The Egyptian Journal of Hospital Medicine (2008) Vol., 33: 544– 558

Evaluation of Hypoglycemic Activity of *Opuntia dillenii* Haw Fruit Juice in Streptozotocin-Induced Diabetic Rats

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Abstract:

Background: *Opuntia dillenii* Haw fruit is used in folk medicine as an antidiabetic agent. The aim of this study was to evaluate the possible curative role of *O. dillenii* fruit juice using the streptozotocin (STZ)-induced diabetic rats. The nutritive value of the edible portion of the fruit was also assessed.

Results: The results showed that *O. dillenii* fruit is a rich source of fiber, carbohydrates, vitamins B_1, B_2 and C, in addition to the minerals, Fe, Zn, Cu, Cr, Mn, Ca, and Mg. Biological results showed that intraperitoneal injection with STZ caused highly significant reduction in body weight gain% , highly significant elevation in blood glucose concentration accompanied by significant reduction in liver glycogen cotent as compared with control group. Diabetic rats also revealed significant elevation in lipid peroxide (MDA) level, highly significant elevation in total cholesterol (TC), triacylglycerols (TAG), low-density lipoprotein cholesterol (LDL-C) and very low-density lipoprotein cholesterol (VLDL-C) concurrent with highly significant reduction in high-density lipoprotein cholesterol (HDL-C) as compared with control group. Oral administration of *O. dillenii* juice had no effect on normal rats. Meanwhile, oral administration of *O. dillenii* juice to diabetic rats induced siginficant improvement in body weight gain % and lipid profile, it reduced significantly blood glucose and MDA levels as compared with non treated diabetic group. Histopathological investigation of the pancreatic tissue of STZ-diabetic rats represented the presence of necrosis, edema and congested blood vessels in the islets of Langerhans cells. *O. dillenii* fruit juice treatment overcome the previous changes, the majority of the cells tend to be normal.The improvement in the cells of Langerhans islets may explain the antidiabetic effect of the fruit juice under study. It also may improve the insulin receptors of β-cells.

Conclusion: It could be concluded that *O. dillenii* fruit juice had a potent hypoglycemic activity, this effect may be attributed to its antioxidant activity and its high content of chromium which was proved in this study. Therefore, it could be recommended that *O. dillenii* should be ingested as fresh fruit to diabetic and hypercholesterolemic patients beside the usual therapy.

Key words: *Opuntia dillenii* Haw fruit juice, Nutritive value, Hypoglycemic activity, Streptozotocin, Diabetic rats.

Introduction

Diabetes mellitus, a metabolic disorder, is characterized by hyperglycemia, altered metabolism of lipids, carbohydrates and proteins with an increased risk of complications of vascular diseases (Petrovsky and Schatz, 2003). Chronic hyperglycemia during diabetes causes permanent tissue damage, notably to the retinas, kidneys and nerve endings (American Diabetes Association, 2007). These may be delayed, decreased or prevented by maintaining blood glucose values close to normal. The increasing number of aging population, consumption of calories rich diet, obesity and sedentary life style have lead to tremendous increase in the number of diabetics worldwide (Wild *et al.,* 2004). According to WHO projections, the prevalence of diabetes is likely to increase by 35%. Currently there are over 150 million diabetics worldwide and this is likely to increase to 300 million or more by the year 2025 (Boyle *et al.,* 2001).

It is apparent that due to the side effects of the currently used drugs, there is a need for safe agents with minimal adverse effects, which can be taken for long duration. Recently, the search for appropriate hypoglycemic agents has been

focused on plants used in traditional medicine, partly because of the fact that natural products may be better treatments than currently used drugs **(**Rates, 2001). Many plants were reported to be useful for the treatment of diabetes mellitus. In Canary Islands folk medicine, *Opuntia dillenii* Haw fruit is used as antidiabetic agent (Perfumi and Tacconi, 1996). Cactus plants are commonly cultivated as ornamentals, some are valued for their edible fruits, others are grown as hedges, while few are used in herbal medicine. Family Cactaceae comprises about 50-150 genera, among which is the genus *Opuntia* which comprises about 250 species (Zomlefer, 1994 and Evans,2002).

