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الصفحة الرئيسية

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جامعة عين شمس، كلية التربية النوعية، القاهرة، مصر
تحية طيبة وبعد،،،

يسر معامل التأثير والاستشهادات المرجعية للمجلات العلمية العربية (ارسیف - ARCIF)، أحد مبادرات قاعدة بيانات "معرفة" للإنتاج والمحتوى العلمي، إعلامكم بأنه قد أطلق التقرير السنوي الثامن للمجلات للعام 2023.

ويسرنا تهنئكم وإعلامكم بأن المجلة المصرية للدراسات المتخصصة الصادرة عن جامعة عين شمس، كلية التربية النوعية، القاهرة، مصر، قد نجحت في تحقيق معايير اعتماد معامل "ارسیف Arcif" المتوافقة مع المعايير العالمية، والتي يبلغ عددها (32) معياراً، وللاطلاع على هذه المعايير يمكنكم الدخول إلى الرابط التالي:

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ويامكانكم الإعلان عن هذه النتيجة سواء على موقعكم الإلكتروني، أو على مواقع التواصل الاجتماعي، وكذلك الإشارة في النسخة الورقية لمجلتكم إلى معامل "ارسیف Arcif" الخاص بمجلتكم.

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محتويات العدد

* بحوث علمية محكمة باللغة العربية:

- العلاج بالفن في التعليم الابتكاري
٧١٧ ا.د/ مصطفى محمد عبد العزيز
- تصور مُقترح لتوظيف الوسائل التكنولوجية الحديثة في التدريس
٧٤١ المُدمج لمُقرر آلة العود بالمعهد العالي للفنون الموسيقية في دولة الكويت
- ا.م.د/ حمد غالب الفضلي
٧٨٥ فاعلية نظام تعليمي قائم على الذكاء الإصطناعي في تنمية مهارات إدارة التعليم الإلكتروني وخفض مستوى قلق المستقبل المهني لدى طلاب تكنولوجيا التعليم وفقاً لمستوى إدارة الذات
- ا.م.د/ سعودي صالح عبد العليم حسن
٨٩٩ اتجاهات الطلاب ذوى الاحتياجات الخاصة نحو ممارسة نشاط المسرح المدرسى
- ا.م.د/ عمرو محمد عبد الله نحلة
٩٦١ ممارسة طلاب المرحلة الإعدادية والثانوية لأنشطة الإعلام التربوى وعلاقتها بتنمية المسؤولية الاجتماعية لديهم
- د/ دعاء محمد عبد المعبود شاهين
١٠٢٩ فاعلية توظيف القيم الملمسية كنقطة جذب محورية في عناصر ديكور التصميم الداخلي (دراسة تجريبية)
- ا.م.د/ سمية عيسى
١٠٧٥ البنية العاملية لمقياس الاندفاعية (UPPS-P) لدى طلاب الجامعة
- ا.د/ نادية السيد الحسينى
د/ أيمن حصافى عبد الصمد
ا/ أحمد عبد السلام على

* بحوث علمية محكمة باللغة الإنجليزية :

- Effects of Bidens Pilosa L Extract on Complete Blood Count and Serum Antioxidant Enzymes Levels in Rats

57

Dr. Batoul N.A. Mohammed

تابع محتويات العدد

- The effect of some plants leaves and pomegranate peel on rats suffering from chronic liver disease 73
Prof. Usama El-Sayed Mostafa
Prof. Safaa Mostafa Abd Elfatah
Eman Sayed Abd ElKhalek
- The Effect of Banana (Musaceae) and Onion (Allium cepa) on Diabetic Rats 97
Prof. Naeem M. Raneh
Prof. Eid Ali ZAKi
Mayada Said Mohamed

The effect of some plants leaves and pomegranate peel on rats suffering from chronic liver disease

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The effect of some plants leaves and pomegranate peel on rats suffering from chronic liver disease

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Abstract

The present study aimed to investigate the effect of different concentrations of olive leaves powder (OLP), guava leaves powder (GLP) pomegranate peel powder (PPP) and their mixture (mix) on rats suffering from chronic liver disease. Sixty male albino rats weighting 150-190g were used in this study, and the rats were divided into two main groups. The first main group (n=6) fed on a basal diet (BD) (as negative control). The second main group (54 rats) was treated with CCl₄ in paraffin oil (50% v/v 2 ml/ kg bwt.) twice a week with subcutaneous injection for two weeks to induce chronic liver disease

Keywords: liver functions, kidney functions, lipid profile, superoxide dismutase, catalase

ملخص:

