THE PROTECTIVE EFFECT OF PECTIN EXTRACTED FROM ORANGE AND POMEGRANATE PEELS AND CELLULOSE EXTRACTED FROM ORANGE ON CARBON TETRACHLORIDE INDUCED HEPATOTOXICITY IN MALE ALBINO RATS

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ABSTRACT

Dietary fibers play an important role in the maintenance of health. In addition, higher dietary fibers intake is correlated with reduced incidence of disease and mortality. Pectin from orange, pomegranate peels, cellulose from orange peels and anthocyanin extracted from pomegranate have shown to possess significant antitoxic activity against CCl4. This study aimed to evaluate the protective effect of pectin extracted from orange, pomegranate peels and cellulose extracted from orange peels and anthocyanin extracted from pomegranate on albino rats induced hepatotoxic stress by treating with CCl4. To accomplish this aim, twenty-five male albino rats were divided into five groups. The groups were normal or negative control group (NC 1), the second, third, fourth and fifth groups were intraperitoneally (i.p.) injected by (1ml/kg) CCl4 dissolved in paraffin oil (1:1 (v/v)) twice every week for two months. Meanwhile groups 3, 4, and 5 were administered orally orange pectin, pomegranate pectin, and cellulose extracted from orange peel (50 mg/kg), respectively. All groups were orally administered with anthocyanin (250 mg/kg) extracted from pomegranate peels twice each week for two

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months as antioxidants supplemented substance except group 1 (normal or negative control) and second group CCl4 group which considered as positive control group. Blood samples after 15, 30 and 60 days at the end of the experiment were taken from the eye artery and collected in vacuum tubes clot activator to get serum, besides EDTA tubes to analyze blood picture. Serum analysis was preferred to measure ALT, AST, T.Protein, S.Albumin, total Cholesterol, Triglycerides, Sodium (Na), Potassium (K) and blood samples in EDTA tubes were used for analyze complete blood picture. The obtained results reveal that ALT and AST levels were increased due to i.p. injection of CCl4, meanwhile, orally administration of pectin extracted from both orange and pomegranate peels besides cellulose from orange peel led to reback ALT and AST levels and other biomarker to be close to normal control rats. This finding shows that dietary fibers play an important protective agent against toxicity induced by CCl4 and attenuate its harmful effect.
Key words: Rats, pectin, dietary fiber, carbon tetra chloride, cellulose, anthocyanin, orange peel, pomegranate peel, blood analysis.

INTRODUCTION

Dietary fibers play an important role in the maintenance of health. In addition, higher dietary fiber intake is correlated with a reduced incidence of disease and mortality (Fardet, 2010 and Renolds *et al*., 2019). The neediness to find alternative fiber sources have been the subject of many researchers worldwide nowadays. Citrus is one of the most consumed types of productivity as well as for their wholesome nutritional properties consisting of vitamin C, A, and B, minerals (Calcium, Phosphorus, Potassium), dietary fibers and many photochemical such as flavonoids, amino acids, terpenes, phenolic acids, and carotenoids (Melendez-Martine *et al*., 2008 & Roussos, 2011).

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However, the consumption of citrus fruits has led to generate residue (peel, pulp, seeds) which accounts approximately for 50% of the fruit weight besides its moisture content (Perez-Alvarez *et al*., 2001) and Castello-Garcia *et al*., (2011). These huge number of wastes are considered as an agricultural waste that contribute to the environmental pollution. Due to its composition being rich in soluble and insoluble carbohydrates units by product shows great potential for the recovery of fibers which can be further used as functional food ingredient. Citrus peel can be divided into two parts namely albedo (inner part or mesocarp) and flavedo (outer part or epicarp). Albedo the white spongy and cellulosic tissue has high fiber content, additionally, it is also possessed good water and oil holding capacity as well good colonic ferment ability and low caloric content (Figuerola *et al*., 2005).

Cellulose is a long chain polymer with repeating units of D-glucose, called pyranose which are joined by single oxygen atoms between the C-1 of one pyranose ring and the C-4 of the next ring called β-1-4 linkages (Kim *et al*., 2006), while each β-1-4-glucopyranose bears three hydroxyl groups and is able to form intra and intermolecular hydrogen bonds that play a major role in determining the physical properties of cellulose (John and Thomas, 2008).

Pectin is a complex mixture of polysaccharides occurring in the primary cell walls terrestrial plants, is a higher eatable functional food ingredient, it consists of linear backbone of α (1-4)–D Galacturonic acid residue partially esterifies with methanol with L-Rhamnose residues that make the backbone

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irregular and with some other neutral sugar present as side chains. The general makeup of the pectin content varies with the ripening of the fruit (Wilkins *et al*., 2005). Pectin is produced commercially in the form of white to light brown powder mainly extracted from citrus fruits and is used in food as gelling agent particular in jams and jellies sweets, milk, drinks and as a source of dietary fiber (Tobias *et al*., 2011). In addition, Srivastava and Malviya (2011) mentioned that pectin did not contribute significantly to the nutrition, but pectin is natural part to the human diet which good to the human intestine furthermore consumption of pectin has been shown to stabilize blood pressure and reduce serum cholesterol level. On the other hand, pomegranate peel has health benefits including antibacterial, antioxidant, anti-inflammatory, and anti-mutagenic properties (Elwej *et al*., 2016).

This study aimed to evaluate the protective effect of pectin extracted from both orange and pomegranate peels and cellulose extracted from orange peels against oxidative stress in male albino rats induced toxicity and hepatotoxicity by carbon tetrachloride (CCl4).

MATERIAL AND METHODS

1) **Chemicals**: All reagents and chemicals used for pectin, cellulose and anthocyanin extraction process were purchased from Oxford Laboratory Reagent India (International Trade Assoc. Egypt) and Ethyl alcohol from Sphinx Chem. Egypt.

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2) Pectin, cellulose and Anthocyanin extraction: Extraction of pectin, cellulose from orange peel and pectin from pomegranate albedo peel were conducted. Orange and pomegranate fruits were obtained from local market and the peels were separated and used to extract all of pectin, cellulose from citrus fruit albedo peel and pectin from pomegranate albedo peel and anthocyanin from pomegranate peel as follow:

Sample preparation: Orange and pomegranate were peeled, then the peels were washed in order to remove dirties, dust and the residues. After that the albedo were separated and cut into small pieces and dried in a convection oven CBM Italy at 50oC for 48 hours. Dried sample was grinded to consistency intermediate by using (dry blander, Mienta) and the powder stored at ambient temperature for further use (Zain, 2014 and Kanmani et al., 2014).