O. dillenii (Ker-Gawl) Haw (family Cactaceae) commonly known as pear bush, prickly pear, mal rachette or tuna, is a succulent shrub growing under desert and dry conditions. It is native to American continent and the West Indies, but recently due to cultivation, it becomes widely distributed throughout Canary Islands, Southern and Eastern Africa, Pakistan, India and Australia (Ross, 1976 and Loro *et al.,* 1999). It has been introduced in Egypt as an ornamental and medicinal plant in the Orman Botanical Garden. *Opuntia* species are rich source of dietary fibers, natural colorants and antioxidant vitamins and therefore, used as a food because of their edible fruit (Saenz, 2002). Pharmacological evaluation of *Opuntia* has shown its efficacy as antihyperlipidemic, antiatheros-clerotic (Choi *et al.,* 2002), antiviral (Ahmed *et al.,* 1996), anti-inflammatory (Park *et al.,* 2001), antidiabetic (Tao *et al.,* 2005), antioxidant and antiulcerogenic agent (Galati *et al.,* 2003). It has also been reported to protect nerve cells and used for the treatment of Alzaheimer's disease, Parkinson's disease and stroke (Saleem *et al.,* 2005). In recent years, there has been a global trend toward the use of natural phytochemicals present in natural resources, such as fruits, vegetables and herbs, as antioxidants and functional foods (Kitts *et al.,* 2000).

Functional components, such as dietary fiber, and antioxidant vitamins, are some of the nutrients which people use in their daily diet. Therefore, the objective of this study was to evaluate the beneficial effect of *O. dillenii* fruit juice on biological, metabolic and antioxidant disorders in streptozotocin induced diabetic rats. The nutritive value of the edible fruit was also assessed.

Material And Methods

Material

Plant material

Plant material was collected in October (2006) from Orman Botanical Garden, Giza, Egypt and identified by Prof. Dr. K.H. Al-Batanony, Professor of Taxonomy, Faculty of Science, Cairo University. Fresh ripe fruit was used in this study.

Drugs and Chemicals

Streptozotocin (STZ) was purchased from Sigma Chemical Co.(St Louis, Mo, USA). Starch and corn oil were obtained from local market. Casein, vitamins, minerals, sucrose and cellulose were obtained from El-Gomhoriya Pharm. and Chem. Ind. Co. Cairo, Egypt.

Experimental animals

Thirty two adult male albino rats of Sprague Dawely strain, weighing (130± 5gm) were used for this study. They were kept in the animal house (National Research Center, Dokki, Egypt) for one week for proper acclimatization before starting the experiment under the same controlled laboratory conditions of illumination, temperature and ventilation. They were housed in stainless steel cages, maintained on standard casein diet (Reeves *et al.,* 1993) and water ad libitum throughout the experimental period.

Methods

Nutritive value

O. dillenii fresh ripe fruit was peeled. The edible portion was cut into pieces and dried by the hybrid solar convective drying system, belonging to the solar Energy Dept., National Research Center, Dokki, Egypt, at 30-40°C. The dried edible portion was ground.

Chemical analysis

Chemical analysis of the edible portion of the fruit was carried out

according to A.O.A.C International (2000).This was done in the Regional Center for Food and Feed, Agriculture Research Center, Giza, Egypt. Analysis included the determination of moisture, crude protein, crude fat, crude fiber and ash. While, total carbohydrates content were calculated by difference.

Study of the mineral content

The dried, powdered edible portion of the fruit was analyzed for micronutrients and macronutrients content. This determination was performed according to A.O.A.C International (2006) in the Regional Center for Food and Feed, Agriculture Research Center, Giza, Egypt. using atomic absorption spectrophotometer against standard elements.

Study of the vitamin content

Vitamins B_1, B_2, C, β - carotene, E and D_3 were determined in the fresh edible portion of the fruit by HPLC in the Regional Center for Food and Feed, Agriculture Research Center, Giza, Egypt. Vitamin B_1 was determined adopting the thiochrome fluorometric procedure, whereas vitamin B_2 was determined adopting the fluorometric method (National Food Agency of Denmark, 1996a). Vitamin C content was determined using the following conditions: suppelco RP C_{18} (5 µ x 250 x 4.6 mm) using acetate buffer as a mobile phase at a flow rate 1ml/min. UV detection was set at λmax 247 nm (National Food Agency of Denmark, 1996b). Vitamin A (β-carotene) and vitamin E were determined by using the following conditions: Si 60, 5 µm x 250 x 4.6 mm, n-heptane-isopropanol mixture flow rate 1ml/min. UV detection of vitamin E was set at 292 nm. Visible detection of β carotene was set at 450 nm (National Food Agency of Denmark, 1996c).Vitamin D_3 was determined by using the following conditions: suppelco C_{18} (5 µ x 250 x 4.6 mm) using methanol acetonitrile as a mobile phase at a flow rate 1.5 ml/min. UV detection was set at λmax 265 nm (National Food Agency of Denmark, 2001).

