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AMELIORATION OF CISPLATIN-INDUCED TOXICITY IN BLOOD DOMESTIC RABBIT'S BY SODIUM THIOSULFATE OR GINGER EXTRACT

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ABSTRACT
Cisplatin is a chemotherapeutic agent that is widely used to treat a variety of malignant tumors. A comparative studies were performed to assess the protective effects of sodium thiosulfate or ginger extract on cisplatin-induced toxicity in some blood hematological and biochemical parameters of twenty eight domestic rabbits. Animals were divide into four groups, the first was comprised 10 rabbits and given 1 ml saline intramuscularly and kept as a control. The second group (6 rabbits) received 5 doses of cisplatin at a dose of 5mg/kg b.wt. i. m. once/day, the third group (6 rabbits) received the previous dose of cisplatin plus sodium thiosulfate at a dose of 0.4gm/kg b.wt. i. m. once/day for 5 days, while the fourth (6 rabbits) given the same dose of cisplatin plus 100mg/ kg b.wt of ginger extract (given orally) once/day for 5 days. In the present study, sodium thiosulfate administration concomitantly with cisplatin reduced cisplatin-induced lesions in a significant manner. These finding suggest that sodium thiosulfate or ginger extract so effectively, reduces cisplatin-induced toxicity and there use concomitantly with cisplatin are highly recommended. It was found in this study that ginger extract more potent against cisplatin than sodium thiosulfate in many biochemical and hematological changes.

KEYWORDS: Cisplatin, sodium thiosulfate, ginger extract, rabbits, blood.

INTRODUCTION
Cisplatin is a chemotherapeutic agent used in the treatment of solid tumors like ovarian, testicular, cervical, lung, head and neck and bladder cancers. The clinical side effects of cisplatin include nausea, vomiting, myelosuppression, allergic reaction, nephrotoxicity, ototoxicity, neurotoxicity, weight loss, nausea, vomiting, seizures, hearing loss, tinnitus and bone marrow suppression (Brock et al., 2012). While nephrotoxicity can to some extent be reversed by increasing saline hydration as well as mannitol diuresis, there are no known cures or preventative treatments available for ototoxicity and neurotoxicity. Cisplatin induced hearing loss is usually bilateral and irreversible, and is particularly serious in the pediatric population with cancers like neuroblastomas, CNS malignancies, head and neck cancers, where irradiation of the base of the skull or brain is also performed (Chen et al. 2006). The major alterations reported in cisplatin toxicity (Yzhang and Lindup,1993) are the occurrence of oxidative stress linked to the accumulation of reactive oxygen species (Chirino, 2009) and decreased efficiency of some antioxidant defense systems including antioxidant enzymes (Sadzuka et al., 1992) and non-enzymatic antioxidants, such as reduced glutathione (GSH). In addition, functional and structural mitochondrial damage, apoptosis, perturbation in Ca2+ homeostasis (Kawaiy etal., 2006; Martins et al., 2008), involvement of pro-inflammatory genes such as cox-2 and inducible nitric oxide synthase (iNOS) may play some important roles in the mechanism of cisplatin hepatotoxicity (Kim et al., 2004). Prevention of cisplatin side effects is one of the major clinical issues and in this respect the use of free radical scavengers has been recently explored (Davis et al., 2001). Thus, there is an imperative need for treatments that will ameliorate ototoxicity. Attenuation of cisplatin ototoxicity has been shown in several scientific research papers by protective agents that are predominantly anti-oxidants like N-acetyl cysteine (NAC), sodium thiosulphate, amifostine, lipoic acid etc. Sulfur containing compounds such as sodium thiosulfateis on the World Health Organization's List of Essential Medicines, it was reported to have many medical uses such as: treatment of calciphylaxis in hemodialysis people with end-stage kidney disease (Cicone et.al., 2004). There is apparently an incompletely understood phenomenon whereby this causes severe metabolic acidosis in some patients. It is used as an antidote to cyanide poisoning. Thiosulfate serves as a sulfur donor for the conversion of cyanide to thiocyanate (which can then be safely excreted in the urine), catalyzed by the enzyme rhodanase (Hall et.al., 2007). It is used in the management of extravasations during chemotherapy. Sodium

