دراسة مقارنة بين أوراق الأفوكادو و الزيتون والمانجوفي الجرزان المصابة بالسكري

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Comparative Study among, Olive , Avocado, and Mango leaves in Diabetic Rats

ABSTRACT

This study was conducted to acquaintance the effectiveness of olive , avocado and mango leaves on experimental rats suffering from diabetes mellitus. Forty-five male albino rats weighing 190 ±10 g were used. After the adaptation period , rats were divided randomly into two main groups; the negative control group (C-) 5 normal rats fed on a basal diet and the second group (n=40) were injected subcutaneously with Streptozotocin (STZ) at (45 mg/kg b.wt) dissolved in 0.01 M citrate buffer (PH 4.5).Then rats were divided into eight groups (each group consisted of 5 rats group of them fed on basal diet and left as positive control group (+) ,the other groups treated with, Metformin at dose 200mg/kg b.wt , olive leaves powder 3%, Mango leaves powder 3%, Avocado leaves powder 3%, olive leaves aqueous extract 20% ,mango leaves aqueous extract 20% and Avocado leaves aqueous extract 20%. Results showed that (STZ) injection led to a significant elevation in the relative weights of the liver, kidney and pancreas ,serum levels of glucose , triglyceride ,total cholesterol, LDL-C, liver enzymes, creatinine and urea nitrogen. Conversely , feed intake , body weight and HDL-c were significantly reduced. In general , it has been shown that the leaves of olive , mango and avocado as a powder or as aqueous extract have antioxidant and hypoglycemic effects ,so they alleviated the studied disorder were resulting from hyperglycemia.

Key words: diabetes mellitus, olive leaves , mango leaves , avocado leaves , metformin , Streptozotocin , rats .

INTRODUCTION

The term “diabetes mellitus” describes a heterogeneous group of disorders that have in common hyperglycemia in the absence of treatment. Although pathogenetic pathways to hyperglycemia include defects in insulin secretion, insulin action, or both, dysfunction or destruction of pancreatic b-cells is the underlying characteristic common to all forms of diabetes **(Colagiuri ,2021)**.The current (2021) estimate of 537 million adults aged 20–79 years worldwide has diabetes. By 2030, 643 million, and by 2045, 783 million adults aged 20–79 years are projected to be living with diabetes.It is a major driver of mortality worldwide **(IDF,2021)**. The Middle East and North Africa region (MENA) carried the highest prevalence of diabetes in 2021 at (18.1%) and Egypt in 10th place with 10.9 million **(Sun *et al* .,2021)**. The growing prevalence of diabetes mellitus is predicted to increase the significant social and economic impact resulting from the disease-associated complications. Nevertheless, prevention is possible when acting on risk factors and making an early diagnosis and adequate treatment that prevent long-term complications **(Urrutia *et al* .,2021)**.

In ancient Egypt, olive leaves were first used to treat several diseases such as fever, cough, and cystitis **(Cedola *et al* .,2020)**. Olive leaves are abundant in a range of known phenolic groups that are broadly clustered into (i) secoiridoids (including oleuropein and oleuropein-aglycone), (ii) flavonoids (such as rutin and luteolin-7-glucoside), and (iii) simple phenols (such as hydroxytyrosol and tyrosol). Of these, secoiridoids are characteristically present in the Oleaceae family that well applies to *Olea europaea L* **(Markhali *et al* .,2020)** .Olive leaves have the highest antioxidant power among the different parts of the olive tree **(Blasi *et al* ., 2016)**.Numerous phenolic components in olive leaf have a strong radical scavenging activity **(Souilem *et al* .,2017)**. **(Kermanshah *et al* .,2020)** reported that OLE improves insulin sensitivity and glucose uptake via increased interleukin-6 also oleuropien accelerates the uptake of glucose by cells.

Mango (*Mangifera indica L.*) is a member of the Anacardiaceae family, which originated in India and traditionally

grows in tropical climates (**Yadav and Singh, 2017)**. Mango leaves extract has antioxidant capacity due to the presence of phenolics and flavonoidAmericanas **(Kumar, 2020)**. **on the other hand, Swaroop et al., (2018) *et al.*,(2018)** studied that mangiferin (7.43%) is a major constituent in ML extract,whereas other compounds reported in higher concentration include quercetin-3-O-β-Dglucoside (0.82%), quercetin-3- O-β-D-galactoside (0.86%), and isoswertisin (1.25%). Mangiferin is a natural xanthonoid polyphenol antioxidant and plays role in ameliorating insulin resistance, modulating glucose metabolism, and lowering cholesterol synthesis.

Avocado (*Persea AmericanaMill*) is an oleaginous fruit highly distributed and consumed worldwide **(Chen *et al*.,2008)**. The avocado leaf is a potential source of saponins, alkaloids, steroids and tannins, these compounds also help to prevent oxidative stress .The flavonoids also freeze the free radicals by donating hydrogen atoms and are useful in treating oxidative stress. As avocado leaves are high in flavanoid content, their consumption meets the daily requirement of flavanoids which is higher than any other component like vitamin E, vitamin C, and carotenoids **(Majid *et al* .,2020)**.

