Al-Azhar Med. J . (Medicine). *Vol. 51(2), April, 2022, 1311 – 1324*
DOI: 10.21608/amj.2022.233226
https://amj.journals.ekb.eg/article_233226.html

COMPARATIVE EFFECTS OF OLIVE LEAVES EXTRACT AND SILYMARIN ON HEPATIC REDOX STATE IN EXPERIMENTALLY CARBON TETRACHLORIDE-INDUCED LIVER INJURY IN ADULT MALE ALBINO RATS

By

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ABSTRACT

Background: Liver cirrhosis is the final common pathway of many pathological conditions that affect hepatic tissue. Its prevalence is increasing globally with the highest age-standardized cirrhosis mortality rates in Egypt. Many drugs that were used in the management of hepatic diseases can induce further liver cirrhosis.

Objective: To assess the possible hepatoprotective activity of olive leaves extract and silymarin on experimentally induced liver damage caused by Carbon Tetra Chloride (CCL4)-induced liver damage.

Materials and methods: Fifty male adult albino groups were randomly assigned into five equal groups: Group (I): control saline (CS) rats were given normal saline orally by gavage as 1 ml/kg/day for 5 weeks, Group (II): control corn oil(CCO) were given corn oil orally by gavage as 1 ml/kg/day twice a week for four weeks Group (III): CCL4 - treated group (CCL4) were given CCL4 at a dose of 1ml/kg body weight 4 weeks, twice weekly orally by gavage, diluted with corn oil (1:1) to induce liver fibrosis, Group (IV): Olive leaf extract and the CCL4 - treated group (OLE+ CCL4) were given olive leaf extract only by oral gavage as100 mg/kg per day for one weak then, olive leaf extract by oral gavage as100 mg/kg per day simultaneously with CCl4 for 4 weeks, and Group (V): Silymarin & CCL4 - treated group (S+ CCL4) were given Silymarin only by oral gavage as100 mg/kg per day for one weak, then sylimarin by oral gavage as100 mg/kg per day simultaneously with CCl4 for 4 weeks.

Results: CCl4 produced marked hepatic injury through inducing oxidative stress in hepatic tissues. This was evidenced by a significant increase in serum AST, serum ALT, and hepatic MDA as well as a significant decrease in serum albumin, hepatic GSD, SOD, and CAT with CCl4 treatment compared to the control. Both Olive leaves extract and Silymarin protected hepatic tissue against the hazardous effects of CCl4 by restoring a balanced redox state of hepatic tissue. This was evidenced by a significant decrease in serum AST, serum ALT, and hepatic MDA as well as a significant increase in serum albumin, hepatic GSD, SOD, and CAT with OLE+ CCL4 and S+ CCL4 treatment compared to CCl4. No significant changes were noted between OLE+ CCL4 and S+ CCL4.

Conclusion: Both Silymarin and olive leaves extract have prophylactic protection and ameliorate the hepatotoxic effects of CCl4 as both have antioxidant properties. No significant difference was noted between OLE+ CCL4 and S+ CCL4.

Keywords: Hepatoprtection, antioxidants, Silymarin, olive leaf extract and CCl4.

INTRODUCTION

Liver diseases are widely recognized as one of the most serious health problems in developing or developed countries *(Araujo et al., 2018).* CCl4 is a classical hepatotoxic substance that induces hepatic pathology that resembles those seen in most cases of human liver diseases by generating oxidative stress *(Niu et al., 2017).*

Although the exact cause of liver fibrosis is unknown, oxidative stress is undeniably important in pathological changes in the liver, especially in instances of alcoholics and toxic liver disorders *(Contreras-Zentella and Hernandez-Munoz, 2016).*

Herbal products are alternative and complementary substances that can be used to ameliorate oxidative stress *(Abirami et al., 2015).*