3) Pectin extraction process: A dry mass of 5gm orange peels was subjected to extract by adding 90 ml of distilled water followed by 10 ml of citric acid with pH value 3 then the mixture was heated at 40 – 90 oC for 1/hour. The hot acid extract was filtered through whatman No 1 filter paper. The filtrate was coagulated using an equal volume of 95% ethanol alcohol and left for 2 hours to allow the pectin to float on the surface. The gelatinous pectin flocculants were then skimmed off. The extracted pectin was then filtrated and washed 2-3 times with ethyl alcohol to remove any remaining

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impurities. Finally, the precipitate was dried at 35-40 oC in hot air oven and yield percentage was calculated (Kanmani *et al*., 2014) as follow:

$$\text{Pectin (\%)} = \frac{P}{Bi} \times 100$$

Where:

P is the amount of extracted pectin in g.

Bi is the initial amount of fruit peel powder

4) Preparation of cellulose: The orange was peeled and albedo was separated and cut into small pieces and dried in a convection oven at 50oC for 48 hours. Dried sample was grinded by using (dry blender, Mienta) and the powder formed was sieved until certain sizes were achieved. Extraction of cellulose was performed by using alkaline treatment followed by bleaching (Zain, 2014) as follow:

A 50g of dried albedo powder were weighed and transferred into a round bottom flask. Alkali solution (4 wt% NaOH) was added and the treatment was performed at reflux condition at 100–120 oC for 2h. The mixture was then filtered and washed with distilled water several times to remove lignin and hemicelluloses that dissolved in the solution. The resultant fiber was dried before used for bleaching treatment.

Bleaching treatment was performed at reflux condition at 110– 130 oC for 4 hours after adding 60 g of fiber into 400 ml of each solution of 1.7% NaClO2, acetic buffer and distilled water. The mixture was then allowed to

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cool before filtered and washed with distilled water until white cellulose was obtained. The cellulose obtained was dried at room temperature (Zain 2014).

5) Extraction of anthocyanin

Sample collection: Pomegranate peel was supplied locally from the market then dried far of sun light, the samples were grounded by blender and kept in polyethylene bags at room temperature until used according to (Abadi, 2015).

Procedure:

Eight grams of pomegranate peels were soaked in (50 ml) methanol containing l% of hydrochloric acid for (5-10) minutes, then the solution was filtered using filter paper (Whatman No.1), then the filtrate was air dried and leave the solution in plate glass (Petri dish) uncovered in the shade at room temperature to dry where they are getting sticky substance amorphous (Harborne, 1984).

6) Experimental rats design: The biological experiment was performed in laboratory of experimental animal in Faculty of Pharmacy, Cairo University. Twenty-five waster Swiss albino rats with mitral weight 100-120 g were used in present study. The rats were divided into five groups, five rats for each group and housed in cages a room temperature 18–20 oC with 40–60% humidity as recommended by Yalcin and Fazilet (2002). The groups were normal untreated control group1, while the second, third, fourth and fifth groups were intraperitoneal injected by (1ml/kg) CCl4 diluted with paraffin oil [1:1 (v/v)] twice every week for two months.

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Meanwhile groups 3, 4 & 5 were administered orally pectin extracted from both orange and pomegranate peels and cellulose extracted from orange (50 mg/kg), respectively. All groups administered orally anthocyanin (250 mg/kg) twice each week for two months antioxidants supplemented substance except groups 1 & 2 normal negative control and positive control of CCl4 groups. Blood sampling was withdrawn after 15, 30 days and at the end of the experiment after 60 days. Blood was taken from the eye artery and collected in vacuum tubes clot activator. Then blood samples were centrifuged at 3500 rpm for 15 min to separate the serum. Rat's diet composition was: corn starch 65%, casein 10%, corn oil 10%, salt mixture 4%, vitamin mixture 1% and Cellulose 10% according to Osfor *et al*., 2013. Temperature of 18 – 20 o C with 40 – 60 % humidity were adopted according to Inan and Fazilet (2002)

7) **Blood analysis methods:** Serum analysis was preferred to measure transaminase enzymes ALT according to Reitman, and Frankel (1957), while AST was determined as mentioned by Sherwin (1984), Albumin as reported by Douma *et al*. (1972), total protein according to Douma *et al*. (1981), total cholesterol according to Ellefson and Caraway (1976), triglycerides (TG) according to Vassault *et al.* (1986). Na and K were determined as described by Henry (1974) and by using Flame Photometer (Model FP8500). However, Both WBC and Platelet were measured using

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Abacus3, Hematology Analyzer, DIATRON MI PLC .H- 1038 Budapest, Papirgyaru.58-59, Hungary.

8) **Statistical Analysis:** Statistical analysis was performed according to Everitt and Anders (2010), the Cambridge Dictionary of statistics and Richard G. Brereton (2018).

RESULTS AND DISCUSSION

1) **Liver transaminase enzymes (ALT & AST)** The enzyme alanine aminotransferase (ALT) is widely distributed with concentration in liver and to a lesser extent in kidney, heart, skeletal, muscles, pancreas and lungs. Elevated serum ALT is found in hepatitis, cirrhosis, obstructive jaundice, carcinoma at the liver and chronic alcohol abuse. Although both serum ALT, AST become elevated whenever disease processes affect liver cell integrity. ALT is the more liver specific enzyme. The enzyme aspartate aminotransferase (AST) is widely distributed in erythrocytes and tissues, principally, heart, liver, muscle and kidney. Elevated serum levels are found in diseases involving these tissues such as viral hepatitis and muscular dystrophy. So, it was hypnotist to know the effect dietary fiber on AST, ALT is more sensitive to acute liver injury test, whereas AST is more sensitive to chronic injury according to Young (1990).

Data in Tables (1 & 2) represent serum ALT & AST values in rats treated with CCl4 and orally administrated cellulose from orange and pectin

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as dietary fiber extracted from two sources of agricultural wastes (orange and pomegranate pectin) and orally were taken anthocyanin twice each week for two months. Results in Table (1) show that serum ALT was increased to 110.88 U/L due to CCl4 treatment compared to non-treated control group 24.27 U/L. Meanwhile administration of pectin either extracted from orange or pomegranate peels led to decrease this elevation to 95.51 & 79.51 and 80.79 U/L compared to 110.88 U/L in group treated only with CCl4. Despite orange or pomegranate pectin or cellulose from orange lower ALT in serum of rats but the level is still higher than in rats non-treated with CCl4 (normal control) 24.27 U/L. However, results in Table (2) show that serum AST was also increased to 103.56 U/L due to CCl4 treatment compared to normal control group 67.67 U/L. Meanwhile administration of pectin either extracted from orange or pomegranate peels or cellulose from orange led to decrease this level to 72.56, 65.78 and 68.44 U/L compared to 103.56 U/L in group treated only with CCl4, despite orange pectin or pomegranate pectin or cellulose from orange lower AST in serum rats but the level is still higher than in rats non-treated with CCl4 (normal control 67.67 U/L.