thiosulfate prevents alkylation and tissue destruction by providing a substrate for the alkylating agents that have invaded the subcutaneous tissues. It may be instilled subcutaneously into multiple sites using a small-gauge needle in measuring the volume of extracellular body fluid and the renal glomerular filtration rate (Selk and Rodby, 2011). It was reported to provide good otoprotection against cisplatin (Otto et al., 1988), but had significantside effects and/or interference with antitumor activity. It was found that sodium thiosulfate prevents cisplatin-induced hypomagnesemia (Wong et al., 1988). The Protection by sodium thiosulfate and thiourea against lethal toxicity of cis-diammine dichloroplatinum (II) in bacteria and mice was reported by (Ishizawa et al., 1981). Reduction of acute cisplatin ototoxicity and nephrotoxicity in rats by oral administration of allopurinol and ebselen was detected by (Lynch et al., 2005), while (Kart et al., 2010) investigate the protective effect of Caffeic acid phenethyl ester ameliorates cisplatin-induced hepatotoxicity in rabbit. The results of (Rybak et.al., 2009) study was shown that the reduced glomerular filtration rate in early cisplatin-induced renal failure is due, in part, to reversible changes in renal blood flow and renal vascular resistance. On the other hand, the first mechanism that can explain the amelioration of the side effects of cisplatin by sodium thiosulfate was reported by (Sooriyaarachchi et al., 2012), they suggest that when plasma was incubated with cisplatin for up to 3 h and sodium thiosulfate was added thereafter the analysis of the obtained mixture revealed that the formation of the same Pt- sodium thiosulfate complex which in turn greatly diminished the plasma protein binding of cisplatin-derived hydrolysis products. Thus, the observed amelioration of the side effects of cisplatin by sodium thiosulfate can be rationalized in terms of the rapid formation of a biologically inactive Pt- sodium thiosulfate complex in the bloodstream.

Ginger belongs to a tropical and sub-tropical family-Zingiberaceae, it has been cultivated for thousands of years as a spice and for medicinal purposes (Park and Pezzuto, 2002). For centuries, it has been an important ingredient in Chinese, Ayurvedic and Tibb-Unani herbal medicines for the treatment of rheumatism, gingivitis, toothache, asthma, stroke, nausea, vomiting and diabetes (Ali et al., 2008). Extracts of the ginger are rich in shagaols and gingerols which exhibit anti-inflammatory, anti-oxidant and anti-carcinogenic proprieties under ‘‘in vitro’’ and ‘‘in vivo’’ systems (Surh, 2002). Ginger was found to have hypocholesterolemia and caused a decrease in blood glucose, body weight and alkaline phosphatase in adult male rat (Bhandari et al., 2005). Several studies showed that ginger extract possesses hypoglycemic and hypolipidemic potential in the diabetic rats (Kondeti et al., 2011).While data is very rare, the goal of this study was to evaluate the protective effect of sodium thiosulfate on cisplatin-induced toxicity in some blood values of domestic rabbits.

MATERIAL AND METHODS

Preparation of plant material

The fresh rhizomes of Chinese ginger (zingiberofficinale) were bought from the local market of Gaza city. After the plant material was identified, the fresh rhizomes were washed with distilled water and then dried at room temperature for two days under the shade, the dried rhizome was cut into small pieces and ground into powder by using electric mill for 3 minutes. 50g of powder were put in the round bottle flask, 200ml of ethanol (70%) were added to the flask and extracted for 12 hours at 70c^{o}. the extract was filtered by using WhatmanNo.31 filter paper, then the extract were put in the Petri dish and left at room temperature under the shade. the resultant was viscous substance with brown color. The collection extracts were kept in tight closed container and stored at 4c^{o} untilusing.