Preparation of the fruit juice

Opuntia dillenii Haw fresh ripe fruit was peeled (freed from cuticle and epidermis), then crushed in a Braun blender without using water. The palatable dense, red juice obtained was carefully filtered and then frozen until use.The juice was orally administered to the animals by an intragastric gavage.

Induction of diabetes

Diabetes was experimentally induced by using a single intraperitoneal (i.p.) injection of 50 mg/kg body weight STZ dissolved in 0.2 ml of 0.05 M citrate buffer pH: 4.5 according to Lutz and Pardridge (1993). Diabetic rats were supplied with 5% sucrose solution orally for the first 48 h., after STZ injection to minimize death from hypoglycemia (Peschke *et al*., 2000). Seventy-two h., later, blood samples were obtained by puncture of retro-orbital plexus with a fine capillary glass tube and blood glucose concentrations were determined to confirm induction of diabetes. Animals with blood glucose levels > 300 mg/dl were considered diabetic and used for the experiment.

Experimental design

After acclimatization period, rats were randomly divided into four groups, each of eight rats as follows:

Group 1: Conrol group, rats i.p. injected with 0.2 ml of 0.05 M citrate buffer pH: 4.5 (negative control).

Group 2: Diabetic rats (positive control).

Group 3: *O. dillenii* fruit juice group, rats i.p. injected with citrate buffer as in group (1), 72 h., later they received a daiy oral dose of the fruit juice at a dose level of 5 ml/kg body weight according to Perfumi and Tacconi (1996).

Group 4: Diabetic rats treated with *O. dillenii* fruit juice at the same dose in group (3).

The experiment lasted for four weeks starting from *O. dillenii* fruit juice administration. Food intake was recorded daily and animals were weighed once weekly. At the end of the experimental period, rats were deprived of food overnight. After ether anesthesia, blood samples were collected from hepatic portal vein in centrifuge tubes, left to clot and the supernatant sera were separated after centrifugation for 10 min., at 3000 r.p.m. for biochemical analysis.The pancreas tissues were collected immediately after scarification of rats in all groups and fixed in 10% formalin and prepared for histopathological examination.

Biochemical analysis

Separated serum samples were used for determination of glucose (Trinder, 1969), malondialdehyde (MDA) (Yoshioka *et al.*, 1979), total cholesterol (TC) (Allain *et al.*, 1974), high density lipoprotein cholesterol (HDL-C) (Lopes-Virella *et al.*, 1977) and triacylglycerols (TAG) (Fossati and Prencipe,1982).While,low-density lipoprotein cholesterol (LDL-C) and very low-density lipoprotein cholesterol (VLDL-C) were calculated according to the equation of Friedwald *et al*. (1972). Glycogen content in liver was determined according to the method of Hassid and Abraham (1957).

Histopathological examination

Specimens from pancreas were fixed immediately in 10% neutral buffered formalin, dehydrated in different grades of alcohol, cleared in xylol, embedded in paraffin wax, sectioned at 4-6 µ thick and stained with Haematoxylin and Eosin (Bancroft *et al.*, 1996) and examined microscopically.

Statistical analysis

Results were expressed as mean ± SE. Data were statistically analyzed for variance using one way analysis of variance (ANOVA) according to Snedecor and Cochron (1989). An IBM computer with a software system SPSS version 15 was used for these calculations.

Results

Chemical characterization and nutritive profile of the fruit

Chemical analysis of the edible portion of *O. dillenii* fruit is illustrated in Table (1). It included the determination of moisture, crude protein, crude fat, crude fiber, ash and total carbohydrates. The edible portion of *O. dillenii* fruit appeared to be rich in mineral elements (Table 2). Regarding micronutrients, the edible portion showed high contents of Fe, Zn and Cu being much higher than 100% of the RDA and high contents of Cr and Mn being much higher than 100% of the AI. For macronutrients, the edible portion showed to be rich source of Ca (65%-78% of the AI) and Mg (58.8%-79.7% of the RDA). In addition, Na and K were present in appreciable amounts representing 24.9%-28.7% and 40.2% of the AI for Na and K respectively. Table (3) revealed that the edible portion of the fruit was shown to be rich in vitamins B_1, B_2 representing 50%-54.5% and 53.8%-63.6% of the RDA, respectively. The content of vitamin C was 36 ± 0.88 mg/100g representing 40%-48% of the RDA. On the other hand, fat soluble vitamins (β - carotene and vitamin E) were detected in trace amounts.

