

Hypoglycemic Effect of Alcoholic Extracts of *Phyllanthus virgatus* and *Juniperus Phoenicea L*, on Streptozotocin-Induced Diabetic in Male Rats

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Abstract: Diabetes mellitus encompasses a heterogeneous group of disorders characterized by insulin hyposecretion and/or insensitivity. Therefore, the present study was designed to investigate the antihyperglycemic effect of alcoholic extracts of *Phyllanthus virgatus* and *Juniperus Phoenicea* leaves in streptozotocin-induced diabetic rats. Forty two Sprague-Dawley rats were randomized into 6 equal groups. Group 1 was kept as a negative control group Group 2, kept as positive control group, groups (3) and (4) were orally administered the extract of *Phyllanthus virgatus* in doses of 200 and 400 mg/kg B.wt/day respectively. While groups (5) and (6) were orally administered the extract of *Juniperus Phoenicea L* in doses of 200 and 400 mg/kg B.wt/day respectively, for four weeks. Blood samples were collected for serum biochemical analyses, also the pancreas was taken for histopathological examination. The results showed that the alcoholic extracts of *Phyllanthus virgatus* and *Juniperus Phoenicea* leaves at doses of 400 mg/kg B.wt/day significantly reduced body weight gain, feed intake and food efficiency ratio; normalized serum levels of liver enzymes and kidney function parameters; improved lipid profile; decreased blood glucose and MDA but increased in levels of insulin, GSH and SOD compared to that of untreated diabetic rats as well as there was an alleviation in the histopathological changes which seen in the pancreases of diabetic rats. The study recommends that administration of *Phyllanthus virgatus* and *Juniperus Phoenicea* Leaves extract can reduce blood glucose level and the incidence of different complications as results of hyperglycemia.

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Key Words: *Phyllanthus virgatus*, *Juniperus Phoenicea L*, antihyperglycemic, biochemical analyses, Histopathology, and Rats.

1. Introduction

Diabetes is a common metabolic disease with many side effects (Sarah et al., 2011). Diabetes increases blood glucose, and impaired metabolism of proteins and lipids. Altered cellular metabolism caused by hyperglycemia has been suggested to play an important role in increasing the risk of cardiovascular, renal, ophthalmic and neurological complications of diabetes mellitus (Anfenan, 2014). Despite considerable progress in the treatment of diabetes by oral hypoglycemic agents, search for newer drugs continues because the existing synthetic drugs have several limitations (Om Prakash et al., 2015). The herbal drugs with antidiabetic activity are yet to be commercially formulated as modern medicines, even though they have been acclaimed for their therapeutic properties in the traditional systems of medicine (Prasad et al., 2009).

The plants provide a potential source of hypoglycemic drugs because many plants and plant-derived compounds have been used in the treatment of diabetes (Bnouham et al., 2006).

Phyllanthus virgatus Forst (Euphorbiaceae) commonly known as Bhuiamla, has great potential as an antioxidant and an antihyperglycemic agent in vitro (Hashim et al., 2013). *Phyllanthus* has been used in Ayurvedic medicine for over 2,000 years and has a

wide number of traditional uses. *P. virgatus* is also known traditionally for its remedial properties and extensively known, for antioxidant property and used in the treatment of intestinal, liver, kidney, bladder problem, intestinal infection, cancer, and diabetes (Shabeer et al., 2009).

The genus *Juniperus* is the main member of family *Cupressaceae*. There are between 50 and 67 species of juniper, Junipers have needle-like leaves, their fruits called berries. The berries of junipers, especially *Juniperus communis*, are used as a spice, particularly in European cuisine (FAO, 1998). Also, some indigenous peoples of America have traditionally used juniper to treat diabetes (McCabe et al., 2005). *Juniperus phoenicea* (*J. phoenicea*) is considered as an important medicinal plant largely used in the Tunisian traditional medicine. Its leaves are used in the form of decoction to cure many diseases such as diarrhea, bronchitis, rheumatism (Madar, 1989), and diabetes (Amer et al., 1994 and Allali et al., 2008).

The present study was carried out to investigate the hypoglycemic effect of alcoholic extracts of *Phyllanthus virgatus* and *Juniperus Phoenicea L*. on streptozotocin-induced diabetic rats in order to explore its use in folk medicine as an antidiabetic herb.

