

EVALUATE THE HEALTH IMPACT OF SOME VEGETABLE WASTES ON LIVER DISORDER OF RATS PRETREATED WITH CCL₄

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ABSTRACT: *This study was conducted to investigate to possible health action of some plant leaves for impaired liver function of injected rats with carbon tetrachloride (CCL₄).Thirty five mature albino rats weighting (150±10) g.B.Wt. each, were used and divided to 7 equal group, one was kept as control- ve group. The other 6 groups were injected s/c by carbon tetrachloride (CCL₄) in olive oil 50% (3ml / kg.B.Wt.). Administration of single dose per day of CCl₄ to a rats, twice a week for 3 weeks to induce hepatotoxicity in laboratory animals was carried out. The used plants leaves were given at 7.5% from (Mango Tree leaves, Pear Tree leaves, Leek Leaves and Pomegranate Peels, and also as a mixture of all). At the end of experimental period (28 day), animal were scarified. Blood samples were collected to determine the following parameters: Antioxidant Enzymes (SOD,CAT,GST), level of serum liver enzymes (ALT,AST,ALP) and AST/ALT, serum total protein, serum albumin, serum globulin ,total bilirubin , total cholesterol ,triglycerides, lipoprotein fractions (HDL-c, LDL-c and VLDL-c), atherogenic index(AI), uric acid ,urea nitrogen, creatinine .Mango tree leaves and Pomegranate peels showed gradual decreases in total cholesterol level and Pear tree leaves recorded the best results in the improvement of liver enzymes while leek leaves showed improvement of liver and kidney function. On the other hand the best results found by using mixture of all plants leaves. While mean according to the results, Mango Tree leaves, Pear Tree leaves, Leek Leaves and Pomegranate Peels could be used single for impaired liver function and protection from hepatotoxicity.*

Key words: *Liver, hepatotoxicity, liver function, kidney function, lipid profile, Plant leaves, Mango Tree leaves, Pear Tree leaves, Leek Leaves, Pomegranate Peels.*

INTRODUCTION

The liver is one of the largest solid organs of the body. Its major functions include processing the food that passes through the gut and converting it into energy that can be utilized by the body. It is also a powerful detoxification center that handles many chemicals, alcohol, poisons and toxins as well as drugs and clears the blood.. They may be of short duration, acute liver disease, or long term, chronic liver disease. An acute liver disease may also convert into a chronic liver disease over time. Some liver diseases are caused by infective viruses like hepatitis virus (A, B and C). Liver diseases also result from taking in some drugs or alcohol over long term. Sometimes the diseased liver over long term becomes shrunken and scarred. Such a condition is called cirrhosis. Like other organs liver can

also be afflicted with cancers (Ananya, 2013). The liver is indispensable in breaking down fats from food. The proper functioning of the eyes, the heart, the brain, the gonads, the joints, and the kidneys, are all dependent on good liver activity. If the liver is impaired from constructing even one of the thousands of enzyme systems the body requires, there is an impairment in overall body function and a resultant greater metabolic stress on the individual .It is also important to note that the herb contains powerful antioxidants which are known to help protect your liver cells from serious damage (Karl,2013). The mangiferin (extract from mango leaves) was investigated for the hepatoprotective effects in two kinds of models of experimentally-induced liver injuries in rats. The models were induced by acetaminophen & carbon tetrachloride. The

results showed that mangiferin caused significant reduction of the elevated enzyme levels of serum glutamate oxaloacetate transaminase, serum glutamate pyruvate transaminase in model rats, and alleviated the pathological damages in livers of the model rats. (Anon2013-a). Pear fruit is also called Naspatti or Nashpati in India. Sometimes it is also called the "gift of the Gods". Its leaves are also loaded with many medicinal health benefits (Anon 2013-b). Pomegranate Peel is possibly effective against prostate cancer, diabetes, and lymphoma disease, possible benefit in dealing with breast cancer, maintain correct blood pressure, possibly helpful in dealing with fertility problems ,helps maintain a smooth healthy skin, possibly prevents premature aging, useful in fevers, can reduce the inflammation of arthritis, by slowing down the enzymes that breakdown cartilage;for dealing with coughs, can be added to smoothies and other food preparations (Anon 2013-c). Leeks contain many noteworthy flavonoid anti-oxidants, minerals, and vitamins that have proven health benefits. In addition, leeks are one of the good sources of flavonoid phenolic anti-oxidants such as carotenes, xanthin, and lutein. They also have some other essential vitamins such as vitamin C, K, and vitamin E. Vitamin C helps the human body develop resistance against infectious agents and scavenge harmful, pro-inflammatory free radicals (Kylenorton ,2011).The plant parts (mango tree leaves, pear tree leaves, pomegranate peels& leek leaves)were selected in present work for treatment of hepatointoxication, since previously no studies were carried out on them.

MATERIALS AND METHODS

MATERIALS

Plants: Mango tree leaves (*Mangifera indica*), Pear tree leaves (*Pyrus Communis*), leek leaves (*Allium ampeloprasum*) and Pomegranate peels (*Punica granatum*), were obtained from Haraze Company for medical herbal plants, dried at 50c^o and milled.

Carbon Tetra Chloride (CCl₄): Was obtained from El-Gomhoria Company for

Chemical Industries, Cairo, Egypt as 10 % liquid solution.

Olive oil: was obtained from Pharaouhs Company for dilution during the induction.

Animals: Thirty five (35) (Spargue – Dawley strain) male albino rats, weighing (150 ± 10 g) were used in this study. Rats were housed in wire cages under the normal laboratory condition and fed on basal diet for 4 consecutive days as adaptation period. Diets were introduced to rats in a special non-scattering feeding cup to avoid loss of food and contamination. Tap water was provided to rats by means of glass tubes projecting through wire cages from inverted bottles supported to one side of the cage.