Polyphenols, flavonoids, flavones, iridoids, and sugars are among the elements found in olive leaves and fruits. These compounds have a significant pharmacological effect while being minimal in toxicity. Total olive leaf extract has stronger antioxidant activity than vitamin C and vitamin E because it contains flavonoids, oleuropeosides, and substituted phenols that have synergetic effects *(Zhang and Tsao, 2016).*

Silymarin is a herbal medicine derived from the dried seeds of the milk thistle plant (Silybum marianum). This extract includes flavono-glignans (silibinin, isosilychristin, silychristin, isosilibinin, and silydianin) as well as a flavonoid (taxifolin) *(Kim et al., 2015).* It has been reported to have many biological activities such as anti-inflammatory, immunomodulatory, anti-fibrotic, anti-proliferative, antiviral properties, and anti-oxidative effects *(Asrani et al., 2019).* Silymarin has been shown to have a potential impact on many liver disorders, including oxidative stress, hepatic injury, and fibrosis *(Taleb et al., 2018).*

Pre-treatment using silymarin altered oxidative stress, cell cycle, cytoskeletal network, cell-cell adhesion, extracellular matrix, inflammation, apoptosis, and cell signaling, with the findings indicating that the effects of silymarin may be attributable to its antioxidant activity *(Abenavoli et al., 2018).*

The present work aimed to study the potential prophylactic hepatoprotective effect of olive leaf extract and compare this effect with silymarin.

MATERIALS AND METHODS

Animals:

Fifty adult male albino rats, of a local strain weighing 180-200 g with an average age of 8-12 weeks, were chosen as animal models for this study. Rats were purchased from the animal house of Nile pharmaceutical company (Cairo-Egypt). The study was done at the experimental laboratory of Pharmacological Department, Faculty of Medicine, Al-Azhar University. They were housed in a well-aerated polypropylene transparent cage (average dimension 20x32x20), and 5 rats in each cage maintained normal room temperature with a natural dark-light cycle. They were provided with rats' food and water ad libitum. They were left 2 weeks before starting the experiment to adapt to the new environmental conditions

and, to detect any visible signs of pathological conditions among them. The design and the procedures of this work were approved by the ethical committee of Al-Azhar University. The national laws and guidelines for use and care of laboratory animals were followed.

Drugs:

1. Carbon tetrachloride (CCL4): (El-Nasr Pharmaceuticals chemical company, Egypt).
2. Silymarin was supplied as a yellow fine powder, then dissolved in normal saline (Sigma-Aldrich Chemical Company, USA).
3. Olive leaf extract: Leaves were washed with tap water for 10 min, then transferred to laminar airflow and submerged in sodium hypochlorite at 0.5% for 2 minutes. They were washed three times in sterilized distilled water for 5 minutes. The dried leaves were powdered using a coffee grinder and then extracted. Fifty grams of the processed plant leaves were extracted in 250 ml of ethanol (70%) using the Soxhlet apparatus. The obtained extract was then evaporated at 37ºC by the incubator, and the resultant crude extract was kept frozen at -20C° until used *(Badawy et al., 2013)*. Olive leaf extract was dissolved as 100 mg in 10 ml of distilled water and given as 10 mg/ 1 ml per day.

Experimental design:

Rats were divided randomly into five equal groups (10 rats each):

Group (I): The control saline (CS) group was treated with normal saline orally by gavage as 1 ml/kg/day for 5 weeks.

Group (II): Control corn oil (CCO) group was treated with corn oil orally by gavage as 1 ml/kg/day twice a week for four weeks.

Group (III): CCL4 - treated group was treated with CCL4 in a dose of 1ml/kg body weight twice weekly for 4 weeks orally by gavage, diluted with corn oil (1:1) *(Prabhu et al., 2010).*

Group (IV): Olive leaf extract (OLE) & CCL4 - treated group was treated with olive leaf extract only by oral gavage (100 mg/kg per day for one weak), then they were treated with olive leaf extract by oral gavage (100 mg/kg per day simultaneously) with CCl4 (1ml/kg body weight) twice weekly orally by gavage, diluted with corn oil (1:1) for 4 weeks *(Dub and Dugani, 2013).*

Group (V): Silymarin (S) & CCL4 - treated group was treated with Silymarin only by oral gavage.