العنوان: تأثير بعض أوراق النباتات وقشر الرمان علي الفئران المصابة بالتهاب كبدي مزمن
المؤلفون: أسامة السيد مصطفى ، صفاء مصطفى عبد الفتاح ، اسمان سيد عبد الخالق
هدفت الدراسة الحالية إلى معرفة تأثير تراكيز مختلفة من مسحوق أوراق الزيتون (OLP) ومسحوق أوراق الجوافة (GLP) ومسحوق قشر الرمان (PPP) وخليطها (الخليط) على الجرذان المصابة بمرض الكبد المزمن. تم استخدام ستين فأراً ألبينو ذكراً تراوحت أوزانهم بين 150-190 جرام في هذه الدراسة، وتم تقسيم الفئران إلى مجموعتين رئيسيتين. المجموعة الرئيسية الأولى (ن = 6) تغذت على النظام الغذائي الأساسي (BD) (كمجموعة سيطرة سلبية). المجموعة الرئيسية الثانية (54 فأراً) عولجت بـ CCl₄ في زيت البارافين (50% حجم/حجم 2 مل/كجم من وزن الجسم) مرتين أسبوعياً بالحقن تحت الجلد لمدة أسبوعين لتحفيز مرض الكبد المزمن.
الكلمات الدالة: أوراق النباتات ، قشر الرومان ، التهاب الكبد المزمن.

Introduction

The liver is the biggest organ in the human body and a multi-functional organ that plays an important part in the preservation and maintenance of the body. Although the liver has excellent regeneration and restorative properties, the liver can be lost permanently because of damage caused by chronic liver disease or viral infections (**Yoon No et al., 2015**).

Liver disease causes about 2 million fatalities worldwide annually, 1 million deaths from cirrhosis complications, and 1 million deaths from hepatitis viral and hepatocellular carcinoma. Cirrhosis is the 11th most common global cause of mortality, while liver cancer is the 16th most common cause of death; combined, 3.5% of all deaths worldwide Cirrhosis (**Asrani et al., 2019**).

Despite recent treatment progress, liver diseases are still an important global health problem. However, some plants have been utilized and shown effective in liver problems ,among plants is the olive leaves (*Olea europaea L.*) that were used as a folk remedy for combating fevers, a lot of scientists have shown this plant's vital involvement in enhancing cardiovascular risk, cancer and other illnesses. An enormous abundance of olive leaves is produced by the cutting of olive trees and the preparation of olive oil. An estimated 25 kg of olive leaves and twigs per trees are produced each year by olive trees **Taamalli et al., (2020)**, and it belongs to the *Oleaceae family*. Its bioactive compounds such as oleuropein, verbascoside, rutin, tyrosol and hydroxytyrosol may be responsible for the biologic activities of this plant (**Guex et al., 2019**).

Guajava leaves (*Psidium guajava Linn.*) from the *Myrtaceae family* has been used as a folk medicine or herbal tea to treat diarrhea and diabetes over the long term in India, China, Pakistan and Mexico due to lower toxicity and good therapeutic function **Luo et al., (2019)** and uses include the treatment of gastroenteritis, dysentery, stomach pain and diabetes mellitus. guava leaves also have phenolic chemicals and flavonoids with strong antioxidant activity. Gallic acid,

caffeic acid, guaijaverin are the primary active compounds in guava leaves (**Seo et al., 2014**).

Pomegranate (*Punica granatum L.*) is a fruit bearing plant well-known for its therapeutic qualities. The production of juice is less than half the weight of the fruits; pomegranate peel being a byproduct of the juices industries, the dried pomegranate powder was quite excellent for the treatment of numerous illnesses, including headaches and stomach issues, and treatment of bleeding gums. Pomegranate peel is known to be a good source of phenolic acids, tannins (ellagitannins such as punicalin and punicalagin) and flavonoids (**Singh et al., 2019**).

Therefore, this study aims to investigate the effect of some plant leaves on rats suffering from chronic liver disease.

Materials and methods

Materials

- Olive leaves and guava leaves were obtained from one of the gardens, Beni Suef, Egypt during the summer of 2021.
- Pomegranate fruits were obtained from the local market, Beni Suef, Egypt during the summer of 2021.
- Casein, Starch, Corn oil, vitamins, minerals were obtained from ElGomhoryia , company Cairo, Egypt.
- CCl₄ was obtained from ElGomhorya Company, Cairo, Egypt.
- Kits of analyzed items were purchased from El - Gomhorya company, Cairo, Egypt.

Animals

Sixty (60) normal male albino rats (Sprague-Dawely Strain) weighting 150-190g were obtained from the agriculture research center , Giza, Egypt.

Methods

Preparation of pomegranate peels

Ripened pomegranate was rinsed with running tap water, the fresh peels were manually separated, peels were cut into

small pieces and dried by solar energy, then dried peels were powdered using a blender, passed through a 110-mesh sieve, and kept in polyethylene bags and stored at -18°C until used according to **(Salem et al., 2020)**.

