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EFFECT OF CRUDE METHANOLIC EXTRACTS OF SOME FRUIT TREE LEAVES ON SOME BLOOD CONSTITUENTS OF DIABETIC RATS.

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ABSTRACT

Crude methanoilc extracts of thompson seedless, flame seedless, zizyphus, pomegranate and fig leaves were prepared. In this work the efficiency of the investigated extracts has been studied to elucidate their activities on some blood constituents of diabetic rats. The most effective treatment was the methanolic extract of zizyphus leaves which decreased blood glucose level to 128.33mg/dl at a dose of 40mg/100g body weight after 30 days where the maximum reduction of about 66.95% was obtained comparing with control diabetic rats. Whereas treatments of streptozocin diabetic rats with methanolic extract of thompson seedless leaves resulted the maximum percentage of reduction in ALT activity with 60.91% after 30 days by using 40mg/100 body weight. Extract of flame seedless leaves was the most effective for reduction of AST enzyme with a percentage of 46.23% after 30 days when 40mg/100 body weight was used. Serum creatinine and urea levels were affected by the methanolic leaves extract of thompson seedless to achieve the maximum reduction of 51.36 and 39.64% comparing with control diabetic rats at a dose of 40mg/100g body weight for 30 days. Also, lipid profile (triglycerides, total cholesterol, HDL, LDL and vLDL) were affected with treatment by the methanolic leaves extract with different values.

Keywords: Diabetes mellitus, ALT, AST, creatinine, urea, triglycerides, total cholesterol, HDL, LDL and vLDL

INTRODUCTION

Diabetes mellitus is a metabolic disease characterized by hyperglycemia caused by defective insulin secretion and action, resulting in long term multi-organ complications (Caughron and Smith, 2002). Chronic hyperglycemia causes damage to the eye, heart, kidney, nerves system and blood vessels (Lebovitz, 2001). Different types of oral hypoglycemic agents are available along with insulin for the treatment of diabetes and there is still need to search for natural products with hypoglycemic effect (Venkatesh *et al.,* 2003). Several studies have shown that some plants (grape, zizyphus, pomegranate and fig leaves) have an antidiabetic activity (Orhan *et al.*, 2006; Abdel-Zaher *et al.*, 2005; Serraclara *et al.,* 1998 and Allam *et al*., 2008).

It is well known that the most common substances inducing diabetes in the rats are alloxan and streptozocin. Streptozocin causes induction of β-cell death via alkylation of DNA by nitrosourea moiety of this compound (Vessal *et al.,* 2003).

Several authors suggested that blood glucose level, liver functions, kidney functions and lipid profile, were affected when experimental animals injected by alloxan in concentrations ranged between (8.0 –17.5mg/100g

body weight) or streptozocin at doses ranged between (4.0 – 6.5mg/100g body weight) Anand *et al.,* (1989); Glombitza *et al*., (1994); Terada *et al*., (1998); Andallu and Vardacharyulu, (2001); Hessien (2003); Pérez *et al.* (2003); Orhan *et al*., (2006); Soltani *et al*., (2007); Allam *et al*., (2008); Ju-Tung *et al*., (2008) and Gokce and Haznedaroglu, (2008).

Grape (*Vitis vinifera* L.) has been used as a food and a beverage, as well as a remedy against various complaints in traditional medicine worldwide since ancient times. Leaves of the plant have been used to stop bleeding and to treat inflammatory disorders and pain (Bombardelli and Morazzonni, 1995 and Baytop, 1999). Leaves are also recorded to reduce blood glucose levels in diabetics as a folk remedy in Turkey. Although the chemical composition of *Vitis vinifera* leaves is known very well, the studies conducted on biological effects of the leaves are limited. Additionally, antihyperglycaemic and hypoglycaemic effects have not been evaluated so far. The aqueous extract of *Vitis vinifera* leaves have antidiabetic activity on normoglycaemic, hyperglycaemic and streptozocin-induced diabetic rats (Orhan *et al.*, 2006).

Zizyphus (Rhamnaceae) species are used in folk medicine for the treatment of some diseases, such as digestive disorders, weakness, liver complaints, obesity, urinary troubles, diabetes, skin infections, fever, diarrhea and insomnia (Kirtikar and Basu, 1984 and Han and Park, 1986). Also, Glombitza *et al.* (1994) found that the butanol extract of *Zizyphus spina-christi* leaves or its main saponin glycoside, christinin-A, improved glucose utilization in diabetic rats and serum insulin level showed a significant increase in diabetic rats treated for a period of 4 weeks.

Fig leaves decoction have a pharmacological properties as hypoglycemic agent in diabetic patients and alloxan-induced diabetic rats (Serraclara *et al.,* 1998 and Allam *et al*., 2008).

The aim of this study is to examine the efficiency of some natural products namely, thompson seedless, flame seedless, zizyphus, pomegranate and fig leaves as hypoglycemic agents in induced diabetic rats. Also, to study their effect on some blood constituents of diabetic rats.

MATERIALS AND METHODS

Sampling:

The present investigation was carried out using leaves of two species of grape *Vitis vinifera* (Thompson and Flame seedless), zizyphus (*Zizyphus spina-christi*), pomegranate (*Punica granatum*) and fig (*Ficus carica*) belong to families Viticeae, Rhamnaceae, Punicaceae and Moraceae, respectively.

The samples were collected from the experimental farm of the Faculty of Agriculture, Mansoura University, Mansoura, Egypt (2007). All samples were air dried in the shade and ground into a fine powder.

Powdered air dried leaves (3 Kg) from each sample were extracted six times by soaking in 5 L methanol overnight followed by filtration. The filtrate of each sample was evaporated to dryness at 45°C under reduced pressure using rotary evaporator. The obtained crude methanolic extract for each sample was kept in refrigerator for investigation.

Determination of antidiabetic activity of methanolic leaves extracts:

Experimental animals:

A number of 85 albino rats (100-120g) were obtained from the animal house of The Nile Company for Pharmaceuticals and Chemical Industries, Cairo, Egypt. The rats were kept for adaptation under normal laboratory conditions for 7 days before the beginning of the experiment. All rats were fed on normal diet and allowed free access of water.

Induction of diabetes mellitus:

The experimental rats (85 rats) were divided randomly into two major groups:

Group 1 (5 rats), represents healthy non diabetic rats "common control". The other 80 rats (Group 2) were fasted for 24 hours then intraperitoneal injected by streptozocin (MP, USA) freshly prepared in 0.10M citrate buffer, pH 4.5 at a dose of 4.5 mg/100g of body weight to induced diabetes mellitus according to Ghasemi *et al.,* (2007). In order to stave off the hypoglycaemia during the first day after streptozocin administration, diabetic rats were given 5% glucose solution orally as reported by Orhan *et al.,* (2006).

Serum glucose level of all rats was determined 72 hours post-injection, rats were fasted for 18 hours before determination. Experimental rats with serum glucose level over 250 mg/dl were considered sufficiently as streptozocin-diabetic rats and ready for treatment with crude methanolic extracts.

Group 2 (80 diabetic rats) were divided randomly to 16 subgroups (5 rats for each) as follows:

Subgroup 1: represent the control diabetic rats and received a normal diet for 30 days (without any treatment).

Subgroups 2, 3 and 4 were treated with the crude methanolic extract of thompson seedless leaves with doses of 10, 20 and 40 mg/100g of body weight, respectively.

Subgroups 5, 6 and 7 were treated with the crude methanolic extract of flame seedless leaves with doses of 10, 20 and 40 mg/100g of body weight, respectively.

Subgroups 8, 9 and 10 were treated with the crude methanolic extract of zizyphus leaves with doses of 10, 20 and 40 mg/100g of body weight, respectively.

Subgroups 11, 12 and 13 were treated with the crude methanolic extract of pomegranate leaves with doses of 10, 20 and 40 mg/100g of body weight, respectively.

Subgroups 14, 15 and 16 were treated with the crude methanolic extract of fig leaves with doses of 10, 20 and 40 mg/100g of body weight, respectively.