Therefore, the present study aims to acquaintance the effectiveness of olive, avocado and mango leaves on experimental rats suffering from diabetes mellitus.

Materials and methods

Plant materials and chemicals

Leaves of olive (*Olea europaea*), mango(*Mangifera indica*)leaves and avocado (*Persea Americana*)leaves were obtained from a fruit garden in Sadat city , Egypt, Metformin, Streptosotozine, formalin, diethyl ether, Casein (> 80% protein), cellulose, vitamins, minerals, cornstarch, DL-methionine, choline chloride and other required chemicals were obtained from Elgomhouria Company for Trading Drugs, Chemical and Medical Appliances, Cairo, Egypt.

Preparation of Olive , Mango and Avocado leaves

Leaves were washed separately, then dried at room temperature in the shade, then ground in powdered form and stored on dark bottles until use **(Yetendje, *et al*,2019)** .

Preparation of Aqueous Extracts Leaves

Finely ground leaves' powder were suspended in deionized water at 75 °C for 15 min, then filtered by a sterilized membrane filter (0.20 µm pore-size). Then the filtrates were concentrated by using a rotary evaporator at 50 °C, followed by drying in an oven at 50 °C **(Abdel-Aziz *et al* .,2020)** .

Induction diabetes mellitus for rats

Forty rats were injected subcutaneously with STZ at a dose of (45mg/kg B.W). Dissolved in 0.01 mml citrate buffer (pH4.5) immediately before use after STZ injection ,rats had free access to food ,water and were given 5% glucose solution to drink over-night to encounter a hypoglycemic shock ,rats were checked daily for the presence glycosuria ,three days after STZ injection ,fasting blood samples were obtained and blood sugar were determined**(Afify, *et al*, 2018)** .

Animals

Forty-five (45) Sprague -Dewley Strain Male albino rats, average weight (190 ±10g) were used in this study. They were obtained from the laboratory Animal Colony, Vaccine, and Immunity Organization of Helwan Farm Cairo, Egypt. All rats were housed in individual cages under hygienic conditions and at controlled temperature (22-24C). All rats were fed the basal diet for 6 days before the beginning of the experiment for adaptation. Rats were deprived of food overnight before the induction of STZ .diets were presented to rats in special containers to avoid loss of food and contamination ,water was provided to rats adlibitum . All rats were fed for 28 days on their experimental diets.

Experimental design

Rats were divided into two main groups as a following :

- **The First main group (C-): (5 rats):** was fed on a basal diet only as a negative control group .
- **The second main group (40rats):** Injected subcutaneously with STZ at a dose of (45mg/kg B.W). Dissolved in 0.01 M citrate buffer (pH4.5) immediately before use after STZ injection, then divided into eight groups (five rats each)

Subgroup (C+): was fed on a basal diet and kept as a positive control group.

Subgroup T1 (Metformin): was fed on basal diet + hypoglycemic drug (metformin) as 200mg/kg bw.
Subgroup T2 (OL.P 3%): was fed on basal diet containing 3% olive leaves powder.
Subgroup T3 (M.P 3%): was fed on basal diet containing 3% mango leaves powder.
Subgroup T4 (A.P 3%): was fed on basal diet containing 3% avocado leaves powder.
Subgroup T5 (OL.E 20%): was fed on basal diet containing olive aqueous leaves extract 20%.
Subgroup T6 (M.E 20%): was fed on basal diet containing mango leaves aqueous extract 20%.
Subgroup T7 (A.E 20%): was fed on basal diet containing avocado leaves aqueous extract 20%.

Biological evaluation

During the experiment period (28 days), the feed intake of rats was recorded every day and body weight gain (BWG) were recorded every week. Biological evaluation of the different diets was carried out by determination of body weight gain (BWG) and feed efficiency ratio (FER) and also relative organs weight % (Liver, kidneys , spleen, lung, pancreas and heart) were calculated According to **(Chapman *et al*., 1959)**.

Biochemical analysis

At the end of the experiment period, the rats were fasted overnight before being sacrificed and the blood samples were collected from each rat and centrifuged for 10 minutes at 4000 revolutions per minute "r.p.m" to obtain the serum. Serum was carefully separated and transferred into dry clean Ebendorf tubes and kept frozen at-20^{o}c for analysis as described by **(Schermer, 1967)**. Glucose was determined in serum according to the method described by **Trinder,(1969)**. Urea was determined according to the method described by **Kaplan,(1984)**. Creatinin was determined according to the method described by **Murray and Kaplan,(1984)**.Serum aspartate aminotransferase (AST) and alanine aminotransferase (ALT) were determined according to the method described by **Murray and Kaplan(1984)**.Alkaline phosphatase (ALP) activity in serum was determined according to **Wenger and Kaplan,(1984)**.Triglycerides were determined in

serum according to the method described by **Trinder and Ann,(1969)**. Total cholesterol was determined in serum according to the method described by **Allain *et al*.,(1974)**. HDL-C was determined in serum according to the method described by **Lopes-Virella *et al* .,(1977)** . Serum VLDLc- and LDLc- were calculated according to **FriedWald ,*et al* .,(1972)**.