Preparation of olive leaves and guava leaves

Olive leaves and guava leaves fresh were rinsed with running tap water, and dried by solar energy, then dried were powdered using a blender according to **Amer, (2014)**, passed through a 110-mesh sieve, and kept in polyethylene bags and stored at -18°C until used according to **(Salem et al., 2020)**.

Preparation of diet

Basal Diets: The rats were fed on a basal diet according to **Reeves, et al., (1993)**. The Salt mixture used in this study according to **Hegsted al., (1941)** and the vitamin mixture prepared according to **Campbell, (1963)**.

Experimental design

Male albino rats weighing (150-190g) were kept under normal healthy conditions and fed on basal diets (BD) for a week for adaptation. After this period, the rats were divided into two main groups:

The first main group

(6) rats fed on basal diets and used as a **(negative control)**

The Second main group

this group consists of (54) rats that are divided into 9 groups, each group of 6 rats. rats were induced with chronic liver by injection of carbon tetrachloride in paraffin oil (50% v/v 2 ml/ kg bwt.) twice weekly for two weeks **Jayasekhar et al., (1997)** to induce chronic liver disease.

Sub-Group (1): (6) rats fed on a basal diet **(BD) (positive control)**.

Sub-Group (2): (6) rats fed on BD containing 2% olive leaves powder (2% OLP).

Sub-Group (3): (6) rats fed on BD containing 4% olive leaves powder (4% OLP).

Sub-Group(4): (6) rats fed on BD containing 2%guava leaves powder (2%GLP).

Sub-Group (5): (6) rats fed on BD containing 4% guava leaves powder (4% GLP).

Sub-Group(6): (6) rats fed on BD containing 2% pomegranate peels powder (2% PPP).

Sub-Group(7): (6) rats fed on BD containing 4% pomegranate peels powder (4% PPP).

Sub-Group(8): (6) rats fed on BD containing 2% (mix of OLP, GLP and PPP) in equal percentage (2% mix).

Sub-Group(9): (6) rats fed on BD containing 4 % (mix of OLP, GLP and PPP) in equal percentage (4% mix).

Each of the above groups was kept in a single cage. The diets were introduced to rats in special non – scattering feeding cups. 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. Rats were weighed before, after and during the experiment.

After completing the treatments: -

The experiment lasted for 8 weeks, the diets were consumed and body weights were recorded twice weekly according to **Chapman et al., (1959)**, after night fasting all rats were sacrificed under anesthesia. Blood samples were withdrawn from the hepatic portal vein in non-heparinized tubes. Blood was centrifuged at 3500 (rpm). for 15 min., serum samples were carefully separated and stored frozen at – 20°C for different biochemical analyses. cholesterol according to **Allain et al., (1974)**, triglycerides **Foster & Dumns, (1973)**, high density lipoprotein HDL-c **Burstein, (1970)**, low density lipoprotein LDL-c and VLDL-c calculated according to **FriedWald et al., (1972)**, Aspartate Amino Transferase (AST) and Alanine Amino Transferase (ALT) by **Reitman and Frankel, (1957)**, Uric Acid and Urea Nitrogen determined according to

Patton & Crouch, (1977), creatinine determined according to **Henry, (1974)** superoxide dismutase (SOD) enzyme assayed according to **Kakkor et al., (1984)**, catalase (CAT) enzyme assayed according to the method of **Sinha (1972)**.

Statistical analysis

The statistical analysis was carried out by using SPSS, PC statistical software (version 10.0; SPSS Inc, Chicago, USA). The results were expressed as mean \pm SD. Data were analyzed by one-way analysis of variance (ANOVA). The differences between means were tested for significance using the least significant difference (LSD) test at ($P < 0.05$) (**Steel & Torri 1980**).

RESULTS AND DISCUSSION

Effect of OLP, GLP, PPP and their mix on feed intake, body weight gain % and liver weight / body weight% in rats suffering from chronic liver disease

Data presented in *table (1)* showed the effect of olive leaves powder, guajava leaves powder pomegranate peel powder and their mix on feed intake, body weight gain % and feed efficiency ratio in rats suffering from chronic liver disease

The mean value of feed intake and body weight gain % in healthy rats (negative control group) showed a significant increase ($P \leq 0.05$), as compared to the chronic liver disease group (positive control group). Treating rats with basal diet containing (4% OLP, 4% GLP and 4% mix of them) caused non-significant differences changed the mean values of feed intake as compared to the negative control group. The mean values of feed intake in all treated groups showed a significant increase ($P \leq 0.05$), as compared to the positive control group. Treating rats with basal diet containing 2% OLP caused non-significant changes ($P \leq 0.05$) in the mean values of feed intake, as compared to the negative control group and positive control group.