All crude methanolic extracts for investigated leaves were dissolved in saline solution (sodium chloride, 0.9%) and given orally by a stomach tube after fasting for 2 hours, daily for 30 days.

Blood samples were collected from the eye canthus by heparinized tubes every 10 days after the beginning of extracts administration. Then, each blood sample was centrifuged to obtain clear serum where serum

glucose levels for fasting animals were determined immediately. Serum blood samples were kept at refrigerator under freezing conditions for the determination of the other parameters included liver functions (ALT and AST), kidney functions (creatinine and urea) and lipid profile (triglycerides, total cholesterol, HDL, LDL and vLDL).

Chemical analysis of blood:

Serum glucose was determined by a colorimetric enzymatic method as described by Ghasemi *et al.,* (2007).

Liver functions: alanine amino transferase (ALT) and aspartate amino transaferase (AST) were determined according to the method of Reitman and Frankel (1957).

Kidney functions: Creatinine and blood urea were determined by a colorimetric method according to Patton and Crouch (1977).

Lipid profile: triglycerides, total cholesterol, HDL, LDL and vLDL were determined by enzymatic colorimetric method of Richmond, (1973).

Statistical analysis of obtained data were done using the statistical software package CoStat (2005). All comparisons were first subjected to one way ANOVA and significant differences between treatment means were determined using Duncan's multiple rang test at $p<0.05$ as the level of the significance (Duncan, 1955).

RESULTS AND DISCUSSION

Effect of crude methanolic extracts of investigated leaves on blood glucose level:

In this study, streptozocin was used at a dose of 4.5 mg/100g body weight to induce diabetes mellitus in rats. Several researchers could use this compound in concentration ranged from 4.0 to 6.5 mg/100g body weight to induce diabetes mellitus in experimental animals (Glombitza *et al*., 1994; Terada *et al*., 1998; Andallu and Vardacharyulu, 2001; Pérez *et al.* 2003; Orhan *et al*., 2006 and Soltani *et al*., 2007).

Data in tables (1, 2, 3, 4 and 5) revealed that the injection of streptozocin at a dose of 4.5 mg/100g body weight caused a highly significant ($P<0.05$) increase in blood glucose level from 124.67 to 388.33 mg/dl for non-diabetic and diabetic rats at zero time, respectively. This increase represents about 67.89% of that obtained value of non-diabetic rats.

In addition, gradual increasing was observed during the experimental periods (10, 20 and 30 days) until reached the maximum level of 461.33 mg/dl for diabetic rats at the end of the experiment.

This increase may be due to the destructive effect of streptozocin on β-cells of islets of Langerhans which lead to insulin deficiency. In addition to the absence of available insulin in blood circulation, these may be the main causes of hyperglycemia which observed in the treated rats with streptozocin as reported by (Vessal *et al*., 2003).

Also, Hecht *et al.,* (1973) reported that the significant increase in the levels of blood glucose in streptozocin-induced diabetic rats could be due to a

beta cytotoxic induces chemical diabetes through damaging insulin-secreting cells.

Table 1: Effect of crude methanolic extract of thompson seedless leaves on levels of blood glucose (mg/dl), liver functions (U/L), kidney functions (mg/dl) and lipid profile (mg/dl) in diabetic rats.

Levels of :		Treatment period	Groups of Experimental Animals					LSD 5%
			Group 1	Group 2				
				Subgroup				
				1	2	3	4	
Serum glucose (mg/dl)		Zero time	124.67 d	388.33 abc	403.33 ab	369.67 abc	371.33 abc	86.57
		10 days	123.67 d	432.67 a	365.33 abc	301.33 c	314.67 bc	
		20 days	123.33 d	425.67 a	178.67 d	163.67 d	171.00 d	
		30 days	124.67 d	461.33 a	167.00 d	138.00 d	131.67 d	
Liver functions (U/L)	ALT	Zero time	25.33 fg	58.00 bcd	55.67 bcd	51.00 de	53.33 cde	6.87
		10 days	24.00 g	62.33 b	59.67 bc	55.00 bcde	48.00 e	
		20 days	23.33 g	59.00 bc	31.00 f	28.33 fg	26.33 fg	
		30 days	23.67 g	71.33 a	29.67 fg	27.00 fg	22.67 g	
	AST	Zero time	36.67 g	70.67 bcd	70.67 bcd	65.33 d	66.33 d	5.31
		10 days	35.00 g	74.33 ab	69.33 bcd	67.33 cd	66.00 d	
		20 days	36.00 g	72.33 bc	46.00 e	45.33 e	43.00 ef	
		30 days	36.33 g	79.33 a	43.33 ef	39.33 fg	38.00 fg	
Kidney functions (mg/dl)	Creatinine	Zero time	1.30 efg	2.20 ab	2.20 ab	2.13 bc	2.20 ab	0.22
		10 days	1.23 efg	2.27 ab	2.10 bc	1.93 cd	1.83 d	
		20 days	1.20 efg	2.20 ab	1.33 e	1.17 efg	1.10 fg	
		30 days	1.23 efg	2.40 a	1.33 ef	1.17 efg	1.07 g	
	Blood Urea	Zero time	50.67 hi	75.67 de	73.67 e	75.00 de	74.67 de	5.23
		10 days	51.33 hi	94.67 b	86.33 c	80.33 d	80.00 d	
		20 days	50.00 i	97.67 ab	63.33 f	51.33 hi	57.33 g	
		30 days	56.00 gh	100.33 a	59.00 g	46.67 i	45.67 i	
Lipid profile (mg/dl)	Triglycerides	Zero time	174.67 gh	288.00 cd	287.00 cd	288.67 cd	284.67 cd	10.58
		10 days	171.00 h	300.67 ab	292.67 bcd	282.33 d	268.33 e	
		20 days	172.67 h	295.00 bc	201.67 f	186.00 g	179.00 gh	
		30 days	181.67 gh	307.00 a	204.33 f	176.67 gh	172.67 h	
	Total Cholesterol	Zero time	194.67 g	382.67 c	358.33 d	385.00 bc	386.33 bc	21.68
		10 days	189.67 g	407.67 ab	395.67 abc	386.00 bc	376.33 cd	
		20 days	194.00 g	410.67 a	242.33 e	233.33 ef	214.33 fg	
		30 days	197.67 g	414.33 a	226.33 f	201.67 g	192.67 g	
	HDL	Zero time	47.00 a	27.67 def	27.00 defg	29.00 de	28.00 de	3.97
		10 days	44.67 ab	24.67 efg	25.00 efg	28.67 de	30.00 d	
		20 days	42.67 b	27.33 def	27.00 fg	30.33 d	34.67 c	
		30 days	43.67 ab	27.00 defg	25.00 efg	22.67 g	31.00 cd	
	LDL	Zero time	101.67 h	287.67 ab	285.67 ab	284.33 ab	286.67 ab	22.50
		10 days	103.67 h	299.00 a	265.00 b	169.00 def	157.00 ef	
		20 days	105.67 h	308.67 a	216.67 c	178.33 de	147.00 fg	
		30 days	107.00 h	305.67 a	233.00 c	186.00 d	131.67 g	
	vLDL	Zero time	34.67 j	57.67 cde	57.33 cde	58.00 cde	56.67 de	1.97
		10 days	34.33 j	60.33 ab	58.67 bcd	56.33 e	53.67 f	
		20 days	34.67 j	59.00 bc	40.33 g	37.00 i	36.00 ij	
		30 days	36.33 ij	61.33 a	41.00 h	35.67 ij	34.67 j	

Group 1: Control non-diabetic rats; Subgroup 1: Control diabetic rats; Subgroups 2, 3 and 4 treated rats with the crude methanolic extract of thompson seedless leaves with doses of 10, 20 and 40 mg/100g of body weight, respectively.

Table 2: Effect of crude methanolic extract of flame seedless leaves on levels of blood glucose (mg/dl), liver functions (U/L), kidney functions (mg/dl) and lipid profile (mg/dl) in diabetic rats.

