Protective Effect of Captopril on Cisplatin Induced Hepatotoxicity in Rat

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Abstract: Cisplatin is one of the most potent anticancer drugs used in chemotherapy. Hepatotoxicity can also occur when cisplatin is administered at high doses. Captopril increases the activity of liver superoxide dismutase and glutathione peroxidase, which are of the main anti-oxidant enzymes found in aerobic organisms. The aim of the present study was to evaluate the possible protective effects of captopril on cisplatin induced hepatotoxicity in rats. 48 male Wistar rats were used in this study. Rats were randomized into 6 treatment groups with 8 animals in each group, as follows: (1) saline solution (NaCl 0.9%); (2) Captopril (100 mg/kg/d); (3) Cisplatin (CPN); (4) CPN + Captopril (50 mg/kg/d); (5) CPN + Captopril (100 mg/kg/d); (6) CPN + Captopril (150 mg/kg/d). Blood sera were isolated for the evaluation of aspartate aminotransferase (AST) and alanine aminotransferase (ALT) activities. Present results demonstrated that captopril protect the liver against toxic effects of cisplatin in a dose dependent manner.

Key words: Cisplatin · Captopril · Hepatotoxicity · Liver Enzymes

INTRODUCTION

Cisplatin (CPN) is one of the most potent anticancer drugs used in chemotherapy [1]. In spite of its significant anticancer activity, the clinical use of cisplatin is often limited by its undesirable side effects such as nephrotoxicity [2]. Hepatotoxicity can also occur when cisplatin is administered at high doses [3]. Oxidative stress appears to play an important role in cisplatin induced hepatotoxicity. For example, metallothionein protects against liver injury induced by high doses of cisplatin in mice [4]; selenium and high dose of vitamin E administration protect against cisplatin-induced oxidative damage to liver [5]; heme oxygenase (HO) and catalase are important protective responses against cisplatin toxicity in the livers of tumour-bearing mice [6].

Angiotensin-converting enzyme (ACE) inhibitors are popular drugs in the treatment of hypertension and congestive heart failure [7]. Other pharmacological effects such as free radical scavenger action reduction of oxidant stress [8] and anti-fibrotic effects have been postulated [9].

It has been shown that captopril, a prototype ACE inhibitor, increases the activity of liver superoxide dismutase and glutathione peroxidase, which are of the main anti-oxidant enzymes found in aerobic organisms, *in vitro* independently of ACE inhibition. This activity protects cells from oxidative damage, although the mechanism is not fully understood [7].

The aim of the present study was to evaluate the possible protective effects of captopril on cisplatin induced hepatotoxicity in rats.

MATERIALS AND METHODS

Animals and Study Design: Forty eight male Wistar rats weighing 364±37 g were used in this study. Animals kept under controlled temperature conditions (22±2°C) with relative air humidity of 60% and 14 hours of light daily. The animals had free access to rat chow and water. All experiments were conducted in accordance with the Animal Research Ethics Committee. 7.5mg/kg b.w. CPN administered intraperitoneal (i.p.) in a single dose. Captopril (Exir Pharmaceutical Company, Iran) doses (50,100 and 150 mg/kg/d drinking water) and administered to the animals by gavage. Treatment with these doses of per day began two days before the application of CPN and continued to be administered until five days after application of the herbicide. Rats were randomized into 6 treatment groups with 8 animals in each group, as follows:

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(1) saline solution (NaCl 0.9%); (2) Captopril (100 mg/kg/d); (3) CNP; (4) CPN + Captopril (50 mg/kg/d); (5) CPN + Captopril (100 mg/kg/d); (6) CPN + Captopril (150 mg/kg/d).

Serum Biochemical Parameters and Oxidative Status: Blood samples were taken via cardio puncture under ether anaesthesia. Blood sera were isolated for the evaluation of aspartate aminotransferase (AST) and alanine aminotransferase (ALT) activities.

Liver tissue used for measurement of antioxidant factors level. Liver were separated and washed in normal saline. Ten percent of homogenised tissue was prepared in phosphate buffer. The homogenate was centrifuged at 3000 rpm for 30 min to remove debris. The supernatant was used for the measurement of catalase (CAT), superoxide dismutase (SOD), glutathione-S-transferase (GST) activities and the levels of reduced glutathione (GSH), lipid peroxidation. SOD activity was determined from the ability of the tissue to delete the superoxide anion generated from the photo-illumination of riboflavin according to the method of Mc Cord and Fridovich [8]. Tissue CAT activity was determined from the rate of degradation of H_2O_2 [9]. Reduced GSH was determined according to the method of Moron *et al*. [10] based on the formation of a yellow colour complex. GST activity was determined from the rate of increase in reduced glutathione and CDNB [11]. The level of lipid peroxidation was evaluated as malondialdehyde (MDA), a thiobarbituric acid reacting substance, using tetra-methoxypropane as standard [12].

Statistical Analysis: Statistical significance of differences between means of groups was evaluated using ANOVA followed by Tukey test with SPSS software. A value of $P < 0.05$ was considered significant.

RESULTS

The damage to the structural integrity of liver is commonly assessed by the determination of serum aminotransferases (ALT and AST) activities After treatment with CNP, levels of serum ALT and AST was significantly (Pb<0.05) increased compared to the control (Table 1). The effect of CNP and its co-administration with different dose of Captopril on chemical parameters of liver are presented in Tables 2 and 3.