Basal Diets: consisted of casein (12%), corn oil (10%), Choline Chloride (0.25%), and vitamins mixture(1%), cellulose (5%), salt mixture (4%) , corn starch (up to100%) and according to Campbell,1963 . The used salt and vitamins mixture was according to Hegested *et al.*, 1941 and Campbell, 1963.

METHODS

Induction of Liver Intoxication Rats:

Thirty (30) male albino rats (Spargue – Dawley strain) weighing (150 ± 10 g) were treated by subcutaneous (s/c) injection of Carbon Tetra Chloride (CCl₄) in Paraffin oil 50% V/V (2ml/ kg.B.wt) twice a week for three weeks to induce chronic damage of the liver according to the method described by Jayasekhar *et al.*, (1997).

Experimental Designs and Animal Groups

Thirty five (35) (spargue – Dawley strain) male albino rats, weighing (150 ± 10 g) fed on basal diet casein (12%), corn oil (10%), Choline Chloride (0.25%), and vitamins mixture(1%), cellulose (5%), salt mixture (4%) , corn starch (up to100%) and according to (Campbell,1963) for four consecutive days as adaptation period. Then, rats were distributed into 7 groups each of 5 rats in which means of rats weight for all groups were nearly equal. All the groups of rats were housed in wire cages

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and fed on the experimental diet for 4 weeks according to the following groups:

- **Group (1):** Control negative group (-ve), in which normal rats were fed on basal diet for 28 days.
- **Group (2):** Control positive (+ve), in which rats were inflicted with hepatotoxicity by injection with CCl_4 , were fed on basal diet for 28 days.
- **Group (3):** CCl_4 hepatotoxicity rats were fed on basal diet containing 7.5 % mango tree leaves for 28 days.
- **Group (4):** CCl_4 hepatotoxicity rats were fed on basal diet containing 7.5% pear tree leaves for 28 days.
- **Group (5):** CCl_4 hepatotoxicity rats were fed on basal diet containing 7.5% pomegranate peels for 28 days.
- **Group (6):** CCl_4 hepatotoxicity rats were fed on basal diet containing 7.5% leek leaves for 28 days.
- **Group (7):** CCl_4 hepatotoxicity rats were fed on basal diet containing 7.5% of combination of all plants for 28 days (1:1:1:1).

Each of the above groups was kept in a single cage. The diets were introduced to rats in special non-scattering feeding cups to avoid loss of feed and contamination. Tap water provided to rats by means of glass tubes projecting through wire cages from inverted bottles supported to one side of the cage. Rats were weighted at the beginning of the experiment then weekly and at the end of the experiment.

Blood Sample and Organs collection:

Blood samples were collected from all groups after 12 hours fasting at the end of experiment. Using the retro-orbital method, by means of a microcapillary glass, blood was collected into a dry clean centrifuge tube, and left to clot at room temperature for half an hour. The blood was centrifuged for 10 minutes at 3000 r.p.m to separate the serum. Serum was carefully aspirated and transferred into clean quit fit plastic tubes

and kept frozen at (-20 c) until the time of analysis. The organs (liver, kidney, heart, lungs and spleen) were removed and washed in saline solution and weighted.

Biological Indices Calculation:

Biological evaluation of the different diets was carried out by determination of feed intake daily, body weight gain g (BWG g/day) and feed efficiency ratio (FER) according to Chapman *et al.*, (1959).

Body Weight Gain = Final weight (g) - Initial Weight (g)

Feed efficiency ratio (FER) = gain in body weight (g) / Feed intake (g).

Biochemical analysis of serum:

Determination of serum total triglyceride was carried out according to Fassati and Prencipe (1982).

Serum total Cholesterol was determined according to Allain (1974).

Serum HDL-Cholesterol was determined by the same method used for total cholesterol, according to Lopez (1977).

Serum VLDL and LDL -Cholesterol were carried out according to the method of Lee and Nieman (1996) as follows.

$\text{VLDL (mg/dl)} = \text{T.G (mg/dl)} / 5$

$\text{LDL} = \text{Total Cholesterol} - [(\text{VLDL-C}) + (\text{HDL-C})]$.

Atherogenic Index was calculated according to Nakabayashi *et al.*, (1995) as follow:

$\text{Atherogenic Index (AI)} = \text{LDL} + \text{VLDL} / \text{HDL}$

Determination of Liver enzymes:

Glutamic-pyruvate transaminase (ALT) was carried out according to the method of Henry (1974) and Yound (1975).

Glutamic oxaloacetic transaminase (AST) was carried out according to the method of Henry (1974) and Yound (1975).

Alkaline phosphatase (ALP) was determined according to IFCC, (1983) methods.

Serum Total Protein was determined as described by Henry, 1974.

Determination of Serum Albumin formation of an albumin bromocresol green complex at pH 4.2 was done followed by photometric measurement of absorbance maximally at 578 nm (Doumas *et al.*, 1971).

Determination of Enzymatic Antioxidant:

Superoxide Dismutase (SOD) was carried out according to the method of (Sun *et al.*, 1988).

Glutathione S Transferase (GST) was carried out according to the method of (John, and Kathryn 1998).

Catalase (CAT) was carried out according to the method of (Diego,2011).

Statistical Analysis: The data were statically analyzed using a computerized costat program by one way ANOVA. The results are presented as mean \pm SD. Differences between treatments tested at $p \leq 0.05$ for significance.

RESULTS AND DISCUSSION

The current study focused on the possible health impacts of some plants leaves for impaired liver function of rats by injection with carbon tetrachloride (CCl₄).

1. Effect of feeding with some vegetables wastes and their combination on Body Weight Gain (BWG %), Feed intake (FI)

and Feed Efficiency Ratio (FER) of rats pretreated with CCl₄.