Levels of :		Treatment period	Groups of Experimental Animals					LSD 5%
			Group 1	Group 2				
				Subgroup				
				1	5	6	7	
Serum glucose (mg/dl)		Zero time	124.67 g	388.33 bc	390.33 bc	468.67 a	436.00 ab	43.49
		10 days	123.67 g	432.67 ab	348.33 c	440.67 a	375.33 c	
		20 days	123.33 g	425.67 ab	257.67 d	212.00 e	193.33 ef	
		30 days	124.67 g	461.33 a	204.67 ef	164.67 fg	141.33 g	
Liver functions (U/L)	ALT	Zero time	25.33 j	58.00 bc	52.67 cdef	55.67 bcde	53.00 cdef	6.30
		10 days	24.00 j	62.33 b	57.67 bcd	50.67 def	50.00 efg	
		20 days	23.33 j	59.00 bc	45.33 fg	43.67 gh	37.67 hi	
		30 days	23.67 j	71.33 a	40.67 hi	35.00 i	28.67 j	
	AST	Zero time	36.67 gh	70.67 bc	67.00 c	68.00 bc	66.00 cd	5.89
		10 days	35.00 gh	74.33 ab	70.33 bc	66.33 cd	60.33 de	
		20 days	36.00 gh	72.33 bc	55.33 e	44.67 f	38.67 fg	
		30 days	36.33 gh	79.33 a	57.67 e	39.33 fg	32.00 h	
Kidney functions (mg/dl)	Creatinine	Zero time	1.30 ij	2.20 abcd	2.10 cde	2.33 ab	2.17 bcd	0.19
		10 days	1.23 ij	2.27 abc	2.00 def	1.90 efg	1.87 fgh	
		20 days	1.20 j	2.20 abcd	1.70 gh	1.70 gh	1.43 i	
		30 days	1.23 ij	2.40 a	1.67 h	1.43 i	1.27 ij	
	Blood Urea	Zero time	50.67 k	75.67 fg	72.67 g	76.67 fg	74.67 g	5.81
		10 days	51.33 k	94.67 ab	93.33 bc	88.33 cd	83.33 de	
		20 days	50.00 k	97.67 ab	78.33 de	65.00 h	57.67 ij	
		30 days	56.00 jk	100.33 a	81.67 ef	62.67 hi	53.67 jk	
Lipid profile (mg/dl)	Triglycerides	Zero time	174.67 ijk	288.00 cd	285.00 d	292.00 cd	283.67 d	7.93
		10 days	171.00 k	300.67 ab	296.33 ab	275.67 e	267.00 f	
		20 days	172.67 jk	295.00 bc	190.33 g	184.00 h	174.00 ijk	
		30 days	181.67 hi	307.00 a	181.33 hij	175.67 hijk	172.33 k	
	Total Cholesterol	Zero time	194.67 ef	382.67 b	382.00 b	389.33 b	384.33 b	10.38
		10 days	189.67 f	407.67 a	405.00 a	388.33 b	381.67 b	
		20 days	194.00 ef	410.67 a	247.33 c	223.33 d	192.33 ef	
		30 days	197.67 ef	414.33 a	223.00 d	201.67 e	192.00 ef	
	HDL	Zero time	47.00 a	27.67 ef	28.00 ef	26.33 ef	28.00 ef	3.92
		10 days	44.67 ab	24.67 f	26.00 ef	25.33 f	27.00 ef	
		20 days	42.67 bc	27.33 ef	28.67 ef	30.00 e	34.67 d	
		30 days	43.67 ab	27.00 ef	40.33 bc	39.00 c	42.33 bc	
	LDL	Zero time	101.67 e	287.67 bcd	316.00 a	289.00 bcd	286.33 bcd	23.0
		10 days	103.67 e	299.00 abc	288.67 bcd	278.00 cd	269.33 d	
		20 days	105.67 e	308.67 ab	127.67 e	118.33 e	117.00 e	
		30 days	107.00 e	305.67 ab	125.67 e	116.00 e	108.00 e	
	vLDL	Zero time	34.67 hi	57.67 cd	56.67 d	58.33 cd	56.67 d	1.55
		10 days	34.33 i	60.33 ab	59.33 bc	55.00 e	53.67 e	
		20 days	34.67 hi	59.00 bc	38.33 f	37.00 g	34.67 hi	
		30 days	36.33 gh	61.33 a	36.00 ghi	35.33 ghi	34.33 i	

Group 1: Control non-diabetic rats; Subgroup 1: Control diabetic rats; Subgroups 5, 6 and 7 treated rats with the crude methanolic extract of flame seedless leaves with doses of 10, 20 and 40 mg/100g of body weight, respectively.

Table 3: Effect of crude methanolic extract of zizyphus leaves on levels of blood glucose (mg/dl), liver functions (U/L), kidney functions (mg/dl) and lipid profile (mg/dl) in diabetic rats.

Levels of :		Treatment period	Groups of Experimental Animals					LSD 5%
			Group 1	Group 2				
				Subgroup				
				1	8	9	10	
Serum glucose (mg/dl)		Zero time	124.67 d	388.33 abc	391.67 abc	410.33 ab	390.67 abc	69.52
		10 days	123.67 d	432.67 a	315.00 c	344.00 bc	331.33 c	
		20 days	123.33 d	425.67 a	196.33 d	176.33 d	159.33 d	
		30 days	124.67 d	461.33 a	185.67 d	124.00 d	128.33 d	
Liver functions (U/L)	ALT	Zero time	25.33 gh	58.00 bcd	53.67 cd	52.33 de	54.00 cd	5.55
		10 days	24.00 h	62.33 b	56.00 cd	47.67 e	47.33 e	
		20 days	23.33 h	59.00 bc	35.00 f	27.33 gh	26.67 gh	
		30 days	23.67 h	71.33 a	30.67 fg	24.67 gh	23.67 h	
	AST	Zero time	36.67 fgh	70.67 bc	67.00 c	68.00 bc	68.67 bc	5.73
		10 days	35.00 fgh	74.33 ab	68.67 bc	60.00 d	57.67 d	
		20 days	36.00 fgh	72.33 bc	42.33 e	39.33 fg	36.33 fgh	
		30 days	36.33 fgh	79.33 a	41.33 ef	31.67 h	33.00 gh	
Kidney functions (mg/dl)	Creatinine	Zero time	1.30 e	2.20 ab	2.17 b	2.20 ab	2.17 b	0.20
		10 days	1.23 e	2.27 ab	1.90 c	1.87 c	1.70 cd	
		20 days	1.20 e	2.20 ab	1.70 c	1.57 d	1.60 d	
		30 days	1.23 e	2.40 a	1.63 d	1.20 e	1.23 e	
	Blood Urea	Zero time	50.67 h	75.67 cde	72.67 ef	74.67 de	74.00 ef	8.769
		10 days	51.33 h	94.67 a	93.00 ab	84.67 bc	83.67 cd	
		20 days	50.00 h	97.67 a	62.67 ef	54.33 h	53.33 h	
		30 days	56.00 gh	100.33 a	64.67 fg	55.33 h	50.67 h	
Lipid profile (mg/dl)	Triglycerides	Zero time	174.67 gh	288.00 bc	286.33 bcd	286.67 bcd	284.33 bcd	11.49
		10 days	171.00 h	300.67 a	294.67 ab	279.00 cde	274.33 de	
		20 days	172.67 h	295.00 ab	232.33 e	174.00 gh	181.33 gh	
		30 days	181.67 gh	307.00 a	214.67 f	186.67 g	179.33 gh	
	Total Cholesterol	Zero time	194.67 i	382.67 b	387.33 b	384.00 b	383.00 b	10.72
		10 days	189.67 i	407.67 a	371.33 c	314.33 d	302.00 e	
		20 days	194.00 i	410.67 a	252.67 f	210.67 h	195.33 i	
		30 days	197.67 i	414.33 a	237.00 g	190.67 i	189.33 i	
	HDL	Zero time	47.00 a	27.67 def	29.00 cdef	28.00 def	26.33 ef	4.97
		10 days	44.67 ab	24.67 f	29.00 cdef	34.00 c	34.00 c	
		20 days	42.67 ab	27.33 def	37.67 cde	41.67 ab	41.00 b	
		30 days	43.67 ab	27.00 ef	33.00 cd	44.00 ab	43.00 ab	
	LDL	Zero time	101.67 g	287.67 b	286.00 b	284.33 b	285.33 b	10.09
		10 days	103.67 g	299.00 a	286.00 b	269.00 c	271.33 c	
		20 days	105.67 g	308.67 a	161.67 d	135.33 e	133.33 e	
		30 days	107.00 g	305.67 a	143.67 e	120.33 f	121.67 f	
	vLDL	Zero time	34.67 h	57.67 bc	57.33 bc	57.00 bcd	57.00 bcd	2.31
		10 days	34.33 h	60.33 a	59.00 ab	56.00 cde	54.67 de	
		20 days	34.67 h	59.00 ab	46.67 e	35.00 gh	36.33 gh	
		30 days	36.33 gh	61.33 a	43.33 f	37.33 g	36.00 gh	

Group 1: Control non-diabetic rats; Subgroup 1: Control diabetic rats; Subgroups 8, 9 and 10 treated rats with the crude methanolic extract of zizyphus leaves with doses of 10, 20 and 40 mg/100g of body weight, respectively.