Statistical analysis:

Statistical analysis was carried out using one –way analysis of variance (ANOVA) test followed by Duncan test through the programme of statistical packages for the social science (SPSS). Results were expressed as mean±SD. The differences among means at $P<0.05$ were considered significant **(Snedecor and Cochran,1989)**.

Results and discussion

Effect of olive, mango, avocado leaves on body weight gain (BWG), feed intake (FI) and feed efficiency ratio (FER) of diabetic rats.

It is clear to notice in the table (1) that the body weight gain values showed a significant decrease in the positive control group compared with the negative control group (-19.4 Vs 38.2 g), respectively. All treated groups showed a significant increase when compared with the positive control group. The best result was recorded for the metformin group (2.80 g), then olive and mango powder(-2.40 for both of them). Regarding feed intake values showed significant decrease in positive control group when compared with negative control group (18.0 Vs. 22.1 g/ /rat , respectively).All supplemented diets showed increase when compared with positive control group except group of (avocado leaves aqueous extract 20%) which recorded (17.69 g /rat) . The best result recorded for four groups that fed on (olive leaf powder 3% , avocado leaf powder 3% , aqueous extract of olive leaf 20% then aqueous extract of mango leaf 20% , being (19.84 , 19.82 , 19.66 , 19.52 g/rat , respectively) Which close negative control group (22.1g /rat).

As for feed efficiency ratio (FER) the highest FER recorded for negative control group while the lowest FER recorded for positive control group with significant differences between them. The mean values were (0.06 and 0.04, respectively).On the other

side , the highest FER of treated groups (diabetic groups) were recorded for (olive leaves extract 20%) group ,the mean value was (-0.019).The result of this study are in accordance with **Elhassaneen, *et al* ., (2021)** who reported that the treated groups with olive and mango leaves powder showed significant increase in values of body weight gain (P≤0.05), food intake (P≤0.05) and FER (P≤0.05) when compared with positive control group. Additionally, **AbuRahma *et al* .,(2018)** showed that the body weight values increased significantly in the diabetic rats treated with mango leaves aqueous extract when compared with untreated diabetic rats, but the increase was below the weight of normal rats**.** Due to that mango has a lot of vitamins and nutrients that help the body feel fuller **(Wahba and Ebrahim ,2015)**.

Table (1): Effect of olive, mango, avocado leaves on body weight gain (BWG), feed intake (FI) and feed efficiency ratio (FER) of diabetic rats.

	BWG g	FI g /rat	FER
	Mean ± SD	Mean ± SD	Mean ± SD
(C-)	38.20^{a} ±15.15	618.32^{a}±2.21	$.0618^{a}$±.022
(C+)	-19.40^{c} ± 8.79	504.11^{b}±1.64	$-.0385^{c}$±.018
T1:(Metformin)	2.80^{b} ± 9.09	536.03^{b}±2.05	$.0049^{b}$±.016
T2:(OL.P 3%)	-2.40^{b} ± 10.92	555.52^{ab}±1.52	$-.0032^{b}$±.018
T3:(M.P 3%)	-2.40^{b} ±12.05	533.23^{b}±1.55	$-.0042^{b}$±.021
T4:(A.P 3%)	-4.80^{bc} ±7.85	555.18^{ab}±2.36	$-.0083^{b}$±.014
T5:(OL.E 20%)	-1.60^{b} ± 9.60	550.59^{ab}±2.02	$-.0019^{b}$±.017
T6:(M.E 20%)	-4.20^{bc} ±10.61	546.67^{ab}±2.30	$-.008^{b}$± .020
T7:(A.E 20%)	-9.40^{bc} ±15.94	495.48^{b}±1.47	$-.019^{bc}$± .033

- Each value represents the mean ± SD. Means with the different superscript letters in the same column were significantly different (P≤0.05).

Effect of olive, mango, avocado leaves on relative weight of liver, kidney , pancreas ,heart ,spleen ,and lung of diabetic rat.

In Table (2) ,the mean values of relative liver , Kidney , pancrease , heart , spleen and lung weights in negative control groups were significant increase as compared to positive control group , respectively .Also, all treated groups recorded decrease for

relative liver , Kidney , pancrease , heart , spleen and lung weights compared with negative control group. Numerically the best results of relative liver weight values recorded for four groups that fed with avocado leaf powder 3% then olive leaf powder 3% then aqueous extract of mango leaf 20% then hypoglycemic drug (metformin 200mg/kg) which they are higher than positive control group with (7.74 , 0.48 , -2.58 and -4.03 %) respectively . As for relative kidney weight, the best result were recorded for two groups that fed with avocado leaf powder 3% then olive leaf powder 3% which they are higher than positive control group with (6.25 and 6.25 %) respectively .Also, the highest mean values of relative pancrease weight recorded for olive leaves powder 3% then mango leaves powder 3% , which they are higher than positive control group with (45 and 36,6 %), respectively. On the other hand , groups treated with avocado leaf powder 3% , olive leaf powder 3% and aqueous extract of olive leaf 20% increased the mean values of relative heart weights compared with untreated group , which they are higher than positive control group with (18.9, 15.5 and 15.5 %) respectively .Also, the best results on relative spleen weights were recorded for two groups that fed on aqueous extract of mango leaf 20% then hypoglycemic drug metformin 200mg/kg , which they are higher than untreated group with (80.5 and 75 %), respectively. As for relative lung weight, the highest value of relative lung weight was recorded for olive leaf powder 3% ,Which it higher than positive control group with 33,7 % . The results of the present study partially agreed with the results obtained by **Eleazu *et al*.,(2013)** who reported that the liver weights of the diabetic rats showed a significant decrease compared with the non diabetic rats. **Abd El-khalik ,(2017)** found that treating diabetic groups with dosage (4ml/rat/day) of aqueous extracts of olive leaves decreased the mean values of liver and kidney weight/body weight %, as compared to untreated group. Also, **Kouame *et al* .,(2019)** reported that the liver to body weight ratio significantly decreased for the group treated with avocado aqueous extract, these results in agreement with our results.