The mean value of Body weight gain (BWG %) of control (+) group was significantly ($p \leq 0.05$) lower than control (-) group, being 13.90 ± 1.19 & 38.90 ± 2.59 g respectively, with percent of increase $+17.98$ (table1). All CCl₄ injected rats fed on various diets showed significant increases in mean values as compared to control (+) group . The best BWG % was recorded for group 7 (rats fed on mixture of all) followed by that fed on pear tree leaves when compared to control (+) group.

The mean value of feed intake (F.I) of control (+) group was significantly lower than control (-) group, being 10.36 ± 1.00 & 12.68 ± 1.50 respectively, control (-) rats showed significant increase $+22.39$ % as compared to control (+).All CCl₄ injected rats fed on various diets showed significant increases in mean values as compared to control (+) group. The best F.I was recorded for group5 (CCl₄ injected rats fed on pomegranate peels) indicating non significant differences when compared to control (-) group (Table1).

Table (1): Effect of feeding with some vegetables wastes and their combination on Body Weight Gain (BWG %), Feed intake (FI) and Feed Efficiency Ratio (FER) of rats pretreated with CCl₄.

Groups	Parameter	BWG% (Mean \pm SD)	Feed Intake (Mean \pm SD) (g/day/rat)	FER (Mean \pm SD)
Control – ve (G1)		$38.90^a \pm 2.59$	$12.68^a \pm 1.50$	$0.024^a \pm 0.0013$
Control + ve (G2)		$13.90^g \pm 1.19$	$10.36^e \pm 1.00$	$0.013^b \pm 0.002$
7.5% Mango tree leaves(G3)		$15.50^f \pm 1.50$	$11.76^c \pm 1.00$	$0.017^{ab} \pm 0.001$
7.5% Pear tree leaves(G4)		$26.50^c \pm 2.52$	$11.50^{cd} \pm 1.00$	$0.019^{ab} \pm 0.010$
7.5% Pomegranate peels(G5)		$22.30^d \pm 1.20$	$12.57^a \pm 1.20$	$0.014^b \pm 0.001$
7.5% Leek leaves(G6)		$18.90^e \pm 1.80$	$12.07^b \pm 1.20$	$0.016^b \pm 0.001$
7.5% Mixture of all plants(G7)		$29.60^b \pm 2.90$	$11.26^d \pm 1.12$	$0.020^{ab} \pm 0.001$

Values denote arithmetic means \pm standard deviation of the mean . Means with different letters (a,b, c, d, etc ,) in the same column differ significantly at $p \leq 0.05$ using DUNCAN test , while those with similar letters are non-significantly different

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Data of Table (1) also illustrated the mean value of FER of CCl₄ injected rats fed on different diets. Data show that the mean value of FER of control (+) group was lower than control (-) group, being 0.013±0.002 & 0.024±0.0013 g respectively, showing significant difference with percent of increase +84.61% of control (-) group as compared to control (+). All CCl₄ injected rats fed on various diets showed significant increases in mean values as compared to control (+) group. The best FER was recorded for group 7 (CCl₄ injected rats fed on mixture of all plants) which revealed nonsignificant differences when compared to control (-) group.

The obtained results are in line with that of Bakr (2009); Faramawy, Asmaa (2010); Shehata, Rehab (2012) who found that injected rats by CCl₄ caused decrease in both BWG, FI and FER.

2. Effect of feeding with some vegetables wastes and their combination on Total Cholesterol (T.C.), Triglycerides (T.G) and Lipids profile of rats pretreated with CCl₄.

Data of Table (2) illustrated the mean value of serum Total Cholesterol (T.C.) (mg/dl) of CCl₄ injected rats fed on different diets. It could be observed that the mean value of T.C. of control (+) group was higher than control (-) group, being 204.00 ±7.7 & 168.00 ±8.00 mg/dl, respectively with a percent of decrease -17.65% of control (-) group compared to control (+) group. All CCl₄ injected rats fed on different experimental diets revealed significant decreases in mean values as compared to control (+) group. The best serum (T.C) showed for group 3 (fed on mango tree leaves) being less even when compared to control (-) group.

The mean value of Triglycerides (T.G.) of control (+) group was significantly (p≤0.05) higher than control (-) group, being 129.00 ±7.80 & 103.00 ±8.80 mg/dl respectively, indicating significant difference with percent of decrease -20.16% of control (-) group as compared to control (+) group. All CCl₄ injected rats fed on different diets revealed significant (p≤0.05) decreases in mean values as compared to control (+) group. Rats fed on mixture of plants (treatment 7) showed the best serum (T.G.) when compared to control (+) group (Table 2).

Table (2): Effect of feeding with some vegetables wastes and their combination on Total Cholesterol (T.C.) and Triglycerides (T.G) of rats pretreated with CCl₄.

Parameter	Total Cholesterol mg/dl (Mean±SD)	Serum Triglycerides mg/dl (Mean±SD)
Control – ve(G1)	168.00 ^c ± 8.000	103.00 ^e ± 8.80
Control + ve(G2)	204.00 ^a ± 7.700	129.00 ^a ± 7.80
7.5% Mango tree leaves(G3)	145.00 ^g ± 10.68	122.00 ^b ± 6.90
7.5% Pear tree leaves(G4)	173.00 ^b ± 6.30	116.00 ^c ± 3.50
7.5% Pomegranate peels(G5)	152.00 ^f ± 9.60	116.00 ^c ± 9.30
7.5% Leek leaves(G6)	156.00 ^e ± 9.25	120.00 ^b ± 7.90
7.5% Mixture of all plants(G7)	160.00 ^d ± 8.00	108.00 ^d ± 9.70

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The mean value of (HDL-c) of control (+) group was lower ($p \leq 0.05$) than control (-) group, being 90.00 ± 7.60 & 115.30 ± 8.30 mg/dl, respectively. showing significant difference with percent of increase +27.78% of control (-) group as compared to control (+) group (table3) . All hepatic rats fed on different experimental diets revealed significant increases in mean values as compared to control (+) group . The best (HDL-c) was observed for group 6 & 7 (hepatointoxicated rats fed on leek leaves & mixture of all plants).