Table 4: Effect of crude methanolic extract of pomegranate leaves on levels of blood glucose (mg/dl), liver functions (U/L), kidney functions (mg/dl) and lipid profile (mg/dl) in diabetic rats.

Levels of :		Treatment period	Groups of Experimental Animals					LSD 5%
			Group 1	Group 2				
				Subgroup				
				1	11	12	13	
Serum glucose (mg/dl)		Zero time	124.67 h	388.33 bc	419.00 ab	373.33 bc	382.00 bc	60.48
		10 days	123.67 h	432.67 ab	401.33 abc	303.67 de	334.33 cd	
		20 days	123.33 h	425.67 ab	261.00 ef	235.00 fg	173.00 gh	
		30 days	124.67 h	461.33 a	202.67 g	187.33 gh	133.67 h	
Liver functions (U/L)	ALT	Zero time	25.33 hi	58.00 bc	54.33 cd	56.67 bc	56.00 cd	5.68
		10 days	24.00 i	62.33 b	54.33 cd	50.00 de	47.33 e	
		20 days	23.33 i	59.00 bc	39.33 f	34.33 fg	30.67 gh	
		30 days	23.67 i	71.33a	39.33 f	30.67 gh	24.00 i	
	AST	Zero time	36.67 f	70.67 b	68.67 b	70.33 b	70.00 b	5.54
		10 days	35.00 f	74.33 ab	71.33 b	69.67 b	68.00 b	
		20 days	36.00 f	72.33 b	59.67 c	55.00 cd	50.00 d	
		30 days	36.33 f	79.33 a	78.33 a	60.67 c	43.00 e	
Kidney functions (mg/dl)	Creatinine	Zero time	1.30 gh	2.20 abc	2.27 ab	2.10 bcd	2.17 abcd	0.22
		10 days	1.23 h	2.27 ab	2.17 abcd	2.03 bcde	2.03 bcde	
		20 days	1.20 h	2.20 abc	1.93 de	1.83 e	1.57 f	
		30 days	1.23 h	2.40 a	2.00 cde	1.50 fg	1.30 gh	
	Blood Urea	Zero time	50.67 j	75.67 fg	73.67 fg	74.33 fg	74.00 fg	5.96
		10 days	51.33 j	94.67 ab	88.00 cd	85.00 de	79.67 ef	
		20 days	50.00 j	97.67 a	75.67 fg	72.67 g	63.67 h	
		30 days	56.00 ij	100.33 a	91.33 bc	73.67 fg	60.00 hi	
Lipid profile (mg/dl)	Triglycerides	Zero time	174.67 e	288.00 abc	286.67 abc	285.67 abc	287.33 abc	23.05
		10 days	171.00 e	300.67 ab	284.00 abc	274.33 bc	272.00 c	
		20 days	172.67 e	295.00 abc	245.33 d	237.67 d	217.33 d	
		30 days	181.67 e	307.00 a	286.33 abc	228.33 d	181.33 e	
	Total Cholesterol	Zero time	194.67 j	382.67 bcd	283.67 g	384.00 bcd	384.00 bcd	11.62
		10 days	189.67 j	407.67 a	393.67 b	378.67 cde	368.00 e	
		20 days	194.00 j	410.67 a	348.67 de	306.00 f	257.33 h	
		30 days	197.67 j	414.33 a	390.00 bc	304.67 f	212.33 i	
	HDL	Zero time	47.00 a	27.67 cde	27.67 cde	28.67 bcde	28.00 cde	4.83
		10 days	44.67 a	24.67 e	26.33 cde	26.00 de	27.00 cde	
		20 days	42.67 a	27.33 cde	29.33 bcde	32.00 bc	31.67 bcd	
		30 days	43.67 a	27.00 cde	24.67 e	25.00 e	34.00 b	
	LDL	Zero time	101.67 h	287.67 b	286.00 b	287.33 b	284.67 b	10.71
		10 days	103.67 h	299.00 a	273.67 c	260.00 d	257.00 d	
		20 days	105.67 h	308.67 a	174.00 e	164.67 e	147.67 f	
		30 days	107.00 h	305.67 a	129.33 g	124.00 g	105.00 h	
	vLDL	Zero time	34.67 d	57.67 ab	57.33 ab	57.00 ab	57.33 ab	4.53
		10 days	34.33 d	60.33 a	57.00 ab	55.00 b	54.33 b	
		20 days	34.67 d	59.00 ab	49.00 c	47.67 c	43.67 c	
		30 days	36.33 d	61.33 a	57.33 ab	45.67 c	36.33 d	

Group 1: Control non-diabetic rats; Subgroup 1: Control diabetic rats; Subgroups 11, 12 and 13 treated rats with the crude methanolic extract of pomegranate leaves with doses of 10, 20 and 40 mg/100g of body weight, respectively.

Table 5: Effect of crude methanolic extract of fig leaves on levels of blood glucose (mg/dl), liver functions (U/L), kidney functions (mg/dl) and lipid profile (mg/dl) in diabetic rats.