Results in Table (1 & 2) show that intraperitoneal injection with CCl4 induces elevation in ALT and AST levels in treated rats compared to control. However, rats kept on pectin as dietary fiber extracted from both orange or pomegranate pectin or cellulose from orange led to decrease this elevation significantly compared to positive group treated with CCl4, but the level is

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still higher than the normal control value. These results indicate that pectin extracted from orange or pomegranate peels or cellulose from orange could be inhibit ALT and AST elevation that happen due to oxidative stress and toxicity induced by CCl4 treatment. Therefore, it could be said that pectin and dietary fiber protect liver from oxidative stress and work as hepato-protective agent against CCl4 toxicity. Moreover, results in Table (1 & 2) appear that pomegranate pectin or cellulose from orange lower ALT value more than orange pectin. This result inversely happens in AST that lower by orange pectin more than pomegranate pectin or cellulose from orange. This finding of pectin in lowering CCl4 oxidative stress and toxicity may be due to that pectin could work as scavenger agent against ROS that produce by CCl4 oxidative stress, besides pectin hydrolysate produces galacturonic acid which works as a detoxifying agent.

These obtained results are in accordance with Middha *et al*. (2013) who noticed the protective effects of pomegranate peel extracts on oxidative stress in mice. Also, Sen *et al*. (2010) indicated to hepato–protective activity of pomegranate on the liver injury. In this regard, Wang *et al*. (2010) noticed that cellulose could significantly decrease ALT, AST in serum levels in rats. Besides, Sadeghipour *et al*. (2014) found that pomegranate peel extract significantly reduced ALT, AST levels in serum of rats fed on heavy lipid diet. Also, Murthy *et al*. (2002) observed that in-vivo study on rats fed pomegranate peel, resulted in protective activity to CCl4 toxicity.

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This mechanism of lowering ALT& AST could be interpreted due to hepato-protective activity of pectin extracts as demonstrated by Renjie *et al*. (2017) who confirmed the protective effect of polysaccharides against carbon tetrachloride that induced liver injury in rats and demonstrated that treatment of rats with Glycyrrhiza glabra polysaccharides significantly prevented the increased activities of ALT & AST. Moreover, Wenfeng *et al*. (2018) reported that in nonalcoholic fatty liver disease in mice, pectin dose dependently generated an increase in acetic acid and propionic acid contents and significantly increased the relative abundance of buteroides, parabacteroides, olsenella, Bifid bacterium in the gut of HF-fed (High fat diet) micro biota and short-chain fatty acids (SCFAS) may thus contribute to the well-established link between pectin consumption and nonalcoholic fatty liver disease (NAFLD). Moreover, Osfor *et al*. (2013) studied the hypocholesterolemia and hypoglycemic effects of orange albedo powder (Citrus aurantium L.) on male albino rats and revealed that, the groups of rats which received diets supplemented with 10% and 20% orange albedo powder significantly decrease ALT, AST compared with positive control. The hepatoprotective effect of extracted sweet orange peel and its biological compound on oxidative stress in vivo, were investigated. This study demonstrated that citrus peel protects rat liver from CCl4 induced injury by attenuating hepatic oxidative stress ad can be used as a therapeutic

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antihepatoxic agent for the treatment of hepatic injury as coincided by Chen *et al*. (2013).

Table (1): ALT (U/L) in serum of rats treated by CCl4 and fed on cellulose extracted from orange and pectin extracted from orange and pomegranate peels.

Groups / Phases blood draw in month	Negative control group	Positive control group (CCl4)	CCl4+Pectin from orange peels	CCl4+ Pectin from pomegranate peels	CCl4+ Cellulose from orange peels	Total	One Way ANOVA test	
							F	P-value
First	17.79 a ±3.06	166.09** ±11.41	137.36**,a ±34.84	144.32**±22.56	155.03**, a ±5.18	124.12 a ±58.31	28.993	0.000
Second	22.22 a ±6.89	101.05** ±43.34	104.17**,a ±28.82	58.50 B ±18.38	39.33 A, a ±3.79	62.29 a,b ±41.38,	8.303	0.001
Third	32.80 a ±4.66	65.50**± 2.12	45.00 B,a ±11.31	37.00 A ±7.07	48.00**, C, a ±12.49	43.07 b ±13.21	6.532	0.009
General mean	24.27±4.87	110.88** ±18.95	95.51**±24.99	79.94**,A ±16.00	80.79**,B ±7.15	76.50± 37.63	20.378	0.000

*: Significant difference from normal control at P <0.05;
**: Highly significant difference from normal control at P <0.010.
Small letters mean significant differences between groups duration.
Capital letters mean significant differences from positive control (CCl4) group

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Table (2): AST (U/L) in serum of rats treated by CCl4 and fed on cellulose extracted from orange and pectin extracted from orange and pomegranate peels.

Groups / Phases blood draw in month	Negative control group	Positive control group (CCl4)	CCl4+Pectin from orange peels	CCl4+Pectin from pomegranate peels	CCl4+Cellulose from orange peels	Total	One Way ANOVA test	
							F	P- value
First	78.00 a ±22.61	99.33 a ±4.04	86.33 a ±8.14	95.33 a ±14.47	94.33 a ±2.31	90.67 a ±13.32	1.343	0.320
Second	53.00 a ±4.36	106.33 a, **±15.31	63.00 a , C ±5.29	39.67 a , A ±8.50	42.67 a , B ±5.03	60.93 a ±26.05	28.836	0.000
Third	72.00 a ±16.82	105.00 a ±5.57	68.33 a ±15.70	62.33 a ±28.92	68.33 a ±25.50	75.20 a ±23.25	2.122	0.153
General mean	67.67±14.60	103.56**±8.31	72.56 C ±9.71	65.78 A ±17.30	68.44 B ±10.95	75.60±20.88	7.867	0.001

*: Significant difference from normal control at P <0.05;
**: Highly significant difference from normal control at P <0.010.
Small letters mean significant differences between groups duration.
Capital letters mean significant differences from positive control (CCl4) group.

2) Serum minerals (Sodium and Potassium): Hyponatremia is a common finding in patients with decompensated cirrhosis due to an abnormal regulation of body fluid homeostasis. In past years, hyponatremia has attracted interest as possible prognostic factor for liver cirrhosis according to Kim *et al*. (2009). Serum potassium concentration range widely in patient with chronic liver disease. Both hypokalemia and hyperkalemia may occur, but usually normokalemia is observed. Early studies revealed a 40% prevalence of hypokalemia in cirrhotic patients, irrespective of the

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disease stage. Aim: to determine the frequency of hypokalemia in patients with chronic liver disease according to Izhar, *et al*. (2022).

Data in Tables (3 & 4) represent serum Na & K values in rats treated with CCl4 and orally administrated cellulose from orange and pectin as dietary fiber extracted from two sources of agricultural wastes (orange and pomegranate pectin) and orally were taken anthocyanin twice each week for two months. Results in Table (3) show that serum Na were 140.91-142.19 mmol/L in negative control & CCl4 groups. These values ascertained that serum sodium in treated rats did not change significantly due to CCl4 treatment and still close to the values of normal rats. These results demonstrate that CCl4 or pectin or cellulose extracts and anthocyanin administration have not affected change sodium level parameter compared to normal control group which non-treated or treated with CCl4 and the value was nearest to control group.