Biological results

The effect of oral administration of *O. dillenii* fruit juice on biological parameters (body weight gain%, food intake and food efficiency ratio (FER) in normal and diabetic rats is represented in Table (4). There were highly significant decrease in body weight gain % in diabetic group as compared with control group. Administration of *O. dillenii* juice to normal rats did not influence on body weight gain%, there was no difference as compared with control group. Treatment of diabetic rats with *O. dillenii* juice induced noticeable increase in body weight gain%, there was significant difference between diabetic group and diabetic group treated with *O. dillenii* juice in body weight gain%.

Regarding food intake and FER, diabetic rats showed non-significant change in food intake meanwhile, FER showed highly significant decrease compared with control group. *O. dillenii* fruit juice group showed slightly increase in food intake when compared with control group, the value of FER tended to match control value. Administration of *O. dillienii* juice to diabetic group showed noticeable improvement in FER as compared with diabetic group, there was significant difference between diabetic treated and non treated groups.

Biochemical results

Table (5) revealed that diabetic rats showed significant elevation in lipid peroxide content meanwhile, *O. dillenii* fruit juice group showed non-significant change in MDA content compared with control group. On the other hand, treatment of diabetic rats with *O. dillenii* juice

showed amelioration in MDA content, its value tended to decrease as compared with non treated diabectic rats.

Data illustrated in table (5) also revealed that serum glucose concentration in diabetic rats showed highly significant elevation accompanied by significant reduction in liver glycogen content as compared with control group. Administration of *O. dillenii* juice to normal rats revealed non significant changes in serum glucose concentration and liver glycogen content, their values tended to match with the control values. Administration of *O. dillenii* juice to diabetic rats ameliorated the elevation in glucose concentration and the reduction in liver glycogen content, there was significant difference in glucose concentration between treated and non treated diabetic groups.

As shown in Table (6) there were highly significant elevation in TC, TAC, LDL-C, VLDL-C and LDL/HDL-C ratio concurrent with highly significant reduction in HDL-C in diabetic group as compared with control group. Administration of *O. dillenii* fruit juice to normal rats induced non-significant changes in all tested lipids parameters compared with control rats. Treatment of diabetic rats with *O. dillenii* juice recorded significant improvement in all tested lipid parameters when compared with diabetic group.

Histopathological results

Microscopical examination of the pancreas tissues from control and normal rats administered *O. dillenii* fruit juice revealed normal islets of Langerhans and pancreatic acini (Fig.1&2). The islets of Langerhans cells in pancreas tissue of streptozotocin diabetic rats showed necrosis and surrounded by mononuclear inflammatory cells infiltration and congested blood vessels (Fig.3). While, (Fig.4) presented edema of the islets of Langerhans which showed necrosis and some cells showed pyknotic nuclei. Oral administration of *O. dillenii* fruit juice to STZ diabetic rats improved the previous changes and partially reversed the damage caused by STZ to pancreas after four weeks of treatment. The majority of the cells consisting the islets of Langerhans in pancreas tissue of diabetic rats treated with *O. dillenii* fruit juice were normal but few cells showed necrosis and some cells contained pyknotic nuclei as illustrated in (Fig.5&6).

Table (1): Chemical analysis of the edible portion of *O. dillenii* fruit

Item	Percentage %
****Moisture**	83.00 ± 0.33
***Crude protein**	7.80 ± 0.20
***Crude fat**	4.20 ± 0.02
***Crude fiber**	29.48 ± 0.29
***Ash**	4.14 ± 0.07
***Total carbohydrates**	54.38 ± 0.54

- Each value represents the mean of 3 replications and expressed as mean ± SE.
- * = % on dry weight basis.
- ** = % on fresh weight basis.
- Data can be converted from dry weight to fresh weight by multiplying by (17/100).

Table (2): Mineral content of the edible portion of *O. dillenii* fruit.

