ISSN NO. 2320-5407

Journal homepage: http://www.journalijar.com

INTERNATIONAL JOURNAL OF ADVANCED RESEARCH

RESEARCH ARTICLE

The Role of *Ficus carica* Leaf Extract in Modulation of the experimentally induced Hepatotoxic Damage in Male Rats

Maher A.Amer, Mohamed A.EL-missiry, Aya Abd rab EL-nabi
Zoology department, Faculty of science, Mansoura university. Mansoura, Egypt.

Manuscript Info

Manuscript History:

Received: 14 October 2015
Final Accepted: 25 November 2015
Published Online: December 2015

Key words:

Ficus carica- Hepatotoxic damage- CCl_4- Oxidative stress markers- Antioxidants-Hepatic enzymes- Lipid profiles- Male rats

Corresponding Author

Maher A.Amer

Abstract

Background: Hepatic diseases remain problems throughout the world. Egypt has the highest country wind prevalence of hepatic C virus, there are no completely effective drugs that stimulate hepatic function and help to regenerate liver cells. Thus, it is necessary to obtain pharmaceutical alternatives for liver diseases. **The aim of this study** was to estimate the hepatoprotective activity of methanolic leaves extract of *Ficus carica* in a rat model of CCl_4-induced liver damage. **Animal grouping:** Adult male rats weighting 100-120g, were divided into five groups as the following (1) Control group. (2) Olive oil treated group(2.5ml/ kg bw)day after day for8 weeks. (3)*Ficus carica* extract treated group (500mg/kg) orally by the same manner and duration. (4)Carbon tetrachloride intoxicated group (CCl_4): CCl_4 dissolved in olive oil (V/V) at a dose (2.5ml /kg body weight) orally by the same manner and duration. *(5)Ficus carica* extract administered orally before CCl_4 –intoxication by the same dose, manner and duration.
Results: CCl_4-significantly altered serum and hepatic enzymes, total protein, albumin, globulin, oxidative stress markers and lipid profiles. The *Ficus* extract showed are notable amelioration in all biochemical alteration comparing with normal control rats. **Conclusion**: It can be concluded that supplementation of *Ficus* extract can exert a hepatoprotective effect in rat model of CCl_4-induced liver damage, which was attributed to the strong antioxidant power.

INTRODUCTION

Hepatotoxicity is main problems worldwide. Thus, it is necessary to obtain pharmaceutical alternatives for liver diseases. This study was conducted to explore possible protective effect of methanolic leaves extract of *Ficus carica* on carbon tetrachloride (CCl_4)-induced hepatotoxicity, haemotoxicity and oxidative stress in experimental animal models.Hepatic fibrosis is a dynamic process caused by chronic liver injury, eventually leading to cirrhosis. It is predominantly characterized by excessive accumulation of extracellular matrix. Egypt one of countries suffering from hepatitis C virus (HCV) in the world, with an estimated 8–10 million among a population of 68 million having been exposed to the virus and 5–7 million active infections **(Frank *et al*., 2000)**. Carbon tetrachloride is one of the most widely used hepatic toxins for experimental induction of hepatic fibrosis and cirrhosis in experimental animals**(Jiang *et al*., 2004; Singab *et al*., 2005)**.CCl_4 is frequently found as a byproduct in drinking water, which remains a potential health hazard to humans. Furthermore, most CCl_4 in food is residual contamination fumigation as a pesticide ***(*Weber *et al*., 2003*)*.**CCl_4 prompted cirrhosis or fibrosis in experimental animals resembles cirrhosis of human in some features of morphology and pathophysiology. CCl_4 are widely used as model compound to induce hepatotoxicity and elucidate its mechanisms of action following exposure to these compounds **(Kim *et al*., 2010)**

There has been a great deal of interest recently in role of alternative and complementary medicines for the treatment of various acute and chronic diseases **(seef *et al*., 2001)**.

Ficus carica Linn. has been used for metabolic, cardiovascular, respiratory, antispasmodic and anti-inflammatory disorders (**Patil Vikas, *et al*., 2010**). It has high minerals, vitamins, dietary fibers and phenolic contents which play an important role in its antioxidant capacity **(Veberic, *et al*., 2008)**.

Analysis of antioxidants in fig revealed that it contains significant amounts of the antioxidant vitamins; vitamin A and vitamin C. **Lattanzio, (2003) reported** that besides antioxidant effects, phenolic compounds have a wide variety of biochemical properties and can also have a useful effect in preventing the development of ailment like cancer and cardiovascular diseases. There is linear correlation between the total content of phenolics and the antioxidant capacity (**Cai *et al*., 2004; kumaran and karunakaran, 2006**).

The present work was mainly conducted to examine the effect of antioxidant and hepatoprotective activity of *Ficus carica* leaves methanolic extract against carbon tetrachloride induced hepatotoxic damage in male rats the biochemical parameters will be assayed to evaluate the protective effect of *Ficus carica* leaves extract.

MATERIALS AND METHODS

1. Materials

1.1. Chemicals:

Carbon tetrachloride (CCl_4) from BDH Laboratory supplies Poole, BH15 1TD England; Methyl Alcohol (pure reagent analysis) was obtained from El-Nasr Pharmaceutical Chemicals Company, Olive Oil.

1.2. Animals and experimental protocol:

Adult male rats, eight weeks old weighing 100-120 g, were kept under normal condition. They were housed in stainless cages with air-conditioned temperature (22-25°C). The animals received standard chow diet and water *ad libitum*. After 2 weeks of acclimatization, rats were divided into five groups each comprising of ten animals. Animal were treated agreeing with "Guide for the Care and Use of Laboratory Animals" prepared by the National Academy of Sciences and published by the US National Institutes of Health (1985).

1.2.1. Animal grouping:

i. **Control group:** Animals did not receive any treatment.

ii. **Olive oil treated group:** Animals were orally received (2.5ml /kg body weight) day after day for 8 weeks.

iii. ***Ficus carica* extract treated group:** Animals were received *Ficus carica* leaf methanolic extract at dose (500mg/kg) orally day after day for 8 weeks (**Mohan *et al*., 2007).**

iv. **Carbon tetrachloride intoxicated group(CCl_4):**Animals were received CCl_4 dissolved in olive oil (V/V) at a dose (2.5ml /kg body wt) orally day after day for 8 weeks***(Fischer-Nielsen et al., 1991).***

v. ***Ficus Carica* extract+ CCl_4intoxicatedgroup**: Animals were received CCl_4 dissolved in olive oil (V/V) at a dose (2.5ml /kg body wt) orally day after day for 8 weeks that previously administered *Ficus carica* leaf methanolic extract at dose (500mg/kg) orally (**Mohan *et al* ., 2007).**

1.2.2. Plant Extract:

Leaves of Ficus carica were dried in shadow at room temperature for 15 days. The dry materials were ground in electric grinder. Dried leaf powder (200 g) was extracted with methanol by maceration in absolute methanol with stirring. After ten days the extract was filtered and concentration. After ten days the sample was filtered with filter paper. The filtrate was then transferred and concentrated which gave a greenish oily crude methanol extract of 16.6 g (**Trease and Evan 1983)**.

2. Methods

2.1. Sampling:

At the end of the experimental period (8 weeks), overnight fasted rats were sacrificed using a sharp razor blade. Two blood samples from each rat were collected into clean centrifuge tube; the first was collected on EDTA for hematological studies, while the second was allowed to clot, then centrifuge at 3000r.p.m for 20 min for biochemical analysis. Blood sera were carefully separated and were kept at - 20◦C for subsequent analysis future estimations. The rats were sacrificed 24 h after the last treatment and the liver of each rat was removed and weighed then frozen for biochemical analysis.

2.2. Preparation of Liver homogenate:

A tissue sample from a known portion of the liver was accurately weighed and homogenized in distilled water to form 10 % (w/v) homogenate, after labeling the samples, there were kept at -20^{o}C.

2.3. Biochemical studies:

Liver index calculate by divide liver weight and body weight then multiply 100 Liver body weight ratio = liver weight / body weight × 100.