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Table (3): Sodium (Na) mmol/L in serum of rats treated by CCl4 and fed on cellulose extracted from orange and pectin extracted from orange and pomegranate peels.

Groups / Phases blood draw in month	Negative control group	Positive control group (CCl4)	CCl4+ Pectin from orange peels	CCl4+ Pectin from pomegranate peels	CCl4+ Cellulose from orange peels	Total	One Way ANOVA test	
							F	P-value
First	135.33 a ±0.58	135.33±0.58	132.33**, B,a ±2.08	129.67**,A ±0.58	133.67±0.58	133.27 a ±2.37	14.912	0.000
Second	148.40 b ±1.34	152.25** ±1.89	150.33 a ±2.31	149.17 B ±2.02	148.33 A ±1.53	149.69 b ±2.22	3.302	0.045
Third	139.00 a,b ±1.58	139.00±1.41	137.50 a ±2.12	141.00±0.00	137.33±1.53	138.71 c ±1.77	2.093	0.164
General mean	140.91±1.17	142.19±1.29	140.06±2.17	139.94±0.87	139.78±1.21	140.56 ±2.12	2.522	0.073

:*Significant difference from normal control at P <0.05 ;
:**Highly significant difference from normal control at P <0.010.
Small letters mean significant differences between groups duration .
Capital letters mean significant differences from positive control (CCl4) group.

Data in Table (4) show potassium level in serum of rats treated i.p. with CCl4 and kept on cellulose from orange and two types of pectin extracted from orange and pomegranate peels as two agricultural wastes and administrated anthocyanin two times every week for two months. Results in Table (4) reveal that serum potassium level was elevated to 6.94 mmol/L in serum rats treated with CCl4 compared to 4.42 mmol/L in normal control group. Meanwhile, this elevation was lowered to 5.49, 5.93 & 6.38 mmol/L in

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serum rats treated with pectin extracted from both orange and pomegranate peels or cellulose from orange respectively. This result cleared that CCl4 resulted in increased serum K level about one and half folds compared to control untreated group while treatment with pectin either extracted from orange peel or pomegranate peel or cellulose from orange led to lower serum K level in treated rats. Also, in this connection orange pectin was lowered K more than pomegranate pectin or cellulose from orange which reflect potentiality of orange pectin in maintaining K serum level near to normal range from alkalinity and protect body from CCl4 toxicity. This phenomenon discloses the protective effect of pectin and cellulose to safe biological system from CCl4 toxicity. These results agree with study of Sayed et al. (2012) who found that 50% fiber, 30 soluble fiber and 20% insoluble fenugreek fiber show that the mechanism under lying the protection effects of fenugreek, levels of urea, creatinine, sodium, and potassium, found that the dietary fiber of fenugreek significantly reduces the high levels of urea, creatinine, sodium, and potassium in serum compared with, diabetic untreated group. The obtained results indicated that the serum contents of monovalent mineral were negatively affected by pectin administration (El-Zoghbi and Sitohy 2001). Moreover, the effect of CCl4 on Na/K-ATPase and Ca2+/Mg2+ATPase activates on erythrocytes membranes and trace element levels of serum erythrocyte and liver tissue of rats. ATPase were significantly reduced in CCl4 treated group ($P<0.01$, $P<0.001$) trace element levels also showed

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statistically significantly differences in CCl4–treated group membrane Na/K-ATPase and Ca2/Mg2ATPase activates were diminished due to CCl4-induced injury on erythrocyte membrane and liver by altering the mineral substance concentration (Fahretin *et al*., 2009). Also, in study of carbon tetra chloride induced hepatotoxicity, the force feeding of carbon tetrachloride resulted in a) reduction in potassium content of rat liver mitochondria, b) an impairment in the ability of potassium depleted mitochondria to accumulate potassium, d) an increased water content of the mitochondria, as shown by Share and Richard (1959). The study of Grudeva and Sirakova (1998) found that water- soluble dietary fibers are widely used in preventive and curative nutrition, especially in cases of hyper lipoproteinemia occurrence of disturbances of the electrolytes and trace elements metabolism causing an effect in long-term treatment with pectin can be theoretically substantiated the results of the comparative assessment of the electrolytes (sodium, potassium, chloride, ionized calcium, total and ionized magnesium) failed to reach statistical significance during the administration of pectin product. Neither did the serum level of trace elements (iron and copper) change significantly during the observed period.

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Table (4): Potassium (k) mmol/l in serum of rats treated by CCl4 and fed on cellulose extracted from orange and pectin extracted from orange and pomegranate peels.

Groups / Phases blood draw in month	Negative control group	Positive control group (CCl4)	CCl_4+ Pectin from orange peels	CCl_4+Pectin from pomegranate peels	CCl_4+Cellulose from orange peels	Total	One Way ANOVA test	
							F	P- value
First	$5.73^{a,b} \pm 1.11$	$10.53^{**,a} \pm 2.00$	$6.43^{A,a} \pm 0.29$	$7.50^{B,a} \pm 1.61$	$7.93^{C,a} \pm 0.12$	$7.63^{a} \pm 2.01$	6.421	0.008
Second	$5.52^{a} \pm 0.31$	$5.60^{a} \pm 0.22$	$5.63^{a,b} \pm 0.25$	$5.37^{a} \pm 0.21$	$6.07^{a,b} \pm 1.06$	$5.62^{b} \pm 0.47$	0.917	0.483
Third	$2.00^{b} \pm 1.87$	$4.70^{a} \pm 3.38$	$4.40^{b} \pm 0.92$	$4.93^{a} \pm 0.90$	$5.13^{b} \pm 0.23$	$4.01^{b} \pm 2.18$	1.817	0.186
General mean	4.42±1.10	6.94**±1.87	$5.49^{*,A}$ ±0.49	5.93**±0.91	6.38**±0.47	5.75±1.55	3.803	0.019

*: Significant difference from normal control at P <0.05;
**: Highly significant difference from normal control at P <0.010.
Small letters mean significant differences between groups duration.
Capital letters mean significant differences from positive control (CCl4) group.

3) Albumin profile: Albumin is the most abundant plasmatic protein. Albumin is an important compound as it constitutes nearly 50% of the total plasma protein, while its role in maintaining osmotic pressure in the plasma is well known, (Walayt et al 2017). Liver cirrhosis associated with decreased levels of albumin as well as disturbed albumin function according to Carvalho and Machado (2018). The serum albumin concentration is used to evaluate chronic liver disease and hepatocellular function (protein syntrhesis) (Gropper, 2020).