Micronutrients	Concentration Mg/100g DW	RDA[a] and AI [b] For adults (amount/day)	Macronutrients	Concentration mg/100g DW	RDA[a] and AI [b] For adults (amount/day
Iron (Fe)	8.65 ± 0.10	8-18[a] mg	**Calcium (Ca)**	780.10 ± 9.58	1000-1200 [b] mg
Zinc (Zn)	9.52 ± 0.49	8-11 [a] mg	**Sodium (Na)**	373.25 ± 5.51	1300-1500 [b] mg
Copper (Cu)	2.09 ± 0.004	0.9 [a] mg	**Potassium (K)**	1890 ± 16.74	4700 [b] mg
Chromium (Cr)	0.064± 0.004	0.02-0.035[b] mg	**Magnesium (Mg)**	246.95 ± 9.96	310-420[a] mg
Manganese (Mn)	7.84 ± 0.002	1.8-2.3 [b] mg			

- Each value represents the mean of 3 replications and expressed as mean ± SE.
- DW = Dry weight.
- [a] RDA = Recommended dietary allowances.
- [b] AI = Adequate intakes **(Food & Nutrition Board, 2004).**

Table (3): Vitamin content of the fresh edible portion of *O. dillenii* fruit.

Vitamin	Content/ 100g FW	RDA[a] and AI [b] for adults (amount/day)
B_1	0.60 ± 0.03 mg	1.1-1.2 [a] mg
B_2	0.70 ± 0.03 mg	1.1-1.3 [a] mg
C	36.00 ± 0.88 mg	75-90 [a] mg
β-carotene	0.40 ± 0.01 µg	700-900 [a] µg
E	0.002 ± 0.0003 mg	15 [a] mg
D_3	Undetected	

- Each value represents the mean of 3 replications and expressed as mean ± SE.
- FW = Fresh weight.
- [a] RDA = Recommended dietary allowances.
- [b] AI = Adequate intakes (Food &Nutrition Board, 2004).

Table (4): Effect of oral administration of *O. dillenii* fruit juice on body weight gain, food intake and food efficiency ratio (FER) in normal and diabetic rats.

Groups / Parameters	Control	Diabetic	*O. dillenii* fruit juice	Diabetic + *O. dillenii* fruit juice
Body weight gain %	36.65±3.21	13.93±1.90[**a]	37.80±3.51	28.03±3.06
Food intake (g/day/group)	91.33±3.26	98.27±2.99	92.90±2.32	88.58±2.02
FER	0.151±0.013	0.047±0.006[**a]	0.151±0.008	0.120±0.017

- Each value represents the mean of 8 rats ± SE.
- * Significant difference from control group at $p < 0.05$ and ** highly significant difference from control group at $p < 0.01$.
- a: Significant difference between diabetic group and diabetic group treated with *O. dillenii* fruit juice at $p < 0.05$.

Table (5): Effect of oral administration of *O. dillenii* fruit juice on serum lipid peroxide as malondialdehyde (MDA, serum glucose and liver glycogen in normal and diabetic rats

Groups / Parameter	Control	Diabetic	*O. dillenii* fruit juice	Diabetic + *O. dillenii* fruit juice
MDA (nmol/l)	70.85±2.74	76.98±0.92^{*a}	69.21±1.29	71.76±1.64
Gucose (mg/dl)	116.68±1.94	325.32±6.65^{**a}	111.37±2.43	143.30±2.40
Liver glycogen (mg/g wet liver)	7.89±0.26	7.18±0.21*	8.25±0.08	7.49±0.13

- Each value represents the mean of 8 rats ± SE.
- * Significant difference from control group at $p < 0.05$ and ** highly significant difference from control group at $p < 0.01$.
- a: Significant difference between diabetic group and diabetic group treated with *O. dillenii* fruit juice at $p < 0.05$.

Table (6): Effect of oral administration of *O. dillenii* fruit juice on serum lipids profile in normal and diabetic rats.

Groups / Parameters	Control	Diabetic	*O. dillenii* fruit juice	Diabetic + *O. dillenii* fruit juice
TC (mg/dl)	84.21±0.87	113.55±1.62^{**a}	83.13±1.27	85.41±0.55
TAG (mg/dl)	96.34±1.25	135.09±2.81^{**a}	94.43±1.00	101.48±1.73
HDL-C (mg/dl)	44.96±1.03	30.20±0.99^{**a}	45.28±0.92	42.79±1.54
LDL-C (mg/dl)	19.98±1.80	56.34±2.26^{**a}	18.97±1.03	22.69±1.96
VLDL-C (mg/dl)	19.27±0.25	27.02±0.56^{**a}	18.89±0.20	20.30±0.35
LDL/HDL-C ratio	0.444±0.014	1.865±0.065^{**a}	0.419±0.019	0.530±0.025

- Each value represents the mean of 8 rats ± SE.
- * Significant difference from control group at $p < 0.05$ and ** highly significant difference from control group at $p < 0.01$.
- a: Significant difference between diabetic group and diabetic group treated with *O. dillenii* fruit juice at $p < 0.05$.

