatase and alkaline phosphatase are now frequently detected to estimate the degree of liver dysfunction due to CP of advanced liver cirrhosis as well as expectation of heart failure development. In the present experimental study, it has been observed after7 and 35 days of CP that GOT, GPT, ACP and ALP were significantly elevated in Rattus norvegicus (0.2 ml/100g b.wt/animal/week) exposure in comparison to control group. The increased levels of these enzymes suggest that CP may induce hepatic and renal toxicity by interfering metabolic activities and protein synthesis. Hepatic activation of CP leading to the formation of toxic metabolites caused damage to the liver tissues as shown by increased GOT and GPT levels.

The changes occurred in liver and kidney by cyclophosphamide in Rattus norvegicus may also suggested that the cyclophosphamide might be modulating the hepatic and renal functions by acting directly or indirectly on these organs and these effects are dose and duration dependents.

4. FIGURE 1, 2, 3 AND 4:

Showing GOT, GPT, ACP and ALP levels in liver and kidney of Rattus norvegicus after 7 and 35 days of the cyclophosphamide exposures.

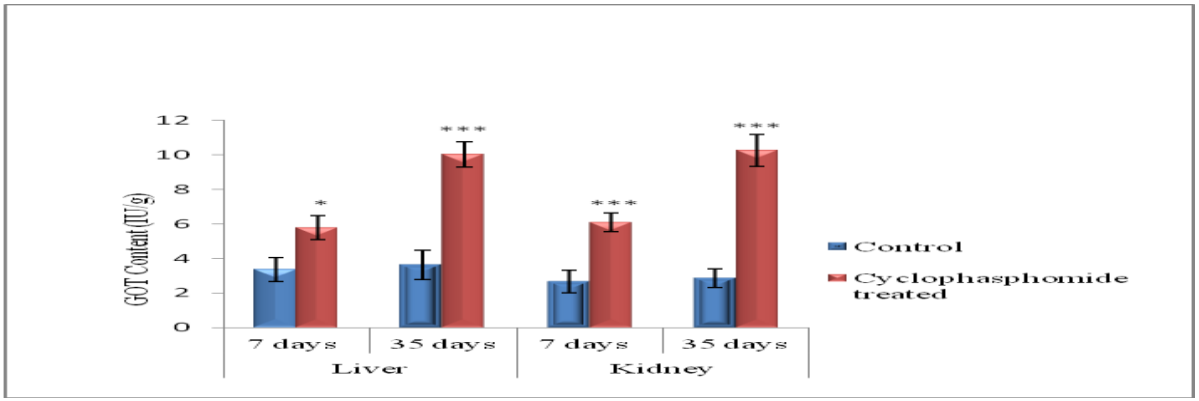

FIGURE 1. Glutamate Oxyaloacetate Transaminase (GOT) (IU/g tissue weight) content in the liver and kidney of control and experimental albino rats treated with CP (0.2 ml/100g b.wt/animal/week) for 7 and 35 days. Data Mean value ± SEM (n=5), significant value *p<0.05, ***p<0.001 in between control vs experimental by Student "t" test.

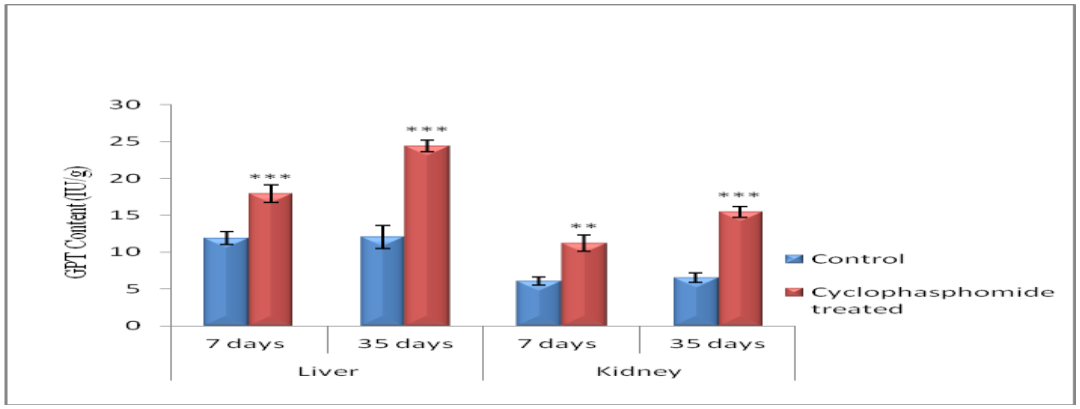

FIGURE 2. Glutamate Pyruvate Transaminase (GPT) (IU/g tissue weight) content in the liver and kidney of control and experimental albino rats treated with CP (0.2 ml/100g b.wt/animal/week) for 7 and 35 days. Data Mean value ± SEM (n=5), significant value **p<0.01, ***p<0.001 in between control vs experimental by Student "t" test.