Table (2): Effect of olive, mango , avocado leaves on relative weight of liver, kidney , pancreas ,heart ,spleen and lung of diabetic rats.

Groups	Liver (Mean ±SD)	Kidney (Mean ± SD)	Pancrease (Mean ± SD)	Heart (Mean ± SD)	Spleen (Mean ± SD)	lung (Mean ±SD)
(C-)	8.02^{b} ±1.37	1.67^{b} ± 0.25	1.06^{d}±0.20	0.74^{b}±0.14	0.83^{c}±0.12	1.30^{b}±0.26
(C+)	6.20^{a} ±0.39	1.28^{a} ±0.11	0.60^{ab} ±0.13	0.58^{a}±0.10	0.36^{a}±0.11	0.89^{a}±0.17
T1:Metformin	5.95^{a}±0.76	1.31^{a}±0.19	0.66^{abc}±0.12	0.64^{ab}±0.10	0.63^{b}±0.16	1.11^{ab}±0.14
T2:OL.P 3%	6.23^{a}±0.90	1.36^{a}±0.13	0.87^{cd}±0.10	0.67^{ab}±0.12	0.61^{b}±0.06	1.19^{ab}±0.11
T3:M.P 3%	5.81^{a} ±0.71	1.24^{a}±0.20	0.82^{c}±0.28	0.60^{ab}±0.05	0.39^{a}±0.11	1.08^{ab}±0.20
T4:A.P 3%	6.68^{a} ±1.67	1.36^{a}±0.21	0.79^{bc}±0.18	0.69^{ab}±0.09	0.53^{ab}±0.08	1.18^{ab}±0.34
T5:OL.E 20%	5.45^{a}±0.67	1.29^{a}±0.06	0.70^{abc}±0.05	0.67^{ab}±0.07	0.59^{b}±0.16	1.11^{ab}±0.21
T6:M.E 20%	6.04^{a} ±0.77	1.30^{a}±0.27	0.58^{ab}±0.11	0.65^{ab}±0.10	0.65^{b}±0.20	1.15^{ab}±0.23
T7:A.E 20%	5.49^{a} ±1.18	1.25^{a}±0.23	0.53^{a} ±0.09	0.60^{ab}±0.14	0.47^{ab}±0.10	1.04^{ab}±0.18

- Each value represents the mean ± SD. Means with the different superscript letters in the same column were significantly different ($P \leq 0.05$).

Biochemical analysis:

Effect of olive , mango , avocado leaves on serum glucose of diabetic rats .

In Table (3) the mean value of the positive control group was higher than the negative control group ,being (476.8 ±12.63 and 89.4 ±9.07 mg/dl),respectively. The mean value of all groups showed a significant decrease compared to positive control group. Data presented in the same table , show that the best result was recorded for groups that were treated with hypoglycemic drug metformin 200mg \kg then aqueous extracts of olive , mango and avocado leaves which are lower than positive control group with (-71.9% , -69% , -65% and -60%) ,respectively. The results in this study revealed that there was a significant elevate on glucose level in the DM group compared to the negative control group, which are in agreement with **Zayed *et al* .,(2018) and Guex *et al* .,(2019)**. Hyperglycemia could be explained by the inhibition of insulin secretion, β-cell destruction, and insulin resistance caused by STZ **(Khattab *et al* .,2020)**. These results are in agreement with **Abunab *et al*.,(2017)** who showed that there was a significant decrease in serum glucose in the group treated with olive leaves aqueous extract. The hypoglycemic activity of olive leaves extract was demonstrated by a reduction in starch digestion and absorption, inhibition of saliva and pancreatin α-amylases activity, increasing peripheral uptake of glucose, and potentiation of glucose-induced insulin release via major phenolic compounds of olive leaves, which have hypoglycemic and antioxidant activities **(Laaboudi,2016).**

These results are in agreement with **Saleem *et al* .,(2019)**who reported that therapy for seven days with 550, 750 or 950 mg/kg mango leaves hydro-alcoholic extract in diabetic mice showed a remarkable decrease in postprandial blood glucose level compared to untreated diabetic mice. The postprandial blood glucose level in experimental groups was comparable to the hypoglycemic drug treated mice. Also **Samanta *et al* .,(2019)** confirmed that the aqueous extract of mango leaves at a dose of 400mg/ kg body weight reduce significantly ($P<0.05$) the blood glucose level.