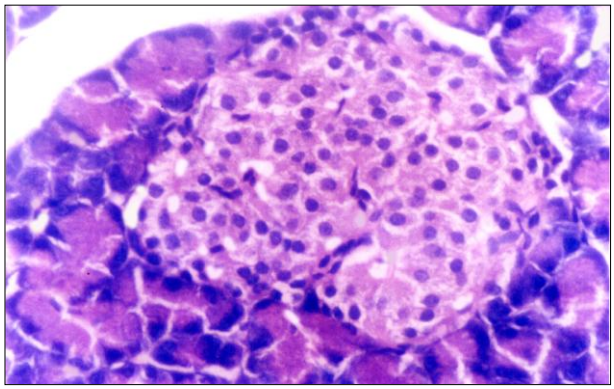


Fig. (1): Pancreas of control rats showing normal islets of Langerhans and pancreatic acini (H & E stain x 400)

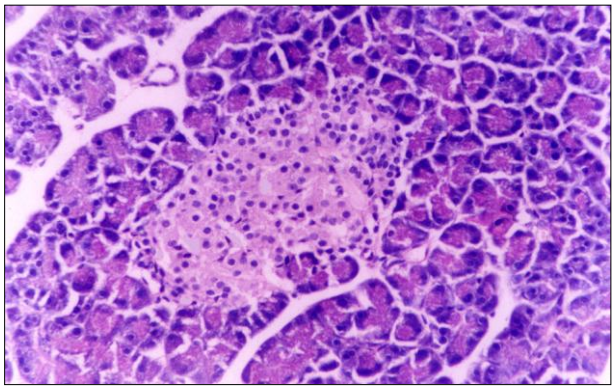


Fig. (2): Pancreas of normal rats administered *O. dillenii* fruit juice showing no histopathological changes (H&E stain x 200)

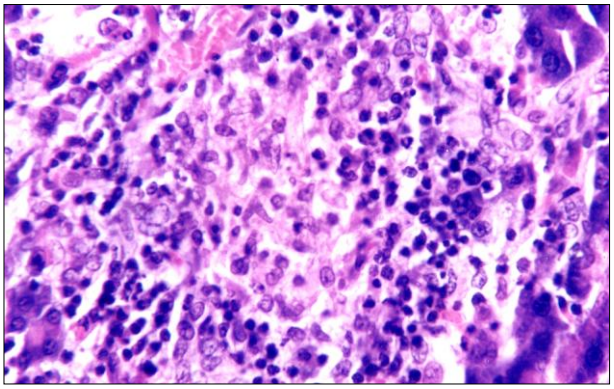


Fig. (3): Pancreas of STZ diabetic rats showing necrosis of the islets of Langerhans cells, surrounded by mononuclear inflammatory cells infiltration and congested blood vessels (H&E stain x 400)

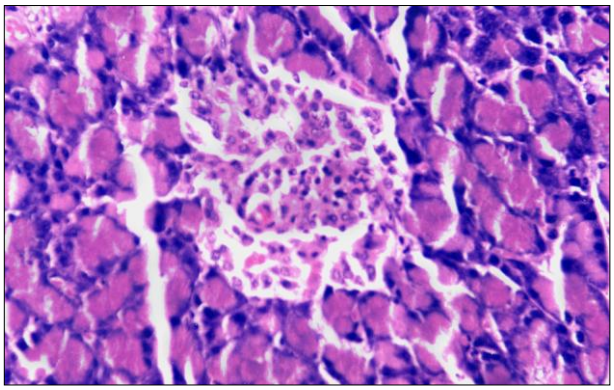
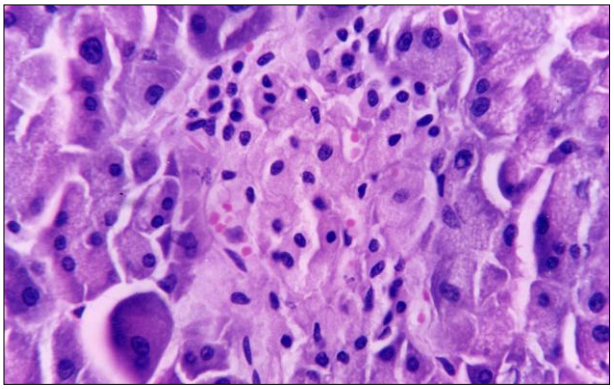
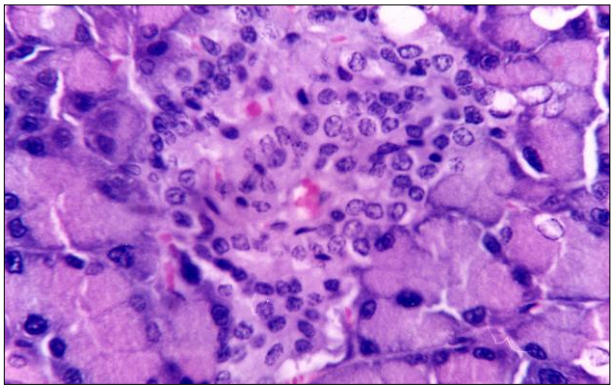