Experimental animals

A total of (28) rabbits weighing 1.5 – 2 kg were used throughout this study. Animal were obtained from the breeding unit of Biochemistry Department (Faculty of Applied Sciences, Al-Aqsa University, Gaza, Palestine) and kept under standard laboratory conditions (12 hour light/12 hour dark and 24 ± 3°C) and fed with standard commercial rabbit chow. Food and water were provided ad libitum. The experimental protocol was approved by the Veterinary Control and Research Institute Ethics Committee. The rabbits were randomly divided into 4 equal groups; the first group was comprised 10 rabbits served as control and received a single intramuscularly dose of 0.9% saline. Rabbits from the second group (6 rabbits) were injected intramuscularly with a 5 mg/kg body weight cisplatin dose for 5 days. The third group (6 rabbits) received the previous dose of cisplatin (5 mg/kg body weight) plus sodium thiosulfate (0.4 gm / kg body weight) intramuscularly once/day for 5 days (Kim et al., 2004). While the fourth group (6 rabbits) received the previous dose of cisplatin (5 mg/kg body weight) plus 100mg/ kg body weight of ginger extract once/day for 5 days. All chemicals used were of analytical grade and were procured from Sigma Chemical Company Germany. Cisplatin and sodium thiosulfate amp. were brought from Shifa Hospital, Gaza.

Animals from both experimental and control groups were decapitated at weekly intervals. Blood samples were collected from all animals by puncture of the jugular vein into sterile microtubes after cisplatin and sodium thiosulfate treatments. After clotting at room temperature for 1 hour, samples were centrifuged at 700g for 10 minutes at room temperature and sera were carefully harvested and stored at -20°Cuntil analysed. . However, determinations of enzyme activities were carried out on fresh serum samples, On the other hand, about 2 mL of blood samples were collected in a tube containing dipotasium ethylene diamine tetra acetate (EDTA) for the hematological tests.

BIOCHEMICAL ANALYSES

Serum glucose, triacylglycerol and total cholesterol, total protein, albumin, globulin, bilirubin, urea, creatinine and uric acid were analyzed by Konelab 60 Auto analyzer in Al Nasser Hospital Clinical Chemistry Laboratory. Serum Glucose was determined by glucose oxidase (GOD) (Trinder, 1969) using Lab Kit kit, Spain. Serum total protein was determined by Biuret method using DiaSys kit spain (Johnson et. al., 1999). On the other hand, Bio Systems kits, Spain were used in determinations of proteins, urea, Creatinine and uric acid. Serum Albumin was determined by Bromocresol green (Doumas, et al., 1971). The concentration of globulins were calculated using the equation: Concentration of globulins (mg/dl) = Total protein – Albumin. Serum urea was determined by urease - glutamate dehydrogenase (GDH) (Gutmann and Bergmeyer, 1974). Serum Creatinine was determined by Alkaline Picrate method (Fabiny and Ertingshausen, 1971). Serum Uric Acid was determined by Uricase /POD method (Fossati et. al., 1980). The activities of serum aspartate aminotransferase (AST) and alanine aminotransferase (ALT) were determined according to the method described by Gloiser and Mager, 1972. The measurement of serum alkaline phosphatase (ALP) activity was based on the method of Perry et al., 1983. Estimation of serum Na and K was conducted by Flame Photometric method (Tietz, 1990) estimation of serum Ca was done by Gitelman method (Gitelman, 1967). Determination of hematological parameters were carried out using an 18 automated parameter hematology analyzer. ABX Micros 60 from Horiba ABX. France.

Statistical Analysis

Data are presented as mean ± standard error of means (SEM). One-way analysis of variance and post hoc Duncan's test were used to determine the differences between groups in terms of all studied parameters using the SPSS/PC computerprogram (version 12.0; SPSS, Chicago, IL, USA). The Kruskal Wallis test was used for the comparison between the control and experimental groups of examination. Differences were considered as significant when P-value was less than 0.05.Percentage change was also calculated.

RESULTS

Serum glucose, cholesterol, triglycerides and bilirubin mean values of rabbits affected by cisplatin (5mg/kg/day), cisplatin plus sodium thiosulfate (5mg/kg/day + 0.4gm/kg/day) and cisplatin plus ginger extract (5mg/kg/day +100mg/ kg/day) during five days are summarized in table1. The data revealed that administration of cisplatin plus sodium thiosulfate compared to cisplatin only, increase in a highly significant way serum glucose level from 21.34% to 13.81%, also administration of cisplatin plus ginger extract increase in a highly significant way at 12.73% as compared to the control level . Cholesterol was highly significant ($P < 0.01$) increased at 70% in for cisplatin only and in a significant way for cisplatin plus sodium thiosulfate at 9.64% and at -6.07% for cisplatin plus ginger extract. While triacylglycerol and bilirubin have shown a highly significant ($P<0.01$) decreased the (% change) from 44.44% to 26.74% and 37.33%, and from 233.33 % to 130.77% and 166.67% respectively by treatment with these drugs.