AST , ALT , ALP, γ-GT and LDH activities in serum and liver were estimated according to the method **Reitman and Frankel***, (1957)*, **Belfield and Goldberg (1971),Persijn and van der Silk (1976), and Koh and Choi (1987)** , respectively.

T.Bil , T.P ,Alb , T.Chol and TG were estimated by a colorimetric method of **Walter and Gerade (1970) ,Henry (1964),***Doumas et al. (***1971***),Kim and Goldnerg***(1969) , Fassati and prencipe (1982)** respectively . The concentration of **(HDL-C)** in serum was estimated by the method of ***(Grove, 1979)***, **(LDL-C)** in serum was calculated from the total cholesterol concentration (TC), the HDL-cholesterol concentration (HDL-c) and triglycerides concentration (TG) according to the method described by ***Friedewald et al., (1972).***Serum very low-density lipoprotein **(VLDL-C)** level was calculated according to the following equation: VLDL-C = TG/5, as described by **Satheesh and Pari (2008)** Where, 5 is a calculation factor**.**

MDA, TAC, GSH, GSH-PX, SOD and **CAT** were estimated by **methods** of **Ohkawa *et al.*, (1982),Koracevic *et al*. (2001)**,**Beutler*et al.*, (1963).**.,**Paglia and Valentine(1967),Nishikimi*etal.*(1972**),and **Aebi (1984); Fossati*et al.* (1980).** respectively.

2.4. Haematological studies:

Hematological parameters including the count of red blood corpuscles (RBCs), white blood cells (WBCs), haemoglobin (Hb) content, platelets count and haematocrit percent were conducted using hematological analyzer (Sysnex Ts-21) Japan **(Dacie and Lewis, 1999).**

2.5. Statistical analysis

Results were expressed as means ± SE. Statistical significance was calculated using one- way analysis of variance (ANOVA) followed by post comparison was carried out with LSD test and tukey test using SPSS program (Statistical Package for Social Science). All the statistical analysis was carried out with the use of SPSS 17.00 software. Differences were considered significant at P ≤0.05 **(Sendecor and Cochran,1980).**

Results

Table (1): Body weight, liver weight and liver index of control and different treated animal groups:

Animal groups	Control	Olive Oil	Ficus. Ext	CCl_4	Ficus Ext. + CCl_4
Initial body weight (g)	110.33 ± 4.04	120.50 ± 2.07 (9.21 %)*	109.33 ± 5.36 (-0.91%)*	112.6 ± 3.71 (2.11%)*	119.17 ± 4.91 (8.01%)* (5.8%)**
Final body weight (g)	237 ± 10.43	194.58 ± 2.70 (-17.9%)*	203 ± 7.72 (-14.38%)*	155.5[a] ± 5.50 (-34.41%)*	175[a] ± 4.85 (-26.19%)* (+12.54%)**
Weight gain (g)	126.75 ± 10.311	74.08 ± 3.09 (-41.55)*	93.6 ± 10.61 (-26.10)*	42.83[a] ± 5.89 (-66.21%)*	55.83[a,b] ± 4.46 (-55.9%)* (+30.35%)**
Liver weight (g)	5.66 ± 0.16	5.88 ± 0.37 (+3.82%)*	5.85 ± 0.19 (+3.24%)*	8.11 ± 0.43 (+43.24%)*	6.2 ± 0.26 (+10.88)* (-22.59)**
Liver index(g)	2.42 ± 0.14	3.04 ± 0.22 (+25.66%)*	2.89 ± 0.06 (+19.52%)*	5.29[a] ± 0.44 (+118.90%)*	3.61[a,b] ±0.20 (+49.24%)* (-31.82%)**

Values are presented as means ±SE. (n=6 rats per each group).

P<0.05, a = significance as compared with control, b = significance as compared with CCl_4 group.

Table (1) illustrate body weights, liver weight and liver index (liver / body weight ratio) in the control and different treated animal groups. Data showed significant decrease in body weight of CCl_4 –intoxicated rat group and significant increase in liver weight as well as, liver index as compared with the control group. On the other hand, a significant decrease in liver weight and liver index was observed in Ficus extract administered as a protective agent for CCl_4-intoxication group, compared with the CCl_4-intoxicated group but, a significant increase in liver weight and liver index was detected when compared with control group.

Table (2): Serum and hepatic biochemical parameters in the control and different treated animal groups.

Parameters	Animal Groups				
	Control	Olive oil	Ficus. Ext	CCl_4	Ficus.Ext+CCl_4
S.ALT (U/L)	10.50±0.43	9.83 ±0.31	10.00 ±0.45	32.83^{a} ±2.82	$19.50^{a,b}$ ±0.41
S.AST (U/L)	19.50±0.99	18.33±0.99	18.67±1.05	58.67^{a}±5.19	31.00^{ab}±1.29
S.ALP (IU/L)	113.80±9.24	123.40±8.92	136.00±6.55	197.20^{a}±8.76	166.80^{ab}±10.59
S.LDH (U/L)	217.69±14.52	219.62 ±10.01	228.27±53.27	390.39^{a}±34.43	$262.74^{a,b}$±23.88
S. γ GT (U/L)	13.17 ±1.32	13.40 ±1.38	12.47 ±0.69	26.64^{a}±0.69	18.55^{b} ±1.55
S .total Bilirubin (mg/dl)	0.53 ±0.03	0.49 ±0.06	0.47 ±0.05	0.71^{a} ±0.05	0.56^{b} ±0.04
S .total Protein (g/dl)	6.12 ±0.09	5.97 ±0.10	6.40 ±0.06	3.38^{a} ±0.17	5.95^{b} ±0.04
S .total Albumin(g/dl)	4.03 ±0.08	3.97 ±0.06	4.07 ±0.06	1.68^{a} ±0.13	3.92^{b} ±0.05
S .total Globulin (g/dl)	2.08 ±0.05	2.00 ±0.04	2.17 ±0.16	1.70^{a}±0.18	2.03^{b} ±0.02
A/G ratio	1.94 ±0.06	1.99 ±0.02	1.95 ±0.21	1.08^{a} ±0.18	1.93^{b} ±0.04
H.ALT (U/g)	58.17±2.09	51.50±2.57	56±2.19	101.67±6.92	77.80±3.56
H.AST (U/g)	82.17±1.40	60.17 ±2.08	62.20 ±2.24	119.50±8.85	85.50^{ab} ±2.69
H. total protein (g/g wet tissue)	4.97 ±0.34	4.87 ±0.11	4.93 ±0.24	4.12^{a} ±0.16	4.75 ±0.22

Values are presented as means ±SE. (n=6 rats per each group).
P<0.05, a = significance as compared with control b = significance as compared with CCl_4 group.

Data on liver function were observed in table (2): Serum levels of ALT, AST, ALP, γ-GT, were significantly increased in CCl_4-intoxicated animal group compared to the control animals. Similar results were obtained in protein, albumin, globulin contents as well as liver -body weight ratio. Liver tissue levels of transaminases was significantly increased in animal treated with CCl_4 compared to the control rats. On the other hand, oral administration of Ficus extract (500mg /kg) for 8 weeks as a protective agent notably diminished CCl_4-induced damage that observed significant reducing levels of all the investigated parameters. The extract also restored protein, albumin and globulin levels compared to the normal rats.

Table (3): Hematological parameters in the control and different treated animal groups

Parameters	Animal Groups				
	Control	Olive oil	Ficus. Ext	CCl_4	Ficus.Ext+CCl_4
RBCS (10^6/mm^3)	5.61±0.95	6.80±0.29	6.11±0.50	2.17^{a}±0.82	5.91^{b}±0.94
WBCS (10^3/mm^3)	5.56±0.98	7.01±0.78	6.36±0.65	9.16^{a}±1.12	8.27±1.48
Platelets count (10^3/mm^3)	718.60±52.72	570.53±64.33	605.60±60.90	403.20^{a}±50.21	$565.60^{a,b}$±43.44
Hb content (g/dl)	12.34±0.40	11.68±0.54	13.06±0.25	10.36^{a}±0.28	11.42^{b}±0.77

Values are presented as means ±SE. (n=6 rats per each group). P<0.05,
a = significance as compared with control, b = significance as compared with CCl_4 group.