(100 mg/kg per day) for one weak then was treated with Silymarin by oral gavage (100 mg/kg) per day simultaneously with CCl4 (1ml/kg body weight) twice weekly orally by gavage, diluted with corn oil (1:1) for 4 weeks.

Induction of hepatic lesion:

Hepatopathy was induced by CCL4 given to rats orally twice a week for 4 weeks at a dose of 1 ml/kg body weight mixed with an equal volume of corn oil (1:1) *(Prabhu et al., 2010).*

Biochemical analysis:

Immediately after the end of the experiment and after overnight fasting,

rats were anesthetized in the morning by light pentobarbitone anesthesia, then blood samples were collected from the retro-orbital venous plexuses by a capillary tube. Collected blood was centrifuged at 5000 rpm for 20 minutes for serum separation. Separated serum was aliquot in an Eppendorf tube and stored frozen at -20O C until analysis of serum alanine aminotransferase (ALT) and serum aspartate aminotransferase (AST) *(Bergmeyer et al., 1985),* and Serum Albumin *(Batholomew and Delaney, 1994).*

Anesthetized animals were sacrificed and a dorsal midline incision was done to excise the liver. Obtained livers were cut into small pieces, minced, and homogenized in 1 ml lysis buffer *(Ahmed et al., 2013).* The cell lysate was stored at -70O C until used to estimate the hepatic superoxide dismutase (SOD) activity *(Nandi and Chatterjee, 1987),* malondialdehyde (MDA) content *(Olatosin et al., 2014),* Hepatic catalase (CAT) activity *(Claiborne, 1985)* and reduced glutathione (GSH) contents *(Ellman, 1959).*

Statistical Analysis:

Data input and analysis were done using SPSS computer program version 20. All results were expressed as the mean ± standard error. Mean values of the different groups were compared using a one-way analysis of variance (ANOVA). Post hoc. analysis was used to identify significantly different mean values. A P value < 0.05 was accepted to denote a significant difference.

RESULTS

CCL4 induced marked hepatic insults as evidenced by a significant increase in ALT (190.1 ± 11.69), AST (229.8 ± 5.61), and a significant decrease in serum albumin level (2.61± 0.124), compared to the control group given corn oil (21.37 ± 1.22, 20.78 ± 1.58 and 3.93 ± 0.039) respectively **(Table 1)**. Also, CCL4 induced severe hepatic oxidative stress as evidenced by a significant increase in MAD (3.72±0.308), and a significant decrease in GSH (29.27 ±1.53), SOD (2.75 ± 0.30), and CAT (0.007± 0.001), when compared with a control group given corn oil (1.39 ± 0 .144) (64.56 ± 2.88), (7.43 ± 0.0005) and (0.040 ± 0.004) respectively **(Table 2)**.

Olive leaf extract ameliorated the toxic effects of CCl4 on hepatic tissue as it produced a significant decrease in ALT (76.30 ± 5.50, % change was -59.8 %), AST (88.200 ± 6.96, % change was -61.61), and a significant increase in serum albumin level (3.4 ± 0.17, % change was +30.26), compared to CCl4-treated group (190.1±11.69, 229.8±5.61 and 2.61±.124) respectively. **Tables (1 and 3)**. Also, it reduced the oxidative stress effect of CCl4 on hepatic tissues as it produced a significant decrease in MDA (1.61±0.176, % change was -56.7), and a significant increase in GSH (56.23±3.46, % change was +92.10), a significant increase in SOD (6.98±0.69, % change was +153.81) and CAT (0.033±0.0024, % change was +371.42), compared to CCl4-treated group (3.72±0.308), (29.27 ±1.53), (2.75 ± 0.30) and (0.007±0.001) respectively **(Tables 2 and 3)**.