Table (1): Effect of cisplatin, cisplatin plus sodium thiosulfate and cisplatin plus ginger extract administration on glucose, cholesterol, triglycerides and bilirubin of rabbits.

Parameters	*Experimental Drug*			
	Control n=10	**Cisplatin n=6**	**Cisplatin/Sodium Thiosulfate n=6**	**Cisplatin/Ginger Extract n=6**
Glucose (mg/dl) % change P value	75.12±0.42	214.44 ± 0.28 21.34 P < 0.01	139.15 ± 0.29 13.81 P < 0.01	128.31± 0.19 12.73 P < 0.01
Cholesterol (mg/dl) % change P value	130.59±0.27	222.0± 0.36 70.00 P < 0.01	143.19± 0.33 9.64 P < 0.05	122.66± 0.07 -6.07 P < 0.05
Triacylglycerol (mg/dl) % change P value	114.40±0.40	165.25±0.36 44.44 P < 0.01	145.0± 0.29 26.74 P < 0.01	157.11± 0.42 37.33 P < 0.01
Bilirubin (mg/dl) % change P value	0.39±0.10	1.30±0.11 233.33 P < 0.01	0.90±0.12 130.77 P < 0.01	1.04± 0.21 166.67 P < 0.01

All values were expressed as mean ± Standard error(S.E) ; P value <0.05 significant; P >0.05 nonsignificant,; P<0.01 highly significant.

Data in Table (2) showed that administration of only cisplatin (5mg/kg/day) for 5 days have increased urea in a highly significant effect ($P<0.01$) by 59.48% compared to control, but when administered combined with sodium thiosulfate or with gineger extract, the percentage of change values were decreased to12.58% and 16.56%. In the same manner, uric acid and creatinine were highly significant ($P<0.01$) increased by 67.07% and 97.53% compared to control, but when administered combined with sodium thiosulfate or with ginger extract, the values were decreased in a highly significant manner ($P<0.01$) to 47.69%, 72.83% and 50.12%, 61.72% respectively.

Total protein and albumin were decreased in a different way, as total protein in a significant ($P < 0.05$) while albumin in a highly significant decreased ($P < 0.01$) with -10.62% and -20.25% with cisplatin only, but when administered combined with sodium thiosulfate or with ginger extract, the values were decreased from -7.15% , -14.11% and from-2.12%(no significant),-22.49% respectively. Meanwhile, globulin was increased signicantly ($P < 0.05$) as the percentage of change values increased from 4.59% to 4.84% with sodium thiosulfate and decreased to -3.06% with ginger extract, all compared to control.

Table: (2) Effect of cisplatin, cisplatin plus sodium thiosulfate and cisplatin plus ginger extract administration on protein profiles of rabbits.

Parameters	Experimental Drug			
	Control n=10	Cisplatin n=6	Cisplatin/ Sodium Thiosulfate n=6	Cisplatin/Ginger Extract n=6
Urea (mg/dl) % change P value	30.19±0.27	48.15 ± 0.20 59.48 P < 0.01	33.99 ± 0.25 12.58 P < 0.01	35.19 ± 0.13 16.56 P < 0.01
Uric acid (mg/dl) % change P value	4.13±0.17	6.90± 0.14 67.07 P < 0.01	6.10± 0.21 47.69 P < 0.01	6.20 ± 0.25 50.12 P < 0.01
Creatinine (mg/dl) % change P value	0.81±0.05	1.60±0.02 97.53 P < 0.01	1.40± 0.03 72.83 P < 0.01	1.31± 0.03 61.72 P < 0.01
Total Protein (gm/dl) % change P value	8.95±0.21	8.00± 0.18 -10.62 P < 0.05	8.31± 0.23 -7.15 P < 0.05	8.76± 0.03 -2.12 P > 0.05
Albumin (gm/dl) % change P value	4.89±0.18	3.90±0.15 -20.25 P < 0.01	4.20± 0.16 - 14.11 P < 0.01	3.79± 0.46 -22.49 P < 0.01
Globulin (gm/dl) % change P value	3.92±0.11	4.10±0.18 4.59 P < 0.05	4.11± 0.13 4.84 P < 0.05	3.80± 0.29 -3.06 P < 0.05