Data in Table (3) also illustrated the mean value of serum(LDL-c) (mg/dl) of CCl_4 injected rats fed on different diets. It could be observed that the mean value of (LDL-c) of control (+) group was higher ($p \leq 0.05$) than control (-) group, being 88.20 ± 6.41 & 32.40 ± 1.18 , respectively. showing significant difference with percent of decrease - 63.27% of control (-) group as compared to control (+) group. All CCl_4 injected rats fed on different diets revealed significant ($p \leq 0.05$) decreases in mean values as compared to control (+) group . Rats fed on group 3(mango tree leaves) recorded the best serum (LDL-c).

The mean value of (VLDL-c) of control (+) group was higher ($p \leq 0.05$) than control (-) group, being 25.80 ± 1.45 & 20.60 ± 2.04 mg/dl, respectively. Showing significant difference with percent of decrease -20.16 % . All CCl_4 injected rats fed on different diets showed significant decreases in mean values as compared to control (+) group (table 3). The best treatment was recorded for group7 (mixture of all plants) considering serum (VLDL-c).

Data of Table (3) revealed also the mean value of serum (A.I.) (mg/dl) of hepatic rats fed on different diets. It could be noticed that the mean value of (A.I.) of control (+) group was higher ($p \leq 0.05$) than control (-) group, being 1.26 ± 0.119 & 0.46 ± 0.025 respectively. With -63.49% percent of decrease of control (-) group when compared to control (+) group. Meanwhile all hepatointoxicated rats fed on various diets showed significant ($p \leq 0.05$) decreases in mean values as compared to control (+) group . The best treatment was recorded for group 6 considering serum (A.I.) which showed less A.I numerically compared to control (-) group.

Table (3): Effect of feeding with some vegetables wastes and their combination on Lipid profile of rats pretreated with CCl_4 .

Groups	Parameter	Serum LDL-c mg/dl (Mean \pm SD)	Serum VLDL-c mg/dl (Mean \pm SD)	Serum HDL-c mg/dl (Mean \pm SD)	Atherogenic Index(A.I.) mg/dl (Mean \pm SD)
Control – ve(G1)		$32.40^c \pm 1.18$	$20.6^e \pm 2.04$	$115.00^a \pm 8.30$	$0.46^{bc} \pm 0.025$
Control + ve(G2)		$88.20^a \pm 6.41$	$25.80^a \pm 1.45$	$90.00^e \pm 7.60$	$1.26^a \pm 0.119$
7.5% Mango tree leaves(G3)		$14.60^f \pm 1.35$	$24.40^b \pm 1.13$	$106.00^c \pm 4.30$	$0.52^{bc} \pm 0.226$
7.5% Pear tree leaves(G4)		$50.80^b \pm 4.02$	$23.20^c \pm 2.17$	$99.00^d \pm 6.30$	$0.74^b \pm 0.065$
7.5% Pomegranate peels(G5)		$28.40^{cd} \pm 1.19$	$23.20^c \pm 1.18$	$105.00^c \pm 7.20$	$0.61^{bc} \pm 0.265$
7.5% Leek leaves(G6)		$22.00^e \pm 1.10$	$24.00^{bc} \pm 1.09$	$110.00^b \pm 8.66$	$0.41^c \pm 0.039$
7.5% Mixture of all plants(G7)		$28.40^{cd} \pm 2.00$	$21.60^d \pm 1.13$	$110.00^b \pm 7.300$	$0.45^c \pm 0.032$

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The obtained results are in agreement with that of Faramawy,Asmaa (20110);Abd Al-Halem, Eman (2012); Shehata, Rehab (2012) who indicated that injected rats by CCl_4 caused increase of T.C. , T.G. ,LDL-c ,VLDL-c & decrease of HDL-c in serum. Guo *et al.*, (2011) found that the mangiferin, a natural polyphenol, has been shown to have hypolipidemic effect in rat and mouse by a high -fat diet. Also they suggested that mangiferin may ameliorate hypertriglyceridemia partly by modulating the expression levels of genes involved in lipid oxidation and lipogenesis. Yucun *et al.*, (2012) reported that the mangiferin is a xanthone glucoside and exists in mango tree leaves reduced the level of of plasma free fatty acids and triglycerides in diabetic animals. Raihan *et al.*, (2013) suggested that mangiferin, the predominant constituent of extracts of the mango plant *Mangifera indica* modulating lipid metabolism and inhibits hyperlipidemia. Also they reported that mangiferin able to reverse elevated plasma total cholesterol, triglycerides, and LDL caused by high fat diet fed rats. Wolfram *et al.*, (2002) studied the effect pear in lowering cholesterol levels in both animals and man with hyperlipidemia because of high content of pectin and a soluble fiber. Mohammad *et al.*, (2011) demonstrated that *Pyrus* leaf extract reduces blood glucose and lipid levels and increases antioxidant status in rats with alloxan-induced hyperglycaemia. Velmurugan and Bhargava (2013) reported that pear reduce the elevated biochemical parameters such as triglycerides (TGL), low density lipoprotein (LDL), very low density lipoprotein (VLDL), and total cholesterol (TC), increased the reduced level of high density lipoprotein (HDL).Johanningsmeier and Harris (2011) and Viladomiu *et al.*, (2013) reported that pomegranate fruit prevents cardiovascular disease. Singh *et al.*, (2002)& Sushil *et al.*, (2013) suggested that pomegranate peel extract decrease the level of TC/HDL-c ratio and serum LDL-c levels and concluding