Silymarin also can protect hepatic tissue from the harmful effects of CCl4. This prophylactic effect of silymarin was evidenced by a significant decrease in ALT (78.17±6.61, % change was -58%), AST (85.17±5.46, % change was -62%), and a significant increase in serum albumin level (3.62±0.117, % change was 38.69%), compared to CCI4 treated group (190.1±11.69), (229.8±5.61) and (2.61±.124) respectively (Tables 1 and 3). Silymarin balanced the disturbed redox state of hepatic tissues produced by CCl4.

This effect was cleared by a significant decrease in MDA (1.72±.180, % change was -53.76%) and significant increase in GSH (48.63±4.13, % change was +66.14), a significant increase in SOD (6.65 ± 0.37, % change was +141.81) and CAT (0.031±0.028, % change was+342.85), compared to CCl4-treated group (3.72±0.308) (29.27 ±1.53), (2.75 ± 0.30) and (0.007±0.001) respectively **Table (2 and 3)**.

Table (1): Effects of olive leaf extract (OLE) and silymarin (S) on serum ALT, AST, and albumin levels in induced hepatic damage by CCl4

Groups / Parameters	Group I (CS) N=10	Group II (CCO) N=10	Group III (CCl_4) N=10	Group I V (OLE+ CCl4) N=10	Group V (S+ CCl4) N=10
ALT (U/L)	20.40± 1.77	21.37± 1.22	190.1±11.6*	76.30 ± 5.5*#	78.17±6.61*#
AST (U/L)	24.63± .936	20.78± 1.58	229.8±5.61*	88.200± 6.96*#	85.17±5.46*#
Albumin (g/dl)	4.04±.092	3.93±.039	2.61±.124*	3.4 ± 0.17#	3.62±0.117#

Data were presented as means ± SE.
*: Significant values compared to CCO. #: Significant values compared to CCL_4.
CS: control group treated with saline. CCO: control group treated with corn oil.
CCL_4: Group treated with CCL_4 to induce hepatic lesion.

OLE +CCl_4: Group treated with olive leave extract before and with CCL_4.
S +CCl_4: Group treated with silymarin before and with CCL_4.

Table (2): Effects of olive leaf extract (OLE) and silymarin (S) on MDA, GSH, SOD, and CAT levels in liver tissue homogenate in induced hepatic damage by CCl4

Groups / Parameters	Group I (CS) N=10	Group II (CCO) N=10	Group III (CCl_4) N=10	Group IV (OLE+CCl_4) N=10	Group V (S+CCl_4) N=10
MDA (nmol/mg protein)	1.42±0.117	1.39±0 .144	3.72±0.308*	1.61±0.176#	1.72±.180#
GSH (µmol/g wet tissue	68.88± 3.43	64.56± 2.88	29.27 ±1.53*	56.23±3.46#	48.63 ±4.13#
SOD (U/mg protein)	7.60± 0.47	7.43± 0.0005	2.75± 0.30*	6.98±0.69#	6.65± 0.37#
CAT (U/mg protein)	0.039±0.003	0.040 ± 0.004	0.007±0.001*	0.033±0.0024#	0.031±0.0028#

Data were presented as means ± SE.
*: Significant values compared to CCO. #: Significant values compared to CCL_4.
CS: control group treated with saline. CCO: control group treated with corn oil.
CCL_4: Group treated with CCL_4 to induce hepatic lesion.
OLE +CCl_4: Group treated with olive leave extract before and with CCL_4.
S +CCl_4: Group treated with silymarin before and with CCL_4.