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Data in Table (5) show Albumin level in serum of rats treated i.p. with CCl4 and kept on cellulose from orange and two types of pectin extracted from orange and pomegranate peels as two agricultural wastes and administrated anthocyanin two times every week. Results in Table (5) reveal that serum Albumin level was slightly decreased in groups treated with CCl4 to 3.63 g/dl in compared to 4.0 g/dl in normal control. Meanwhile, this lowering was found in rats serum treated with pectin extracted from both orange and pomegranate peels and cellulose from orange to record 3.56, 3.43 & 3.78 g/dl respectively. This result clear that CCl4 resulted in decrease serum Albumin level while treatment with pectin either extracted from orange peel or pomegranate peel could not return serum Albumin level to normal range for untreated rats. Generally, administration of pectin from orange and pomegranate peels or cellulose from orange could not increase this slightly lowering compared to normal control group and the level was not equal to control group. These results are in concomitant with Linyan et al. (2020) who studied the effects of barley insoluble fiber (Bif) and barley soluble fiber (Bsf) isolated from barley on serum lipids, liver function and cecal short–chain fatty acids in type 2 diabetic and normal rats. Results showed that effect treatment of insoluble and soluble Fibers reduced FBS in diabetes condition with cecal level of propionic acid due to insulin sensitivity improved by (Bsf) respectively. The two treatments further ameliorative liver function (ALT, AST, S.Alb, T. protein). These results are in according with Rotenberg and

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Jakobsen (1978) who reported that replacing 9.6% of starch in rat's diet with pectin led to lower, but still within the normal limits, serum concentration of total protein, albumin and α1 globulin. They attributed these results to that swelling of pectin inside the intestines may be responsible for the fact that rats did not consume larger amounts of food though the food had a lower energy value.

Table(5): Albumin g/dL in serum of rats treated by CCl4 and fed on cellulose extracted from orange and pectin extracted from orange and pomegranate peels.

Groups / Phases blood draw in month	Negative control group	Positive control group (CCl4)	CCl_4+Pectin from orange peels	CCl_4+Pectin from pomegranate peels	CCl_4+Cellulose from orange peels	Total	One Way ANOVA test	
							F	P-value
First	4.27^{a} ±0.32	3.97±0.06	4.03^{a} ±0.68	3.83±0.15	4.40^{a} ±0.36	4.10^{a} ±0.39	1.094	0.411
Second	4.10^{a} ±0.16	3.48** ±0.21	3.43**^{a} ±0.25	3.50**±0.30	3.29**^{a} ±0.26	3.62^{a,b} ±0.37	8.258	0.002
Third	3.62^{a} ±0.31	3.45±0.07	3.20^{a} ±0.57	2.95±0.35	3.63^{a} ±0.15	3.44^{b} ±0.37	2.212	0.148
General mean	4.00±0.26	3.63±0.11	3.56±0.50	3.43±0.27	3.78±0.26	3.72± 0.38	2.549	0.071

*: Significant difference from normal control at P <0.05;
**: Highly significant difference from normal control at P <0.010.
Small letters mean significant differences between groups duration.
Capital letters mean significant differences from positive control (CCl_4) group.

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4) Total protein profile: Plasma proteins are synthesized predominantly in the liver plasma cells lymph nodes, spleen and in bone marrow. In the case of disease, the total protein concentration and also percentage represented by individual fractions can significantly deviate from normal values. Hyperproteinemia can be observed in cases of severe dehydration and illnesses such as myeloma and cirrhosis of liver so total protein measurements are used in the diagnosis and treatment of variety of diseases involving the liver according to Dumas *et al*. (1981).

Data in Table (6) show total protein level in serum of rats treated with i.p CCl4 and fed on cellulose from orange and two types of pectin extracted from orange and pomegranate peels as two agricultural wastes and administrated anthocyanin two times every week. Results in Table (6) reveal that serum total protein level was elevated to 8.25g/dl in rats serum treated with CCl4 compared to 7.34 g/dl in normal control. Meanwhile, this elevation was lowered to 7.42 & 7.0 & 7.2 g/dl in rats serum treated with pectin extracted from orange and pomegranate peels and cellulose from orange respectively. This result clear that CCl4 resulted in increased serum total protein level about 12.3% compared to normal control while, treatment with pectin either extracted from orange peel or pomegranate peel and cellulose from orange led to maintain serum T. protein level in normal range for untreated rats. Also in this trend pectin extracted from orange and pomegranate peels as well as cellulose from orange peel give approximately T.protein value close to NC

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group which reflect the ability of pectin and cellulose from orange peel in maintaining total protein serum level at normal range and protect body from CCl4 toxicity. These results disclose the protective effect of pectin to safe liver from CCl4 toxicity. These results are in accordance with Rotenberg and Jakobsen (1978) who reported that replacing 9.6% of starch in rats' diet with pectin led to reduced body weight and reduced carcass fat lower, but still within the normal limits, serum concentration of total protein, they attributed these results to that swelling of pectin inside the intestines may be responsible for the fact that rats did not consume larger amounts of food though the food had a lower energy value. Also, these results are agreed with Quazi *et al.* (1983) they noticed that pectin depressed the elevation serum total protein, triglycerides, cholesterol, and it was not for cellulose any emotional effect on these indicators while in current study cellulose was decreased the ratio of rat protein. In the current study, pectin extracted from both orange and pomegranate peels and orange cellulose ameliorated serum Albumin and Total Protein. This finding is agreed with study of Linyan *et al*. (2020) who reported that insoluble and soluble fiber both two treatments feather ameliorated liver function, judged by the recovered serum level of albumin, and total protein.

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Table (6): T. protein g/dl in serum of rats treated by CCl4 and fed on cellulose extracted from orange and pectin extracted from orange and pomegranate peels

Groups / Phases blood draw in month	Negative control group	Positive control group (CCl4)	CCl4+ Pectin from orange peels	CCl4+Pectin from pomegranate peels	CCl4+ Cellulose from orange peels	Total	One Way ANOVA test	
							F	P-value
First	7.53 a,b ±0.46	11.00**± 1.00	8.87 C,a ±0.23	7.67 A ±1.53	8.37 B,a ±0.32	8.69 a ±1.49	7.956	0.004
Second	8.02 a ±0.23	6.95**±0. 29	6.83**,a ±0.35	7.10**±0.30	6.39 **,A,b ±0.26	7.16 a,b ±0.64	19.000	0.000
Third	6.48 b ±0.22	6.80±0.85	6.55 a ±0.49	6.25±0.07	6.87a,b ±0.64	6.59 b ±0.45	0.694	0.614
General mean	7.34±0.30	8.25**±0. 71	7.42 C ±0.36	7.01 A ±0.63	7.21 B ±0.41	7.48± 0.86	4.385	0.010

*: Significant difference from normal control at P <0.05;
**: Highly significant difference from normal control at P <0.010.
Small letters mean significant differences between groups duration.
Capital letters mean significant differences from positive control (CCl4) group.

5) Lipid profile (Total cholesterol and Triglycerides): Measurement of serum cholesterol levels is important as an indicator of liver function, intestinal absorption, biliary function, and in the diagnosis and classification of hyperproteinemias according to Roeschlau et al. (1974).