2. Materials and Methods

Materials

Plant Material:

Dried leaves of *Phyllanthus virgatus* and *Juniperus Phoenicea* were purchased from Haraz Market for Herbs and Medicinal Plants, Cairo, Egypt.

Rats:

Forty two male albino rats (Sprague Dawley strain) weighting 200 ± 5 g were obtained from the animal house of National Research Center, Giza, Egypt.

Streptozotocin and Biochemical Kits:

Streptozotocin (STZ) was purchased from El-Gomhoryia Company for Chemicals; Cairo, Egypt. Kits for estimating serum lipid profile, Glucose enzymatic kits for estimating blood glucose (BG) and radioimmunoassay kits for leptin and insulin hormones were procured from Gamma Trade Company, Egypt. The other biochemical kits were obtained from Gama Trade Company, Dokki, Egypt.

Methods

Preparation of the basal diet:

The basal diet was prepared according to recommended dietary allowances for rats (American Institute of Nutrition, AIN) adjusted by **Reeves et al. (1993)**. Basal diet consisted of 14 % casein, 10 % sucrose, 5 % corn oil, 0.25% choline chloride, 1% vitamin mixture (**Campbell, 1963**), 3.5 % salt mixture (**Hegsted et al., 1941**), 5% fibers and the remainder was corn starch up to 100 %.

Preparation of Alcoholic Extract of *Phyllanthus virgatus* and *Juniperus Phoenicea* leaves:

Leaves of *Phyllanthus virgatus* and *Juniperus Phoenicea* were finely grounded into a fine powder using a mechanical grinder. Dried plant (10 g) was extracted twice with 100 ml of 50% methanol in distilled water. The pooled extract was filtered and concentrated at 40 ° C using a rotary evaporator under low pressure. The residue was freeze-dried in a lyophilizer and stored at -20°C until used (**Kanchana and Nuannoi, 2012**).

Induction of Diabetes:

Diabetes was induced by a single intraperitoneal (i.p.) injection of freshly prepared STZ (at a dose level of 65 mg/kg of body weight dissolved in 0.02 ml of 0.05 M citrate buffer pH 4.5) according to (**Nafiu et al., 2011**). After i.p. injection, the animals allowed drinking 5 % glucose solutions overnight to overcome the death from hypoglycemia shock. Seventy-two hrs later, the blood samples obtained from orbital plexus vein of each injected rat by a fine capillary glass tube and the blood glucose concentration was determined to confirm induction of diabetes, the non-diabetic rats excluded from the study, and diabetes established with no- fasting blood glucose levels of ≥ 200 mg/dl (**Al-Malki et al., 2015**).

Experiment and grouping of rats:

Forty two mature male Sprague-Dawley rats were housed in a well-ventilated animal room under controlled hygienic conditions of 24°C temperature, 50% relative humidity, and 12 h light/12 h dark cycles. After acclimatization, rats were randomized into 5 groups of 7 rats each. Group (1) was fed on basal diet and kept as a normal control, while the other four groups were injected i.p. with STZ to induce diabetes. Blood glucose levels will be monitored on the 3rd day after STZ injection until confirmation of DM (> 200 mg/dl) according to a method of (**Al-Malki et al., 2015**). Group (2) was left as diabetic untreated rats (positive control group), animals fed standard diet and diabetic rats of groups (3) and (4) were orally administered the extract of *Phyllanthus virgatus* in doses of 200 and 400 mg/kg B.wt/day respectively, for four weeks. While groups (5) and (6) were orally administered the extract of *Juniperus Phoenicea* L in doses of 200 and 400 mg/kg B.wt/day respectively, for four weeks. At the end of the experimental period, blood samples were collected from orbital plexus of the vein, left to clot, and centrifuged at 3000rpm for serum separations. Rats were euthanized using deep ether anesthesia method; pancreas tissue was dissected out for determination of antioxidant biomarkers and histopathological examination.