Table (3): % change in different parameters produced by OLE and silymarin in comparison with CCl4

Groups / Parameters	CCl_4 N=10	OLE +CCl_4 N=10	S+CCl_4 N=10
ALT (U/L)	190.1±11.69	76.30 ± 5.5#	78.17±6.61#
% change		-59.8 %	-58.8
AST (U/L)	229.8±5.61	88.200± 6.96#	85.17±5.46#
% change		-61.61	-62.93
Albumin (gm/dl)	2.61±.124	3.4 ± 0.17#	3.62±0.117#
% change		+30.26	+38.69
MDA (nmol/mg protein)	3.72±0.308	1.61±0.176#	1.72±.180#
% change		-56.7	-53.76
GSH (µmol/g wet tissue)	29.27 ±1.53	56.23±3.46#	48.63 ±4.13#
% change		+92.10	+66.14
SOD (U/mg protein)	2.75± 0.30	6.98±0.69#	6.65± 0.37#
% change		+153.81	+141.81
CAT (U/mg protein)	0.007±0.001	0.033±0.0024#	0.031 ±0.0028#
% change		+371.42	+342.85

Data were presented as means ± SE.
*: Significant values compared to CCO. #: Significant values compared to CCL_4.
CS: control group treated with saline. CCO: control group treated with corn oil.
CCL_4: Group treated with CCL_4 to induce hepatic lesion.
OLE +CCl_4: Group treated with olive leave extract before and with CCL_4.
S +CCl_4: Group treated with silymarin before and with CCL_4

DISCUSSION

CCl4 is a classical hepatotoxin that induced hepatic pathologies resembling those seen in most cases of human liver diseases, Therefore, the periodic administration of carbon tetrachloride in rats was used in our work to induce toxin-mediated experimental hepatic lesions *(Liedtke et al., 2013).*

In the present study, periodic administration of CCL4 produced severe hepatic injury as evidenced by a significant increase in the serum levels of liver enzymes, AST, and ALT as well as a significant decrease in serum albumin. These changes are early markers that more specific for detecting liver damage *(Badrick and Turner, 2016).* These results were in agreement with *Althnaian et al. (2013)* who reported that rising activities of specific liver enzymes, ALT and AST, are due to hepatocellular necrosis induced by CCl4 or its metabolites. A significant decrease in serum albumin indicates a progressive loss of synesthetic function of the liver as a result of hepatic fibrogenesis *(Scholten et al., 2015).* CCl4 is converted in the liver by the cytochrome P450 to the trichloromethyl radical (CCl3). This hepatotoxic radical reacts with nucleic acids and proteins in hepatocytes, thus reducing cellular processes responsible for protein synthesis resulting in lowered protein quantities, especially albumin *(Ismail et al., 2016).* In addition, CCL4 and its metabolites cause fragmentation of hepatic endoplasmic reticulum and disruption of ribosomes into subunits with subsequent disengagement of the 40S subunit from mRNA, thus impairing protein synthesis *(Saad et al., 2014).*

There was strong direct evidence that refers to oxidative stress as a cause of hepatic injury in our work such as an increase in hepatic lipid peroxidation (MDA concentration), decreased CAT, SOD activity, and reduced GSH content in hepatic tissue. Trichloromethyl radical (CCl3) reacts with oxygen to form trichloromethylperoxy radicals (CCl3OO) that initiates lipid peroxidation and destruction of polyunsaturated fatty acids to form thiobarbutric reactive substances, MDA *(Vuda et al., 2012).* CCl4 -induced hypoalbuminemia deprives the body of the antioxidant activity of the albumin, that further increases oxidative stress insults *(Zheng et al., 2017).* The peroxidation of unsaturated fatty acids in biological membranes of hepatocytes leads to a decrease in membrane fluidity and disruption of membrane integrity causing leakage of the hepatic enzymes (ALT and AST) from the cytosol of hepatic cells to the extracellular compartment *(Erturk et al. 2014* and *Yan et al. 2015).* Reduced glutathione (GSH) comprises the major non-enzyme antioxidant system that protects the cells against free radicals. GSH acts directly as a free radical scavenger, reduces peroxides, and maintains protein thiols in the reduced state. The low level of GSH content in CCl4– treated group was in agreement with *Goodarzi et al. (2017)* who stated that exposure to CCl4 caused GSH depletion, combined with induction of a high level of lipid peroxidation in hepatic tissue, implying down-regulation of numerous antioxidative reactions in the liver. Chronic administration of CCL4 led to mitochondrial DNA (mtDNA) alterations and depletion of (GSH)