The effect of CCl_4-intoxication and the protective effect of *Ficus carica* leaves extracts on hematological parameters of the rats are showed in table (3):There was a significant reduction in Hb concentration, RBCs, platelets count, while WBCs, was higher in the rats that were exposed to CCl_4 without pretreatment with *Ficus* extracts where compared to the normal control group. However there was a moderate increase in these parameters in the rats that were pretreated with the extract except WBCs count that observed a significant decrease in number.

Table (4): Lipid profile in control and different treated animal groups.

Parameters	Animal Groups				
	Control	Olive oil	Ficus. Ext	CCl_4	Ficus.Ext+CCl_4
S Total Cholesterol(mg/dl)	107.00 ±2.39	107.33 ±2.60	102.50 ±2.00	120.50^{a} ±1.88	112.00^{b} ±1.24
S. Triglycerides(mg/dl)	85.33 ±2.54	86.50±2.26	85.17±2.80	105.67^{a}±3.27	97.17^{ab}±3.44
S.HDL- Cholesterol (mg/dl)	35.50±1.39	33.67±0.95	34.00±1.73	24.33^{a}±1.63	$31.00^{a,b}$ ±0.77
S.LDL-Cholesterol (mg/dl)	54.43±2.95	56.37±1.83	53.13±1.65	75.03^{a}±3.04	$61.57^{a,b}$±2.27
S.VLDL-Cholesterol(mg/dl)	17.07±0.51	17.30±0.45	17.03±0.56	21.13^{a}±0.65	$19.43^{a,b}$±0.69
H. Total Cholesterol(mg/g)	100.67 ±3.90	107.17 ±3.87	105.60 ±3.41	115.92^{a} ±3.77	108.00^{b} ±2.57
H. Triglycerides(mg/g)	287.83±13.94	273.17±16.35	286.83±7.87	332.67±17.18	288.67±6.32

Values are presented as means ±SE. (n=6 rats per each group). P<0.05, a = significance as compared with control, b = significance as compared with CCl_4 group. Table (4): showed the effect of *Ficus* extract on lipid profile in control and different treated animal groups.CCl_4-intoxicated group showed a significant elevation in level of hepatic total cholesterol and triglycerides. *Ficus* supplementation resulted in a significant decrease compared to normal control rats. On the other hand, serum total cholesterol, triglycerides, HDL cholesterol, showed significant increase in CCl_4-intoxicated rats but S. HDL cholesterol observed increase in the same group. The administration of *Ficus* extract as a protective agent against CCl_4-induced group restored the levels of these parameters.

Table (5): Malondialdehyde (MDA) levels and Antioxidant parameters in control and different treated animal groups

Parameters	Animal Groups				
	Control	Olive oil	Ficus. Ext	CCl_4	Ficus.Ext+CCl_4
MDA(n mol/g Tissue)	931.23±203.65	1057.95±193.93	622.91±67.26	2591.28^{a}±329.13	1887.11^{ab} ±294.27
Total antioxidant capacity(m M/g)	1.49 ±0.00	1.46 ±0.01	1.59^{a} ±0.07	1.14^{a} ±0.10	1.48 ±0.01
GSH (mg/g tissue)	156.20±8.33	165.09±7.99	263.18±9.07	83.99^{a}±9.30	125.07^{ab}±13.58
GPX (U/g tissue)	833.28±14.47	851.24±13.53	869.14±10.55	680.21^{a}±15.10	$743.70^{a,b}$±27.30
Catalase (U/g tissue)	130.00 ±13.42	115.17±11.55	160.00±5.16	73.33^{a}±8.43	126.67^{b}±15.20
SOD (U/g tissue)	188.33 ±13.52	181.83±17.51	208.67±22.28	100.50^{a}±19.29	$160.00^{a,b}$±7.30

Values are presented as means ±SE. (n=6 rats per each group). P<0.05,
a = significance as compared with control, b = significance as compared with CCl_4 group.

Table (5): showed the effect of *Ficus* leaves methanolic extract on oxidative stress marker and antioxidants in control and different treated animal groups .Among the CCl_4-intoxicated group a significant increase in MDA levels in liver suggest enhanced lipid peroxidation leading to tissue damage as well as decreasing the antioxidants. On the other hand, supplementation of Ficus extract resulted in significant decrease in the level of hepatic MDA, where as it resulted in higher GSH, GPX, CAT and SOD levels in the liver compared to the normal control group.

DISCUSSION

Herbal medicine is progressively achievement acceptance from the public and medical professionals due to advances in the understanding of the mechanisms by which herbs positively influence health and quality of life **(Panda and Naik , 2009).**The pathogenesis of liver diseases as well as the role of oxidative stress and inflammation there is well recognized **(Tacke, et al., 2009),** and consequently, blocking or retarding the chain reactions of oxidation and inflammation development could be promising therapeutic strategies for prevention and treatment of liver damage.

Hepatoprotective results showed that plants have active ingredients that are capable of free radical scavenging in living system (**Nikolova *et al* ., 2011 ; Kumar ., 2012).** Phenolic compound are promising bioactive secondary metabolites playing an important role in detoxification of free radicals **(Shanmugas undaram *et al* ., 2006; Chon *et al* ., 2008).**

Antioxidative action plays an important role in protection against CCl_4- induced liver injury **(Xiong *et al*., 1998).**There is increasing evidence for the hepatoprotective role of hydroxyl- and polyhydroxy organic compounds, particularly from vegetables, fruits and herbs (**Bass, 1999)**.

Rats intoxicated with CCl_4 developed a significant liver damage and showed elevated serum levels of hepato-specific enzymes as well as severe alteration in other biochemical parameters, oxidative damage and histological changes. Values of the biochemical parameters were observed to be increased in the CCl_4 intoxicated rats. Liver damage induced by CCl_4 is commonly used model for the screening of hepatoprotective drugs **(Slater, 1965)**. CCl4 is known to undergo reductive metabolism by CYP2E1 into a highly reactive trichloromethyl radical (.CCl3) that initiates lipid peroxidation, disrupts membrane integrity and causes cell death (**Pohl *et al.*, 1984; Fadhel and Amran, 2002; Basu, 2003)**. Literature shows that oxidative stress has been implicated in the aetiopathogenesis of CCl_4 organ injury and carcinogenesis (**Slater, 1984; Stal and Olston, 2000).** However, when the oxidative stress is overwhelming, the various inherent defense mechanisms (such as the antioxidant defense mechanisms, intracellular concentration of glutathione, superoxide dismutase (SOD) and catalase (CAT) activities become significantly impaired and insufficient (**Szymonik-Lesiuk *et al.*, 2003).**

In the present experimental conditions, the methanolic leaves extract of Ficus carica, administered prophylactically at a dose 500mg/kg b.w for 8 wks. It was found to be practically non-toxic since no mortality was observed, the Ficus extract exhibited significant protection against CCl4-induced liver injury as manifested by the reduction in toxin mediated rise in serum transaminases, ALP and total bilirubin in rats.

Liver /body weight ratio:

The results of the present study showed that CCl_4 caused an increase in liver weight and a decrease in body weight. So it is clear that, the weights ratios (LW/BW) significantly increased in rats treated with CCl_4, also it could be noticed that the toxicity of CCl_4 is related to loss in weight and in-crease in liver weight in rats. These results go in parallel with **Honma (1990)** who **observed** that liver to body weight ratio and liver weight were raised with increase doses of CCl_4, furthermore the liver enlargement appeared to be a sensitive marker of hepatotoxicity related to the changes in lipoproteins. Also, the increase in body weight after drug administration usually indicate to absence of toxicity , but decrease in body weight is an index of toxic effect of a compound or drug (**Kiran *et al* ., 2012).** On the other hand weights ratios (LW/BW) reversed significantly in rats treated with ficus extract prior the CCl_4-intoxication. These results are agreed with **Rajkapoor *et al* .(2002)** who Found that the liver was pale reddish brown and enlarged in CCl_4-intoxicated rats and normal in other groups , Also Chronic administration of CCl_4 caused a significant increase in relative liver weight this may be related to liver triglycerides accumulation as reported by **Ohta *et al.* (1997)** .