Rahman *et al* .,(2019) and Sintowati *et al* .,(2016) demonstrated that the ethanol extract of avocado leaves can reduce the fasting blood glucose level . Due to the antioxidant content of avocado leaf extracts. It contained flavonoid compounds, saponins, tannins, triterpenoids, steroids **(Rahman *et al* .,2018)** . Flavonoids contained in avocado leaves (*Persea Americana Mill*) acts as hypoglycemic agents **(Ghorbani *et al* .,2017)** Saponins work by inhibiting the action of the enzyme "-glucosidase, an enzyme in the intestine which functions to convert carbohydrates into glucose **(Bhushan *et al* .,2010)** . The role of tannins is to capture free radicals and reduce increased oxidative stress in diabetics to control blood glucose level **(Cosan *et al* .,2015)**.

Table (3): Effect of olive , mango , avocado leaves on blood glucose of diabetic rats .

Groups	Glucose mg/ dl	Glucose/(C+)	%Resistance to high blood glucose
	Mean ± SD	**Mean ± SD**	**Mean ± SD**
(C-)	89.4^{g} ±9.07	-81.2	Not infected
(C+)	476.8^{a} ±12.63		-
T1:Metformin	134.2^{f} ±12.75	-71.8	71.85
T2:OL.P 3%	216^{c} ±19.51	-54.6	54.69
T3:M.P 3%	268^{b} ±20.61	-43.7	43.79
T4:A.P 3%	282^{b} ±23.48	-40.8	40.85
T5:OL.E20%	147.8^{ef} ±22.42	-69.001	69
T6:M.E 20%	163.8^{e} ±21.89	-65.6	65.64
T7:A.E 20%	189.2^{d} ±7.08	-60.3	60.31

- Each value represents the mean ± SD. Means with the different superscript letters in the same column were significantly different ($P \leq 0.05$).

Kidney functions

Effect of olive, mango , avocado leaves on Urea and Creatinine levels for diabetic rats.

Table (4) show the effect of supplemented diets with leaves of olive , mango and avocado on urea and creatinine levels for diabetic rats. All treated groups were decreased significantly the mean values of urea and creatinin. Treating rats with aqueous extract of olive leaves 20% gives the lowest mean value of urea and

creatinin (20.6 and 0.67 mg/dl),respectively then metformin 200mg/kg group (19.2 and 0.56 mg/dl), compared to positive control group in the same parameters (98.6 and 1.34mg/dl) .Also mango and avocado aqueous extracts 20% decreased the mean values of urea level with (-68% and -59% a positive control group),also they lowers creatinin level with (-46.8% and -43.6%) compared to C+ group

The present results are in agreement with **Al-Hayaly *et al*.,(2020)** who reported that the treatment with olive leaf water extract for animals exposed to alloxan showed a marked decrease in the levels of glucose, urea, and creatinine compared with diabetic animals without treatment .As well as, **Laaboudi, *et al*. (2016)** who found that the treatment of diabetic mice by using olive leaf extracts led to a decrease in the serum of creatinine, uric acid, and urea.

The reason for the decrease in blood serums from (glucose, urea and creatinine) may be due to the improvement of the kidney filtration process as a result of renal glomerular regeneration **(Mohammed *et al*.,2018))**. And since the compounds of flavonoids are rich in antioxidants or inhibitors of free radicals that lead to nephron protection **(Helal, *et al* 2013)**. Also, **Al-Attar and Alsalmi,(2019)** proposed the probable therapeutic use of olive leaf aqueous extract as a new nephron protective factor against severe renal failure. Additionally **Gürbüz and Öğü , (2020)** confirmed that the olive leaves ethanol extract may possibly reduce urea reabsorption by suppressing urea transporters in collecting ducts .

These results agree with **Okafor *et al* ., (2017)** who found that the blood urea and creatinine levels increased significantly in hyperglycaemic control group and reversed in avocado leaves aqueous e extract and metformin treated groups. Also these results supported by **Elhassaneen *et al* .,(2021)** who reported that there was decrease on mean values of kidney functions (plasma urea, creatinine, and uric acid) of diabetic rats treated with mango leaves powder.

The results of the study are in line with **Rahman *et al* .,(2020)** who showed that the treatment of the diabetic group with a dose of 200 mg/kg body weight avocado leaves ethanol

extract , gives a relatively effective influence on regenerating STZ induced renal cell rats.

Table (4) :Effect of olive, mango , avocado leaves on Urea and Creatinine levels for diabetic rats.