Results in Table (7) reveal that total cholesterol level was elevated to 102.44 mg/dl in rats' serum treated with CCl4 compared to 90.22 mg/dl in normal control. Meanwhile, pectin was extracted from orange and pomegranate and cellulose from orange peels led to decrease this elevation to

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86.50, 89.22 & 90.11 mg/dl compared to positive and negative control groups which treated and untreated with CCl4. On the other hand, the level values in cellulose group show the same lowering effect compared with CCl4 and control group values. This finding could be interpreted due to anti-cholesterolemic activity of pectin extract. In addition, these results are in accordance with Parmar and Kar (2007), they noticed that pomegranate peel extract reduced the triglycerides, total cholesterol, LDL, VLDL levels. Also, Sadeghipour *et al*. (2014) notice that pomegranate peel extract in most measured factors such as blood cholesterol, triglycerides, LDL, HDL. The pomegranate peel extract significantly reduced total cholesterol triglycerides, LDL HDL. In the same connection Srivastava and Malviya (2011) reported that although pectin did not contribute significantly to the nutrition, but pectin is natural part to the human intestine, furthermore consumption of pectin has been shown to stabilize blood pressure and reduce serum cholesterol level. Moreover, Indahsari *et al*. (2020) noticed that pectin depressed the elevation in serum cholesterol. Also, Osfor *et al*. (2013) studied the hypocholesterolemia and hypoglycemic effects of orange albedo powder (Citrus aurantium L.) on male albino rats and revealed that the groups of rats which received diets supplemented with 10% and 20% orange albedo powder significantly decrease total lipids, total cholesterol, TG, LDL compared with positive control. It could be concluded that the mechanism by which dietary pectin lowers plasma and liver cholesterol levels in cholesterol-fed rats were

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represent as follow: pectin feeding increased focal bile acid excretion in cholesterol-fed rats. In vitro studies with inverted intestinal sacs demonstrated that pectin decreased taurocholic acid transport by approximately 50%. Rats responded to dietary pectin and cholesterol amine, allow a known inhibitor of bile acid absorption, similarly. Cholesterol–4–C14 absorption was somewhat depressed by dietary pectin as evidenced by fecal radioactive cholesterol excretion and deposition of cholesterol–4–C14 in liver. The effect of pectin on plasma and liver, cholesterol was not altered by dietary succinic sulfathiazole. These data are interpreted to suggest that the hypocholesterolemic effects of pectin are not mediated by an alteration of the intestinal microflora. The results of this study indicate that pectin lowers plasma and liver cholesterol levels in cholesterol-fed rats primly by inhibiting bile acid absorption and by reducing cholesterol absorption (Leveille and Sauberlich 1966).

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Table (7): Cholesterol mg/dl in serum of rats treated by CCl4 and fed on cellulose extracted from orange and pectin extracted from orange and pomegranate peels.

Groups / Phases blood draw month	Negative control group	Positive control group (CCl4)	CCl4+ Pectin from orange peels	CCl4+ Pectin from pomegranate peels	CCl4+ Cellulose from orange peels	Total	One Way ANOVA test	
							F	P-value
First	93.67 a ±4.93	112.33±7.51	89.67 a ±2.08	98.00±15.10	97.67 a ±9.81	98.27a ±11.02	2.688	0.093
Second	87.60 a ±10.97	106.00±14.21	100.33 a ±8.50	91.67±15.31	85.33 a ±20.23	94.11a ±14.68	1.474	0.266
Third	89.40 a ±8.56	89.00±4.24	69.50 a ±2.12	78.00±12.73	87.33 a ±7.37	84.43a ±10.02	2.759	0.095
General mean	90.22±8.15	102.44±8.65	86.50±4.24	89.22±14.38	90.11±12.47	92.27± 11.90	1.837	0.161

*: Significant difference from normal control at P <0.05;
**: Highly significant difference from normal control at P <0.010.
Small letters mean significant differences between groups duration.
Capital letters mean significant differences from positive control (CCl4) group.

Triglycerides are the main lipids present in the human plasma; the others are the cholesterol, phospholipids and non-esterified fatty acids. They are formed in the intestinal mucosa by the esterification of glycerol and fatty acids. Triglyceride measurements are used in the diagnosis and treatment of patients with diabetes mellitus, liver obstruction according to Roeschlau *et al*. (1974).

Results in Table (8) reveal that triglycerides level was elevated to 102.56 mg/dl in serum rats treated with CCl4 compared with 69.67 mg/dl in negative control group. Meanwhile, pectin from orange, pomegranate and cellulose

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from orange led to decrease this elevation to 76.56, 74.00 & 82.22 mg/dl respectively compared with positive and negative control groups which treated and non-treated with CCl4. On the other hand, the level values in rats treated with pectin from orange were close to negative control group. This finding could be interpreted due to anti hyperlipidemia activity of orange pectin extract. In this connection, it discloses that orange pectin was more potent than pomegranate pectin and cellulose from orange in lowering triglycerides and plays as protective effect against CCl4 oxidative stress.The current results are in accordance with Parmar and Kar (2007) who noticed that pomegranate peel extract reduce the triglycerides, total cholesterol, LDL, VLDL levels. Also, Sadeghipour *et al*. (2014) observed that pomegranate peel extract in most measured factors such as blood cholesterol, triglycerides, LDL, HDL significantly reduces total cholesterol triglycerides, LDL HDL. Also, Indahsari *et al*. (2020) revealed that pectin depressed the elevation of serum triglycerides. Moreover Osfor *et al*. (2013). studied the hypo-cholesterolemic and hypoglycemic effects of orange albedo powder (Citrus aurantium L.) on male albino rats and revealed that the groups of rats which received diets supplemented with 10% and 20% orange albedo powder significantly decrease TG compared with positive control. Also serves to improve blood picture. In this correction, the hypolipidemic mechanism of triglycerides could be attributed to bifidogenic potentials in the dietary fiber prepared from mikan (Japanese mandarin orange citrus unshin). Albedo was

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obtained from mikan albedo contained arabinose 37.21%, galactose 16.05%, Xylose 18.30% and glucose 13.94%. Rats fed a diet containing 1% albedo for 4 weeks showed significantly decreased serum triglycerides concentration due to TDF. Consumption of albedo TDF also increased the number of Bifidobacterium in the cecum. In this report they have demonstrated that consumption of albedo TDF increased the levels Bifidobacterium in the rat cecum and decreased S.TG due to the accelerated lipid excretion into the feces caused by the inhibition of pancreatic lipase (Iwata *et al*., 2012). Orange peel had very pronounced hypothermia effects as compared to cellulose. It could significantly decrease the levels of serum triglyceride, serum total cholesterol, liver total lipid, and liver cholesterol. The hypo cholesterol elemental action. The dietary fiber that is so effective in helping to reduce cholesterol is present in large amounts in the white limit of etuis fruit (albedo) pectin is also helpful in stabilizing blood sugar and dietary fiber has been associated with a wide range of health benefits are agreed with Osfor *et al*. (2013). In addition, Ghadir *et al*. (2010) reported an abnormal lipid profile in patients with severe liver dysfunction besides a prominent decline in plasma cholesterol and triglyceride (TG) levels in patients with severe hepatitis and hepatic failure because of reduction of lipoprotein due to reduced liver biosynthesis capacity, and low levels of TG and cholesterol is usually observed in chronic liver diseases.