The feed intake was calculated daily and the body weight gain (BWG) was recorded twice a week. Feed efficiency ratio (FER) was calculated, according to the method described by **Chapman et al., (1959)** using the following equations:

$$\text{BWG} = \text{final weight (g)} - \text{initial weight (g)}.$$

$$\text{FER} = \text{BWG (g/day)} / \text{Feed Intake (g/day)}.$$

Biochemical Analyses:

Blood glucose (BG) was determined using glucose enzymatic kits according to **Siest et al., (1981)**. Serum insulin was estimated using a specific antibody radioimmunoassay kit (**Yellow and Bauman, 1983**). Total cholesterol (TC) (**Richmond, 1973**), triglycerides (TG) (**Friedewald et al., 1972**), atherogenic index (**Kumari et al., 1995**) and high density lipoprotein cholesterol (HDL) (**Richmond, 1973**) were chemically determined using specific diagnostic kits and measured using a spectrophotometer. Low-density lipoprotein (LDL) cholesterol was calculated according to **Friedewald et al., (1972)**. Serum aspartate aminotransferase (AST) and alanine aminotransferase (ALT) (**Bergmeyer et al., 1978**), alkaline phosphatase (ALP) (**Roy, 1970**). Blood urea nitrogen was determined using Bio Mérieux kits according to (**Patton and Crouch 1977**), Serum uric acid was determined using the enzymatic colorimetric method as described by (**Fossati et al., 1980**). Serum creatinine concentrations were

calorimetrically determined by (Husdan and Rapoport, 1968). Malondialdehyde (MDA) (Yoshioka *et al.*, 1979), reduced glutathione (GSH) (Beutler *et al.*, 1963), and superoxide dismutase (SOD) (Giannopolitis and Ries, 1977) were measured in homogenized pancreas.

Histopathological Examination:

Pancreas of the scarified rats were taken and immersed in 10% formalin solution. The fixed specimens were then trimmed, washed and dehydrated in ascending grades of alcohol. Specimens were then cleared in xylol, embedded in paraffin, sectioned at 4-6 microns thickness and stained with Haematoxylin and Eosin stain for histopathological examination as described by Carleton, (1979).

Statistical Analysis

Statistical analysis was carried out using Statistical Package for the Social Sciences (SPSS) for

Windows, version 20 (SPSS Inc., Chicago, IL, USA). The collected data were presented as a mean± standard deviation (SD). Analysis of Variance (ANOVA) test was used for determining the significances among different groups according to Armitage *et al.* (2002). All differences were considered significant if $P < 0.05$.

3. Results

The statistical data in Table 1 showed that untreated diabetic rats (control +ve group) significantly decreased ($P < 0.05$) FI, BWG, and FER when compared to negative control rats. Oral administration of an alcoholic extract of *Phyllanthus and Juniperus Phoenicea* leaves at doses 200 and 400 mg/kg B.wt., given to diabetic rats induced significant increases in BWG and FER compared to the positive control group.

Table (1): The effect of oral administration of *Phyllanthus* leaves (PLAE) and *Juniperus Phoenicea* (JPAE) alcoholic extracts on feed intake (FI), body weight gain (BWG) and food efficiency ratio (FER) in diabetic rats.

Parameters	FI (g/day)	BWG (g/day)	FER
G1: Control (C-ve)	23.69±0.67a	2.23±0.29a	0.094±0.002 a
G2: Control (C+ve)	16.97±0.99d	1.77±0.13d	0.104±0.011d
G3: PLAE (200 mg/kg b.wt.)	19.88±0.77c	1.82±0.62c	0.092±0.017c
G3: PLAE (400 mg/kg b.wt.)	22.69±1.33b	2.17±0.14b	0.096±0.009b
G5: JPAE (200 mg/kg B.wt.)	19.01±1.05c	1.89±0.32c	0.099±0.018c
G6: JPAE (400 mg/kg B.wt.)	21.23±0.75b	2.12±0.34b	0.099±0.016b

Data are presented as means ± standard deviation, (n = 7 for each group) Values with different superscripts within the column are significantly different at $P < 0.05$. Values with similar or partially similar superscripts are non-significant.

Data recorded in Table 2 showed that hyperglycemic rats had significant increases ($P < 0.05$) in BG, while there was a significant decrease in serum insulin levels when compared to the negative control group. Oral administration of an alcoholic extract of

Phyllanthus and Juniperus Phoenicea leaves at different doses had significant decreases in BG and a significant increase in serum insulin level when compared to the positive control group.

Table (2): The effect of oral administration of *Phyllanthus* leaves (PLAE) and *Juniperus Phoenicea* (JPAE) alcoholic extracts on blood glucose and insulin hormone levels in diabetic rats.