Groups	Urea Nintrogen mg/dl	Urea Nintrogen /(C+)	Creatinin mg/dl	Creatinin / (C+)
	Mean ± SD		Mean ± SD	
(C-)	17.8^{g} ±3.49	-81.9	0.55^{f} ± 0.10	-59.07
(C+)	98.6^{a} ±7.02		1.34^{a} ±0.21	
T1:Metformin	19.2^{g}±4.86	-80.5	0.564^{ef}±0.11	-58.03
T2:OL.P 3%	50.2^{d}±3.11	-49.08	0.822^{cd}±0.09	-38.8
T3:M.P 3%	56.2^{c} ±4.91	-43.002	0.868^{bc}±0.10	-35.4
T4:A.P 3%	79.6^{b} ±4.15	-19.2	1.008^{b}±0.05	-25
T5:OL.E 20%	20.6^{g} ±3.43	-79.1	0.672^{def}±0.11	-50
T6:M.E 20%	31.4^{f} ±3.36	-68.1	0.714^{cde}±0.09	-46.8
T7:A.E 20%	40.4^{e} ±3.50	-59.02	0.758^{cd}±0.09	-43.6

- Each value represents the mean ± SD. Means with the different superscript letters in the same column were significantly different ($P \leq 0.05$).

Liver functions

Effect of olive, mango, avocado leaves on (liver enzymes) for diabetic rats.

Data given in Table (5) show effect of olive, mango, avocado leaves on Alanine aminotransferase (ALT), Aspartate aminotransferase (AST) and Alkaline phosphatase (ALP) of diabetic rats.

If could be observed that the mean values of AST & ALT of positive control group were highest than negative control group, with significant differences between them. While ALP showed non-significant changes between these groups, the best result in ALT enzyme were recorded for groups that Treated with aqueous extract of olive leaf 20% then aqueous extract of mango leaf 20% then hypoglycemic drug metformin 200mg/kg being (14.6 ±3.13 , 24.8 ±6.72 , 28 ±3 IU/l) ,respectively which they were lower

than positive control group with (-70.9% , -50.5% and -44.2) , respectively . Also, numerically the best result of AST values were recorded for aqueous extract of olive leaf 20% then aqueous extract of mango leaf 20% then hypoglycemic drug metformin 200mg/kg being (83±6.32 , 93.8 ±6.53 , 120.4±6.58 IU/l). In the same table, data showed that the best result of ALP values were recorded for aqueous extract of mango leaf 20% then aqueous extract of olive leaf 20% then hypoglycemic drug metformin 200mg/kg being (234±7.35 , 243.4±6.31, 253.8 ±3.35 IU/l ,respectively) which they were lower than positive control group with (-44.01% , -41.7% and -39.2) ,respectively .

The results are supported by **Gürbüz and Öğü ,(2020)** who reported that olive leaf therapy exhibited hepatoprotective effects by decreasing ALT and AST levels due to its antioxidant properties . Also **Aggül, *et al* .,(2021**) who found that , in the group given a high dose of olive leaves ethanol extract to treat STZ-induced diabetes, AST, ALP, and ALT levels decreased remarkably compared to group DM and approximated the levels of the control group**.** Additionally**,** **Aggul *et al* .,(2020)** mentioned that the oral administrations of olive leaves ethanol extract at the low and high doses significantly decreased serum AST and ALT levels of the treated diabetic rats compared with the diabetic rats ($p<0.05$, at all groups). Also **Al-Sahib and Alsaadi, (2020)** showed that 4% and 8% olive leaves improved ALT and AST enzymes activities compared to (diabetic) positive control group. The decrease in the liver enzymes may be due to the presence of some active constituent like flavonoids and terpenoids in the Olive leaves which have hepatoprotective effect against hepatotoxins .

Similar result were reported by **Abd Elhameed *et al* .,(2021)** who found that the treatment with total extract of *M. indica* leaves group showed a significant decrease in AST and ALP activities by about 30 and 31%, respectively, compared untreated group . Also, **Okafor *et al* .,(2017)** observed a significant reduction in AST,ALT and ALP in hypoglycemic rats treated with both metformin and graded doses of the avocado leaves aqueous extract.

Table (5): Effect of olive, mango , avocado leaves on (ALT-AST-ALP) of diabetic rats.

Groups	ALT IU/l	ALT IU/l /(C+)	AST IU/L	AST IU/L /(C+)	AlP IU/L	AlP IU/L /(C+)
	Mean ± SD		Mean ± SD		Mean ± SD	
(C-)	19.8^{ef} ±5.01	-60.5	80^{f} ± 4.64	-60	213.6h±6.27	-48.8
(C+)	50.2^{a} ±5.54		200^{a} ±5.52		418^{a} ±14.12	
T1:Metformin	28^{cd} ±3	-44.2	120.4^{d}±6.58	-39.8	253.8^{f} ±3.35	-39.2
T2:OL.P 3%	35.2^{b}±4.60	-29.8	136.2^{c}±5.97	-31.9	321.4^{d}±11.39	-23.11
T3:M.P 3%	35.6^{b} ±3.57	-29.08	154.4^{b}±6.80	-22.8	333.2^{c}±12.46	-20.2
T4:A.P 3%	36.4^{b} ±3.57	-27.4	160.8^{b}±6.14	-19.6	367.8^{b}±5.40	-12.009
T5:OL.E 20%	14.6^{f} ±3.13	-70.9	83^{f}±6.32	-58.5	243.4^{fg}±6.31	-41.7
T6:M.E 20%	24.8^{de} ±6.72	-50.5	93.8^{e}±6.53	-53.1	234^{g}±7.35	-44.01
T7:A.E 20%	31.4^{bc} ±3.84	-37.4	127.4^{d}±3.36	-36.3	292.6^{e} ±7.77	-30

- Each value represents the mean ± SD. Means with the different superscript letters in the same column were significantly different (P≤0.05).