(Zarezade et al., 2018). The principal causes of CCL4-induced hepatic damage are lipid peroxidation, decreased activities of antioxidant enzymes and, generation of free radicals *(Saad et al., 2014)*.

In the current study, CCl4 intoxication decreased hepatic SOD and CAT activities. These findings are in agreement with *Ismail et al. (2016)* who reported decreased activities of CAT, SOD in CCl4 treated rats. This decrease in hepatic SOD and CAT activities could be due to the over-utilization of these enzymatic antioxidants to scavenge the excess products of lipid peroxidation and free radical production. These results confirmed that the innate antioxidants hepatic mechanisms cannot protect the body against the severe oxidative insult of CCL4 *(Ristow, 2014)*.

As this protection isn't comprehensive, particularly when oxidants (ROS and MDA) are produced in excess, antioxidants' other defensive processes may contribute to the health advantages. As a result, a variety of natural and synthetic antioxidative medicines have been suggested to prevent and cure hepatopathies caused by oxidative stress *(Rebeirio et al., 2019)*.

Conventional or synthetic drugs, that are used in the management of liver diseases have serious adverse effects *(Dara et al., 2017*; *Light et al., 2018* and *Noureddin and Kaplowitz, 2018)*.

Back to nature is a fantastic concept that attempts to utilize natural products to counteract the hazards of synthetic drugs. Many herbal remedies, particularly those containing Polyphenolic compounds, are utilized to treat a variety of liver ailments. These polyphenolic compounds are recognized for their high antioxidant activity, as well as their ability to scavenge free radicals and protect antioxidative defense systems *(Cory et al., 2018)*. The olive tree has been widely accepted as one of the species with the highest antioxidant activity *(Antunes et al., 2020)*. The results of the present study showed that oral administration of olive leaf extract has hepatoprotective effect against CCL4. This was evidenced by a significant decrease in the level of ALT and AST as well as a significant increase in albumin levels in an olive-treated group compared to the CCL4 group. It improved the redox state of hepatic tissue and reduced hepatic oxidative insult, as evidenced by a significant reduction in hepatic MDA and significant increases in GSH, CAT, and SOD. These results were in agreement with the study done by *Badawy et al. (2013)* who proved that olive leaf extract has the power to ameliorate trichloroacetic acid (TCA)-induced hepatopathy. It possesses a strong antioxidant activity that protects hepatocytes against CCl4-induced hepatotoxicity by scavenging free radicals *(Ismail et al., 2015)*. This effect preserves the integrity of hepatocyte’s biological membrane and increases the vitality of hepatocyte’s organelles *(Cory et al., 2018)*. A physiological positive feedback mechanism, that maintains redox homeostasis, was created by olive leaves extract. It improves the synthetic power of the liver and produces more albumin that has antioxidant activity, with subsequent reduction of oxidative insult. This increase the vitality of hepatocyte with more albumin formation with more reduction of oxidative damage. Oleuropein, a well-known phenolic compound that has a high

concentration in olive leaves and fruits, possesses diverse healing properties (vasodilator, anti-inflammatory and antioxidant effects) *(Alirezaei et al., 2012)*. Its antioxidant activity can scavenge superoxide anion, hydroxyl radicals, and hypochlorous acid-derived radicals *(Cumaoğlu et al., 2011* and *Domitrović et al., 2012)*.