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Table (8): Triglycerides mg/dl in serum of rats treated by CCl4 and fed on cellulose extracted from orange and pectin extracted from orange and pomegranate peels.

Groups / Phases blood draw in month	Negative control group	Positive control group (CCl4)	CCl4+ Pectin from orange peels	CCl4+ Pectin from pomegranate peels	CCl4+ Cellulose from orange peels	Total	One Way ANOVA test	
							F	P-value
First	77.67a ±40.28	92.33a ±21.22	107.00±46.70	87.33a ±7.09	112.33a ±44.06	95.33a ±32.65	0.486	0.746
Second	63.33a ±5.51	99.67a ±62.69	69.67±17.56	83.33a ±15.28	82.00a ±9.64	79.60a ±28.72	0.641	0.645
Third	68.00a ±10.15	115.67 a *±53.31	53.00 a , C ±1.73	51.33 a , A ±5.77	52.33 a , B ±7.57	68.07 a ±32.88	3.727	0.042
General mean	69.67±18.64	102.56±45.74	76.56±22.00	74.00±9.38	82.22±20.43	81.00± 31.42	1.209	0.338

*: Significant difference from normal control at P <0.05;
**: Highly significant difference from normal control at P <0.010.
Small letters mean significant differences between groups duration.
Capital letters mean significant differences from positive control (CCl4) group.

6) Hematological parameters: The white blood cell (WBC) count is a simple and convenient marker of inflamtion for use in medical practice. according to Wang,S. *et al* (2016). Also white blood cell (WBC) counts are routinely measured in clinical protice as markers of systemic inflammation. according to (Lee Y.J *et al* 2010). Acute on chronic liver failure (ACLF) is always associated with thrombocytopenia or leukocytosis. Therefore, the white blood cell ratio in ACLF patients is always reduced according to Xu *et al*. (2021).

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Data in Table (9) represent white blood count in rats treated with CCl_4 and orally administrated cellulose from orange and two types of pectin as dietary fibers extracted from two sources of agricultural wastes (orange and pomegranate peels) and orally were taken anthocyanin twice each week for two months. Results in Table (9) show that white blood cells (WBC) count was increased in rat's group treated with CCl_4. Meanwhile, rats administrated with pectin either from orange or pomegranate or cellulose from orange peels led to decrease this elevation compared with group treated only by CCl_4. On the other hand, pectin extracted from orange administration significantly changes WBC and the value was close to control group, but it is still higher than normal values. Also, the same trend was found in rats' group treated by CCl_4 and administrated pectin from pomegranate which reveal lowering in WBC compared with positive control group but also it is still higher than untreated negative control group. But pectin from pomegranate was more potent than other treatments. This finding could be interpreted due to hepatic-protective activity of pectin or cellulose extract against oxidative stress induced by CCl_4. In addition, it is pointed out that pectin either from orange or pomegranate or cellulose from orange could attenuate oxidative stress induced by CCl_4. These results are in accordance with Elwej et al. (2016) who showed a decrease platelet count 46% and 80% increase of white blood cells in $BaCl_2$-treated rat's in vivo study. Besides they mentioned that this decrease

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in hemotoxic and genotoxic effects induced by pomegranate peel is due to its powerful antioxidant capacity.

Table (9): Mean of three draws for white blood count × 103 / UL in serum of rats treated by CCl4 and fed on cellulose extracted from orange and pectin extracted from orange and pomegranate peels.

Groups / Phases / blood draw in month	Negative control Group	Positi ve control group (CCl4)	CCl_4+ Pectin from orange peels	CCl_4+Pectin from pomegranate peels	CCl_4+ Cellulose from orange peels	Total	One Way ANOVA test	
							F	P-value
First	7.72 [a] ±1.29	19.5** [a] ± 3.25	8.93 [A,a] ±1.49	10.00 [**,B,a] ±1.67	14.70 [**,C,a] ±2.45	12.17 [a] ±2.03	25.588	0.000
Second	4.88 [b] ±0.81	12.2** [b] ± 2.03	12.20** [,b] ±2.03	8.15 [**,A,a] ±1.36	9.80 [**,B,b] ±1.63	9.45 [b] ±1.57	17.635	0.000
Third	8.74 [a] ±1.46	14.8** [b] ± 2.47	11.50 [**,C,b] ±1.92	10.20 [A,a] ±1.70	10.40 [*,B,b] ±1.73	11.13 [a,b] ±1.86	7.273	0.001
General mean	7.11±1.19	15.5** ± 2.58	10.88 [**,B] ±1.81	9.45 [**,A] ±1.58	11.63 [**,C] ±1.94	10.91 ±1.82	13.544	0.000

*: Significant difference from normal control at P <0.05;
**: Highly significant difference from normal control at P <0.010.
Small letters mean significant differences between groups duration.
Capital letters mean significant differences from positive control (CCl4) group.

Hepatic steatosis is associated with high platelets counts. It has been observed that hepatic steatosis was with platelets count according to Chao yu lin *et al.* (2022). Platelets are small fragments of the cytoplasm and are detached from the cytoplasm of mature megakaryocytes. They play an important role in the function of blood coagulation in humans and animals.

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Platelets play a critical role in liver regeneration liver tumors, injury and other liver diseases (Ramadori *et al*, 2019).

Data in Table (10) represent platelets values in rats treated with CCl_4 and orally administrated cellulose extracted from orange and two types of pectin as dietary fiber extracted from two sources of agricultural wastes (orange and pomegranate pectin) and orally were taken anthocyanin twice each week for two months. Results in Table (10) show that the platelets count was increased due to treatment the rats by CCl_4. Meanwhile, pectin or cellulose administration led to decrease this elevation compared to positive group which treated with only CCl_4. In addition, pectin from both orange or pomegranate peels inhibits CCl_4 stress but pectin from pomegranate was better in lowering platelets count than pectin was extracted from orange peels and the value was comparable to untreated rats (negative control group). This finding could be related to hepato-protective activity of pectin or cellulose extract. The obtained results agree with Zahra *et al*. (2014) they reported that CCl_4-induced chronic liver injury in male rats in their study on rats received CCl_4 and found that the results of rats received Platelet-Rich Plasma (PRP) 0.5 ml/kg, s.c. two days a week for three weeks twenty-four hours after last CCl_4 injection showed that PRP itself was not toxic for liver and could protect the liver from CCl_4- induced histological damage and attenuated oxidative stress by increase in glutathione content and decrease in lipid peroxidative marker of liver tissue. In addition, these results are in accordance with Elwej

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et al. (2016) who showed a decrease in platelet count by 46% in $BaCl_2$-treated rat's *in vivo* study. Also, they mentioned that this decrease in hemotoxicity and genotoxicity effects induced by pomegranate peel is due to its powerful antioxidant capacity. Besides, Osfor *et al*. *(*2013) reported that orange Albedo powder (*Citrus aurantium* L.) serves to improve blood picture.