All values were expressed as mean ± Standard error(S.E) ; P value <0.05 significant; P >0.05 nonsignificant,; P<0.01 highly significant.

Results demonstrated in Table (3) indicated that administration of cisplatin (5mg/kg/day) was increased in a highly significant way ($P<0.01$) the activities of ALP, ALT and AST at 102.20%, 104.37% and 76.72% respectively, but when sodium thiosulfate (0.4gm/kg/day) administered plus cisplatin the increments of ALP, ALT and AST were also highly significant ($P<0.01$) at 30.98%, 32.44 % and 32.88% respectively as compared to control level. While administration of cisplatin plus ginger extract have increased the value of ALP, ALT and AST in a highly significant way ($P < 0.01$) at 71.03% , 5.54% and 58.35% respectively. On the other hand, it was found that administration of cisplatin only was highly significant($P < 0.01$) increase the percentage of calcium, sodium and potassium level at 72.41%, 20.05% and 37.63% respectively, however when sodium thiosulfate was administered with cisplatin the increment were highly significant($P<0.01$)for calcium, sodium and potassium at 21.29%, 15.75% and 32.75% respectively, but when ginger extract was administered with cisplatin the increment were highly significant ($P < 0.01$) for calcium and potassium at 53.63% and 30.66%, while a significant increase ($P < 0.05$) for sodium at 7.48% was observed.

Table(3):Effect of cisplatin, cisplatin plus sodium thiosulfate and cisplatin plus ginger extract administration on enzymes activity and electrolytes of rabbits.

Parameters	Experimental Drug			
	Control n=10	Cisplatin n=6	Cisplatin/Sodium Thiosulfate n=6	Cisplatin/Ginger Extract n=6
ALP (U/L) % change P value	29.89±0.31	60.44 ± 0.28 102.20 P < 0.01	39.15 ± 0.29 30.98 P < 0.01	51.12±0.27 71.03 P < 0.01
ALT (U/L) % change P value	41.15±0.19	84.10±0.21 104.37 P < 0.01	54.50±0.17 32.44 P < 0.01	59.89±0.34 45.54 P < 0.01
AST (U/L) % change	27.28±0.22	48.21± 0.26 76.72	36.25± 0.18 32.88	43.20± 0.16 58.35

P value		P < 0.01	P < 0.01	P < 0.01
Calcium mg/dl % change P value	11.13±0.13	19.19±0.16 72.41 P < 0.01	13.50±0.14 21.29 P < 0.01	17.10±0.16 53.63 P < 0.01
Sodium meq/l % change P value	120.95±0.14	145.20±0.17 20.05 P < 0.01	140.0±0.12 15.75 P < 0.01	130.0±0.02 7.48 P < 0.05
Potassium meq/l % change P value	2.87±0.17	3.95±0.14 37.63 P < 0.01	3.81±0.11 32.75 P < 0.01	3.75±0.14 30.66 P < 0.01

All values were expressed as mean ± Standard error(S.E); P value <0.05 significant; P >0.05 nonsignificant,; P<0.01 highly significant.