Hematological parameters:

In the present study, oral administration of CCl_4 greatly affected all hematological parameters, as decrease in RBCs count, Hb content, and platelets count, these data go in parallel with the results observed by (**Mandal *etal.*, 1998)**. The depression in RBCs count and Hb content recorded in the present work could be attributed to disturbed hematopoiesis, destruction of erythrocytes, and reduction in the rate of their formation and/or their enhanced

removal from circulation **(Tung *et al.*, 1975)**. Moreover, administration of CCl_4 induced macrocytic, hypochromic anemia as CCl4 caused a significant increase lipid peroxidation, degradation of membrane proteins, alteration of membrane-bound enzymes as well as erythrocyte osmotic fragility (**Makni *et al.*, 2012)**. The destruction of red cells may be supposed that the free radicals resulting from CCl4 metabolism caused liver injury resulting in free radicals liberation from the liver into the blood **(Sherlock and Dooley, 1993).**

The protective effect of *Ficus* extract administered as protective agent for CCl_4 intoxicated group may be due to its role to decrease the production of reactive oxygen species **(Perez-Garcia *et al.*, 2000)**, the oxidation of low-density, lipid peroxidation and protein oxidation **(Jimenez-Escrig *et al.*, 2003).**However, *Ficus* elevated the number of these parameters compared with CCl_4 intoxication group including the platelets which suggest that the extract has anti- anemic property which may be due to its high iron content **(Saliu, et al., 2012)** and the ability to improve bone marrow functions, a major site for erythropoiesis **(Orhue, et al., 2008).**

The present results showed that CCl_4- intoxicated rats significantly increase the level of WBC as compared to normal. Our findings are in agreement with that reported **by Sule *et al*. 2012 and Elshater *et al* 2013**, who found that the administration of CCL4 to rats led to significant decrease of RBC counts and Hb level and significant increase of WBC counts in respect to normal control. Also, the abnormal hematologic parameters caused by CCl4 were decreased by *Ficus* extract administered as protective agent for CCl_4 intoxicated group may be due to the role of flavanoids one of its active components, are known to be vasculo-protector and powerful antioxidant **(Sule *et al*., 2012) ,**as well as the flavanoids probability did so by reducing the accumulation of toxic CCl4 derived metabolites**(Mada *et al* ., 2014)**. Blood parameters were found to be positively affected by using *ficus extract* as a therapeutic agent.

In the present study, it was found that CCl_4-intoxication for 8 weeks has a significant effect on the alteration of liver function, since the activities of serum and hepatic AST, ALT and ALP were significantly increase than those of normal value. These views are in agreement with those of **Zhao *et al*. (2002) ,Wang *et al.* (2009), Motawi *et al* .(2011)**they observed high activities of AST &ALT and ALP after CCl_4-intoxication. Leakage of large quantities of enzymes into the blood stream is often associated with massive fibrosis of the liver***(Shymal et al., 2006).***Higher level of marker enzymes notice that CCl_4 induced liver dysfunction and denotes damage to the hepatic cells (**Pari and Murgavel, 2004 and Ohta *et al*., 2005*).***This elevation in serum hepatic enzymes indicated deterioration in hepatic function due to parenchymal injury after CCl_4intoxication. Elevation may be explained by the basis of increase in hepatic cell membrane fluidity and permeability that lead to enzymes release into circulation **(El-Hawary *et al.*, 2011).** The rise in serum levels of AST, ALT, ALP and LDH has been attributed to the damaged structural integrity of the liver, because they are cytoplasmic in location and released into circulation after cellular damages **(Sallie, *et al*., 1991)**.

Lactate dehydrogenase is a well-known marker for cell toxicity. When cells are damaged due to oxidative stress, the membrane becomes permeable or may rupture which results in the leakage of this enzyme. This enzyme enters into the blood stream thus increasing its concentration in the serum **(Ganie, *et al*., 2011)**. In the present study, LDH leakage is significantly increased in the serum of rats exposed to CCl_4. *Ficus carica* extract administration to significantly decreased LDH level in serum compared to rats treated with CCl_4 alone. Further, the results of our study showed that coadministration of *Ficus carica* extract diminished CCl_4-induced oxidative stress by increasing antioxidant status. In addition, our result indicated that *Ficus carica* extract administration significantly alleviated the increased serum enzymes induced by CCl_4 indicating improvement of the functional status of the liver. Also, *Ficus carica* leaves highly antioxidative characters, which reduce the leakage of these enzymes in blood, this might be due to accelerated regeneration of parenchymal cells that help to prevent membrane fragility and subsequently decrease the leakage of marker enzymes into circulation.

The present results demonstrated also that there was a significant decrease in the serum and hepatic total protein content after CCl_4- intoxication for 8 weeks as compared to control group. This result agrees with the result of **Ohta *et al. (*1999 *)***who reported that the concentration of serum albumin and hepatic protein synthesis decrease post treatment with CCl_4 for 8 weeks. Similarly, impairment of protein synthesis in CCl_4-intoxicated rats was reported by other investigators **(Lamb *et al*., 1984 and Pappas *et al*., 1994).** It was thought that the inhibition of protein synthesis may be involved in cell injury or death mediated by free radicals ***(*Halliwall and Gutteridge, 1984).** CCl_4 intoxication leads to damage of Golgi apparatus damage this in turn adversely affects packaging and release of protein from the hepatocytes. In the present study, co administration of *Ficus* extract with CCl_4 protected the intoxicated animals against hepatotoxic effect of CCl_4 as proved by a significant elevation in serum and liver total protein as well as serum albumin contents when compared to rats intoxicated with CCl_4.

Albumin synthesis is extremely sensitive to CCl_4 probably secondary to the alterations in the cytoplasmic protein-synthesizing system **(Rui *et al.*, 2012)**. The liver loss its ability to synthesize albumin, as albumin is

produced on a polysome bound to the endoplasmic reticulum **(Kaneko, 2008)**. The significant depression of serum albumin levels due to CCl_4 intoxication was possibly associated with the apparent dissociation of albumin protein into smaller subunits (**Folmar *et al.*, 1993).**

The present study demonstrated a significant elevation in oxidative stress markers causing in a cell, tissue, or organ damage can affect a specific molecule such as proteins, lipids and DNA. In the current study, rats that treated with CCl_4 showed a significant increase in malondialdehyde (MDA), the end product of lipid peroxidation levels in liver. Consistent evidences supported the hypothesis that progressive accumulation of oxidative damage to the important cellular molecules is a fundamental mechanism involved in most cellular alterations.

The present results also run in parallel with the findings of **Fruehauf and Meyskens (2007*)*** who reported that MDA appears at significant higher concentrations in the liver and alters antioxidant levels .According to the present data, hepatic GSH content as well as GST activity were significantly decreased by CCl_4-intoxication .These results run in parallel with that of **Cabre *et al. (*2000*)*** they reported that induction of cirrhosis by using CCl_4 produces a decrease in the components of hepatic glutathione antioxidant system. Generation of reactive oxygen species via LPO is one of the mechanisms involved in tissue damage. The detoxication pathway involves GSH depletion by conjugation with trichloromethyl-free radicals (**Biosset *et al.*, 2004*).*** Previous studies was mentioned that mechanism of CCl_4-induced hepatotoxicity have shown that GSH plays a key role in eliminating the reactive toxic metabolites of CCl_4. Liver fibrosis commences when the GSH stores are substantially depleted and this depletion of mitochondrial GSH is a crucial determinant for cell survival or death in oxidative stress conditions (**Hidaka *et al.*, 2007*).***

This decrease might be due to the increased utilization of the hepatocytes in scavenging toxic radical of CCl_4. It has been reported that most covalent binding of toxicant to hepatic protein occurs only after depletion of GSH, and the severity of hepatic necrosis was related to the degree of covalent binging (**Jollow *et al.*, 1973*).***

In addition the antioxidant activity and/or the inhibition of free radical generation is one of the important steps in terms of protecting the liver from CCl_4-induced damage ***(*Borek, 2001 and Manibusan et al., 2007).*** Generally, antioxidant enzymes such as catalase, SOD and GST were easily inactivated by lipid peroxides or reactive oxygen species, as observed in decreased activities of these enzymes in CCl_4 toxicity. Free radicals in CCl_4 intoxication are a major pathway of non-enzymatically induced lipid peroxidation, which subsequently affect various enzyme activities in the body and therefore may also be linked to enzymatically induced lipid peroxidation (**Basu, 2003*).***

Administration of Ficus extract inhibited LPO as an effective chain breaking antioxidant and helped in preventing CCl_4 -induced peroxidative damage. Also, the present results indicated that the treatment with Ficus extract cause an increase in the activity of antioxidant enzymes.