pomegranate peel polyphenolic extract is effective in lowering serum and hepatic lipids in rats fed on high -fat diet. Neyrinck *et al.*, (2013) reported that pomegranate peel extract reduced the serum level of cholesterol(total and LDL) induced by high fat feeding in mice. Durak *et al.*, (2002) demonstrated that cholesterol supplementation leads to dense plaque formation in the aortas. leek extract supplementation ameliorates blood lipid profile and, increases antioxidant potential. Ashraf *et al.*, (2011) said that the Combination of (*Allium sativum*) improve glycemic control in addition to antihyperlipidemic activity. Thomas *et al.*, (2013) evaluated that leek reduce total cholesterol (TC), triglycerides (TG), low-density lipoprotein cholesterol (LDL-C), VLDL-C and elevation in the level of high-density lipoprotein cholesterol (HDL-C).

3. Effect of feeding with some vegetables wastes and their combination on liver function of CCl_4 rats pretreated.

Table (4) illustrated the mean value of serum Glutamic oxaloacetic transaminase (AST) (U/L) of CCl_4 injected rats fed on various diets. It could be noticed that the mean value of (AST) of control (+) group was higher ($p \leq 0.05$) than control (-) group, being 36.00 ± 3.28 & 17.00 ± 0.40 U/L respectively, with -52.77 % percent of decrease of control (-) group when compared to control (+) group. Generally all CCl_4 injected rats fed on different diets revealed significant ($p \leq 0.05$) decreases in mean values as compared to control (+) group..Mango tree leaves (group3) revealed the best ($p \leq 0.05$) treatment when compared to the other treatments.

The mean value of Glutamic-pyruvate transaminase (ALT) (U/L) of control (+) group was higher ($p \leq 0.05$) than control (-) group, being 18.00 ± 0.60 & 10.00 ± 0.30 U/L, respectively. with a decreasing rates of (-44.44 %). All CCl_4 injected rats fed on

various diets revealed significant decreases in mean values as compared to control (+)

group. Group 7 was the best ($p \leq 0.05$) treatment considering the (ALT).

Table (4): Effect of feeding with some vegetables wastes and their combination on liver enzymes (U/L) (AST, ALT, ALP) of rats pretreated with CCl₄.

Groups	Parameter	AST (U/L) (Mean±SD)	ALT (U/L) (Mean±SD)	Alkaline phosphatase (U/L) (Mean±SD)	AST / ALT (U/L) (Mean±SD)
Control – ve(G1)		17.00 ^f ±0.4	10 .00 ^e ± 0.30	147.00 ^g ± 3.40	1.70 ^{bc} ± 0.13
Control + ve(G2)		36.00 ^a ±3.28	18.00 ^a ± 0.60	315.00 ^a ± 4.00	2.00 ^{abc} ± 0.27
7.5% Mango tree leaves(G3)		25.50 ^e ±2.44	16.00 ^b ± 0.90	276.00 ^c ± 2.50	1.59 ^c ± 0.11
7.5% Pear tree leaves(G4)		28.00 ^{cd} ± 1.80	15.00 ^b ± 1.3	159.00 ^f ± 3.80	1.86 ^{abc} ± 0.46
7.5% Pomegranate peels(G5)		31.00 ^b ±2.10	13.00 ^{cd} ± 0.70	229.00 ^d ± 3.18	2.38 ^a ± 0.23
7.5% Leek leaves(G6)		29.50 ^{bc} ± 2.94	14.5 ^{bc} ± 1.25	283.50 ^b ± 4.25	2.03 ^{abc} ± 0.39
7.5% Mixture of all plants(G7)		27 .00 ^{de} ± 1.3	12 .00 ^d ± 0.75	233.50 ^e ± 2.17	2.25 ^{ab} ± 0.18

Values denote arithmetic means ± standard deviation of the mean . Means with different letters (a,b, c, d, etc ,) in the same column differ significantly at $p \leq 0.05$ using DUNCAN test , while those with similar letters are non-significantly different

Data of Table (4) illustrated the mean value of serum Alkaline phosphatase (ALP) (U/L) of hepatointoxicated rats fed on various diets. It could be noticed that the mean value of (ALP)of control (+) group was higher than control (-) group, being 315.00 ±4.00 & 147.00 ±3.40 U/L respectively, indicated significant difference with percent of decrease -53.33 % of control (-) group when compared to control (+)group. All hepatointoxicated rats fed on various diets revealed significant decreases in mean values as compared to control (+) group. Pear tree leaves diet recorded the better ($p \leq 0.05$) treatment of serum ALP.

The mean value of (AST)/ (ALP) ratio of control (+) group was higher than control (-) group (Table 4), Except for group 5 (rats fed on pomegranate peels) all other treatments had no significant ($p > 0.05$) compared to control –ve group (healthy rats). The best treatment considering the (AST)/ (ALP) ratio was recorded for group “3” which showed

nonsignificant difference ($p > 0.05$) in comparison with group 1 (healthy rats).

Data of Table (5) show the mean value of serum total protein (T.P.), albumin and globulin (g/dl) of hepatic rats fed on various diets. The mean value of (T.P.) and albumin of control (+) group were lower ($p \leq 0.05$). All CCL4 injected rats fed on different diets showed significant correction in mean values as compared to control (+) group..The best treatment considering the serum T.P. and albumin were recorded for group “7” and for globulin group (3) numerically.