All treated groups showed non-significant changes in BWG% , except the group which treated with 2% mix., as

compared to the positive control group. On the other hand, the mean value of BWG% of chronic liver disease group which treated with 5% mix. increased as compared to other treated groups. this treatment showed non-significant difference as compared to the negative control group.

The mean value of liver weight / body weight% in healthy rats (negative control group) showed significant decreased ($P \leq 0.05$), as compared to positive control group. The mean value of liver weight / body weight% all treated groups showed non-significant differences, as compared to the positive control group. All treated groups recorded the best results in the mean value of liver weight / body weight % , except the groups which treated with 4% GLP and 2% mix., because these treatments showed non-significant changes in this organ, as compared to the negative control group.

Table (1):Effect of OLP, GLP, PPP and their mix on feed intake, body weight gain % and liver weight / body weight%in rats suffering from chronic liver disease

Parameters Groups	Feed intake (g/day/each rat)	Body weight gain %	Liver weight / body weight%
Negative (-)	18.50 ^{ba} ± 1.05	60.00 ^{ba} ± 18.46	2.57 ^c ± 0.17
Positive (+)	16.67 ^c ± 1.97	27.00 ^{dc} ± 12.84	3.60 ^{ba} ± 0.82
2%OLP	17.00 ^{cb} ± 1.55	40.33 ^{dcb} ± 22.28	3.27 ^{cba} ± 0.40
4%OLP	18.83 ^a ± 1.17	32.67 ^{dcb} ± 20.29	2.97 ^{cb} ± 0.98
2%GLP	19.17 ^a ± 1.17	38.67 ^{dcb} ± 36.06	3.06 ^{cba} ± 0.44
4%GLP	18.50 ^{ba} ± 1.05	15.00 ^d ± 8.51	3.64 ^{ba} ± 0.76
2%PPP	17.67 ^{cba} ± 1.51	43.33 ^{cba} ± 24.46	3.18 ^{cba} ± 0.78
4%PPP	18.67 ^{ba} ± 1.21	31.50 ^{dc} ± 20.59	3.12 ^{cba} ± 0.38
2%mix	19.17 ^a ± 1.17	69.67 ^a ± 21.57	3.96 ^a ± 1.03
4%mix	18.50 ^{ba} ± 1.52	51.00 ^{cba} ± 16.83	2.97 ^{cb} ± 0.98

OLP: Olive leaves powder GLP: Guajava leaves powder

PPP: Pomegranate peel powder

Least significant differences at ($P < 0.05$).

Mean values in each column with same letters are not significant differences.

In the similar study **Saleh & Mahmoud, (2019)** indicated that CCl_4 caused a significant decrease in feed intake, weight and liver index gain. **Lee et al.,(2016)** reported that the average weight gain of rats in the control positive group which treated

with CCl₄ increased than that of normal rats group. **Ullah et al., (2020)** showed that the CCL₄-induced group revealed marked increase in the liver weight compared the negative control.

Omar & Abd EL-khalik, (2018) reported that consumption 6% Pomegranate Peels led to significant reduction in feed intake. Concerning body weight gain (%) of chronic liver disease group results showed a significant decrease and showed significant ($P \leq 0.05$) decrease in liver weight/body weight%, as compared to chronic liver disease group.

Amer (2014) indicated that the hepatotoxic group fed on basal diet plus 3%, 6% or 9% guajava leaves increased in daily feed intake as compared to rats in the control positive group (hepatotoxic rats) and showed that there is an increase in daily feed intake and BWG% for the negative group (non-hepatotoxic) as compared to the positive group and showed significant decrease in the relative weight of the liver as compared to the positive control group.

These results also in line with **Elhassaneen et al., (2022)** noticed that CCl₄ induced a significant decrease ($p \leq 0.05$) in feed intake BWG% and FER as compared to normal control and noticed that supplementation of the diet with PPP induced significant ($p \leq 0.05$) increasing in feed intake, BWG% and FER. **Mansour (2017)** and **Tahoon (2019)** found that injected rats with CCl₄ caused a decrease in both FER and BWG and improved by consumption of plant parts. On the other hand, **Ludwing, (2000)** reported that pomegranate peel consists of high dietary fiber therefore, the increased consumption of fiber is associated with decreased body weight and reduction in other cardiovascular risk factors.

Effect of OLP, GLP, PPP and their mix on superoxide dismutase and catalase in rats suffering from chronic liver disease

Data presented in *table (2)* showed the effect of olive leaves powder, guajava leaves powder pomegranate peel powder and their mix on superoxide dismutase and catalase in rats suffering from chronic liver disease.