Parameters	BG (mg/dl)	Insulin (ng/ml)
G1: Control (C-ve)	113.68 ± 1.24d	3.17 ± 0.12a
G2: Control (C+ve)	227.07 ± 1.06a	0.87 ± 0.17 ^c
G3: PLAE (200 mg/kg b.wt.)	149.14 ± 0.65 ^b	2.16 ± 0.13 ^b
G3: PLAE (400 mg/kg b.wt.)	116.26 ± 1.01d	3.12 ± 0.11 ^a
G5: JPAE (200 mg/kg B.wt.)	125.58 ± 2.43c	3.49 ± 0.36 ^a
G6: JPAE (400 mg/kg B.wt.)	112.08 ± 1.04d	3.24 ± 0.11a

Data are presented as means ± standard deviation, (n = 7 for each group) Values with different superscripts within the column are significantly different at $P < 0.05$. Values with similar or partially similar superscripts are non-significant.

As demonstrated in Table (3), hyperlipidemic rats had significant increases ($P < 0.05$) in the serum levels of total cholesterol (TC), triglycerides (TG), low density lipoprotein (LDL -c), and atherogenic index (AI) while, there was a significant decrease in the serum level of HDL -c when compared to those fed on basal diet only. Oral administration of alcoholic extract

of *Phyllanthus* (PLAE) and *Juniperus* Phoenicea (JPAE) leaves at doses 200 and 400 mg /kg b.wt., to diabetic rats significantly decreased ($P < 0.05$) the elevated serum levels of TC, TG, LDL -c, and increased serum levels of HDL-c and improved AI when compared with control positive rats.

Table (3): The effect of oral administration of *Phyllanthus* leaves (PLAE) and *Juniperus* Phoenicea (JPAE) alcoholic extracts on serum levels of total cholesterol (TC), (TG), LDL -c, HDL -c and Atherogenic index (AI) in rats in diabetic rats.

Parameters Groups	TC (mg/dL)	TG (mg/dL)	LDL (mg/dL)	HDL (mg/dL)	AI (LDL/ HDL)
G1: Control (C-ve)	89.89 ± 0.27dc	56.15 ± 0.41c	28.52 ± 0.87d	50.14 ± 0.22a	0.569
G2: Control (C+ve)	127.13 ± 1.14a	122.05 ± 1.26a	89.24 ± 0.75a	13.48 ± 0.54c	6.620
G3: PLAE (200 mg/kg b.wt.)	101.79 ± 1.18b	63.17 ± 1.29b	49.03 ± 0.54b	38.86 ± 1.28b	1.262
G3: PLAE (400 mg/kg b.wt.)	88.82 ± 1.62c	55.99 ± 2.71c	27.89 ± 0.18d	49.16 ± 0.17a	0.567
G5: JPAE (200 mg/kg B.wt.)	100.46 ± 0.03b	59.06 ± 0.21c	39.32 ± 0.46c	48.52 ± 1.42a	0.810
G6: JPAE (400 mg/kg B.wt.)	87.67 ± 0.23c	57.21 ± 0.47c	26.62 ± 0.87d	49.07 ± 0.22a	0.542

Data are presented as means ± standard deviation, (n = 7 for each group) Values with different superscripts within the column are significantly different at $P < 0.05$. Values with similar or partially similar superscripts are non-significant.

Hyperglycemic rats had significant increases ($P < 0.05$) in the serum level of liver enzymes AST, ALT, and ALP when compared with negative control rats. Alcoholic extract of *Phyllanthus* and *Juniperus Phoenicea* leaves when orally given to diabetic rats at

different dosage levels significantly lowered ($P < 0.05$) serum levels of AST, ALT, and ALP enzymes compared to the positive control group, in a dose-dependent manner, as illustrated in Table 4.

Table (4): The effect of oral administration of *Phyllanthus* leaves (PLAE) and *Juniperus* Phoenicea (JPAE) alcoholic extracts on serum liver enzyme AST, ALT and ALP in diabetic rats.