Levels of :		Treatment period	Groups of Experimental Animals					LSD 5%
			Group 1	Group 2				
				Subgroup				
				1	14	15	16	
Serum glucose (mg/dl)		Zero time	124.67 g	388.33 bc	471.00 a	424.33 ab	426.00 ab	49.51
		10 days	123.67 g	432.67 ab	417.67 ab	349.67 cd	352.67 cd	
		20 days	123.33 g	425.67 ab	310.33 d	250.33 e	226.67 ef	
		30 days	124.67 g	461.33 a	258.67 e	206.00 ef	183.00 f	
Liver functions (U/L)	ALT	Zero time	25.33 j	58.00 bcde	55.67 cdef	51.67 f	53.00 def	5.26
		10 days	24.00 j	62.33 b	58.67 bcd	58.00 bcde	52.33 ef	
		20 days	23.33 j	59.00 bc	48.67 g	51.00 f	31.00 i	
		30 days	23.67 j	71.33 a	39.67 h	50.67 f	30.67 i	
	AST	Zero time	36.67 h	70.67 abc	68.67 bc	65.33 bcd	64.00 cd	8.90
		10 days	35.00 h	74.33 ab	65.67 bcd	54.33 efg	50.33 fg	
		20 days	36.00 h	72.33 abc	67.33 bcd	62.67 cde	49.33 fg	
		30 days	36.33 h	79.33 a	57.00 def	51.33 fg	46.67 g	
Kidney functions (mg/dl)	Creatinine	Zero time	1.30 i	2.20 bc	2.33 ab	2.10 cd	2.27 abc	0.16
		10 days	1.23 i	2.27 abc	2.10 cd	1.93 de	1.90 e	
		20 days	1.20 i	2.20 bc	1.87 de	1.80 ef	1.60 gh	
		30 days	1.23 i	2.40 a	1.80 ef	1.70 fg	1.50 h	
	Blood Urea	Zero time	50.67 f	75.67 de	77.00 cde	73.33 e	74.00 e	6.52
		10 days	51.33 f	94.67 a	95.00 a	84.67 b	83.00 bc	
		20 days	50.00 f	97.67 a	93.33 a	84.67 b	77.33 cde	
		30 days	56.00 f	100.33a	93.67 a	82.00 bcd	71.33 e	
Lipid profile (mg/dl)	Triglycerides	Zero time	174.67 jk	288.00 cde	286.00 de	284.67 de	284.00 e	9.31
		10 days	171.00 k	300.67 ab	297.33 abc	287.67 cde	284.00 e	
		20 days	172.67 jk	295.00 bcd	202.33 fg	203.33 f	193.00 gh	
		30 days	181.67 ij	307.00 a	192.33 gh	187.00 hi	181.33 ij	
	Total Cholesterol	Zero time	194.67 i	382.67 c	390.00 bc	387.33 bc	383.67 c	14.67
		10 days	189.67 i	407.67 a	400.67 ab	386.00 bc	364.33 d	
		20 days	194.00 i	410.67 a	287.33 e	285.67 ef	272.67 f	
		30 days	197.67 i	414.33 a	274.67 f	254.67 g	235.00 h	
	HDL	Zero time	47.00 a	27.67 cde	27.00 cde	28.33 cde	27.67 cde	3.86
		10 days	44.67 ab	24.67 e	25.33 de	25.67 de	27.33 cde	
		20 days	42.67 b	27.33 cde	24.33 de	26.00 de	29.33 cd	
		30 days	43.67 ab	27.00 cde	30.67 c	29.00 cde	30.67 c	
	LDL	Zero time	101.67 k	287.67 e	289.33 de	284.67 e	284.33 e	8.60
		10 days	103.67 k	299.00 bc	297.67 bcd	292.33 cde	291.00 cde	
		20 days	105.67 k	308.67 a	241.67 f	239.33 g	232.00 g	
		30 days	107.00 k	305.67 ab	217.67 h	208.67 i	197.33 j	
	vLDL	Zero time	34.67 i	57.67 cde	57.33 cde	56.67 e	57.00 de	1.90
		10 days	34.33 i	60.33 ab	59.33 bc	57.33 cde	56.67 e	
		20 days	34.67 i	59.00 bcd	40.67 fg	41.00 f	38.67 g	
		30 days	36.33 hi	61.33 a	38.67 g	37.67 gh	36.33 hi	

Group 1: Control non-diabetic rats; Subgroup 1: Control diabetic rats; Subgroups 14, 15 and 16 treated rats with the crude methanolic extract of fig leaves with doses of 10, 20 and 40 mg/100g of body weight, respectively.

From previous tables, it could be reported that the most effective concentration for reducing blood glucose level was 40 mg/100g body weight for all extracts. The antidiabetic activity was observed after ten days then gradually increased for all groups during the experimental periods.

Data in tables (1, 2, 3, 4 and 5) showed that the most effective treatment was the methanolic extract of zizyphus leaves. It decreased blood glucose level to 128.33 mg/dl at a dose of 40mg/100g body weight after 30 days of administration. The maximum reduction percentage was 66.95 comparing with 388.33 in control diabetic rats.

Obtained data for zizyphus leaves were higher than those of Anand *et al.,* (1989) who used 50% of aqueous ethyl alcohol extract at the same doses. Serum glucose levels were decreased by 10.44, 12.53 and 13.43% comparing that of diabetic control rats after 21 days, respectively. Moreover, Glombitza *et al*., (1994) found that serum glucose levels decreased to 127.52 and 190.14 mg/dl after four weeks of administration of zizyphus butanol extract and christinin-A, respectively.

Crude methanolic extracts of thompson seedless, flame seedless, pomegranate and fig leaves decreased serum glucose levels with different extents i.e. 131.67, 141.33, 133.67 and 183.00 mg/dl, respectively, at the same dose of 40 mg/100g of body weight after 30 days. It is clear that the effective role of extracts was more pronounced after 30 days where the percentage of reduction were 66.09, 63.60, 65.58 and 52.87% for thompson seedless, flame seedless, pomegranate and fig methanolic leaves extracts, respectively comparing with blood glucose level for control diabetic rats.

These results agreed with large extents with those reported by several authors, for instance, Orhan *et al*., (2006) suggested that the aqueous extract of *vitis vinifera* leaves at doses of 250 and 500 mg/kg of body weight reduced reduced blood glucose concentration with a percentage of 32.4 and 27.8%, respectively, after 15 days of administration.

The reduction in serum glucose levels using methanolic extract of fig leaves at doses of 10, 20 and 40mg/100g body weight (33.39, 46.95 and 52.87%, respectively) were higher than those found by Allam *et al.,* (2008) who presented a lower reduction value of 30.67, 36.78 and 43.01% using aqueous extract of fig leaves at doses of 20, 40 and 60mg/100g body weight, respectively after the same period.

Several studies reported that investigated leaves contain polyphenols as bioactive compounds such as querecetin which produces an increase in the number of pancreatic islet, probably increase insulin release in streptozocin-diabetic rats and induces the hepatic glucokinase enzyme. The lowering property of plasma glucose, cholesterol and triglyceride could also be attributed to the ability of querecetin to regenerate pancreatic β-cells and to increase insulin release (Vessal *et al.,* 2003).

It could be concluded that lowering of blood glucose levels which was observed in diabetic animals may be due to the stimulation of β-cells of pancreatic islets and mediated through stimulation of insulin release resembling the oral hypoglycemic drugs or peripheral glucose utilization as reported by Esmaeili and Yazdanparast, (2004).

Effect of crude methanolic extracts of investigated leaves on liver functions:

Alanine amino transferase (ALT) and aspartate amino transferase (AST) activities are known as cytosolic marker enzymes reflecting hepatocellular necrosis as they are released into the blood after damaging of the cell membrane, therefore both enzymes are used as indicators for hepatic damage (Andallu and Vardacharyulu, 2001). So, ALT and AST activities in serum of non-diabetic and streptozocin-diabetic rats. The effect of methanolic extracts of thompson seedless, flame seedless, zizyphus, pomegranate and fig leaves of streptozocin-diabetic rats were also studied.

From tables (1, 2, 3, 4 and 5), it is clear that at the beginning of the experiment ALT and AST increased significantly from 25.33 and 36.67 U/L in non-diabetic rats to 58.00 and 70.67 U/L in diabetic ones. Such significant increase suggest a possible necrotic injury of the liver or cholestasis with hepatocellular necrosis as reported by Van Hoof and De Broe, (1994).

ALT activity:

Data in tables (2, 3, 4 and 5) showed that flame seedless, zizyphus, pomegranate and fig leaves decreased ALT activity to the values of 28.67, 23.67, 24.00 and 30.67, respectively. It could be observed that the most effective role of all extracts was recorded after 30 days by using 40 mg/100g body weight. The maximum percentage of reduction in enzyme activity reached to 60.91, 50.57, 59.19, 58.62 and 47.12% for ALT activity when thompson seedless, flame seedless, zizyphus, pomegranate and fig leaves extracts were used, respectively.

Our data showed that the reduction in ALT activity using methanolic extract of fig leaves at a dose of 40mg/100g body weight (47.12 %) which was higher than those found by Allam *et al.,* (2008) who offered a lower reduction value of 20.78 % using aqueous extract of fig leaves at the same dose and the same period.

Dahiru *et al.* (2005), found that the pretreatment of rats with ethanol extract of *Zizyphus mauritiana* leaves protected rats against carbon tetrachloride liver injury. ALT activity was lowered significantly from 321 U/L for animals administrated with carbon tetrachloride to reach 163 and 102 U/L for animals treated with 200 and 300 mg/kg body weight of *Zizyphus mauritiana* leaf extract, respectively.

From tables (1, 2, 3, 4 and 5), it could be concluded that the treatment with methanolic extract of thompson seedless leaves was the most effective which decreased ALT activity to 22.67 U/L after 30 days at 40 mg/100g body weight comparing with 58.00 U/L for streptozocin-diabetic rats. Our finding was in the same line with Pari and Suresh (2008) who found that ethanolic grape leaves extract significantly restored ALT values from 63.02 U/L for alcoholic affected rats to reach 52.13, 39.60 and 26.60 U/L for treated with doses of 25, 50 and 100 mg/kg body weight, respectively.