On the other hand, the present data showed that Ficus extract administered as a protective agent for CCl_4 -intoxication, prior to CCl_4 exposure for 8 weeks caused a significant protection observed by reduction in MDA when compared with the high significant elevation of these in CCl_4intoxicated rats. The rise in lipid peroxides in liver tissue homogenate was prevented significantly. The decrease in lipid peroxides may be due to the antioxidant effect of the extract. A possible mechanism of the *Ficus carica* extract as hepatoprotective may be due to its anti-oxidant effect or inhibition of cytochrome P450s which impair the bioactivation of CCl_4 **(Recknagel *et al* ., 1989)** into their corresponding reactive species.

The hepatoprotective expected mechanism of the effect of *Ficus carica* leaf, against CCl_4-induced liver damage, is the ability of its flavonol glycoside content to act as strong free radical scavengers intercepting those radicals involved in CCl_4 metabolism by microsomal enzymes. Consequently, the flavonol glycosides of *Ficus* extract could hinder the free radical interaction with polyunsaturated fatty acids and would abolish the enhancement of the lipid peroxidative process leading to hepatic cell damage. The present results agree with reported data **(Hewawasam *et al*., 2004; Orhan *et al.*, 2007)**.

In order to verify this finding the antioxidant activity of *Ficus* extract Leaves was determined the significant amelioration in the antioxidant parameters might result in the hepatoprotective action of flavonol glycosides against oxidative stress induced by CCl_4. Consequently, the flavonol glycosides content of *F. carica* leaf are responsible for the abolishment of CCl4-induced hepatic damage through their strong antioxidant activity. Also, **Pukkumani *et al*., (2004)** reported that ferulic acid (FA) is an efficient hepatoprotective agent against alcohol and heated polyunsaturated fatty acid-induced liver damage. **El-Shobaki *etal*., (2010)** reported that ferulic acid a most

abundant natural phenolic compound in Ficus carica leaves.Ferulic acid is an effective scavenger of free radicals exerts its protective effect by modulating lipid peroxidation and augmenting antioxidant defense system in tissues (**Sudheer *et al*., 2007)**.

Serum level of γ-GT has been employed for the measurement of hepatic damage. Elevation of γ -GT level might occur due to CCl_4 induced physiological imbalance in liver. These results run in parallel with the study of **Lee, (2004),** who reported that, increased γ-GT activity may be a response to oxidative stress, one which can increase the transport of glutathione precursors into cell. In the present experiment, *Ficus* extract inhibited the level of γ-GT activity confirming the preventing CCl_4 induced peroxidative damage.Also, the present results indicated that the treatment with *Ficus* extract cause an increase in the activity of antioxidant enzymes.

The results of the present study showed that administration of CCl_4 to the normal rats induced a significant disturbance in the various lipid components. These findings are in agreement with the observation of **Dwived *et al*. (1990**) who reported that CCl_4 causes a significant increase in the hepatic lipids. The inhibition of protein synthesis and nucleic acid hypomethylation after CCl_4 exposure may play a role in the impairment of lipid components (**Weber *et al*., 2003*).***According to the present results, significant increase in serum total cholesterol was accompanied with accumulation of total cholesterol in the liver. This observation may be due to the oxidative modification of LDL which is the major carrier of cholesterol in the blood **(Aviram, 1996).**

On the other hand, the present data showed also a significant decrease in the serum triglycerides after CCl_4-exposure for 8 weeks. These results run in parallel with **Ohta *et al*., (2000*)***who reported that rats injected with CCl_4-show a decrease in serum (TG) concentration .Since, CCl_4 is metabolized in liver to yield highly reactive trichloromethyl peroxyl radical which can react with phospholipids of hepatocyte membrane therefore, a chain of extensive intrahepatic destructive peroxidation reactions is initiated (**Yamammato, 1990** and **Weber *et al*., 2003*).***Moreover, the present data showed that, CCl_4-intoxication causes a significant decrease in HDL associated cholesterol (HDL-cholesterol), while, a significant increase in low-density lipoprotein (LDL-cholesterol) is observed. This might be supported with the findings of **Bauer *et al*. (1990)** and **Gergely *et al*. (1995)** who found an increase in very low density lipoprotein cholesterol concentration and decrease in high-density lipoprotein cholesterol concentration after hepatocellular damage using a nonlethal dose of CCl_4. This may be attributed to the covalent binding of CCl_4 metabolites, $CCl_3^{\bullet}$ and $CCl_3OO^{\bullet}$, to cell constituents .In addition, the inhibition of phospholipids (**Weber *et al*., 2003)** as well as increased activities of AST and ALT ***(*Honma, 1990*)***played a role in the disturbances of lipoprotein secretion. In the present experimental conditions, the methanolic extract of *Ficus carica* leaves, administered prophylactically, exhibited significant amelioration of lipid profile.

In this respect **Asadi *et al*., (2006)** reported that the extract of Ficus carica leaves, could be used to decrease hepatic Triglycerides content and secretion of Triglycerides and Cholesterol from the liver. Also, **Lee *et al*., (2004)** indicated that Cinnamic acid related flavanoids are present in Ficus carica leaves. A series of Cinnamic acid derivatives possess useful biological activity as an antiatherosclerotic agent. Many study showed that total phenolics were important antioxidants in *Ficus carica* extract. Moreover, these findings supported the beneficial effect of *Ficus carica* in maintaining the hepatocytes integrity and function. It is conceivable that these effects may be due, at least in part, to its antioxidant activity. The antioxidant enzyme (SOD, CAT, and GPx) activities were increased by *Ficus* extract in liver tissues when compared with those administered with ccl_4 only. In another study, **Patil *et al* (2011)** also found that *Ficus carica* extract possess anti-inflammatory effects. In another study, *Ficus carica* fruit ethanolic extract was also shown to have anti-inflammatory effects (**Patil *et al*., 2011 Koka, *et al*., 2013).**

In conclusion, CCl_4- intoxication developed liver damage and showed an elevated serum levels of hepato-specific enzymes which lead to severe alteration in biochemical parameters and oxidative stress markers. In the present experimental conditions, the *Ficus* extract of administered prophylactically, exhibited significant protection against liver injury as evidenced by remarkable amelioration in all investigated parameter of serum and liver. Presence of flavonoids, triterpenoids and steroids has protective effect on liver due to its antioxidant properties. *Ficus* extract may be responsible for the protective effect on CCl_4 induced liver damage in rats.

Pre-treatment of the animals with *Ficus* extract improve the deleterious effect that previously produced with experimental liver damage and indicate to their potential role in improving the quality of life of hepatic patients.

REFERENCES

Aebi, H. (1984): Colorimetric method for determination of catalase. Methods,Enzymol.,105:121-126.

Asadi ,F.; Pourkabir, M.; Maclaren, R.; and , Shahriari,A.; (2006): Alterations to lipid parameters in response to fig tree (*Ficuscarica*) leaf extract in chicken liver slices. Turkish Journal of Veterinary and Animal Sciences., (30) 3: 315-318.

Aviram, M.(1996): Oxidized low density lipoprotein (OX-LDL) interrelation with macrophage in artherosclerosis and antiatherogenicity of antioxidants. European J. Clin. Chem. Biochem., 34: 599-608.