Parameters Groups	AST (U/L)	ALT (U/L)	ALP (U/L)
G1: Control (C-ve)	59.28 ± 1.45d	54.64 ± 0.26d	68.71 ± 2.90c
G2: Control (C+ve)	149.82 ± 1.21a	130.21 ± 0.32a	116.73 ± 1.55a
G3: PLAE (200 mg/kg b.wt.)	80.45 ± 2.14b	88.01 ± 0.13db	89.51 ± 1.34 b
G3: PLAE (400 mg/kg b.wt.)	71.03 ± 1.86b	72.42 ± 0.18 bc	71.91 ± 1.47c
G5: JPAE (200 mg/kg B.wt.)	79.15 ± 0.46b	81.21 ± 0.17b	87.51 ± 0.27 b
G6: JPAE (400 mg/kg B.wt.)	60.84 ± 0.27 c	58.21 ± 0.03 c	70.07 ± 0.21 c

Data are presented as means ± standard deviation, (n = 7 for each group) Values with different superscripts within the column are significantly different at $P < 0.05$. Values with similar or partially similar superscripts are non-significant.

Data recorded in Table 5 showed that diabetic rats had a significant increase ($P < 0.05$) in urea nitrogen, uric acid and creatinine compared to the negative control group. Oral administration of an alcoholic extract of *Phyllanthus* and *Juniperus Phoenicea* leaves at doses 200 and 400 mg /kg B.wt., to diabetic rats significantly decreased ($P < 0.05$) the elevated serum levels of urea nitrogen uric acid and creatinine when compared with the positive control group.

Hyperglycemic rats had a significant increase (P

< 0.05) in the serum level of lipid peroxide malondialdehyde (MDA), while, decreases in levels of reduced glutathione (GSH) and superoxide dismutase (SOD) when compared with the negative control group. Alcoholic extract of *Phyllanthus* and *Juniperus Phoenicea* leaves when orally given to diabetic rats at different dosage significantly decreased ($P < 0.05$) serum levels of MDA and increased levels of GSH and SOD compared to the positive control group, in a dose-dependent manner, as illustrated in Table 6.

Table (5): The effect of oral administration of Phyllanthus leaves (PLAE) and *Juniperus Phoenicea* (JPAE) alcoholic extracts on urea nitrogen, uric acid and creatinine in diabetic rats.

Parameters Groups	Urea nitrogen (mg/dl)	uric acid (mg/dl)	creatinine (mg /dl)
G1: Control (C-ve)	31.04 ± 0.12 ^d	3.29 ± 1.02 ^d	0.9 ± 1.05 ^d
G2: Control (C+ve)	69.08 ± 0.28 ^a	8.27 ± 0.24 ^a	5.2 ± 0.17 ^a
G3: PLAE (200 mg/kg b.wt.)	53.25 ± 1.03 ^b	6.52 ± 0.36 ^b	4.1 ± 0.26 ^b
G3: PLAE (400 mg/kg b.wt.)	30.29 ± 0.71 ^d	3.07 ± 0.87 ^d	1.1 ± 1.24 ^d
G5: JPAE (200 mg/kg B.wt.)	43.09 ± 1.54 ^c	4.89 ± 0.61 ^c	2.1 ± 0.91 ^c
G6: JPAE (400 mg/kg B.wt.)	33.04 ± 0.12 ^d	3.09 ± 1.02 ^d	0.79 ± 1.05 ^d

Data are presented as means ± standard deviation, (n = 7 for each group) Values with different superscripts within the column are significantly different at P < 0.05. Values with similar or partially similar superscripts are non-significant.

Table (6): The effect of oral administration of Phyllanthus leaves (PLAE) and *Juniperus Phoenicea* (JPAE) alcoholic extracts on serum levels of Malondialdehyde (MDA), reduced glutathione (GSH) and superoxide dismutase (SOD) levels in diabetic rats.

Parameters Groups	MDA (µmol/dl)	GSH (µmol/dl)	SOD (U/mg)
G1: Control (C-ve)	16.12 ± 1.32 ^d	35.06 ± 1.35 ^a	67.04 ± 1.06 ^a
G2: Control (C+ve)	34.21 ± 0.19 ^a	12.36 ± 0.71 ^c	30.74 ± 0.21 ^c
G3: PLAE (200 mg/kg b.wt.)	22.17 ± 1.27 ^b	29.21 ± 4.13 ^b	61.03 ± 1.56 ^b
G3: PLAE (400 mg/kg b.wt.)	16.02 ± 0.72 ^d	34.89 ± 0.29 ^a	66.79 ± 1.63 ^a
G5: JPAE (200 mg/kg B.wt.)	19.05 ± 0.36 ^c	30.27 ± 1.25 ^b	63.25 ± 0.24 ^b
G6: JPAE (400 mg/kg B.wt.)	17.01 ± 1.32 ^d	35.22 ± 1.35 ^a	66.32 ± 1.06 ^a

Data are presented as means ± standard deviation, (n = 7 for each group) Values with different superscripts within the column are significantly different at P < 0.05. Values with similar or partially similar superscripts are non-significant.