The mean values of serum total direct and indirect bilirubin (mg/dl) of hepatic rats fed on various diets are illustrated in table 6. It could be noticed that the mean value of these fraction of control (+) group was higher ($p \leq 0.05$) than control (-) group. All CCL4 injected rats fed on different diets showed significant decreases in mean

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values as compared to control (+) group. The best treatment considering the serum

bilirubin fractions when compared to control (+) group was that of group 7 and group 3.

Table (5): Effect of feeding with some vegetables wastes and their combination on total protein albumin and globulin (g/dl) of rats pretreated with CCl₄.

Groups	Parameter	Total. protein (g/dl) (Mean±SD)	Albumin (g/dl) (Mean±SD)	Globulin (g/dl) (Mean±SD)
Control – ve(G1)		11.00 ^a ± 0.57	3.6 0 ^a ± 0.29	7.4 0 ^{bcd} ± 0.66
Control + ve(G2)		9.00 ^f ± 0.6	1.10 ^d ± 0.5	7.90 ^a ± 0.73
7.5% Mango tree leaves(G3)		9.30 ^e ± 0.77	2.20 ^c ± 0.18	7.10 ^d ± 0.69
7.5% Pear tree leaves(G4)		10.30 ^{bc} ± 0.75	2.70 ^b ± 0.18	7.60 ^{ab} ± 0.74
7.5% Pomegranate peels(G5)		10.00 ^d ± 0.60	2.80 ^b ± 0.26	7.20 ^{cd} ± 0.70
7.5% Leek leaves(G6)		10.10 ^{cd} ± 0.75	2.80 ^b ± 0.24	7.3 ^{bcd} ± 0.62
7.5% Mixture of all plants(G7)		10.40 ^b ± 0.99	2.90 ^b ± 0.15	7.50 ^{bc} ± 0.19

Values denote arithmetic means ± standard deviation of the mean . Means with different letters (a, b, c, d, etc ,) in the same column differ significantly at p ≤0.05 using DUNCAN test , while those with similar letters are non-significantly different

Data of Table (6) indicate the mean value of serum Glutathione S Transferase (GST) (ng/mL) of CCl₄ injected rats fed on various diets. The mean value of GST of control (+) group was lower than control (-) group, being 0. 97 ±0.09& 2.54 ±0.21 ng/mL respectively, showing significant (p≤0.05) percent of increase +161.85 % of control (-) group when compared to control (+) group. All hepatic rats fed on different diets revealed significant increases in mean values as compared to control (+) group. Nonsignificant (p≤0.05) difference was observed among rats groups fed on pear tree leaves and pomegranate peels. Group 3 recorded the best treatment for increasing GST.

The mean value of Superoxide Dismutase (SOD) (U/L) of control (+) group was lower than control (-) group, being 50.00 ±2.77& 75.00 ±4.55 U/L respectively table 7.All hepatic rats fed on different diets indicate significant (p≤0.05) increases in mean values as compared to control (+) group. Group 4 revealed maximum efficiency as regards SOD enzyme.

Data of Table (7) also illustrated the mean value of serum catalase enzyme (mmol/L) of hepatic rats fed on various diets.

It could be observed that the mean value of catalase of control (+) group was lower than control (-) group, being 52.50 ±2.15& 70.00 ±3.658 mmol/L respectively, indicating significant difference with percent of increase +33.33 % of control (-) group when compared to control (+) group. All hepatic rats fed on different diets showed significant increases in mean values as compared to control (+) group. Group 3(rats fed on diet containing mango tree leaves) showed maximum improvement as regards catalase enzyme.

The obtained results are in the line with that of *Faramawy, Asmaa (2010) and Shehata,Rehab(2012)*who indicated that injected rats by CCl₄ caused increase of AST , ALT ,ALP ,T bili.,direct bili,indirect bili. & decrease of TP.,Alb.,Glb.,GST,SOD,Catalase enzymes in serum. Guha *et al.*, (1996) found that mangiferin a C-glucosylxanthone purified from mango tree leaves was shown to have in vivo growth-inhibitory activity against ascetic fibrosarcoma (anti-HIV effect) in Swiss mice. Muruganandan *et al.*, (2002) suggested that mangiferin increase antioxidant defence (SOD, catalase) mechanism in cardiac tissue in rats –

induced oxidative damage. Rodeiro *et al.*, (2007) reported that the mangiferin had antioxidant properties and strongly inhibited the decrease of GSH levels in rats induced by t-butyl- hydroperoxide dose. Shah *et al.*, (2010) reported that the various part of plant of mango used as treatment of liver disorder. Pal *et al.*, (2013) demonstrate that mangiferin exhibit both antioxidative and antiapoptotic properties and protects the organ in Pb induced hepatic dysfunction and reduce the levels of serum marker enzymes ALT,ALP. Joseph (2004) reported that pear have antioxidant compounds in extracts from pear fruit. Conjugated flavonoids (quercetin, kaempferol and isorhamnetin), ascorbic acid, carotenoids from the extracts, ascorbic acid and carotenoid contents which protect cells from damages by free radicals. Joana *et al.*, (2010) found that pear presented the highest content of total phenolics., chlorogenic, syringic, ferulic and coumaric acids, arbutin and (-)-epicatechin were detected as major components. Li *et al.*, (2012) found that 8 pear cultivars had antioxidant activity total phenolics, total flavonoids, total anthocyanins, and total triterpenes of eight pear samples, and the monomeric compounds could be a useful source to prevent diseases related to oxidative stress. Singh *et al.*, (2002) showed that feeding rats with pomegranate peels (PP) provided protective effect against