Bass ,N.M.; (1999): Is there any use for nontraditional or alternative ther -apies in patients with chronic liver disease? Curr Gastroenterol Rep1: 50–56.

Basu ,S.;(2003): Carbon tetrachloride-induced lipid peroxidation: eicosanoid formation and their regulation by antioxidant nutrients. Toxicology. 189: 113 - 127.

Basu S, 2003. Carbon tetrachloride-induced lipid peroxidation: eicosanoid formation and their regulation by antioxidant nutrients. Toxicology 189: 113 - 127.

Bauer, J.E.; Meyer, D. J.; Campbell, M. and McMurphy, R. (1990): Serum lipid and lipoprotein changes in ponies with experimentally induced liver disease. Am. J. Vet. Res.,51(9):1380-1384.

Belfield, A. and Goldberg, D.M. (1971): Normal ranges and diagnostic value of serum 5, nucleotidase and alkaline phosphatase activities in infancy. Arch. Dis. Child, 46:842 –846**.**

Beutler, E.; Duron, O. and Kelly, M. B. (1963): Colorimetric method for determination of GSH. J. Lab. Clin. Med., 61: 882.

Borek, C. (2001): Antioxidant health effects of aged garlic extract. J. Nutr., 131: 10105-10155.

Cabre, M.;Camps, J.; Paternain, J. L.; Ferre, N.; Joven, J. (2000): Time-course of changes in hepatic lipid peroxidation and glutathione metabolism in rats with carbon tetrachloride-induced cirrhosis. Clinical and Experimental. Pharmacol. and Physiol., 27: 694–699.

Cai, Y.; Luo, Q.; Sun, M.; Corke, H.; (2004): Antioxidant activity and phenolic compounds of 112 Chinese medicinal plants associated with anticancer. Life Sci., 74, 2157–2184.

Chon, S.U, Heo ,B.G.; Park, Y.S.; Cho, J.Y., Gorinstein ,S.;(2008):Characteristics of the leaf parts of some traditional Korean salad plants used for food. J Sci Food Agric ., 88: 1963-1968.

Dacie, J. V. and Lewis, S. M. (1991):Practical Haematology. Churchill Livingstone. Edinburgh. Seventh, edition,Pp 521-534.

dehydrogenase efflux assay. J of neuroscience methods, 20, 83-90.

Doumas, B.T.; Watson, W. and Biggs, H. (1971): Albumin standards and the measurements of serum albumin with bromocresol green. Clin. Chem. Acta., 31: 87-96.

Dwived, Y.; Rastogi, R.; Chonder, R. and Dhawan, I.V. (1990): Hepatoprotective activity of picroliv against carbon tetrachloride induced liver damage in rats. Indian J. Med. Res., 92: 195-200.

El-Hawary, S.A.;Sokkar, N.M.;Ali, Z.Y.;Yehia ,M.M.;(2012): A profile of bioactive compounds of RumexvesicariusL. J Food Sci 76: C1195-C1202.

Elshater, A.A.,Salman, M.M.A.;Moahmed ,S.;(2013): The hepato- ameliorating effect ofSolanum nigrumagainstCCL4induced liver toxicity in Albino rats. Egypt Acad J Biolog Sci ., 5(10): 59-66.

El-Shobaki, F.A.; El-Bahay, A.M, Ismail, R.S.A.; Abd El-Megeid, A.A. and Esmail, N.S. (2010): Effect of Figs fruit (Ficus Carica L.) and its leaves on hyperglycemia in alloxan diabetic rats. World J of Dairy & Food sciences., 5 (1): 47 –57.

Fadhel, Z.A.; Amran, S.;(2002): Effects of black tea extract on tetrachloride-induced lipid peroxidation in liver, kidneys and testes of rats. Phytotherapy Research, 16(Supp 1): S28 – S32.

Fassati, P. and Prencipe, L. (1982): Serum triglycerides determined colorimetricallywithanenzymethatproduceshydrogenperoxide.Clin.Chem.,28: 2077-2080.

Fischer-Nielsen, A.; Poulsen, H. E.; Hansen, B. A.; Hage, E. and Keiding, S. (1991): J. hepatol.,12: 112-117.

Folmar, L., Bonomelli, S.; Moody ,T.; and Gibson, J. ;(1993): The effect of short-term exposure to three chemicals on the blood chemistry of the pinfish (Lagodon rhomboides). Arch Environ Contam Toxicol., 24(1):83–86.

Fossati,P.;Prencipe,L. andBerti,G. (1980): Use of 3,5 dichloro-2-hydroxybenzenesulfonic acid/4-aminophenazone chromogenic system in direct enzymic assay of uric acid in serum and urine. Clin.Chem., 26:227-231.

Frank, C; Mohamed, M.K, Strickland, G.T; Lavanchy, D; Arthur R.R.; And Magder, L.S. et al (2000): The role of parenteral antischistosomal therapy in the spread of hepatitis C virus in Egypt**.** Lancet.,355: 887–91**.**

Friedewald, W.T.; Levy, R.I. and Fredrickson, D.S. (1972): Estimation of the concentration of low-density lipoprotein cholesterol in plasma, without use of the preparative ultracentrifuge. Clin. Chem., 18: 499-502.

Fruehauf, J.P.; Meysrkens, F.L. (2007): Reactive oxygen species: a breath of life or death? Clin. Cancer Res**.,** 13: 789-794.

Ganie, S. A.; Haq, E. ; Masood,A.; Hamid, A. and Zargar, M. A.;(2011): Antioxidant and protective effect of ethyl acetate extract of podophyllum hexandrum rhizome on carbon tetrachloride induced rat liver injury, Evidence-based Complementary and Alternative Medicine, Article ID 238020.

Gergely, J.; Kulcsar, A. and Harsfalvi, J. (1995): Changes in fat metabolism in acute carbon tetrachloride intoxication of rats. Acta Pharm. Hung., 65(1): 3-4.

Grove, T, (1979): Effect of reagent pH on determination of HDL cholesterol by precipitation with Sodium Phosphotungstate-magnesium. Clin. Chem**.,** 25:560-564.

Halliwell, B. and Gutteridge, J.M. (1984): Free radicals in biology and medicine. Biochem.**,** 219: 1-14.

Henry, R.J. (1964): Clinical Chemistry, Principles and Technics, 2nd Edition, Harper and Row., P.525,1974.

Hewawasam ,R.P.; Jayatilaka ,K.A.;Pathirana ,C.; Mudduwa, L.K.;(2004): Hepatoprotective effect of *Epaltes divaricata* extract on carbon tetrachloride induced hepatotoxicity in mice. Indian J Med Res.120:30.

Hidaka, I.; Hino, K.; Korenaga, M.; Gondo, T.; Nishina, S.; Ando, M.; Okuda, M.; Sakaida, I. (2007): Stronger Neo-Minophagen C, a glycyrrhizin –containing preparation protects liver against carbon tetrachloride- induced oxidative stress in transgenic mice expressing the hepatitis C virus polyprotein. Liver Int., 27: 845-853.

Honma, T. (1990): Effects of trichloroethylene, 1,1,1-trichloroethane and carbon tetrachloride on plasma lipoprotein of rats. Ind. Health, 28(4): 159-174.

Honma, T. (1990): Effects of trichloroethylene, 1,1,1-trichloroethane and carbon tetrachloride on plasma lipoprotein of rats. Ind. Health, 28(4): 159-174.

Jiang, Y.; Liu ,J.; Waalkes, M.; Kang ,Y.J.; (2004): Changes in the gene expres-sion associated with carbon tetrachloride-induced liver fibrosis persist after cessation of dosing in mice. Toxicol Sci.,79: 404–410.

Jimenez-Escrig, A.; Dragsted, L.O.; Daneshvar, B.;Pulido, R.; and Saura-Calixto, F.;(2003): In vitroantioxidant activities of edible artichoke(*Cynara scolymus* L.) and effect on biomarkers

Jollow, D. J.; Thogeirsson, S. S.; Potter, W. Z.; Hashimoto, M. and Mitchell, J. R. (1973): Acetaminophen-induced hepatic necrosis. IV metabolic disposition of toxic and non-toxic doses of acetaminophen. Pharmacol., 12: 251-271.