Histopathological Examination.

Microscopical examination of pancreas of the control group revealed a normal histopathological structure of pancreas cells Photo. (1). Meanwhile, pancreas of the positive control group showed vacuolar degeneration of epithelial lining pancreatic acini associated with pyknosis of their nuclei, and vacuolation of sporadic of \square -cells of Langerhans islets Photo. (2). Pancreas of diabetic rats treated with PLAE at a dose of 200 mg/kg b.wt., revealed vacuolation \square -cells of islets of Langerhans Photo. (3).

While, pancreas cells of diabetic rats treated with 400 mg/kg b.wt. PLAE showed improvement in histopathological changes, Photos (4). While Pancreas of diabetic rats treated with the alcoholic extract of *Juniperus Phoenicea* leaves at a dose of 200 mg/kg B.wt., revealed slight hypertrophy of islets of Langerhans Photo. (5). Pancreas sections of rats orally given 400 mg/kg B.wt., an alcoholic extract of *Juniperus Phoenicea* leaves showed no histopathological changes Photo (6).

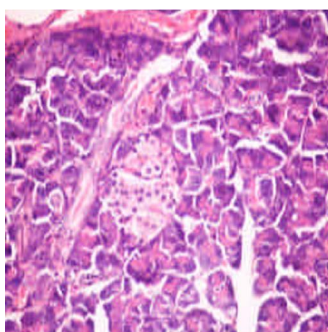


Photo:1

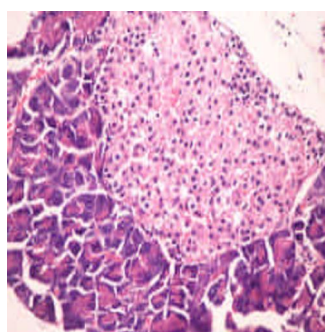


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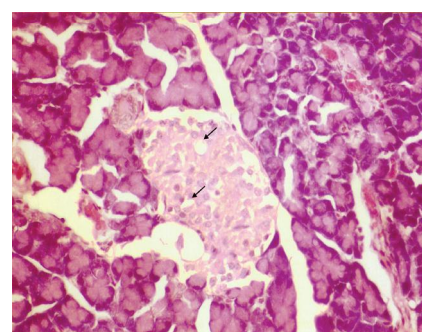


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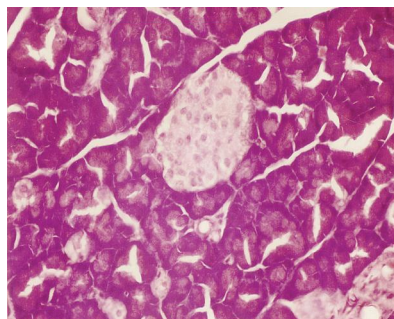


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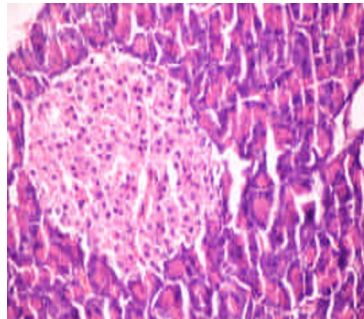


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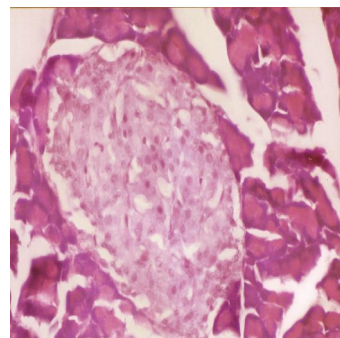


Photo: 6

Photo. 1: Pancreas of normal control rats showing no histopathological changes (H and E x 400).

Photo. 2: Pancreas of diabetic control rats showing hypertrophy and hyperplasia of beta- cells islets of Langerhans associated with pyknosis of their nuclei (H and E x 400).

Photo. 3: Pancreas of hyperlipidemic diabetic rats orally given PLAE at a dose of 200mg/kg b.wt. showing vacuolation □-cells of Langerhans islets. (H & E ×400).