carbon tetra chloride toxicity in rats. Toklu *et al.*, (2007) reported that the level of AST, ALT ,LDH, and cytokine in the serum which were elevated in liver fibrosis models ,considerably reduced and brought to near – normal levels after pomegranate peel treatment..Studies in rats with CCl4 induced liver damage demonstrated pretreatment with PP enhance or maintained the free radical scavenging activity of the hepatic activity of the hepatic enzyme catalase ,superoxide dismutase, catalase and peroxidase and resulted in 54% reduction of lipid peroxidation values compared to controls confirming the antioxidant property of the Pomegranate Peels (Osama *et al.*,2010). Sushil *et al.*, (2013) reported that PP contained phytochemical components that contributed to hepatoprotective. Souri *et al.*, (2004) found that leek and other vegetable showed an antioxidant activity comparable with those of dl-alpha-tocopherol and quercetin. Berneart *et al.*, (2012) indicated that the green leek leaves generally have stronger antioxidant properties. Nasir (2012) showed that effect of leek extract in CCL₄ -intoxicated rats decreased the levels of liver enzymes, total cholesterol, triglycerides and improved lipid profile. Sadeghi *et al.*, (2013) investigated a phytochemical of the leek which protects liver from oxidative stress and damage by free radicals.

Table (6): Effect of feeding with some vegetables wastes and their combination on total bilirubin, direct and indirect bilirubin of rats pretreated with CCl₄.

Groups	Parameter	Total Bilirubin (Mean±SD) (mg/dl)	Direct bilirubin. (Mean±SD) (mg/dl)	Indirect. Bili. (Mean±SD) (mg/dl)
Control – ve(G1)		0.4 ± 0.02 ^e	0.08 ^b ± 0.006	0.32 ^f ± 0.019
Control + ve(G2)		0.7 ± 0.04 ^a	0.11 ^a ± 0.009	0.59 ^a ± 0.043
7.5% Mango tree leaves(G3)		0.45 ± 0.015 ^d	0.09 ^{ab} ± 0.006	0.36 ^e ± 0.029
7.5% Pear tree leaves(G4)		0.54 ± 0.043 ^c	0.10 ^{ab} ± 0.0003	0.44 ^c ± 0.037
7.5% Pomegranate peels(G5)		0.52 ± 0.043 ^c	0.08 ^b ± 0.042	0.42 ^d ± 0.035
7.5% Leek leaves(G6)		0.62 ± 0.058 ^b	0.08 ^b ± 0.006	0.54 ^b ± 0.045
7.5% Mixture of all plants(G7)		0.43 ± 0.019 ^d	0.01 ^{ab} ± 0.007	0.42 ^d ± 0.032

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Values denote arithmetic means \pm standard deviation of the mean . Means with different letters (a,b, c, d, etc ,) in the same column differ significantly at $p \leq 0.05$ using DUNCAN test , while those with similar letters are non-significantly different

Table (7): Effect of feeding with some vegetables wastes and their combination on enzymatic antioxidant of rats pretreated with CCl₄.

Groups \ Parameter	GST Mean \pm SD (ng/ml)	S.O.D (Mean \pm SD) (U/L)	Catalase (Mean \pm SD) (mmol/L)
Control – ve(G1)	2.54 ^a \pm 0.21	75.00 ^a \pm 4.55	70.00 ^a \pm 3.68
Control + ve(G2)	0.97 ^d \pm 0.09	50.00 ^e \pm 2.77	52.50 ^e \pm 2.15
7.5% Mango tree leaves(G3)	1.68 ^b \pm 0.149	52.40 ^d \pm 3.15	63.00 ^b \pm 4.19
7.5% Pear tree leaves(G4)	1.13 ^c \pm 0.103	57.50 ^b \pm 3.29	53.90 ^{de} \pm 3.22
7.5% Pomegranate peels(G5)	1.20 ^c \pm 0.110	53.00 ^d \pm 4.30	57.00 ^c \pm 4.68
7.5% Leek leaves(G6)	1.59 ^b \pm 0.136	55.00 ^c \pm 3.12	55.00 ^d \pm 5.33
7.5% Mixture of all plants(G7)	1.64 ^b \pm 0.059	54.30 ^c \pm 3.21	62.60 ^b \pm 3.11

Values denote arithmetic means \pm standard deviation of the mean . Means with different letters (a, b, c, d, etc ,) in the same column differ significantly at $p \leq 0.05$ using DUNCAN test , while those with similar letters are non-significantly different

4. Effect of feeding with some vegetables wastes and their combination on Kidney function of CCl₄ rats pretreated.

Data of Table (8) indicated the mean value of serum creatinine (mg/dl) of hepatic rats fed on various diets. It could be observed that the mean value of creatinine of control (+) group was higher than control (-) group, being 0.75 \pm 0.033 & 0.60 \pm 0.04 mg/dl respectively, showing significant difference with percent of decrease -20.00% of control (-) group when compared to control (+) group. All hepatic rats fed on different diets revealed significant ($p \leq 0.05$) decreases in mean values as compared to control (+) group. The best treatment considering serum creatinine recorded for group 4(rats fed on diet containing pear tree leaves).

On the other hands the mean value of urea of control (+) group was higher than control (-) group, being 95.00 \pm 8.60 & 66.50 \pm 6.6 mg/dl respectively (table 8). Indicating significant decrease percent of -30.00 % of control (-) group when compared to control (+) group. All hepatic rats fed on different diets revealed significant ($p \leq 0.05$) decreases in mean values as compared to control (+) group. Rats fed on groups 4(pear tree leaves) showed nonsignificant differences with group 1(healthy rats) and recorded the best treatment considering serum urea.

The mean value of uric acid of control (+) group was higher than control (-) group, being 3.10 \pm 0.173 & 2.20 \pm 0.046 mg/dl, respectively (table 8). . All CCl₄ injected rats fed on various diets revealed significant ($p \leq 0.05$) decreases in mean values as compared to control (+) group. The best treatment was recorded for group 7(mixture

of all plants) when compared to control (-) group.