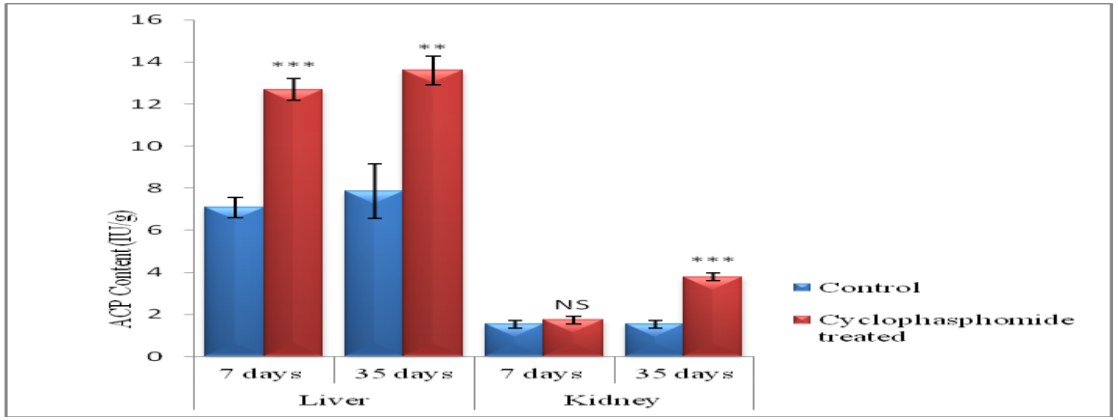

FIGURE 3. Acid Phosphatase (ACP) (IU/g tissue weight) content in the liver and kidney of control and experimental albino rats treated with CP (0.2 ml/100g b.wt/animal/week) for 7 and 35 days. Data Mean value ± SEM (n=5), significant value NS not significant, **p<0.01,***p<0.001 in between control vs experimental by Student "t" test.

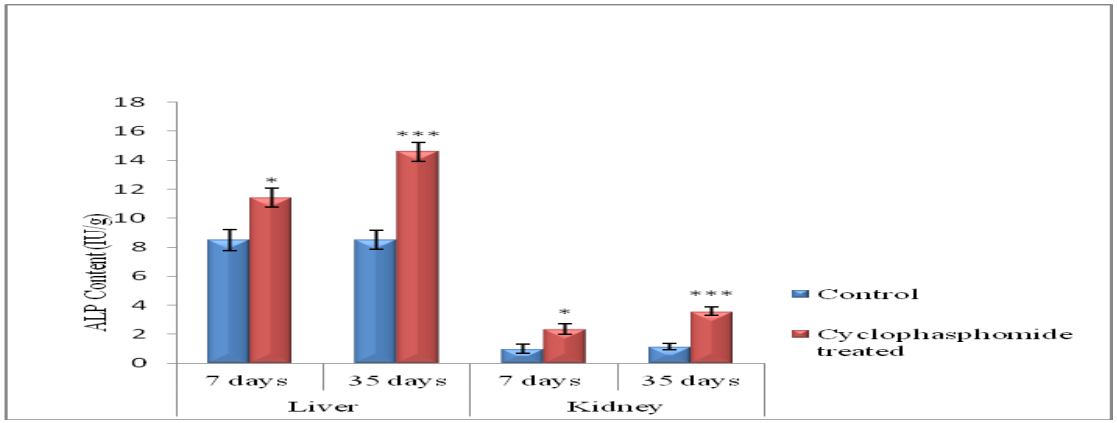

FIGURE 4. Alkaline Phosphatase (ALP) (IU/g tissue weight) content in the liver and kidney of control and experimental albino rats treated with CP (0.2 ml/100g b.wt/animal/week) for 7 and 35 days. Data Mean value ± SEM (n=5), significant value *p<0.05, ***p<0.001 in between control vs experimental by Student "t" test.

5. CONCLUSIONS

Our results may suggest that CP displayed a substantial increase in the activities of diagnostic marker enzymes in liver and kidney tissue which obviously reflect a significant damage in the structural integrity of liver and kidney. So in respect to this we may also suggest that the use of CP drug should be introduce to the patient in the proper pharmacological dose to avoid its toxic effects.

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