Photo. 4: Pancreas of hyperlipidemic diabetic rats orally given PLAE at a dose of 400 mg/kg b.wt. showing no apparent histopathological changes. (H & E ×400).

Photo. 5: Pancreas of rats orally given 200 mg/kg B.wt. JPAE showing slight hypertrophy of islets of Langerhans (H and E x 400).

Photo. 6: Pancreas of rats orally given 400 mg/kg B.wt. JPAE showing no histopathological changes (H and E x 400).

4. Discussion

Diabetes mellitus encompasses a heterogeneous group of disorders characterized by insulin hyposecretion and/or insensitivity (Hashemnia *et al.*, 2012). Herbal therapies have been used in patients with insulin-dependent and noninsulin-dependent diabetes (Mukherjee *et al.*, 2006). The herbal drugs have been prescribed widely because of their effectiveness, fewer side effects and relatively low cost (Venkatesh *et al.*, 2003).

In the current study, STZ- induced diabetic rats showed a significant decrease in final body weight which also accompanied with increased feed intake and accordingly decreased in BWG%, as well as FER when compared with control group (-ve). The obtained results were in agreement with (Suryanarayana *et al.*, 2005 and Gupta *et al.*, 2012) who reported that STZ-induced diabetic rats showed signs of loss weight compared with rats non-injected with STZ. Kota *et al.*, (2012) found that there was an association between hyperglycemia and decreased body weight of diabetic animals, DM-induced reduction in body weight, and the body's inability to store or use glucose causes hunger and weight loss.

Previous studies have reported that there was an improvement in body weight and feed intake in diabetic rats treated with *Juniperus Phoenicea* and *Phyllanthus virgatus* Leaves extract compared with diabetic untreated rats. Our results agreement with the previous results and showed significantly decrease in body weight and increase in FI and consequently decrease in BWG% and FER as compared with control

(-ve) group, at the same time our results showed improvement in final body weight and FI accompanied with an increase in BWG% and FER when compared with untreated diabetic rats. The effect of *Juniperus Phoenicea* Leaves extract treatment may be explained by its ability to inhibit angiogenesis in adipose tissue and decrease differentiation of preadipocytes (Amer *et al.*, 1994). In addition, Saswata *et al.*, (2013) suggested that the gradual increase in the body weight was observed in the STZ diabetic rats treated with *Juniperus Phoenicea* Leaves extract may be due to the retained levels of glucose and insulin levels because of the antioxidant effects of *Juniperus Phoenicea* Leaves extract. Moreover, the obtained results were in agreement with Arshya *et al.*, (2014) who reported that *Phyllanthus virgatus* suppress the accumulation of body fat in rats via the inhibition of lipid synthesis or lipogenesis.

In the present study, STZ- induced diabetic rats showed a significant increase in serum glucose concentration accompanied by significant decrease in serum insulin level when compared with control group (-ve). Histopathological examination of pancreas sections from positive control rats showed the same results. A similar result was reported by Kumar *et al.*, (2012) who reported that rat injected with streptozotocin had been shown a marked raise in plasma glucose level and a decrease in insulin level. Diabetes syndromes characterized by increased blood glucose, altered lipids, carbohydrate and an increased risk of diabetic complications and oxidative stress (Al-Assaf, 2012). Moreover, STZ induced destruction

of β -cells of islets of Langerhans and causing degranulation and reduction of insulin secretion as proposed by **Zhang and Tan, (2002)**.

After treatment with plants extract at different doses, a significant reduction was observed in serum glucose concentration accompanied by significant increase in serum insulin level when compared with the positive control group, this may be due to the ability of the pancreas to revert to its normal condition during the experimental diabetic state. During this condition, the blood glucose level rises due to insulin deficiency but it has been seen that *Juniperus Phoenicea* Leaves extract significantly reduces the fasting blood glucose level in a dose-dependent manner probably by increasing the insulin release from the remnant beta cells (**Karuppusamy and Thangaraj, 2012**). In the current study, we observed that there was a significant decrease in serum glucose concentrations while insulin level demonstrated a significant increase in diabetic groups treated with alcoholic extract of *Phyllanthus virgatus* forst leaves as compared with control diabetic group. These results confirmed the result of **Arshya et al., (2014)** who reported that this may be due to the hypoglycemic effect of the alcoholic extract of *Phyllanthus virgatus* forst leaves due to proactive and pharmacological compounds.