In this context white blood cells count and platelets count were increased in groups in rats treated with CCl4. Meanwhile treated rats with either orange or pomegranate pectin or orange cellulose led to decrease this elevation compared to group treated with CCl_4.according to (Chae Yu-Lin *et al*., 2022).

Table (10): Means of three draws for platelets count × 103 / UL in serum of rats treated by CCl4 and fed on cellulose extracted from orange peels and pectin extracted from orange & pomegranate peels.

Groups / Phases: blood draw in month	Negative control Group	Positive control group (CCl4)	CCl4+ Pectin from orange peels	CCl4+ Pectin from pomegranate peels	CCl4+ Cellulose from orange peels	Total	One Way ANOVA test	
							F	P-value
First	665.00a ±110.83	661.00 a ±110.17	592.00 a ±98.67	455.00**,A,a ±75.83	600.00 a ±100.0	594.60 a ±99.10	3.618	0.022
Second	679.00a ±113.17	1083.00**,b ±180.5	655.00 C,a ±109.17	377.00**,A,a, b ±62.83	556.00*,B,a, b ±92.67	670.00 a ±111.67	24.137	0.000
Third	301.00b ±50.17	994.00**b ±165.67	600.00 **,C,a ±100.0	319.00 A,b ±53.17	439.00**,B,b ±73.17	530.60 a ±88.44	42.247	0.000
General mean	548.33±91 .39	912.67**±1 52.11	615.67 C ±102.61	383.67**,A ±63.94	531.67 B ±88.61	598.40± 99.73	17.622	0.000

*: Significant difference from normal control at P <0.05;
**: Highly significant difference from normal control at P <0.010.

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Small letters mean significant differences between groups duration.
Capital letters mean significant differences from positive control (CCl4) group.

CONCLUSION

In current study, it is pointed out that pectin either from orange or pomegranate peels and cellulose from orange peels besides anthocyanin extracts show potent protective effect against CCl_4 induced liver damage in rats associated with remarkable decrease in hepatic serum enzymes ALT and AST levels. Also, pectin is occurred commonly in most of plant tissues besides it is a high valuable functional food ingredient and considered as the best agent to reduce ALT and AST activity after higher level due to oxidative stress induced by CCl_4 treatment not only that but also pectin is a safe substance could attenuate and reduce the environmental pollution.

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التأثير الوقائي لكل من البكتين المستخلص من قشور البرتقال والرمان، والسليلوز المستخلص من قشور البرتقال على التسمم الكبدي في ذكور الجرذان البيضاء المعاملة برابع كلوريد الكربون

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المستخلص

تلعب الألياف الغذائية دوراً هاما في الحفاظ على الصحة وسلامة الخلايا من عوامل التلوث والإجهاد التأكسدي نتيجة زيادة الملوثات البيئية الكيميائية كالمبيدات وغيرها، بالإضافة إلى ذلك، يرتبط ارتفاع الألياف الغذائية المتناولة مع انخفاض معدل الإصابة بالأمراض والوفيات.

لذلك يهدف هذا البحث إلى دراسة التأثير الوقائي لكل من البكتين المستخلص من قشور البرتقال والرمان، والسليلوز المستخلص من قشور البرتقال والأنثوسيانين المستخلص من قشور الرمان كمضادات أكسدة طبيعية ضد التسمم الذي يسببه رابع كلوريد الكربون CCl_4 وذلك في ذكور فئران التجارب البيضاء. ولتحقيق هذا الهدف تم اختيار خمس وعشرون فأراً من ذكور فئران التجارب البيضاء المتقاربة في الوزن والعمر ١٢٠ جرام، وتم تقسيمها إلي خمس مجموعات كل مجموعة خمسة فئران وتم الاحتفاظ بالمجموعة الأولي كمجموعة مقارنة ضابطة كنترول بدون معاملة وتم حقن المجموعات ٢، ٣، ٤، ٥ داخل الغشاء البريتوني (الصفاق) بمزيج من ١ مل/كجم من وزن الجسم بمادة رابع كلوريد الكربون مع زيت البرافين المعقم بنسبة ١:١ (حجم/حجم) مرتين في الأسبوع لمدة شهرين والمجموعة الثانية اعتبرت كنترول موجب والمجموعات ٣، ٤، ٥ تم تجريعها يوميا عن طريق الفم بكتين البرتقال، وبكتين الرمان، وسليلوز البرتقال ٥٠ ملجم/كجم من وزن الجسم لمدة شهرين وكذلك مادة الأنثوسيانين ٢٥٠ ملجم/كجم مرتين في الأسبوع لمدة شهرين كمادة مكملة ومضاد أكسدة طبيعي.

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وخلال مدة التجربة والتي استمرت شهرين تم سحب عينات الدم من شريان العين ثلاث سحبات الأولي بعد ١٥ يوم من بداية التجربة والثانية بعد شهر والثالثة في نهاية التجربة بعد شهرين. وقد تم سحب عينات الدم في أنابيب مفرغة للتجلط لفصل السيرم وكذلك عينات أخري أيضاً من الدم تم تجميعها في أنابيب بها مادة الإديتا لمنع تجلط الدم لعمل تحاليل صورة الدم. وأجريت تحاليل للسحبة الأولي والثانية والثالثة كل علي حدة وتم قياس نشاط إنزيمات الكبد (ALT، AST) وتقدير مستويات الكوليسترول، والدهون الثلاثية، والبروتين الكلي، والألبيومين وجلوكوز الدم وعناصر الصوديوم والبوتاسيوم.

وكرات الدم الحمراء والصفائح الدموية، وقد أظهرت النتائج أن مستويات ALT، AST قد زادت نتيجة الحقن i.p في الغشاء البريتوني بمادة رابع كلوريد الكربون وفي الوقت نفسه أدي تناول البكتين المستخلص من البرتقال والرمان والسليلوز المستخلص من قشر البرتقال إلي ظهور مستوياتALT، AST قريبة من المستويات الطبيعية للفئران غير المعاملة برابع كلوريد الكربون كما أوضحت هذه الدراسة أن الألياف الغذائية مثل بكتين البرتقال وبكتين الرمان وسليلوز البرتقال والأنثوسيانين المستخلصة من قشورالرمان أظهرت نشاطا كبيراً كمستخلصات طبيعية آمنة تلعب دورا هاما كعوامل وقائية ضد التسمم الذي يسببه رابع كلوريد الكربون في ذكور فئران التجارب البيضاء.

الكلمات المفتاحية: الجرذان، البكتين، الألياف الغذائية، رباعي كلوريد الكربون، السليلوز، الأنثوسيانين، قشر البرتقال، قشر الرمان، تحليل الدم.