Lipid profile

Effect of olive, mango , avocado leaves on Lipid profile of diabetic rats.

Tables (6 and 7) show the effect of supplemented diets with leaves of olive , mango and avocado on Total cholesterol (TC) , Triglyceride (TG) , High-density lipoprotein (HDL) , low-density lipoproteins (LDL) and Very Low-density Lipoprotein (VLDL) of diabetic rats .It could be noticed that the mean values of serum (total cholesterol and triglyceride) for the negative control group decreased significantly as compared to positive control group. Regarding the mean values of serum (total cholesterol and triglyceride),it could be observed that, all treated groups showed significant decrease, as compared to the positive control group. As for total cholesterol value , the best result for cholesterol and triglycerides were recorded for groups that treated with aqueous extract of olive leaf 20% then aqueous extract of mango leaf 20% then hypoglycemic drug metformin 200mg/kg then aqueous extract of avocado leaf 20%, which they are lower than positive control group with (-54.03% , -49.2% , -40 .5% and -35 .5% in serum cholesterol) and (-51.01% , - 45.9% , - 41.9% and -38.3% in triglycerides), respectively. It was found that serum levels of HDL in untreated group positive control group was significantly ($P<0.05$) lower than normal group. Diabetic groups treated by olive , mango , and avocado leaves extracts then metformin increased HDL levels compared with positive control group , with (75.7% , 58.9% , 46.5% , 43.4%), respectively. On the other hand, it was observed that all treated groups decreased significantly the mean values of LDL and VLDL as compared to untreated groups. Treating rats with aqueous extract of olive leaf 20% gave the lowest mean values of LDL and VLDL (10.00 and 15.00 mg /dl) then aqueous extract of mango leaf 20% group

(21.87 and 16.60 mg /dl),compared to positive control group in the same parameters(96.48 and 30 .72 mg /dl).

These results are in agreement with **Sakr *et al* .,(2016)** who showed that the levels of lipids (Cholesterol, Triglycerides, LDL) are raised in Diabetes group. Also, **Mansouri *et al*., (2015)** reported that diabetic patients were characterized by significant increase in lipid profile and decrease of HDL. Because insulin has an antilipolytic effect that inhibits the hormone-sensitive lipase in the adipose tissue, as a result, insulin reduces the secretion of free fatty acids from adipose tissue. It also significantly influences lipid metabolism by suppressing intestinal chylomicron and hepatic lipoprotein synthesis, increasing lipoprotein lipase activity and LDL receptor expression in peripheral tissues **(Vergès,2020)**. Hyperglycemia that induced by STZ may be aggregated the oxidative stress in liver tissues. The increase of lipid peroxidation (LPO) and H_2O_2 levels in diabetic group may be explain the oxidative stress status in liver of diabetic rats. Olive leaves extracts are rich in phenolic constituents and oleuropein is the most prominent phenolic compound. The antioxidant potential of olive leaf extracts has further verified the inhibitory potential of dietary olive leaves on LPO **(Elsaid *et al* ., 2018)**

These results are supported by **Abd El-Baky *et al* .,(2020)** who concluded that the diet supplemented with olive leaves extract significantly decrease ($P<0.05$) the mean value of serum TC, TG, VLDL-C and LDL-C, however, serum HDL-C level was increased significantly ($P<0.05$), compared to the positive control group. Also, **Fki *et al* .,(2020)** reported that treatment with olive leaves significantly decreased serum cholesterol and triglyceride levels in rats fed a high fat diet. Additionally, **Saleem *et al* ., (2019)** showed that the extract of Mngo Leaves significantly lowered (LDL), cholesterol and triglycerides and raised (HDL) dose dependently in diabetic mice compared to the untreated diabetic mice . Catechin, epicatechin, chlorogenic acid, gallic acid and mangiferin were responsible for decreasing hyperlipidemia in diabetic animals **(Begum and Srivalli ,2019). ,**Furthermore **Kouame *et al* .,(2019)** showed that extracts of *Persea. americana*

were generally lipid lowering. The extracts lead to a fall in the LDL- level and an increase in HDL. Because it contain flavonoid and saponin, which interference in biosynthesis lipid fraction in the liver by flavonoid and disruption of lipid absorption by saponin **(Dita *et al* .,2019)** .

Table (6) Effect of olive, mango , avocado leaves on (Total cholesterol and Triglyceride) of diabetic rats.