Table 1: Effect of CNP and its co-administration with different doses of Captopril on serum ALT and AST

| | Treatment | | | | | |
Factor	T1	T2	T3	T4	T5	T6
ALT (U/L)	45±0.3ᵃ	43±0.4ᵃ	84±1.2ᵉ	70±0.8ᵈ	62±0.9ᶜ	54±0.8ᵇ
AST (U/L)	77±2.2ᵃ	78±2.4ᵃ	159±4.3ᵈ	132±5.2ᶜ	95±3.5ᵇᶜ	89±4.7ᵇ

a-e in each row show significant difference (P>0.05)

Table 2: Effect of CNP and its co-administration with different doses of Captopril on liver CAT, SOD and MDA levels

| | Treatment | | | | | |
Factor	T1	T2	T3	T4	T5	T6
CAT	45±3.3ᵇᶜ	46±2.5ᶜ	32±2.3ᵃ	37±2.9ᵃᵇ	42±3.5ᵇ	43±4.1ᵇ
SOD	20±1.2ᶜ	22±1.0ᵈ	14±0.8ᵃ	15.8±0.4ᵃᵇ	17.5±1.1ᵇ	18.8±0.7ᵇᶜ
MDA	55±3.6ᵃ	52±2.7ᵃ	89±4.4ᵉ	74±4.1ᵈ	66±3.1ᶜ	59±2.7ᵃᵇ

a-e in each row show significant difference (P>0.05)

Table 3: Effect of CNP and its co-administration with different doses of Captopril on liver GST and GSH levels

| | Treatment | | | | | |
Factor	T1	T2	T3	T4	T5	T6
GST	73±5.2ᵉ	74±6.4ᵉ	44±2.4ᵃ	53±4.7ᵇ	60±3.6ᶜ	68±5.6ᵈ
GSH	23±1.3ᵈ	22±1.2ᶜᵈ	14±0.8ᵃ	16±0.8ᵃᵇ	18±1.1ᵇᶜ	19±0.9ᶜ

a-e in each row show significant difference (P>0.05)

DISCUSSION

In our study, hepatoprotective activities of captopril were evaluated against CNP induced oxidative stress cytotoxicity. Angiotensin-converting enzyme inhibitors are considered a rather safe group of therapeutic agent with no serious side effects. Captopril inhibits the ACE that catalyses the conversion of angiotensin I to the vasoconstrictor peptide, angiotensin II. It is generally recommended for the treatment of hypertension, congestive heart failure, acute myocardial infarction and renal complications of diabetes mellitus. It also has beneficial experimental effects in hindering the progression of chronic renal failure, diabetic nephropathy and development of atherosclerosis [13, 14]. Also there is increasing evidence that the broader pharmacological properties of ACE inhibitors encompass the anti-oxidant ability through scavenging free radical because of its terminal-SH group in a variety of organ systems [15-17]. The anti-oxidant activity of captopril possibly ameliorates the oxidative stress. The -SH group in the structure is a crucial requirement for free radical scavenging activity but not the proline part [18].

Cisplatin at 45 mg/kg body weight can induce mouse hepatotoxicity 24 h after administration. Cisplatin-induced hepatotoxicity was enhanced by pretreatment with the CYP2E1 inducer acetone, as reflected by the increase in ALT, AST, caspase-3 activity and TUNEL staining and the histopathology [19]. Acetone increased CYP2E1 activity, but not ECOD activity, which is catalyzed by many forms of cytochrome P450, suggesting it was elevated CYP2E1 induced by acetone that enhanced the mouse liver damage induced by cisplatin. Future studies with inhibitors of CYP2E1 or CYP2E1 knockout mice are proposed to validate the role of CYP2E1 in the cisplatin potentiated toxicity [19].

Our results revealed that under the effect of CPN, lipid peroxidation was increased by MDA concentration enhancement and GSH was reduced as well as the total antioxidant capacity (TAC) also was altered to the reversible effect by MDA decreased, GSH content and TCA were increased by variable values according to the type of each extracts and the presence of SN in combination with each extract. These results are in agreement with Ahmed [20]. Glutathione reduced is thought to be an important factor in cellular function and defence against oxidative stress. It was found that dietary GSH suppresses oxidative stress in-vivo in prevention of diabetic complications [21, 22]. The analysis on antioxidant status and biomarkers along with lipid peroxidation in rat after cisplatin treatment was investigated [23]. The investigation revealed a significant increase in lipid peroxidation status and decrease in glutathione level in hepatic tissue of rat after cisplatin treatment, which indicates that it might cause inactivation of enzymes. Our results exerted that cisplatin caused significant increases in serum ALT and AST. This result was in agreement with the results of Iseri *et al.* [21, 24]. They found that CPN caused a marked reduction in liver function by increase the activity of transaminases. Cisplatin is one of the most active cytotoxic agents in the treatment of cancer. Toxic effects, as nephrotoxicity and neutrotoxicity and less frequent toxic effects as hepatotoxicity was generally observed after administration of high doses of cisplatin [25]. CPN has been demonstrated to generate active oxygen species, such as superoxide anion and hydroxyl radical [26] and to stimulate lipid peroxidation in the kidney tissues [27]. Our results revealed that administration of CPN to the rats caused a significant decrease in the level of SOD and CAT in liver. The lipid peroxidative degradation of the biomembrane is one of the principle causes of toxicity of CCl4. This is evidenced by the elevation of TBARS and decreases in the activity of free radical scavenging enzymes same as SOD and CAT in the CCl4 treated animals [28]. SOD is the key enzyme in scavenging the superoxide radicals. Catalase (CAT) is also another key enzyme in the scavenging, which helps in cleaning the H_2O_2 formed during incomplete oxidation.

In conclusion, present results demonstrated that captopril protect the liver against toxic effects of cisplatin in a dose dependent manner. However, further investigations will be done to elucidate the mechanism of protection and potential usefulness of captopril as a source of protective agents against drugs or xenobiotics toxicity in clinical trials.

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