AST activity:

Data in tables (1, 2, 3, 4 and 5), clearly indicated that the most effective role of all extracts was recorded after 30 days when 40 mg/100g body weight was used. The maximum percentage of reduction represent about 46.23, 54.72, 53.30, 39.15 and 33.96 for AST activity for thompson

seedless, flame seedless, zizyphus, pomegranate and fig leaves, respectively when compared with diabetic control rats.

Our data showed that the most effective extract for decreasing AST activity was flame seedless leaves which decrease it to 32.00 U/L after 30 days at a dose of 40 mg/100g body weight comparing with 70.67 U/L for diabetic rats (table 2).

While, the methanolic extracts of thompson seedless, zizyphus, pomegranate and fig leaves decreased AST activity to 38.00, 33.00, 43.00 and 46.67 U/L after 30 days using 40 mg/100g body weight, respectively, tables (1, 3, 4 and 5).

The reduction in AST activity using methanolic extract of fig leaves at a dose of 40mg/100g body weight were 33.96%. This result was higher than those found by Allam *et al.,* (2008) who offered a lower reduction value of 16.67% using aqueous extract of fig leaves at the same dose and the same period.

Dahiru *et al.* (2005), suggested that the pretreatment of rats with 200 and 300 mg/kg body weight of ethanolic extract of *Zizyphus mauritiana* leaves protected rats against carbon tetrachloride liver injury by significantly lowering AST activity from 497 U/L to 296 and 243 U/L, respectively.

Pari and Suresh (2008), found that ethanolic grape leaves extract significantly restored AST levels from 161.91 U/L for alcohol administrated rats to 151.07, 123.53 and 83.81 U/L for treated rats at doses of 25, 50 and 100 mg/kg body weight, respectively.

Effect of crude methanolic extracts of investigated leaves on kidney Functions:

Determination of serum creatinine and urea were used as indicators for kidney functions. The effect of methanolic extracts of all samples under investigation on serum creatinine and urea levels in streptozocin-diabetic rats during the experimental periods are tabulated in tables (1, 2, 3, 4 and 5).

It could be noticed that the injection with streptozocin induced a significantly increase in serum creatinine and urea levels from 1.30 and 50.67 to 2.20 and 75.67 mg/ml compared with non-diabetic rats (tables 1, 2, 3, 4 and 5). These increasing may be attributed to the divers hormonal and metabolic changes that accompany diabetic and the toxic effect of streptozocin on kidney (Rogers *et al.*, 1986 and Gokce and Haznedaroglu, 2008).

Creatinine:

From tables (1, 2, 3, 4 and 5), it could be noticed that creatinine decreased with increasing the concentration of methanolic extracts and the period of experiment for all samples. Crude methanolic extract of thompson seedless, flame seedless, zizyphus, pomegranate and fig leaves reduced serum creatinine to 1.07, 1.27, 1.23, 1.30 and 1.50 mg/dl, respectively, after 30 days at a dose of 40 mg/100g body weight compared with 2.20 mg/dl for diabetic rats.

Previous data revealed that methanolic leaves extract of thompson seedless was the most effective treatment on serum creatinine level, where value of creatinine decreased to 1.07 mg/dl to achieve the maximum reduction of 51.36 % comparing with diabetic rats. While, flame seedless,

zizyphus, pomegranate and fig leaves reduced serum creatinine by the percentages of 42.27, 44.09, 40.91 and 31.82% after 30 days at a dose of 40 mg/100g body weight compared with 2.20 mg/dl for diabetic rats, respectively.

Also, data show that crude methanol extract of fig leaves decreased serum creatinine value from 2.40 mg/dl to 1.80, 1.70 and 1.50 mg/dl after 30 days using doses of 10, 20 and 40 mg/100g body weight, respectively. It is worthy to state that these data were more effective than those reported by Allam *et al*., (2008) who mentioned that aqueous extract of fig leaves decreased serum creatinine value from 2.33 mg/dl to 1.69, 1.64 and 1.59 mg/dl after 30 days using doses of 20, 40 and 60 mg/100g body weight, respectively.

Blood urea:

Investigated data indicated that methanolic leaves extract of thompson seedless, flame seedless, zizyphus, pomegranate and fig leaves at a dose of 40 mg/100g body weight after 30 days reduced the blood urea to 45.67, 53.67, 50.67, 60.00 and 71.33 mg/dl, respectively, compared with 75.67 mg/dl for diabetic rats (tables 1, 2, 3, 4 and 5).

Percentages of reduction for blood urea represent 29.07, 33.04, 20.71 and 5.73% for flame seedless, zizyphus, pomegranate and fig leaves extracts, respectively, compared with 75.67 mg/dl for diabetic rats. In addition, blood urea value was reduced from 100.33 mg/dl to 93.67, 82.00 and 71.33 mg/dl after 30 days at doses of 10, 20 and 40 mg/100g body weight, respectively, when crude methanol extract of fig leaves was used. Whereas, Allam *et al*., (2008) mentioned that aqueous extract of fig leaves decreased blood urea from 93.57 mg/dl to 89.44, 76.39 and 74.57 mg/dl at doses of 20, 40 and 60 mg/100g body weight, respectively, after the same period.

Previous data revealed that methanolic leaves extract of thompson seedless (table 1) was the most effective treatment in decreasing blood urea level. It decreased to 45.67 mg/dl, giving a reduction ratio of 39.64% comparing with diabetic rats.

Effect of crude methanolic extracts of investigated leaves on lipid profile:

Data recorded in tables (1, 2, 3, 4 and 5) revealed that serum triglycerides, total cholesterol, LDL and vLDL values increased from 174.67, 194.67, 101.67 and 34.67 in non-diabetic rats to 288.00, 382.67, 287.67 and 57.67 mg/dl in diabetic rats, respectively, by injection with streptozocin at a dose of 4.5 mg/100g body weight.

On the other hand, the same tables declare that there is a highly significant decrease in serum high density lipoprotein (HDL) level since it reached 27.67 mg/dl for streptozocin-diabetic rats, comparing with 47.00 mg/dl for non–diabetic rats at the beginning of experiment.

Many compositional abnormalities of the lipoproteins have been found in diabetic patients and the major cause of hypertriglyceridemia appeared to be the over production of vLDL which is attributed to hyperglycemia and/or increased influx of free fatty acids into the liver (Tilvis *et al.,* 1988).

Also, Fernandez *et al.,* (2001) suggested that increasing in LDL level may be attributed to some reasons such as an increase of intestinal absorption of lipid, an increase of cholesterol synthesis and increase of liver lipid synthesis or liver disfunction. In addition, the decreasing in serum HDL level may be due to the decrease of lecithin cholesterol acetyl transferase which responsible for estrification of cholesterol in HDL.

On the contrary, obtained data were not agreed with those by Canal *et al.* (2000), who reported that the rats treated with streptozocin-induced diabetes obtained a decline in the levels of total cholesterol compared with the control group with a reduction of the hyperglycaemia.

Triglycerides:

From tables (1, 2, 3, 4 and 5), it could be noticed that triglycerides decreased with increasing the concentration of methanolic extracts and the experimental period for all samples under investigation. Accordingly, the treatment of diabetic rats with 40mg/100g body weight of methanolic extract of thompson seedless and flame seedless leaves have the most effective, when the reduction of triglycerides levels achieve about 172.67 and 172.33 mg/dl. Also, pomegranate and fig leaves have the lowest effect with a same reducing value for triglycerides levels which were 181.33 and 181.33 mg/dl, respectively. While, zizyphus leaves have a moderate value for reducing triglycerides levels from 288.00 to 179.33 mg/dl at the end of the experiment.

The same tables showed that crude methanolic extract of fig leaves decreased triglycerides values from 307.00 to 192.33, 187.00 and 181.33 mg/dl after 30 days by using doses of 10, 20 and 40 mg/100g body weight, respectively.