Groups	TC mg/dl	TC/(C+)	TG mg/dl	TG/(C+)
	Mean ± SD		Mean ± SD	
(C-)	66.25^{f}±4.43	-56.6	69.40^{f}±9.07	-54.8
(C+)	153.00^{a}±20.31		153.60^{a}±22.67	
T1:Metformin	91.00^{de}±9.24	-40.5	89.20^{cde}±11.64	-41.9
T2:OL.P 3%	104.60^{cd}±13.31	-31.6	102.75^{c}±10.96	-33.1
T3:M.P 3%	110.60^{c}±14.97	-27.7	123.40^{b}±17.70	-19.6
T4:A.P 3%	134.60^{b}±15.24	-12.02	129.80^{b}±16.81	-15.4
T5:OL.E 20%	70.33^{f}±1.78	-54.03	75.00^{ef}±7.21	-51.1
T6:M.E 20%	77.67^{ef}±3.55	-49.2	83.00^{def}±8.51	-45.9
T7:A.E 20%	98.60^{cd}±10.01	-35.5	94.75^{cd}±9.33	-38.3

- Each value represents the mean ± SD. Means with the different superscript letters in the same column were significantly different ($P \leq 0.05$).

Table (7) Effect of olive, mango , avocado leaves on (HDL-c , LDL-c and VLDL-c) of diabetic rats.

Groups	HDL mg/dl	HDL/ C+	LDL mg/dl	LDL/ C+	VLDL mg/dl	VLDL/C+
	Mean ± SD		Mean ± SD		Mean ± SD	
(C-)	35.60^{cd}±4.61	37.9	16.77^{ef}±1.57	-82.6	13.88^{a}±1.81	-54.8
(C+)	25.80^{e}±3.19		96.48^{a}±8.93		30.72^{a}±4.53	
T1:Metformin	30.60^{de}±3.20	18.6	42.56^{d}±3.85	-55.8	56.36^{a}±88.13	83.4
T2:OL.P 3%	37.00^{bc}±4.06	43.4	47.05^{d}±8.64	51.2	20.55^{a}±2.19	-33.1
T3:M.P 3%	30.60^{de}±2.88	18.6	55.32^{c}±3.65	-42.6	24.68^{a}±3.54	-19.6
T4:A.P 3%	29.40^{e}±3.84	13.9	79.24^{b}±9.41	-17.8	25.96^{a}±3.36	-15.4
T5:OL.E 20%	45.33^{a}±3.48	75.7	10.00^{f}±.82	-89.6	15.00^{a}±1.44	-51.1
T6:M.E 20%	41.00^{b}±4.84	58.9	21.87^{e}±2.52	-77.3	16.60^{a}±1.70	-45.9
T7:A.E 20%	37.80^{bc}±3.56	46.5	41.85^{d}±4.07	-56.6	18.95^{a}±1.86	-38.3

Each value represents the mean ± SD. Means with the different superscript letters in the same column were significantly different ($P \leq 0.05$).

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دراسة مقارنة بين أوراق الأفوكادو و الزيتون والمانجو في الجرزان المصابة بالسكري

المستخلص

أجريت هذه الدراسة للتعرف على فاعلية أوراق الزيتون والأفوكادو والمانجو على فئران التجارب المصابة بداء السكري. تم استخدام خمسة وأربعين من ذكور الجرذان البيضاء وزنها 190 ± 10 جم. بعد فترة التكيف ، تم تقسيم الفئران بشكل عشوائي إلى مجموعتين رئيسيتين. مجموعة التحكم السلبية 5 فئران تم تغذيتها على النظام الغذائي الأساسي والمجموعة الثانية (ن = 40) تم حقنها تحت الجلد باستخدام الستربتوزوتوسينSTZ بجرعة (45 مجم / كجم من وزن الجسم) مذابة في محلول سيترات 0.01 مولار4.5 PH ثم قسمت الفئران إلى ثماني مجموعات كل مجموعة تتكون من 5 فئران وتتغذى على الوجبة الأساسية كما يلي تركت المجموعة الأولي كمجموعة ضابطة إيجابية والمجموعة الثانية مجموعة الميتفورمين بجرعة 200 مجم / كجم من وزن الجسم والثالثة مسحوق أوراق الزيتون 3٪ والرابعة مسحوق أوراق المانجو 3٪ والخامسة مسحوق أوراق الأفوكادو 3٪ والسادسة المستخلص المائي لأوراق الزيتون 20٪ والسابعة المستخلص المائي لأوراق المانجو 20٪ والثامنة المستخلص المائي لأوراق الأفوكادو 20٪. أظهرت النتائج أن حقن STZ أدى إلى ارتفاع معنوي في الأوزان النسبية للكبد والكلى والبنكرياس ومستويات الجلوكوز والدهون الثلاثية والكوليسترول الكلي والكوليسترول الضار–LDL C وأنزيمات الكبد والكرياتينين واليوريا النيتروجين في الدم. على العكس من ذلك ، انخفض المتناول من الغذاء ووزن الجسم و HDL–c بشكل كبير. بشكل عام فقد ثبت أن أوراق الزيتون والمانجو والأفوكادو كمسحوق أو مستخلص مائي لها تأثيرات مضادة للأكسدة ونقص سكر الدم ، لذلك فهي تخفف من الاضطراب الناتج عن ارتفاع السكر في الدم.

الكلمات المفتاحية: داء السكري ، أوراق الزيتون ، أوراق المانجو ، أوراق الأفوكادو ، الميتفورمين ، الستربتوزوتوسين ، الجرذان