Changing in blood indices after administration of cisplatin (5mg/kg/day), sodium thiosulfate (0.4gm/kg/day) and ginger extract (100mg/ kg/day)(are summarized in table 4. Data indicated that administration of cisplatin increased in a highly significant (P<0.01) way the counts of WBC, RBC, Hb, MCV, MCH,MCHC, Lymphocyte and platelets at 56.81%, -38.04%, -25.26%, 123.25%, 159.02%, 18.81%, 43.22% and -41.93% respectively, while hematocrit increased in a significant way (P<0.05) by 10.14%. But when sodium thiosulfate administered plus cisplatin the increments of WBC, RBC, Hb, MCV, MCH,MCHC, Lymphocyte and platelets were highly significant (P<0.01) at 21.11%,-33.82%, -12.45%, 66.57%, 81.10%, 13.12%, 12.81% and -28.20% respectively, and again hematocrit increased in a non significant way (P>0.05) by 0.42%. It was very clear that the increments of the percentage values were lowered when cisplatin administered with sodium thiosulfate. However, when ginger extract was administered with cisplatin the increment were highly significant (P < 0.01) for MCV, MCH, MCHC and platelets at 76.74% ,14.91%, 15.72% and -33.88% respectively, while the increments of WBC, RBC and Lymphocyte were significant (P<0.05) at 7.67%, 9.10% and 4.87% respectively. It was found that the increments of Hb and hematocrit were non significant at 0.35% and 1.84% respectively. It was also very clear that the increments of the percentage values were lowered when cisplatin administered with ginger extract more than when cisplatin administered only.

Table (4): Effect of cisplatin, cisplatin plus sodium thiosulfate and cisplatin plus ginger extract administration on total blood counts of rabbits.

	Experimental Drug			
	Control n=10	**Cisplatin n=6**	**Cisplatin/ Sodium Thiosulfate n=6**	**Cisplatin/Ginger Extract n=6**
WBC count ($x10^3$cell/ul) % change P value	5.21±0.14	8.17±0.22 56.81 P < 0.01	6..31±0.26 21.11 P < 0.01	5.61±0.19 7.67 P < 0.05
RBC count ($x10^6$ cell/ul) % change P value	6.15±0.13	3.81±0.22 -38.04 P < 0.01	4.07±0.11 -33.82 P < 0.01	6.71±0.33 9.10 P < 0.05
Hb (g/dl) % change P value	14.05±0.16	10.50±0.12 -25.26 P < 0.01	12.30±0.17 -12.45 P < 0.01	14.10±0.57 0.35 P > 0.05
Hematocrit (%) % change P value	30.97±0.15	34.11±0.26 10.14 P < 0.05	31.10±0.13 0.42 P > 0.05	31.54±0.18 1.84 P > 0.05
MCV (fi) % change P value	72.41±0.17	161.66±0.15 123.25 P < 0.01	120.62±0.19 66.57 P < 0.01	127.98±0.39 76.74 P < 0.01
MCH (pg) % change P value	15.35±0.18	39.76±0.19 159.02 P < 0.01	27.80±0.16 81.10 P < 0.01	17.64±0.12 14.91 P < 0.01
MCHC (g/dl) % change P value	27.21±0.19	32.33±0.18 18.81 P < 0.01	30.78±0.13 13.12 P < 0.01	31.49±0.58 15.72 P < 0.01
Lymphocyte % change	34.27±0.15	49.07±0.11 43.22	38.66±0.35 12.81	35.94±0.41 4.87

P value		P < 0.01	P < 0.01	P < 0.05
Platelets(x10^3cell/ul)	363.0±29.90	210.80±33.39	260.60±30.66	240.00±40.11
% change		-41.93	-28.20	-33.88
P value		P < 0.01	P < 0.01	P < 0.01

All values were expressed as mean ± Standard error(S.E) ; P value <0.05 significant; P >0.05 nonsignificant,; P<0.01 highly significant.