In the present work, we compared the effect of olive leave extract and silymarin. Concomitant oral intake of silymarin with CCl4 ameliorated all the deleterious effects induced by CCl4 as shown by the studied biochemical parameters. Silymarin treatment decreased the hepatotoxicity as manifested by the correction of liver serum enzymes AST, ALT, and albumin. These results can be explained by *Siegel et al. (2013)* who reported that silymarin can stabilize the hepatocyte's biological membrane structure, thereby preventing toxins from entering the cell through it. In addition, it has the ability to promote liver regeneration, by stimulating nucleolar polymerase A, and increasing ribosomal protein synthesis *(Kondylis et al., 2017)*.

Silymarin intake significantly reduced CCl4-induced lipid peroxidation and significantly enhanced SOD and CAT activities, and GSH content in the hepatic tissue. These results were in accordance with previous studies that reported that silymarin significantly reduced lipid peroxidation, and liver enzymes and increase glutathione content in rats exposed to CCl4 *(Kwon et al., 2013)*. Also, these results were supported by *Surai (2015)* who stated that Silymarin significantly decreased lipid peroxidation and increased endogenous antioxidants, such as SOD, CAT, and GSH. Biologically, silibinin (silybin) is the main ingredient and the most active flavonolignan in silymarin *(Abenavoli et al., 2018)*. Silibinin is a polyphenolic flavonoid hepatoprotective substance. It can protect intact liver cells, or cells not yet irreversibly damaged by CCL4 by many mechanisms. It is a free radical scavenger that can balance toxic–induced disturbance in redox homeostasis and reduce oxidative stress and consequent hepatotoxicity *(Surai, 2015)*. It can modulate the enzymes responsible for the development of hepatocyte damage, fibrosis and cirrhosis *(Abenavoli et al., 2018)*. It selectively prevents glutathione depletion in hepatocytes protecting cells from damage in vitro *(Kwon et al., 2013)*.

When we compared the percentage changes of studied parameters in both olive leave extract and silymarin -treated groups in comparison with CCL4, biochemical markers (serum level of ALT, AST and albumin) of hepatic lesion were nearly similar and hepatic redox state's parameters (hepatic MDA, CAT, SOD and GSH) in olive treated group was insignificantly better than in silymarin treated group especially GSH.

Finally, both olive leave extract and silymarin had hepato-protictive effects and can ameliorate the CCL4-induced hepatopathy. The main explanation for their benefits is the "biochemical scavenger effect. Both of them have polyphenolic compounds that can reduce free radicals by forming stabilized chemical complexes, thus preventing further oxidative injury *(Cory et al., 2018)*. Furthermore, there is also an evidence of an additional mechanism by which polyphenols protect against

oxidative stress by producing hydrogen peroxide (H202), which can then help to regulate immune response actions, like cellular growth *(Jakobek, 2015).*

CONCLUSION

CCl4 caused hepatotoxicity via oxidative stress. This was revealed by significant elevation of ALT, AST, and MDA as well as a significant reduction in albumin, CAT, SOD, and GSH in the CCl4-treated group in comparison with a control group. Treating the hepatotoxic rats with olive leaves extract and silymarin protected the liver against CCl4 toxicity in the fourth and fifth groups, respectively. This hepatoprotective activity may be attributed to the biologically active compounds that exist in both olive leaves extract and silymarin which work to scavenge free radicals.

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مقارنة تأثير خلاصة أوراق الزيتون والسيليمارين على حالة الأكسدة الاختزالية الكبدية فى حالة الكبد المتلف تجريبيا بواسطة مادة رباعي كلوريد الكربون لدي ذكور الجرذان البيضاء البالغة

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خلفية البحث: إن التليف الكبدي هو النهاية الأكثرحدوثا في العديد من الحالات المرضية الكبدية.ولقد زاد معدل انتشاره علي مستوي العالم كما زاد معدل الوفيات المعياري لدي المصريين بسببه. وأن الأدوية التي تستخدم في معالجة أمراض الكبد قد تزيد من معدل إنتشاره.