Kaneko, J.J.; (2008): Hepatic function. In: JJ Kaneko, JW Harvey and ML Bruss (eds), Clinical Biochemistry of Demostic Animals, 6thEd, (EAcedemic Press, London).,356-365.

Kim, E. and Goldnerg, M. (1969): Direct method for cholesterol determination. Clin. Chem., 15**:** 1171-1172.

Kim; Hyo-Yeon; Joon-Ki Kim; Jun-Ho Choi; Joo-Yeon Jung,;Woo-Yong Oh;Dong Chun Kim; Hee Sang Lee; YeongShik Kim; Sam Sik Kang; Seung-Ho Lee; and Sun-Mee Lee.; (2010): Hepatoprotective Effect of Pinoresinol on Carbon Tetrachloride–Induced Hepatic Damage in Mice. J PharmacolSci .,112:105 –112.

Kiran, P.M.; Raju, A.V.; Rao, B.G.;(2012):Investigation of hepatoprotective activity of Cyathea gigantea(Wall. ex. Hook.) leaves against paracetamol-induced hepatotoxicity in rat. Asian Pac J Trop Biomed. 5: 352-356.

Koh, J. Y. and Choi, D. W.(1987): Quantitative determination of glutamate mediated cortical neuronal injury in cell culture by lactate

Koka, S., et al.; (2013): Effect of Ficus carica fruit extract on experimentally induced inflammation and nociception.

Koracevic, D.; Koracevic, G.; Djordjevic, V.; Andrejevic, S. and Cosic, V. (2001): Method for the measurement of antioxidant activity in human fluids. J. Clin. Pathol., 54: 356 –361.

Krishna Mohan.G.; Pallavi.E.; Ravi Kumar.B.;Ramesh.M.; Venkatesh,S.;(2007): Hepatoprotective activity of Ficus carica Linn. leaf extract againstcarbon tetrachloride induced hepatotoxicity inrats.DARU., 15(3): 162-166.

Kumar, S.V.; Sanjeev ,T.;Ajay S,A. ;(2012): Review on hepatoprotective activity of medicinal plant. IJARPB 1: 31-38.

Kumaran, A.; Karunakaran, J.; (2006): In vitroantioxidant activities of methanol extracts of five Phyllanthus speciesfrom India. LWT-Food Sci. Technol., 40, 344-352.

Lamb, R.G.; Mc Cue, S. B.; Taylor, D. R. and Mcguffin, M. A. (1984): The role of phospholipids metabolism in bromobenzene and carbon tetrachloride dependent hepatocyte injury. Toxicol.Appl. Pharmacol.,75: 510-520.

Lattanzio, V.; (2003): Bioactive polyphenols: Their role in quality and storability of fruit and vegetables. Journal of Applied Botany., 77: 128-146.

Lee, D.H., Ha, M.H., Kim, K.Y., Jin, D.G. and Jacobs, D.R. (2004): G-glutamyltransferase, an effect modifier in the association between age and hypertension in 4-year follow-up study. J. Hum. Hypertens. 18.803-807.

Lee, D.H., Ha, M.H., Kim, K.Y., Jin, D.G. and Jacobs, D.R. (2004): G-glutamyltransferase, an effect modifier in the association between age and hypertension in 4-year follow-up study. J. Hum. Hypertens. 18.803-807.

Mada, S. B.; Inuwa, H. M.; Abarshi, M. M.; Mohammed, H. A. and Aliya, A. (2014): Hepatoprotectiveeffect of *Momordica charantia* extract against CCL4 induced liver damage in rats. British Journal ofPharmaceutic Research.,4(3): 368-380.

Makni, M.; Yassine, C.; Hamadi, F.; El Mouldi, G.;Mohamed, B.; Chama ,M,.;Choumous, K.; andNajiba, Z.; (2012): Erythrocyte oxidative damagein rat treated with CCl4 protective role of vanillin, Saga Journal., 28 (10): 908-916.

Mandal ,A.; Karmakar, R.; Bandyopadhyay, S.; and Chatterjee, M.; (1998): Antihepatotoxic potentialof Trianthema portulacastrum in carbontetrachloride-induced chronic hepatocellula injury biochemical characteristics. Arch. Pharm.Res.,;21: 223- 230.

Manibusan, M. K.; Odin, M.; Eastmond, D. A. (2007): Postulated carbon tetrachloride mode of action: a review. J. Environ. Sci. Health C Environ. Carcinog. Ecotoxicol. Rev., 13: 539-553.

Motawi ,T.K.; Hamed, M.A.; Shabana ,M.H,; Hashem, R.M.; Aboul- Naser A.F.;(2011): Zingiberofficinaleacts as a nutraceutical agent against liver fibrosis. Nutr Metab ., 8: 40-51.

Nikolova, M.; Evstatieva, L.; Nguyen ,T.D.;(2011): Screening of plant extracts for antioxidant properties. Botanica Serbica ., 35: 43-48.

Niskikimi, M.; Rao, N. A. and Yog, K. (1972): Colormetric determination of superoxide dismutase activity. Biochem. Biophys. Res. Commun., 46:849-851.

Ohkawa, H.; Wakatsuki, A. and Kaneda, C. (1982): Assay for lipid peroxides in animal tissues by thiobarbaturic acid reaction. AnalBiochem., 95: 351-358.

Ohta, Y.; Imai, Y.; Matsura, T.; Kitagawa, A. and Yamada, K. (2005): Preventive effect of neutropenia an carbon tetrachloride- induced hepatotoxicity in rats. J. Appl. Toxicol. 26:178-186**.**

Ohta, Y.; Kongo, M.; Sasaki, E.; Nishda, K. and Ishigura, I. (2000): Therapeutic effects of melatonin on carbon tetrachloride-induced acute liver injury in rats. J. Pineal. Res.,28(2): 119-126**.**

Ohta, Y.;Sahashi, D.; Sasaki, E. and Ishiguro, I. (1999): Alleviation of carbon tetrachloride-induced chronic liver injury and related dysfunction by L- tryptophan in rats. Ann. Clin. Biochem., 36(4): 405-510.

Ohta, Y.; sasaki , E. ; Nishida, K.; Hayashi , T .; Nagata , M.; and Ishiguro, I .;(1997): preventive effect of oran-gedoku-to (Huanglian –Jie-Du –Tang) extract on progression of Commun. Mol. Pathol. Pharmacol.,95: 191-207.

Orhan, D.D.; Orhan, N.; Ergun, E.; Ergun, F.;(2007): Hepatoprotective effect of *Vitis vinifera* L. leaves on carbon tetrachloride-induced acute liver damage in rats. J Ethnopharmacol .,112:145-51.

Orhue, E.G.; Idu, M.; Atamari ,J.E.; Ebite, L.E.; (2008): Haematological and Histopathological Studies of *Jatropha tanjorensis* Leaves in Rabbits. Asian: Journal of Biological Sciences. 192: 84-89.

Paglia, D.E.; and Valentine, W. N.; (1967):UV method for determination of glutathione peroxidase. J. Lab. Clin. Med., 70: 158-169.

Panda V. S. ; andNaik, S. R. ;(2009): "Evaluation of cardioprotective activity of Ginkgo biloba and Ocimum sanctum in rodents," Alternative Medicine Review., 14(2) : 161–171.

Pappas, N.; John, J.R.; Wisecarver, J. and Becker, S. (1994): Effect of cycloheximide on increased aspirate aminotransferase in carbon tetrachloride hepatotoxicity. Annal. Clin. Lab. Sci.,14: 40-46.

Pari, L.and Murgavel, P. (2004): Protective effect of alpha lipoic acid against cholorquine induced hepatotoxicity in rats. J. Appl. Toxicol., 24: 21-26.