The mean value of superoxide dismutase (SOD) and catalase (CAT) in the effect liver disease group (positive control) showed a significant decrease ($P \leq 0.05$), as compared to the negative control group. The mean values of SOD and CAT in treated groups with (2%GLP, 4%GLP, 2%PPP, 4%PPP, 2% mix of them and 4%mix of them) showed significantly increased ($P \leq 0.05$), as compared to the positive control group. While all treated groups showed a significant decrease ($P \leq 0.05$), as compared to the negative control group. On the other hand (2%OLP and 4%OLP) showed non-significant differences in SOD, as compared to the positive control group.

The least amounts of SOD and CAT were recorded for the group treated with two levels from olive leaves followed by the group treated with two levels from guajava leaves. While The highest amounts of SOD and CAT were recorded for the group treated with two levels from a mix of them followed by the group treated with two levels from pomegranate peel. The best results in SOD and CAT were recorded for the group treated with 4% mix of them followed by the groups treated with 2% mix of them, 4%PPP, 2%PPP and 4%GLP), respectively.

Table (2): Effect of OLP, GLP, PPP and their mix on superoxide dismutase and catalase in rats suffering from chronic liver disease

Parameters Groups	superoxide dismutase (u/mg/ protein)	catalase (u/mg/ protein)
Negative (-)	4.07 a \pm 0.06	5.66 a \pm 0.14
Positive (+)	0.83 g \pm 0.01	1.14 f \pm 0.06
2%OLP	0.92 g \pm 0.02	1.63 e \pm 0.05
4%OLP	0.98 gf \pm 0.01	1.61 e \pm 0.15
2%GLP	1.09 f \pm 0.06	2.02 d \pm 0.03
4%GLP	1.26 e \pm 0.12	2.13 d \pm 0.09
2%PPP	1.37 e \pm 0.24	2.11 d \pm 0.07
4%PPP	1.71 d \pm 0.09	2.41 c \pm 0.01
2%mix	1.94 c \pm 0.08	2.51 c \pm 0.21
4%mix	2.43 b \pm 0.28	3.09 b \pm 0.08

OLP: Olive leaves powder GLP: Guajava leaves powder

PPP: Pomegranate peel powder

Least significant differences at ($P < 0.05$).

Mean values in each column with same letters are not significant differences

Similar observation was obtained by **Saeed Abd El-Lateef et al., (2022)** reported that SOD, and CAT with CCl₄ showed a significant decrease compared to the control. Olive leaves extract protected hepatic tissue against the hazardous effects of CCl₄ by restoring a balanced redox state of hepatic tissue. This was evidenced by a significant increase in serum SOD and CAT with Olive leaves extract treatment compared to CCl₄. **El Sayed et al., (2014)** demonstrated that the ethanolic extract of pomegranate peel revealed the highest total phenolic content approximately 2 fold more than the ethanolic extract of olive leaves.

Abdel Moneim et al., (2013) demonstrated that pomegranate peel has a potent antioxidative effect which significant increases in SOD and CAT activities of liver and kidney samples of rats that received pomegranate were observed in this study. **Debnath et al., (2020)** suggested oxidative stress markers were significantly reduced and antioxidant enzyme activity was significantly improved by Psidium guajava leaves supplementation in CCl₄ administered rat.

Chuanoi et al., (2009) reported that the extract from Psidium guajava, L. leaves contain important phyto-constituents mainly phenolic, flavonoids, carotenoids, terpenoid and triterpene. The major component of the antioxidant system in mammalian cells consists of three enzymes, namely, superoxide dismutase (SOD), catalase (CAT) and glutathione peroxidase

Effect of OLP, GLP, PPP and their mix on liver functions in rats suffering from chronic liver disease

Data presented in *table (4)* showed the effect of olive leaves powder, guajava leaves powder, pomegranate peel powder and their mix on liver function in rats suffering from chronic liver disease

The mean value of ALT and AST in positive control group showed significant increased ($P \leq 0.05$), as compared to negative control group. Feeding rats which suffering from

chronic liver disease On diets containing the two levels from olive leaves, guajava leaves and pomegranate peel and mixture an equal amount from them led to significant decreased ($P \leq 0.05$) the mean values of ALT and AST as compared to the positive control group While these treatments recorded significant increase, as compared to the negative control group.

On the other hand, no significant difference was observed between groups fed on diet containing (2%OLP, 4%OLP and 2%GLP) also between groups (2% Mix of them and 4%PPP) in ALT. While, no significant difference were observed between groups (4%GLP and 2%PPP) in AST. The best results in ALT and AST recorded for the group treated with 4% mix of them followed by the groups treated with 2% mix of them, 4%PPP, 2%PPP and 4%GLP), respectively. While the least amount of ALT and AST recorded for the group treated with 2% OLP followed by the groups treated with 4%OLP.