الهدف من البحث: مقارنة تأثير خلاصة أوراق الزيتون والسيليمارين على حالة الأكسدة الاختزالية الكبدية فى حالة الكبد المتلف تجريبيا بواسطة مادة رباعي كلوريد الكربون لدي ذكور الجرذان البيضاء البالغة.

مواد وطرق البحث: إستخدم فى هذا البحث خمسون جرذا أبيضا بالغا من فصيلة محلية تتراوح أوزانها بين 180 – 200 جم .وقد قسمت هذه الجرذان الي خمسة مجموعات متساوية وتم معالجتها كما يلي:

- **المجموعة الأولي:** مجموعة ضابطة تم معالجتها بواسطة محلول ملح طبيعي عن طريق الفم (الجرعة 1 مل|كجم) يوميا لمدة خمسة أسابيع.
- **المجموعة الثانية:** مجموعة ضابطة تم معالجتها بواسطة زيت القمح عن طريق الفم (الجرعة 1 مل|كجم يوميا) مرتين في الأسبوع لمدة أربعة أسابيع.
- **المجموعة الثالثة:** تم معالجتها بواسطة رباعي كلوريد الكربون المخفف بزيت الزيتون، وكانت نسبة التخفيف 1:1 (الجرعة 1 مل|كجم يوميا) مرتين أسبوعيا لمدة أربعة أسابيع لمدة خمسة أسابيع.

- **المجموعة الرابعة:** تم معالجتها بمحلول مستخلص أوراق الزيتون فقط (الجرعة 100 مجم|كجم) يوميا لمدة أسبوع ثم تم معالجتها بمحلول مستخلص أوراق الزيتون (الجرعة 100 مجم|كجم) بالتزامن مع معالجتها بمادة رباعي كلوريد الكربون (الجرعة1 مل|كجم) يوميا لمدة أربعة أسابيع أخري.
- **المجموعة الخامسة:** تم معالجتها بالسليمارين فقط (الجرعة 100 مجم|كجم) يوميا لمدة أسبوع ثم تم معالجتها بالسليمارين (الجرعة 100 مجم|كجم) بالتزامن مع معالجتها بمادة رباعي كلوريد الكربون (الجرعة1 مل|كجم) يوميا لمدة أربعة أسابيع أخري.

النتائج: أدت مادة رباعي كلوريد الكربون الي ارتفاع ذو دلالة احصائية في مستوي كل من إنزيمات الكبد بالدم ومستوي مادة المالونديالدهيد بالكبد و إنخفاض ذو دلالة إحصائية في كل من نسبة الألبومين بالدم ومستوي كل من الجلوتاثيون والسوبر اكسيد ديسموتازو الكاتلاز بالكبد. وكان لكل من مستخلص اوراق الزيتون والسليمارين القدرة علي حماية خلايا الكبد ضد التاثير المدمر لمادة رباعي كلوريد الكربون عن طريق تحسين حالة الإختلال التي حدثت في حالة الاكسدة الإختزالية الكبدية وحدوث إنخفاض ذو دلالة إحصائية في مستوي كل من إنزيمات الكبد بالدم ومستوي مادة المالونديالدهيد بالكبد. كما كان هناك إرتفاعا ذو دلالة إحصائية في كل من نسبة الألبومين بالدم ومستوي كل من الجلوتاثيون والسوبر اكسيد ديسموتازو الكاتلاز بالكبد. لم يوجد اختلاف ذو دلالة احصائية بين مستخلص اوراق الزيتون والسليمارين.

الإستنتاج: مستخلص أوراق الزيتون والسيليمارين لهم نفس القدرة الوقائية علي حماية خلايا الكبد ضد الأثار المدمرة لمادة رباعي كلوريد الكربون وكلاهما يحتوي علي مواد مضادة للاكسدة.