These findings were at the same line of those reported by Pérez *et al.* (2003), who conducted that triglycerides values were reduced to 639 and 317 mg/dl by intraperitoneal injection with aqueous and chloroform extracts of *Ficus carica* leaves after 24 hours. Also, Allam *et al*., (2008) mentioned that triglycerides values were reduced from 125.78 to 86.44, 83.93 and 68.60 mg/dl after 30 days by using doses of 20, 40 and 60 mg/100g body weight of aqueous extract of fig leaves, respectively.

Total cholesterol:

From tables (1, 2, 3, 4 and 5), it could be shown that treatment of diabetic rats with 40mg/100g body weight of methanolic extract of zizyphus leaves for 30 days led to the highest reducing effect for total cholesterol levels. It reached to the level of 189.33 mg/dl with a reduction percentage of 50.52% comparing with total cholesterol value for diabetic rats at zero time. Although, methanolic extracts of thompson seedless and flame seedless leaves have a moderate effect with the same reducing value for total cholesterol levels which accomplished to 192.67 and 192.00 mg/dl with a reduction percentage of 49.82%. While, the diabetic rats which were treated with methanolic extracts of pomegranate and fig leaves at a dose of 40mg/100g body weight have the values of 212.33 and 235.00 mg/dl for total cholesterol with a reduction percentage of 44.51 and 38.59%, respectively.

Presented data for crude methanolic extract of fig leaves showed that total cholesterol values decreased from 414.33 to 274.67, 257.67 and 235.00 mg/dl after 30 days when doses of 10, 20 and 40 mg/100g body weight was

used, respectively. These findings were at the same line of those reported by Allam *et al*., (2008) who illustrated that total cholesterol values were reduced from 100.55 to 93.90, 88.55 and 84.57 mg/dl after 30 days by using doses of 20, 40 and 60 mg/100g body weight of aqueous extract of fig leaves, respectively.

High density lipoprotein (HDL):

Data in tables (1, 2, 3, 4 and 5) showed that oral administration with methanolic extracts of all samples leads to a gradual increase in serum HDL. Raising both of concentration of extracts and period of the experiment caused an increase in serum HDL to 31.00, 42.33, 43.00, 34.00 and 30.67 mg/dl for thompson seedless, flame seedless, zizyphus, pomegranate and fig leaves, respectively, at a dose of 40 mg/100g body weight at end of the experiment.

Reported data for crude methanol extract of fig leaves showed that HDL values increased from 27.00 to 30.67, 29.00 and 30.67 mg/dl after 30 days when doses of 10, 20 and 40 mg/100g body weight were used, respectively. On the other hand, our findings were not agreed with those reported by Allam *et al*., (2008) who found that HDL values were reduced from 50.84 to reach 42.98, 35.32 and 32.92 mg/dl after 30 days by using doses of 20, 40 and 60 mg/100g body weight of aqueous extract of fig leaves, respectively.

It could be concluded that the most effective increase were obtained for zizyphus leaves extract which acquired to 55.40% followed by flame seedless leaves extract which reached to 52.98%. While, thompson seedless, pomegranate and fig leaves extract increased HDL to the values of 12.03, 22.87 and 10.84% comparing with diabetic control rats at zero time, respectively.

Low density lipoprotein (LDL):

From tables (1, 2, 3, 4 and 5), it could be shown that all concentrations of methanolic extracts of all samples under investigation during experimental periods caused a decrease in serum LDL which became 131.67, 108.00, 121.67, 105.00 and 197.33 mg/dl for thompson seedless, flame seedless, zizyphus, pomegranate and fig leaves after 30 days at a dose of 40 mg/100g body weight, respectively. The percentage values of decreasing clarified that pomegranate leaves was the most effective extract in reduction of LDL which realized 63.49% followed by flame seedless which acquired to 62.46%. Though, the reduction of LDL reached to 57.70 and 54.23% for zizyphus and thompson seedless leaves extract, respectively.

Obtained data for crude methanolic extract of fig leaves showed that LDL values decreased from 305.67 to 217.67, 208.67 and 197.33 mg/dl after 30 days by using doses of 10, 20 and 40 mg/100g body weight, respectively.

These findings were agreed with those reported by Allam *et al*., (2008) who found that LDL values reduced from 25.16 to 17.29, 16.79 and 13.72 mg/dl after 30 days by using doses of 20, 40 and 60 mg/100g body weight of aqueous extract of fig leaves, respectively.

Finally, it could be reported that the crude methanolic extract of fig leaves have the lowest reduction of LDL to achieve 31.40% comparing with diabetic control rats.

Very low density lipoprotein (vLDL):

Tables (1, 2, 3, 4 and 5) showed that oral treatment with crude methanolic leaves extracts of thompson seedless, flame seedless, zizyphus, pomegranate and fig leaves have the same capability for decreasing vLDL from 57.67 mg/dl for streptozocin-diabetic rats to 34.67, 34.33, 36.00, 36.33 and 36.33 mg/dl at a dose of 40 mg/100g body weight after 30 days, respectively.

Data for vLDL values as a result of treatment with thompson seedless, flame seedless methanolic extracts pointed to extremely normal level which equal to 34.67 mg/dl for non-diabetic rats and have the most effective reduction of vLDL (39.88%). While, vLDL values by treatment with zizyphus, pomegranate and fig leaves extracts have slightly reduction which was about 37.00% comparing with vLDL value of diabetic rats.

Our findings for crude methanol leaves extract of fig showed that vLDL values decreased from 61.33 to 38.67, 37.67 and 36.33 mg/dl after 30 days by using doses of 10, 20 and 40 mg/100g body weight, respectively. On the other hand, these results were not agreed with those recommended by Allam *et al*., (2008) who found that vLDL values raised from 24.55 to 33.63, 36.44 and 37.93 mg/dl after 30 days by using doses of 20, 40 and 60 mg/100g body weight of aqueous extract of fig leaves, respectively.

REFERENCES

Abdel-Zaher, A. O.; Salim, S. Y.; Assaf, M. H. and Abdel-Hady, R. H. (2005). Antidiabetic activity and toxicity of Zizyphus spina-christi leaves. Journal of Ethnopharmacology, 101: 129–138.

Allam, Sahar O.; Nematalla, Kh. M. and Ensaf M. Khalil (2008). Effect of aqueous fig leaves extract as hypoglycemic agent. J. Agric. Sci. Mansoura Univ., 33: 8617-8629.

Anand, K. K.; Singh, B.; Chand, D.; Chandan, N. K. and Gupta, V. N. (1989). Effect of *Zizyphus sativa* leaves on blood glucose levels in normal and alloxan-diabetic rats. Journal of Ethnopharmacology, 27: 121-127.

Andallu, B. and Vardacharyulu, N. (2001). Effect of mulberry leaves on diabetes. Int. J. Diab. Dev. Countries, 21: 147-151.

Baytop, T. (1999). Bitkiler İle Tedavi (Gec¸mis¸te ve Bugün). Nobel Tıp Kitabevleri, Istanbul, pp. 357–358.

Bombardelli, E. and Morazzonni, P., (1995). Vitis vinifera L. Fitoterapia 66: 291–317.

Canal, J. R.; Torres, M. D.; Romero, A. and Pérez, C. (2000). A chloroform extract obtained from a decoction of *Ficus carica* leaves improves the cholesterolaemic status of rats with streptozocin-induced diabetes. Acta Physiol Hung., 87: 71-76.

Caughron, K. F. and Smith, E. L. (2002). Definition and description of diabetes mellitus. South Med. J, 95: 35-49.

CoStat program, Version 6.311 (2005). CoHort Software, 798 Lighthouse Ave. PMB 320, Monterey, CA, 3940, USA. http://www.cohort.com

Dahiru, D., William, E. T. and Nadro, M. S. (2005). Protective effect of Zizyphus mauritiana leaf extract on carbon tetrachloride-induced liver injury. African Journal of Biotechnology, 4: 1177-1179.

Duncan, D. B. (1955). Multiple range and multiple F test. Biometrics, 11: 1-42.

Esmaeili, M. A. and Yazdanparast, R. (2004). Hypoglycemic effect of Teucrium polium: studies with rat pancreatic islets. J. Ethnopharmacology, 95: 27–30.