Table (4): Effect of OLP, GLP, PPP and their mix on liver functions in rats suffering from chronic liver disease

Parameters Groups	ALT (u/ml)	AST (u/ml)
Negative (-)	32.71 g \pm 0.55	43.68 h \pm 0.76
Positive (+)	93.31 a \pm 1.23	108.07 a \pm 4.63
2%OLP	90.47 ba \pm 1.42	96.28 b \pm 5.18
4%OLP	88.77 b \pm 1.54	96.14 b \pm 2.71
2%GLP	87.92 b \pm 0.33	91.24 c \pm 0.22
4%GLP	70.32 c \pm 1.04	86.29 d \pm 0.42
2%PPP	66.78 d \pm 5.05	82.83 d \pm 1.95
4%PPP	61.15 e \pm 0.39	76.39 e \pm 2.08
2%mix	58.92 e \pm 3.14	71.24 f \pm 5.44
4%mix	53.90 f \pm 5.15	54.34 g \pm 1.91

OLP: Olive leaves powder GLP: Guajava leaves powder

PPP: Pomegranate peel powder

Least significant differences at ($P < 0.05$).

Mean values in each column with same letters are not significant differences

These results are in line with **Ullah et al., (2020)** who showed that CCL₄ groups were associated with a significant increase in the ALT and AST. **El Gamel (2022)** showed that positive rats suffering from acute nephritis using CCL₄ have a

significant increase in serum levels of alanine aminotransferase (ALT) and aspartate aminotransferase (AST) compared with those of normal control rats. In contrast, rats feeding on a supplemented diet with olive leaves had significantly decreased serum levels of ALT and AST when compared to positive rats. And notice that olive leaf has a high antioxidant activity owing to the presence of phenolic substances such as hydroxytyrosol, oleuropein and verbascose.

Abozid & Farid, 2013 showed that pomegranate peel and guajava leaves extract treatments protect the liver by significantly inhibiting the ethanol-induced ALT and AST activities. And notice that pomegranate peel extract showed a highly significant effect compared with guajava leaves extract due to: high phenolic and flavonoids contents and highly antioxidant activities.

Mamun et al., (2019) investigated the treatment with guajava leaf significantly normalized ALT and AST activities in the plasma of a high carbohydrate high-fat diet compared to the control rats.

In the same context, **Moustafa (2021)** reported that the treated groups with guajava leaves powder showed a significant reduction of liver enzymes level when compared to the positive control group (rats Induced by Carbon Tetrachloride). This was due to the phenolic content of guajava leaves that possess antioxidant and hepatoprotective activity (**Uboha et al., 2010**).

Effect of OLP, GLP, PPP and their mix on kidney functions in rats suffering from chronic liver disease

Data presented in *table (4)* showed the effect of olive leaves powder, guajava leaves powder, pomegranate peel powder and their mix on kidney function in rats suffering from chronic liver disease

The mean value of creatinine, urea and uric acid significantly increased ($P \leq 0.05$) in rats suffering from chronic liver disease (positive control group), as compared to the negative control group. All treated groups with (2%OLP, 4%OLP, 2%GLP, 4%GLP, 2%PPP, 4%PPP, 2% mix of them and 4%mix

of them) showed significantly decreased ($P \leq 0.05$) in creatinine, urea and uric acid, as compared to the positive control group. While all treated groups showed significant increases ($P \leq 0.05$) in creatinine, urea and uric acid, as compared to the negative control group. On the other hand no significant differences were observed between groups fed on diets containing (2%OLP and 4%OLP) also between groups (2%GLP and 4%GLP) in creatinine, no significant difference were observed between groups (2%OLP and 4%OLP), (2%PPP and 4%GLP) also between groups (2% mix of them and 4%mix of them) in urea and no significant difference were observed between groups (4%OLP and 2%GLP) also between groups (2%PPP and 4%PPP) in uric acid.

The best results in creatinine, urea and uric acid were recorded for the group treated with 4% mix of them followed by the groups treated with 2% mix of them, 4%PPP, 2%PPP and 4%GLP), respectively. While the least amount of creatinine, urea and uric acid was recorded for the group treated with 2% OLP followed by the groups treated with 4%OLP.