In the present study, STZ- induced diabetic rats showed a significant increase in serum TC, TG and LDL-C levels accompanied by significant decrease in serum HDL-C level when compared with control group (-ve). A similar result reported that STZ-induced diabetic rat in a dose of 30 mg/kg had a negative effect on lipid profile levels when compared with normal rats (**Kota et al., 2012**). While treatment of diabetic rats with *Juniperus Phoenicea* Leaves extract exhibited remarkably ameliorated effects, there was a significant improvement in TC, TG, LDL-C and HDL-C levels when compared with the untreated diabetic group, these results are conforming with the results of **Saswata et al., (2013)**. The effect of *Juniperus Phoenicea* Leaves extract treatment may be explained by inhibiting pancreatic lipase activity and by inhibiting or delaying lipid absorption (**Ono et al., 2006**). The inhibitive capacities of the extract of *J. phoenicea* against the lipase activity might be perfectly coincident with their total phenolics compounds (**Henda et al., 2014**). After treatment with plant extract at different doses, a significant reduction was observed in TG, TC and LDL While there was a significant increase in HDL-C level in diabetic rats. The finding of this study is in agreement with previously published data that illustrated the antioxidant and antihyperglycemic effect of *Phyllanthus* methanol extract (**Shabeer et al., 2009**).

In the current study, STZ- induced diabetic rats

showed a significant increase in liver enzymes (AST, ALT, and ALP) compared to the negative control group. The hepatoprotective effect of *Juniperus Phoenicea* Leaves extract reported in this study was evident from the significant decrease in serum level of liver enzymes (AST, ALT, and ALP) in diabetic rats. This effect was in accordance with the previous reports for *Juniperus Phoenicea* Leaves extract and its polyphenols (**Salma et al., 2015**). Treatment of hyperglycemic rats with alcoholic extract of *Phyllanthus virgatus* leaves improves liver enzymes compared to the control positive group. This hepatoprotective activity may be attributed to the antioxidant activity of the plant. **Paritala, (2015)** demonstrated that Treatment with methanolic extract of the levels of *phyllanthus virgatus* have brought back the altered levels of the biochemical marker enzymes like AST, ALT, ALP, Total bilirubin, Total Cholesterol, tissue GSH and lipid peroxidation levels to near normal levels this may be due to scavenging free radicals.

The present study indicated that untreated diabetic rats had a significant increase in serum levels of BUN, UA, and Cr as compared to that of the normal control rats. These results agreed with **Ana et al., (2009)** who indicated that increased kidney functions are signs of kidney dysfunctions in the diabetic disease. After treatment with plant extract at different doses, a significant reduction was observed compared to the positive control group, this may be related to the antioxidant properties of alcoholic extract of *Juniperus Phoenicea* Leaves and its effect in reducing blood glucose level and radical scavenging effect. These results were agreed with **MAII et al., (2016)** who reported that hypoglycemic and antioxidant of *Juniperus Phoenicea* Leaves extract prevent oxidative stress which in turn led to decrease the serum levels of BUN, UA, and Cr. The results of the study showed that there was an improvement in kidney functions by the oral administration of the alcoholic extract of *Phyllanthus virgatus* leaves (compared to the positive control group, this may be related to the its antioxidant properties and its effect in reducing blood glucose level and radical scavenging effect. These results agreed with **Abdulla et al., (2012)** who reported that hypoglycemic and antioxidant of *Phyllanthus virgatus* prevent oxidative stress and preserve kidney function as was observed by low rates in BUN, UA and Cr.

Increasing ROS production plays an important role in the development and progression of hyperglycemia (**Suryanarayana et al., 2007**). Our results show that hyperglycemia increases pancreatic MDA and decreases GSH and SOD activities. These effects might be due to the fact that hyperglycemia increases oxidative stress through ROS overproduction

(Hussein and Abu-Zinadah, 2010). Treatment of hyperglycemic rats with *Juniperus Phoenicea* Leaves extract improves pancreas tissue contents of MDA, GSH, and SOD. Barrero *et al.*, (2004) reported that *Juniperus Phoenicea* Leaves extract reduces oxidative stress by reducing peroxide formation and increasing antioxidant enzyme activities.

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