Patil ,V.V.; Bhangale, S.C.; Patil, V.R.; (2010): Evaluation of anti-pyretic potential of Ficus caricaleaves. Int J Pharm Sci Rev Res 2: 48-50.

patil, V.V. ;and V.R. Patil.;(2011): *Evaluation of anti-inflammatory activity of Ficus carica Linn.Leaves.* Indian Journal of Natural Products and Resources., **2**(2): p. 151-155.

Perez-Garcia, F.; Adzet, T .;and Canigueral, S.; (2000): Activity of artichoke leaf extract on reactiveoxygen species in human leukocytes. FreeRadical Res;33:661–665.

Persijn, J.P. and Van der Silk, W. (1976): New method for the determination of gamma-glutamyltransferase in serum. J.Clin.Chem.Biochem., 14:421-427.

Pohl, L.; Schulick, R.; George. J.;(1984): Reductive oxygenation mechanism of metabolism of carbon tetrachloride to phosgene and carbon dioxide formation. Biochemical and Biophysical Research Communication., 25: 318 – 324.

Pukkumani, R.; Aruna, K; Varma, P.S. and Menon, V.P. (2004): Ferulic acid, a natural phenolic antioxidant modulates altered lipid profiles during alcohol and thermal oxidized sunflower oil induced toxicity. J. Physiol pharmacol., 54: 448-51.

Rajkapoor, B.; Jayakar, B.; Kavimani, S. and Murugesh, N. (2002): Effect of dried fruits of Carica papaya Linn. on hepatotoxicity. Biol. Pharm. Bull., 25 (12): 1645-1646.

Recknagel, R. O.; Glende, E. A.; Dolak, I. A. and Waller, R. L. (1989): Mechanism of carbon tetrachloride toxicity. Pharmacol. Ther., 43: 139-154.

Reitman, S. and Frankel, S. (1957): A colorimetric method for the determination of serum glutamic oxaloacetic and glutamic pyruvic transaminases. Am. J. Clin**.** Path**.,** 28: 56-63.

Rui ,Jia.; Liping.; Cao.; Pao Xu, Galina Jeney and Guojun Yin.; (2012): In vitro and in vivo hepatoprotective and antioxidant effects of Astragalus polysaccharides against carbon tetrachloride- induced hepatocyte damage in common carp (Cyprinus carpio). Fish Physiology and Biochemstry.,38(3):871-881.

Saliu, J.A.; Elekofehinti, O.O.; Komolafe, K.; Oboh ,G.; (2012): Effects of Some Green Leafy Vegetables on the Haematological Parameters of diabetic rats. Journal of Natural Product Plant Resources. 2(4): 482-485.

Sallie, R. Tredger, J. M. ; and Williams, R. ;(1991): Drugs and the liver. Part 1: testing liver function,"Biopharmaceutics and Drug Disposition.,12(4) : 251–259,

Satheesh, M. and Pari. L. (2008): Effect of pterostilbene on lipids and lipid profiles in streptozotocin–nicotinamide induced type 2 diabetes mellitus. J. Appl. Biomed., 6: 31–37.

Shanmugasundaram. P.; Venkataraman ,S.;(2006): Hepatoprotective and antioxidant effects of Hygrophilaauriculata(K. Schum) Heine Acanthaceae root extract. J Ethnopharmacol ., 104: 124-128.

Sherlock, S.; and Dooley, J.; (1993): Disease of the Liver and Biliary System. Blackwell Scientific Publications, p.128.

Shymal, S.; Latha, P.G.; Shine, Y.J.; Suja, S.R.; Rajase Kharan, S. and Ganga Devi, T. (2006): Hepatoprotective effects of pittosporum neelgherrense a popular Indian ethnomedicine. J. Ethnopharmcol.,107: 151-155.

Singab, ANB. Youssef, DTA. Noaman, E. Kotb, S. (2005): Hepatoprotective effect of flavonol glycosides rich fraction from Egyptian Vicia calcarata Desf. against CCl4-induced liver damage in rats. Arch Pharm Res28:791–798.

Slater, T. F.; (1965): Biochemical mechanism of liver injury. London: Academic Press.

Slater, T.F.; (1984): Free radical mechanisms in tissue injury. Biochem J, 222: 1-15.

Snedecor ,G Cochran, W .(1980).In Statistical Method , 7 th ed . The Iowa State University press , Ames ,IA, USA , ISBN :0-81381560-6.P.507.

Stal, P.; Olson, J.;(2000): Ubiquinone: Oxidative stress, and liver carcinogenesis. In: Coenzyme Q: Molecular Mechanisms in Health and Disease, Eds. Kavan VE, Quinn DJ. Boca raton: CRC Press, pp: 317 - 329.

Sudheer, A.R.; Srinivasan, M.; Devipriya, N. and Menon, V.P. (2007): Dose-dependent inhibitory effect of ferulic acid, a dietary antioxidant on nicotine-induced tissue oxidative stress in experimental rats. IJPT., 6 : 177-187.

Sule, O.J.; Elekwa ,I.; Ayalogu, EO.;(2012): Effect of *Acalypha wilkesiana muell arg*. on haematologicalparameters in wistar albino rats. Int J Biol. Med Res., 3(1): 1234-1237.

Symonik-Lesiuk, S.; Czechowska, G.; Stryjecka- Zimmer, M.; Slomka ,M,.;Madro ,A.; Celinski ,K.; Wielosz, M.;(2003): Catalase, superoxide dismutase, and glutathione peroxidase activities in various rat tissues after carbontetrachloride intoxication. Journal of Hepatobiliary and Pancreatic Surgery., 10: 309 – 315.

Tacke, F.; Luedde, T.; andTrautwein, C. ;(2009):"Inflammatory pathways in liver homeostasis and liver injury,"Clinical Reviews in Allergy and Immunology., 36(1): 4–12.

Trad, M.; et al. ;(2012): *Does pollination affect aroma development in ripened fig [Ficus carica L.] fruit?*Scientia Horticulturae.,134: p. 93-99.

Trease, E.G.; Evan, W.C.; (1983): Textbook of pharmacognosy.12thed, Oxford: Alden Press, 1983. p. 539 .

Tung ,H.T.; Cook, F.W.; Wyatt, R.D .;and .;Hamilton, P.B.;(1975): The anemia caused by aflatoxin.Poult. Sci; 54: 1962-1969**.**

Veberic, R.M.; Colaric, and Stampar, F. ;(2008): Phenolic acids and flavonoids of fig fruit (Ficus carica L.)in the northern Mediterranean region. Food Chemistry., 106(1): p. 153-157.

Waller, R. A. and Duncan, D. B. (1969): Bayes rule for the symmetric multiple comparision problems. Am.J. Stat. Assoc., 64: 1484-1503.

Walter, M. and Gerade, H. (1970):Colorimetric method for totalbilirubin. Microchem., j., 15:231.

Wang, L.; Cheng, D.; Wang, H.; Di, L.; Zhou, X. and Xu, T. (2009): The hepatoprotective and antifibrotic effects of (I) saururus chinensis against carbon tetrachloride induced hepatic fibrosis in rats. J. Enthnopharmacol.,126: 487-491.

Weber, L.W.; Boll, M. and Stampfl, A. (2003): Hepatotoxicity and mechanism of action of haloalkanes: carbon tetrachloride as a toxicological model. Crit. Rev. Toxicol**.,** 33: 105-136.

Weber, L.W.; Boll, M. and Stampfl, A. (2003): Hepatotoxicity and mechanism of action of haloalkanes: carbon tetrachloride as a toxicological model. **Crit. Rev. Toxicol.,** 33: 105-136.

Xiong, Q.; Hase ,K.; Tezuke ,Y.;Tani, T.; Namba ,T.; Kadota, S .;(1998): Hepatoprotective activity of phenylethanoids from Cistanche deserticola. Planta Med.64: 120–125.

Yamammato, H. (1990): Relation of calcium accumulation and lipid peroxidation with CCl_4-induced toxicity in the rat liver. Pharmacol. Toxicol.,66: 213-216.

Zhao, J.; Lu, Z.; Wang, X. and Zhang, X. (2002): The study on the anti-oxidation effect of root of Mallouts apelta in rat model of liver fibrosis. Zhong Yao cai.,25(3): 185-187.