Table (4): Effect of OLP, GLP, PPP and their mix on kidney functions in rats suffering from chronic liver disease

Parameters Groups	Creatinine (nmol/ml)	Urea(mg/dl)	Uric acid(mg/dl)
	Mean \pm SD	Mean \pm SD	Mean \pm SD
Negative (-)	1.55 g \pm 0.17	2.66 g \pm 0.22	0.71 h \pm 0.08
Positive (+)	7.18 a \pm 0.42	12.14 a \pm 0.62	3.84 a \pm 0.56
2%OLP	6.37 b \pm 0.33	10.70 b \pm 0.47	3.48 b \pm 0.36
4%OLP	6.53 b \pm 0.46	10.64 b \pm 0.66	3.05 c \pm 0.03
2%GLP	5.85 c \pm 0.25	8.86 c \pm 0.46	2.89 c \pm 0.07
4%GLP	5.47 c \pm 0.26	8.08 d \pm 0.55	2.49 d \pm 0.09
2%PPP	4.66 d \pm 0.28	7.65 d \pm 0.62	2.29 ed \pm 0.22
4%PPP	4.24 e \pm 0.18	6.63 e \pm 0.40	2.16 e \pm 0.30
2%mix	3.94 fe \pm 0.14	5.20 f \pm 0.12	1.80 f \pm 0.34
4%mix	3.76 f \pm 0.69	4.85 f \pm 0.64	1.30 g \pm 0.13

OLP: Olive leaves powder GLP: Guajava leaves powder

PPP: Pomegranate peel powder

Least significant differences at ($P < 0.05$).

Mean values in each column with same letters are not significant differences

This finding agreed **Azab et al., (2019)** reported that the serum urea, creatinine, and uric acid levels in rats injected with CCl₄ were significantly higher in the control group. **El Gamel (2022)** showed that positive rats suffering from acute nephritis using CCL₄ have a significant increase in serum creatinine, urea and uric acid compared with those of normal control rats. In contrast, rats feeding on a supplemented diet with olive leaves had significantly decreased serum levels of creatinine, urea and uric acid when compared to positive rats. Polyphenols, flavonoids, flavones, iridoids, and sugars are among the elements found in olive leaves. These compounds have a significant pharmacological effect while being minimal in toxicity (**Zhang & Tsao, 2016**).

Omar & Abd EL-khalik, (2018) reported that injected rats with CCl₄ (positive control) induced a significant increase ($P \leq 0.05$) in serum uric acid, urea nitrogen and creatinine. Treating rats which were suffering from chronic liver disease with pomegranate peels led to a significant ($P \leq 0.05$) decrease in serum uric acid, urea nitrogen and creatinine. These effects are assumed to be related to the antioxidant property of pomegranate, through scavenger of free radicals released because of oxidative damage, and contain a wide variety of phytochemical compounds like gallotannins, ellagic acid, gallic acid and punicalins (**Singh et al., 2019**).

Moustafa (2021) showed that the treatment with guajava leaves with different concentrations (2 and 4%) resulted in a marked reduction in urea, uric acid, and creatine when compared to positive control group (rats Induced by Carbon Tetrachloride). **Radwan et al., (2018)** mentioned that the antioxidant activity of guajava leaves extract may be an effective factor in improving kidney functions.

Effect of OLP, GLP, PPP and their mix on serum cholesterol, triglycide and lipid profile in rats suffering from chronic liver disease

Data presented in *table (5)* showed effect of olive leaves powder, guajava leaves powder, pomegranate peel powder and

their mix on serum cholesterol, triglyceride and lipoprotein-cholesterol in rats suffering from chronic liver disease.

The mean value of HDL-c in positive control group showed significant decreased ($P \leq 0.05$), as compared to negative control group. While, The mean value of cholesterol, triglyceride, LDL-c and VLDL-c in positive control group showed significant increased ($P \leq 0.05$), as compared to negative control group.

Table (5): Effect of OLP, GLP, PPP and their mix on serum cholesterol, triglyceride and lipid profile in rats suffering from chronic liver disease

Parameters Groups	Cholesterol (mg/dl)	Triglyceride(mg/dl)	HDL-c(mg/dl)	LDL-c(mg/dl)	VLDL-c(mg/dl)
	Mean \pm SD	Mean \pm SD	Mean \pm SD	Mean \pm SD	Mean \pm SD
Negative (-)	55.00 f \pm 2.20	73.88 g \pm 2.04	35.16 a \pm 1.14	11.73 i \pm 0.44	14.78 g \pm 0.41
Positive (+)	112.52 a \pm 6.57	131.89 a \pm 0.62	13.16 g \pm 1.56	63.17 a \pm 1.70	26.38 a \pm 0.12
2%OLP	102.53 b \pm 2.09	127.34 b \pm 3.46	17.30 f \pm 0.89	60.58 b \pm 0.24	25.47 b \pm 0.69
4%OLP	97.32 c \pm 3.39	124.32 b \pm 1.77	18.70 fe \pm 2.03	58.95 b \pm 1.61	24.86 b \pm 0.35
2%GLP	97.51 c \pm 2.17	113.74 c \pm 1.74	20.88 ed \pm 0.29	54.29 c \pm 2.47	22.75 c \pm 0.35
4%GLP	92.98 c \pm 5.81	101.97 d \pm 4.55	21.56 d \pm 4.90	50.10 d \pm 0.32	20.40 d \pm 0.91
2%PPP	78.55 d \pm 3.32	97.30 e \pm 2.37	26.73 c \pm 1.00	47.32 e \pm 2.36	19.46 e \pm 0.47
4%PPP	75.12 d \pm 3.63	94.24 e \pm 4.20	28.05 cb \pm 0.58	42.18 f \pm 1.60	18.85 e \pm 0.84
2%mix	67.07 e \pm 2.20	81.06 f \pm 0.71	29.87 b \pm 0.86	38.77 g \pm 0.89	16.21 f \pm 0.14
4%mix	64.28 e \pm 3.70	84.19 f \pm 3.87	29.33 b \pm 1.20	27.01 h \pm 0.08	16.84 f \pm 0.77