DISCUSSION

The present study is a comparative one, which performed to assess the protective effect of sodium thiosulfate or ginger extract on cisplatin-induced toxicity in some blood parameters of twenty eight domestic rabbits. It was thought that data on biochemical and hematological alterations on rabbits are limited, therefore, the present study was designed to identify the effects of these drugs on some rabbits' hematological and biochemical changes. The first mechanism that can explain the amelioration of the side effects of cisplatin by sodium thiosulfate was reported by (Sooriyaarachchi et al., 2012), they suggest that when plasma was incubated with cisplatin for up to 3 h and sodium thiosulfate was added thereafter the analysis of the obtained mixture revealed that the formation of the same cisplatin- sodium thiosulfate complex which in turn greatly diminished the plasma protein binding of cisplatin -derived hydrolysis products. Thus, the observed amelioration of the side effects of cisplatin by sodium thiosulfate can be rationalized in terms of the rapid formation of a biologically inactive cisplatin- sodium thiosulfate complex in the blood stream. In the present data, it was found that in comparison with the respective control rabbits, there was a clear decrease in rabbits serum glucose levels in response to sodium thiosulfate or ginger extract administration compared to treatment with cisplatin alone. It was found that cisplatin may indirectly, increase the glycogen content of the hepatocytes and impairs ATP synthesis by the mitochondria (Kademir et al., 2012). The present results in table (1) revealed a high significant increase in the level of glucose concentration in rabbits treated with cisplatin compared with normal control, this result is in concordance with (Han et al., 2003) who found that the cisplatin had decreased expression of the glucose transporter (GLUT4) and reduce glucose transport activity when cultured a dipoccytes that were isolated from rats previously exposure to cisplatin. (Han et al., 2003) demonstrated that there is a beneficial effect of ginger extract on serotonin induce hyperglycemia and hypoinsulinemia in normoglycemic rats and reported that ginger extract inhibited this inductive effect. In support of this hypothesis, it was found that ginger extract has a potential hypoglycemic properties. This hypoglycemic action of ginger may be due to effects involving serotonin receptors and an increase in pancreatic secretion of insulin from B-cell or release ofbound insulin.

The high significant decrements observed in serum triacylglycerol, cholesterol and bilirubin contents in response to treatment by sodium thiosulfate support the finding of Otto et al., 1988. (Agrawal and Sharma, 1999) concluded that cisplatin causes hyper cholesterolemia due to reduced lipoprotein lipase activity which plays a role in the increment of plasma lipid. However, in a Co-treatment of ginger extract with cisplatin there was a significant decrease of total cholesterol, these results are concordant with ours which reported that triacylglycerol, cholesterol and bilirubin levels were high significantly decrease in response to ginger administration (Kondeti et.al., 2011). They stated that ginger treatment can reduce total serum cholesterol by enhancing the activity of liver cholesterol -7-a- hydrolase or inhibition of hydroxy- methyl- glutaryl- Co- enzyme–A (HMG-CoA) reductase, either by bile – acid conversion or fecal excretion of cholesterol .

Sodium thiosulfate amelioration to cisplatin-induced toxicity in rabbits protein profiles was very clear as it decrease urea, uric acid, creatinine, total protein, albumin and globulin levels significantly. This finding was consistent with (Kim et al.,1995) study which found that, urine flow was decreased following cisplatin treatment, which was accompanied by marked reduction in GFR. Cisplatin induced glucosuria, phosphaturia, and aminoaciduria. These results suggest that cisplatin results in impaired proximal tubular reabsorptive function and the renal concentrating defect. The increments of Na^+, K^+ and Ca^{2+} concentrations were very obvious in our study, which was consistent with others(Kawaiy etal., 2006; Martins et al., 2008), who suggest that cisplatin caused an increase in fractional excretion of Na^+, K^+ and Ca^{2+} and a decrease in urine osmolality, free-water reabsorption and urine to plasma creatinine ratio (Kim et al.,1995). These results are also in agree with (Wong et al., 1988), who suggest that concurrent injections of sodium thiosulfate intravenously prevented the hypomagnesemic and the nephrotoxic effects of cisplatin and can be of clinical significance. The present data of liver enzymes activities also show the protective effect of sodium thiosulfate on cisplatin hepatotoxicity. There was a highly significant increase of ALP, ALT and AST values after administration of cisplatin alone, but these alterations were less severe in rabbits receiving cisplatin -sodium thiosulfate. These results were supported by (Kart et al., 2010), who suggest that, parallel to histopathology, cisplatin increased serum AST and ALT levels, whereas sodium thiosulfate treatment significantly reduced cisplatin-induced AST and ALT rise in the serum. These results suggest that cisplatin causes oxidative and nitrosative damage to hepatocytes. Cisplatin-induced increase in XO and NO could contribute oxidative stress in the hepatotoxicity. Sodium thiosulfate shows partial protection against cisplatin-associated biochemical and histopathological alterations.