Fig. (4): Pancreas of STZ diabetic rats showing edema of the islets of Langerhans which showed necrosis and some cells showed pyknotic nuclei (H&E stain x 400).





Figs. (5&6): Pancreas of STZ diabetic rats treated with *O. dillenii* fruit juice showing that the majority of the cells consisting the islets of Langerhans were normal but few cells showing necrosis and some cells contained pyknotic nuclei (H&E stain x 400)

Discussion

In the present study, the chemical analysis of the edible portion of *O. dillenii* fruit revealed high percentage of fiber (29.48% ± 0.29) and carbabydrates (54.38% ± 0.54). The last values were similar to the values obtained by Ahmed *et al*. (2005b). Moreover, the investigated edible portion showed high contents of mineral elements. These results were in line with the resutls reported by Ahmed *et al*. (2005b), but higher than those reported by Díaz Medina *et al*. (2007) for the plant growing abroad. This variation may be due to different soils, thus the mineral content can be quite variable depending on the characteristics of the soil. In addition, the edible portion of the fruit was shown to be rich in vitamins B_1, B_2 and C. On the other hand, β- carotene and vitamin E were detected in trace amounts. These results were similar to the results of Ahmed *et al*. (2005b), while the content of vitamin C was higher than that reported by Chang *et al*. (2008) for the plant growing abroad (15.1±0.6 mg/100g fresh sample). The variation may be attributed to the variety of growth condition, genetic, environmental, handling and cultural factors.

In this study, it was found that diabetic rats revealed highly significant decrease in body weight gain% and FER, and non significant increase in food intake in diabetic group as compared with control group. The decrease in body weight in diabetic group may be attributed to different side effects of inability to use carbohydrates including lypolysis, glycogenolysis and acidosis (Ganang, 1995). In addition, Sjoslrom *et al*. (1998) reported that diabetes mellitus causes decrease in lipogenesis followed by increase in lipolysis, thus causes weight loss. The increase in food intake in diabetic rats may be attributed to polyphagia and polydipsia (Chen and Dawing, 1991). Treatment of diabetic rats with *O. dillenii* juice revealed noticeable improvement in body weight gain% and FER, there were significant differences between treated and non treated diabetic groups. This finding may be explained by the results of Rahman and Zaman (1989) who reported that the fruit of *O.dillenii* may contain an orally active insulin-like compound, which in turn inhibits epinephrine induced lipolysis and decreased body weight.

The current study elicited marked significant elevation in the lipid peroxidation product (MDA) level in diabetic group as compared with control group. This was in accordance with the observation of Maritim *et al*. (2003) who reported that induction of diabetes in rats with STZ uniformly results in an increase in lipid peroxidation (MDA), an indirect evidence of intensified free radical production. Ravi *et al*. (2004) reported significant elevation in plasma MDA level of diabetic rats when compared to control rats. The increase in the levels of lipid peroxides in plasma generally is thought to be the consequence of increased production and liberation into the circulation of tissue lipid peroxides due to pathological changes (Selvam and Anuradha, 1990).

Concerning diabetic rats treated with *O. dillenii* juice exhibited significant improvement in MDA level as compared with non treated diabetic rats. This effect was explained by Lee and Lim (2000) who stated that the aqueous and ethanol extracts of *O. dillenii* Haw have positive roles in scavenging reactive oxidants as natural antioxidants. Butera *et al*. (2002) also added that the methanolic extracts of *O. ficus indica* edible pulp inhibited lipid oxidation induced by organic hydrope-roxide in isolated human red blood cells and by either azo-compound-derived free radicals, or copper ions, in isolated human LDLs. In a further study made by Dok-Go *et al*. (2003), the isolated flavonoids, quercetin, dihydroquercetin and quercetin 3-methyl ether were reported to be the active antioxidant principles in the fruits of the *O. ficus-indica* var. saboten, exhibiting neuroprotective actions against the oxidative injuries induced in primary cultured rat cortical cells. Chang *et al*. (2008) also suggested that the phenolic acids and flavonoids of the methanolic extracts of *O. dillenii* Haw fruit play an important role in antioxidant activity and anti LDL peroxidation.