OLP: Olive leaves powder GLP: Guajava leaves powder

PPP: Pomegranate peel powder

Least significant differences at ($P < 0.05$).

Mean values in each column with same letters are not significant differences

Treating chronic liver disease groups with (2%OLP, 4%OLP, 2%GLP, 4%GLP, 2%PPP, 4%PPP, 2% mix of them and 4%mix of them) showed significant decreased ($P \leq 0.05$) in cholesterol, triglyceride, LDL-c and VLDL-c, while HDL-c which showed significant ($P \leq 0.05$) increase, as compared to the positive control group.

All treated groups showed significant increased ($P \leq 0.05$) in cholesterol, triglyceride, LDL-c and VLDL-c, as compared to the negative control group. While HDL-c decreased as compared to the negative control group. The best results in cholesterol, triglyceride HDL-c, LDL-c and VLDL-c recorded for the group treated with 4% mix of them followed by the groups treated with 2% mix of them, 4%PPP, 2%PPP and 4%GLP), respectively. While the least amount of LDL-c and VLDL-c

recorded for the group treated with 2% OLP followed by the groups treated with 4% OLP.

The current study was in agreement with **Saleh & Mahmoud, (2019)** who indicated that CCl_4 caused a significant decrease in high-density lipoprotein (HDLc). CCl_4 also caused an increase in serum total cholesterol (TC), triglyceride (TG), low-density lipoprotein (LDLc) and very low-density lipoprotein (VLDL-c).

El Gamel (2022) indicated that olive leaves revealed a marked significant decrease in serum levels of (TC, TG, low density lipoprotein cholesterol (LDL-c) and very low density lipoprotein cholesterol (VLDL-c) and significant increase at serum HDL-c, that nearly returned toward the normal levels as comparing infected rats(positive group) in rats suffering from acute nephritis using CCL_4 .

Omar & Abd EL-khalik, (2018) reported that Treating chronic liver disease groups with the two levels (3 and 6%) from pomegranate peels the mean values of all lipid parameters significant ($P \leq 0.05$) decreased, except HDL-c which showed significant ($P \leq 0.05$) increase, as compared to the positive control group. This treatment decreased the mean value of serum (cholesterol, triglyceride, LDL-c and VLDL-c) than that of the positive control group, respectively. While HDL-c increased.

Mathur et al., (2015) reported that guajava leaves intake can reduce plasma total cholesterol, triglycerides, and low-density lipoprotein cholesterol. Furthermore, **Tella et al., (2019)** reported that guajava leaves aqueous extract showed an improved serum lipid profile via reducing of triglycerides, total cholesterol, and LDL-cholesterol levels in addition to increasing of HDL-cholesterol levels in diabetic rats.

CCl_4 produced a significant elevation in triglycerides and cholesterol. Pomegranates are a source of polyphenols and other antioxidants and seem to possess a significant antiperoxidative activity (**Prakash et al., 2008**). **Oh et al., (2004)** suggest that guava leaf extract are a good source of water soluble natural antioxidants. It contains a mixture of phenolic compounds such as gallic, quercetin, procatechuic acid.

Chlorogenic acid, caffeic acid, kampferol and ferulic acid, it could be estimated that the phenolic compounds present in the guava leaves played an important role in antioxidant activity.

Conclusion

In conclusion, the present study showed the protective effects of olive leaves powder, guava leaves powder, pomegranate peel powder and their mixture against suffering from chronic liver disease in rats at both levels of supplementation of olive leaves powder, guava leaves powder, pomegranate peel powder and their mixture (2% & 4%).

Recommendations

The study recommends using the powder of both olive leaves powder, guava leaves powder, pomegranate peel powder and their mixture in food products such as cakes, baked goods and biscuits to benefit from the health benefits.

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