The Co- administration of ginger extract with cisplatin restored the levels of the enzymes in serum of the rabbits as an indication of the protective effect of ginger extract against liver damage induced by cisplatin, which consistent with our finding that ginger has a highly protective effect on the hepatotoxicity. The results of this study demonstrated that ginger extract improve the elevated levels of the serum AST, ALT and ALP when compared to the cisplatin treated group, and these may be attributed to ginger components which may stabilize hepatocytes plasma membrane and prevent delivery of AST and ALT to the extracellular fluid (Ajith et. Al., 2007). (Bhandari et.al., 2003) found that ginger is useful and lowers the serum AST, ALT and ALP enzymes this reduction could be attributed to the fact that ginger contains high content of antioxidant that makes it a free radical scavenger.

The present study aimed to investigate the possible protective effect of sodium thiosulphate or ginger extract against cisplatin-induced disturbance in hematological indices in rabbits. Hematopoietic system is one of the most sensitive systems to evaluate the hazards effects of poisons and drugs in humans and animals (Lijuv et.al., 2013). The toxic effect of cisplatin on blood parameters was demonstrated by the significant reduction in RBCs, Hb and platelets counts with subsequent proceed in the values of WBC, MCV, MCH, MCHC, Hematocrit and lymphocytes. The previous results suggested that there was an etiological relationship between anemia and cisplatin treatment. Such relationship could be explained through different mechanisms including destruction of bone marrow cells or increase osmotic fragility of RBCs. Thus, cisplatin intoxication might lead to anemia as a result of either suppression the activity of hematopoietic tissues, impaired erythropoiesis, and accelerated RBCs destruction because of the altered RBCs membrane permeability, increased RBCs mechanical fragility, and/or defective (Fe) metabolism, thus the disturbances in RBCs could reflect an imbalance between its production and loss (Yuan et.al., 2014 ; Hassan et.al., 2010).

The obtained results indicate that exposure to cisplatin - sodium thiosulfate combination, produces a clear increment of WBCs, MCV, MCH and MCHC values after a equivalent increase when the animals injected with cisplatin alone. This Highly significant increase in WBCs count indicated the activation of defense mechanism and immune system of rabbits. This induction of white blood cells is a positive response for survival due to cell mediated immune response of animals (Sooriyaarachchi et. Al., 2012). Leukocytosis was manifested by lymphocytosis, which was the main features of the differential leukocytic count. It was found that exposure to cisplatin-sodium thiosulfate combination produced a significant response in RBCs, Hb, Hematocrit and platelets counts. However, when cisplatin was applied concurrently with ginger extract the animals treated with ginger revealed significant modulation in most of the hematological indices changed in cisplatin-treated rabbits. Ginger was found to have beneficial effects against cisplatin -induced suppression in most of hematological parameters and RBCs indices as it increased number of RBCs and Hb concentration about to normal levels. Also, ginger extract induced great improvement in the reduction of MCV, MCH, MCHC levels in cisplatin -treated animals. Ginger-induced increase in erythrocytes count might be linked either to an increase in erythropoiesis or to the ability of ginger in decreasing membrane rigidity inherent to its cholesterol lowering effect. These results indicated that ginger had preventive and protective effects against cisplatin induced hematological changes. The ameliorative effect of ginger extract could be due to reduction of lipid peroxidation level in cell membrane with subsequent prevention of free radicals induced damage through its antioxidant activity achieved by its active compounds (Hamlaoui et.al., 2012). This finding may be explained on the basis of inhibitory effect of these drugs on histogenesis. The decreased in RBC count and hemoglobin (Hb) lowered the oxygen supply to different tissues thus resulting in low energy production. Decrease in Hb contained MCHC can be explained due to decreased in size of RBCs or impaired biosynthesis of heme in bone marrow (Sooriyaarachchi et. al., 2012 and Kim et.al., 2004).

CONCLUSION

Sodium thiosulfate and/or ginger extract administration concomitantly with cisplatin plays an effective role in ameliorating cisplatin-induced toxicity. They reduced cisplatin-induced lesions in a significant manner. Ginger extract is found to be more potent as a protective against cisplatin than sodium thiosulfate in many plasma biochemical and hematological changes.

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