Fernandez, M. D. ; Garcia, J. L. and Garcia, F. D. (2001). Upper gastrointestinal toxicity of rofecoxib and naproxen. N. Engl. J. Med., 334: 1398-1399. [C.F. (Hessien, Rania, A. A. (2003). Biochemical studies on mulberry (Morus alba) and prickly-pear (Opuntia sp.). M.Sc.Thesis, Agric. Biochem. Dept., Fac. of agric; Ain Shams, Univ. Cairo].

Ghasemi, M.; Sadeghipour; H.; Asadi; S. and Dehpour, A. R. (2007). Time-dependent alteration in romakalim-induced relaxation of corpus cavernosum from streptozocin-induced diabetic rats. Life Sciences, 81: 960–969.

Glombitza, K. W.; Mahran, G. H.; Mirhom, Y. W.; Michel, K. G. and Motawi, T. K. (1994). Hypoglycemic and antihyperglycemic effects of *Zizyphus spina-christi* in rats. Planta Medica., 60: 244-247.

Gokce, G. and Haznedaroglu, Z,M. (2008). Evaluation of antidiabetic, antioxidant and vasoprotective effects of *Posidonia oceanica* extract. Journal of Ethnopharmacology, 115: 122–130.

Han, B. H. and Park, M. H., (1986). Folk Medicine: The Art and Science. The American Chemical Society, Washington, DC, 205-211.

Hecht, A.; Geishberg, H. and Halse, M. (1973). Effect of chloropamide treatment on insulin secretion in diabetes, its realtioship to the hypoglycemic effect. Metabolism, 22: 723-727.

Hessien, Rania, A. A. (2003). Biochemical studies on mulberry (Morus alba) and prickly-pear (Opuntia sp). M.Sc. Thesis, Agric. Biochem. Dept., Fac. of Agric., Ain Shams Univ., Cairo, Egypt.

Ju–Tung, B. ; Kima Ji Su ; Chang, W.C. ; Hae K. L. ;Tae-Kyun Oha and Sei Chang Kim. (2008). Comparison between ethanolic and aqueous extracts from Chinese juniper berries for hypoglycemic and hypolipidemic effects in alloxan-induced diabetic rats. Journal of Ethnopharmacology, 115: 110–115.

Kirtikar, K.R. and Basu, B.D., (1984). Indian Medicinal Plants, Lalit Mohan Basu, Allahabad, p. 593.

Lebovitz, H. E. (2001). Diagnosis, classification and pathogenesis of diabetes mellitus. J. Clin. Psychiatry., 62: 27, 5-9.

Orhan, N.; Aslan, M.; Orhan, D. D.; Ergun, F. and Yeşilada, E. (2006). In-vivo assessment of antidiabetic and antioxidant activities of grapevine leaves (*Vitis vinifera*) in diabetic rats. Journal of Ethnopharmacology, 108: 280–286.

Pari, L. and Suresh, A. (2008). Effect of grape (Vitis vinifera L.) leaf extract on alcohol induced oxidative stress in rats. Food and Chemical Toxicology 46: 1627-1634.

Patton, C. J. and Crouch, S. R. (1977). Enzymatic determination of urea. Anal. Chem., 49: 464-469.

Pérez, C.; Canal, J. R. and Torres, M. D. (2003). Experimental diabetes treated with *Ficus carica* extract: effect on oxidative stress parameters. Acta Diabetol, 40: 3–8.

Reitman, S. and Frankel, S. (1957). Colorimetric methods for determining GOT and GPT. Amer. J. Clin. Path. 28: 56-63.

Richmond, W. (1973). Preparation and properties of cholesterol oxidase from Nocardia sp. And its application to the enzymatic assay of total cholesterol in serum. Clin. Chem., 19: 1350-1356.

Rogers, K.S. ; Friend, W.H. and Higgins, E.S. (1986). Metabolic and mitochondrial disturbance in STZ-treated Sprague-dawely and Sherman rats. proc. Soc. Exp. Bio. Med., 182: 167–176. [C.F. (Hessien, Rania, A. A. (2003). Biochemical studies on mulberry (Morus alba) and prickly-pear (Opuntia sp.). M.Sc. Thesis, Agric. Biochem. Dept., Fac. of Agric., Ain Shams Univ., Cairo, Egypt.].

Serraclara, A.; Hawkins, F.; Pérez, C.; Dominguez, E.; Campillo, J. E. and Torres, M. D. (1998). Hypoglycemic action of an oral fig-leaf decoction in type-I diabetic patients. Diabetes Research and Clinical Practice, 39: 19-22.

Soltani, N. ; Keshavarz, M. and Dehpour, A. (2007). Effect of oral magnesium sulfate administration on blood pressure and lipid profile in streptozocin diabetic rat. European Journal of Pharmacology, 560: 201–205.

Terada, M. ; Yasuda, H. and Kikkawa. (1998). Delayed Wallerian degeneration and increased neurofilament phosphorylation in sciatic nerves of rats with streptozocin-induced diabetes. Journal of Neurological Sciences, 155: 23–30.

Tilvis, R. S.; Tuskincn, M. R. and Mieltinen, T. A. (1988). Effect of insulin treatment on fatty acids of plasma and erothocyte membrane lipids in type 2 diabetes. Clinica Chimica Acta, 171: 293-303.

Van Hoof, V. O. and De Broe, M. E. (1994). Interpretation and clinical significance of alkaline phosphate isoenzyme patterns. Cht. Rev. Clin. Lab. Sci., 31: 193-197.

Venkatesh, S.; Reddy, G. D.; Reddy, B. M.; Ramesh, M. and Apparao, A. V. N. (2003). Anti-hyperglycemic activity of Carulluma asttenuate. Fitoterapia, 74: 274-277.

Vessal, M. ; Hemmatia, M. and Vasei, M. (2003). Antidiabetic effects of quercetin in streptozocin-induced diabetic rats. Comparative Biochemistry and Physiology Part C, 135: 357–364.

تأثير المستخلص الميثانولى الخام لأوراق بعض أشجار الفاكهة على بعض مكونات دم فئران التجارب المريضة بالسكر

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تم فى هذه الدراسة إستخلاص المركبات الفعالة لأوراق العنب بصنفيه طومسون عديم البذور ، فلام عديم البذور ، السدر ، الرمان ، التين عن طريق النقع فى الميثانول ثم التركيز تحت تفريغ بغرض الحصول على المستخلص الميثانولى الخام للأوراق تحت الدراسة . ثم دراسة تأثير تلك المستخلصات على بعض مكونات دم فئران التجارب المصابة بمرض السكر وكان مستخلص أوراق السدر بتركيز 40 ملجم/100جم وزن الجسم أفضل المستخلصات فى تخفيض سكر الدم حيث كانت نسبة التخفيض 66,95% مقارنة بالفئران المريضة بعد 30 يوم من المعاملة المستخلص . بالنسبة لوظائف الكبد كان مستخلص أوراق العنب طومسون عديم البذور بتركيز 40 ملجم/100جم وزن الجسم أفضل المستخلصات فى تحسين نشاط إنزيم ALT بنسبة 60,91% ، بينما كان أفضل تحسن فى نشاط إنزيم AST (بنسبة 46,23%) بعد المعاملة بمستخلص أوراق عنب فلام عديم البذور بتركيز 40 ملجم/100جم مقارنة بالفئران المريضة بعد 30 يوم من المعاملة . أما وظائف الكلى فإن أوراق العنب طومسون عديم البذور بتركيز 40 ملجم/100جم كان أفضل المستخلصات حيث قام بتخفيض مستوى الكرياتينين واليوريا فى سيرم الدم بنسبة 51,36 و 39,64% على التوالى مقارنة بالفئران المريضة بعد 30 يوم من المعاملة . وكذلك تأثرت مستويات الجلسريدات الثلاثية والكولسترول الكلى والكولسترول عالى الكثافة والكولسترول منخفض الكثافة فى سيرم الدم عند المعاملة بالمستخلص الميثانولى الخام للأوراق تحت الدراسة .

قام بتحكيم البحث

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