It seems possible that the used plants could correct the changes in kidneys function due to injection of rats with CCL₄.

Table (8): Effect of feeding with some vegetables wastes and their combination kidney function of rats pretreated with CCl₄.

Groups	Parameter	Creatinine (mg/dl) (Mean±SD)	Urea (mg/dl) (Mean±SD)	Uric Acid mg/dl (Mean±SD)
Control – ve(G1)		0.60 ^e ± 0.04	66.50 ^e ±6.60	2.20 ^d ±0.046
Control + ve(G2)		0.75 ^a ± 0.033	95.00 ^a ± 8.60	3.10 ^a ± 0.173
7.5% Mango tree leaves(G3)		0.65 ^c ± 0.043	85.50 ^b ±8.30	2.9.00 ^b ± 0.19
7.5% Pear tree leaves(G4)		0.55 ^f ± 0.026	65.33 ^e ±6.53	2.30 ^d ± 0.21
7.5% Pomegranate peels(G5)		0.60 ^e ± 0.033	76.00 ^d ± 0.76	2.70 ^c ± 0.26
7.5% Leek leaves(G6)		0.67 ^b ± 0.055	80.70 ^c ± 7.97	2.60 ^c ± 0.25
7.5% Mixture of all plants(G7)		0.63 ^d ± 0.052	61.00 ^f ±3.80	2.20 ^d ± 0.19

Values denote arithmetic means ± standard deviation of the mean . Means with different letters (a,b, c, d, etc ,) in the same column differ significantly at p ≤ 0.05 using DUNCAN test , while those with similar letters are non-significantly different

The obtained results are in agreement with that of Faramawy, Asmaa (2010) and Shehata, Rehab (2012) who indicated that injected rats by CCl₄ caused increase of creatinine , urea & uric acid in serum. Prabhu *et al.*, (2005) reported that mangiferin from mango leaves had cardiotoxic and diuretic properties. Abolfazl *et al.*, (2011) found that Hydroalcoholic extract of Mango reduce plasma Cr and urea concentrations significantly. Naya (2008) reported that pear stimulates the urine elimination, and suitable in those cases in that it is necessary to stimulate the kidneys to increase micturition, in illnesses like: kidney pain (nephritis), renal calculation, renal inadequacy and inflammation of urinary bladder. Cekmen *et al.*, (2012) reported that pomegranate extract had a protective effect against gentamicin induced nephrotoxicity in rats. Anon-g (2013) found that leek is listed among natural diuretics because it contains a

lot of potassium (300mg potassium in 100g leek) and little sodium . On the other hand Anon-f (2013) suggested that people with untreated or existing kidney and gall bladder problems should avoid the consumption of leeks on a regular basis . This is because of the fact that leek contains considerable amount of oxalates, which, when becomes too concentrated in the human body crystallize and create health problems.

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تقييم الاثر الصحى لبعض مخلفات الخضر على خلل الكبد للفئران التى سبق معاملتها برابع كلوريد الكربون

فاطمه الزهراء امين الشريف ، أيمن السيد العدوى ، شيماء مصطفى المصيلحى ،
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قسم التغذية وعلوم الاطعمه . كلية الاقتصاد المنزلى - جامعه المنوفية

الملخص العربى

أجريت هذه الدراسة لمعرفة التأثير الصحى المحتمل لبعض اوراق النبات على الخلل الفسيولوجى فى كبد الفئران المصابه المحقونه برابع كلوريد الكربون . تم استخدام ٣٥ فأر ابيض بالغ يتراوح وزن كل منها (١٥٠ ± ١٠) جم وتم تقسيمهم الى الى ٧ مجموعات متساويه , وتركت احداها كمجموعه ضابطه سالبه اما المجموعات الست الاخرى فتم احداث تسمم للكبد فيها باستخدام رابع كلوريد الكربون بجرعه ٣ مل / كجم من وزن الجسم مرتين اسبوعيا لمدة ٣ اسابيع . واضيف مسحوق اوراق النباتات المختلفه بنسبه ٧.٥% من : اوراق شجر المانجو , واوراق شجر الكمثرى , واوراق نبات الكراث , وقشر الرمان واعطيت فى صورته خليط . فى نهايه فتره التجربه (٢٨يوما) تم تجميع عينات الدم من الفئران لقياس العوامل التالفيه انزيمات الاكسده (SOD,CAT,GST) وانزيمات الكبد (ALT,AST,ALP) والبروتين الكلى , والبيومين السيرم , وجلوبيولين السيرم وحساب نسبة الالبيومين / الجلوبيولين , والكوليسترول الكلى , والجليسيريدات الثلاثيه , والبروتينات الدهنيه (HDL-c, LDL-c, and VLDL-c) , ودليل تصلب الشرايين (AI) وحمض اليوريك , و نيتروجين اليوريا , والكرياتينين فى السيرم . سجلت كلا من اوراق شجر المانجو وقشر الرمان انخفاضاً تدريجياً فى معدل الكوليسترول الكلى واطهرت اوراق شجر الكمثرى افضل النتائج فى تحسين انزيمات الكبد بينما حققت اوراق الكراث تحسينات لوظائف الكبد والكلى . و كانت افضل هذه النتائج باستخدام خليط اوراق هذه النباتات ووفقاً للنتائج فانه يمكن ايضا استخدام اوراق شجر المانجو واوراق شجر الكمثرى واوراق نبات الكراث وقشر الرمان, لتحسين وظائف الكبد والوقايه من التسمم الكبدى. **الكلمات المفتاحيه :** الكبد , تسمم الكبد , وظائف الكبد , وظائف الكلى , البروتينات الدهنيه , اوراق النبات , اوراق شجر المانجو ,اوراق شجر الكمثرى , اوراق نبات الكراث , قشر الرمان