The obtained results revealed marked highly significant elevation in serum glucose concentration and significant reduction in liver glycogen content in diabetic rats when compared to control rats. Administration of *O. dillenii* juice to diabetic rats ameliorated the elevation in glucose concentration and the reduction in liver glycogen content, there was significant difference in glucose concentration between treated and non treated diabetic groups. These findings were in coincidence with Abd El Razek (2004) who found highly significant hyperglycemia and decrease in liver glycogen content in alloxan diabetic group as compared with non diabetic group. The hyperglycemia and decrease in liver glycogen content in diabetic rats may be due to lack of insulin, increased gluconeogenesis and/or glycogenolysis (Defronzo and Simonson, 1992).

O.dillenii juice failed to produce hypoglycemic activity in normal rats. This was in coincidence with the observation of Perfumi and Tacconi (1996) who found that single or repeated oral doses of *O. dillenii* juice did not alter blood glucose level in normoglycemic rabbits. The hypoglycemic effect of *O. dillenii* fruit juice on diabetic rats was different from that obtained by Perfumi and Tacconi (1996), they detected the hypoglycemic effect of *O. dillenii* juice when a glucose load was given orally to diabetic rabbits. On the other hand, the antihyperglycemic effect of the fruit juice under investigation was in agreement with that reported by Afifi *et al.* (1996) in diabetic rats and Tao *et al.* (2005) in diabetic mice. The previous authors reported that oral administration of the alcoholic extract, mucilage and pectin of *O. dillenii* to the previous animals significantly decreased the blood glucose level. Phytochemical analysis of *O. dillenii* alcoholic extract showed the presence of flavonoids (Ahmed *et al.*, 2005a), which are known to be bioactive antidiabetic principles (Rao *et al.*, 1997). Flavonoids are known to regenerate the damaged beta cells in the alloxan diabetic rats (Chakkravarthy *et al.*, 1980). *O. dillenii* fruit juice may act as hypoglycemic agent by stimulating insulin receptors of β-cells. Moreover, the antihyperglycemic activity of *O. dillenii* fruit juice may be attributed to its high content of chromium, an essential mineral involved in carbohydrate and lipid metabolism. Ample intake of chromium appears to promote insulin sensitivity and improve glycemic control (McCarty, 2005).

In the present results, diabetic rats showed highly significant elevation in TC, TAG, LDL-C and VLDL-C and LDL/HDL-C ratio concurrent with highly significant reduction in HDL-C as compared with control group. Meanwhile, diabetic rats treated with *O. dillenii* fruit juice exhibited significant improvement in these parameters when compared with non treated diabetic group. Diabetes Mellitus is a metabolic disorder affecting carbohydrate, fat and protein metsabolism. Colca *et al.* (1991) reported that hypercholesterolemia, hypertriglyceridemia and reduced HDL-C levels were commonly seen in diabetes. Similar results were obtained by Cameron-Smith *et al.* (1994) who stated that STZ-induced diabetic rats had elevated fasting TC and TAG levels relative to non-diabetic rats. Moreover, Kameswararao *et al.* (2003) found significant higher values of serum TC, TAG, LDL-C and significant lower values of HDL-C in diabetic rats compared to normal rats, which were fed the same diets. Yousri *et al.* (2002) suggested that increasing the level of serum TAG may be occurred as consequence of lipoprotein lipase inactivation in adipose tissue, which reduced the ability to uptake the TAG from serum leading to accumulation in serum. Fernandez *et al.* (2001) demonstrated that, increasing serum LDL-C level may be related to increase of intestinal absorption of lipid due to increased cholesterol synthesis and increased liver lipid synthesis. While, decreasing serum HDL-C level may be attributed to decrease of lecithin-cholesterol acetyl transferase (LCAT), which is responsible of estrification of cholesterol in HDL.

The therapeutic effect of *O. dillenii* juice on diabetic rats was in coincidence with the observations of Lee and Lim (2000) who found a marked statistical reduction in plasma TC, TAG, LDL-C and VLDL-C in diabetic rats after receiving the aqueous and ethanol extracts of *O. dillenii* Haw.The hypocholesterolemic action may be partly explained by the soluble fiber (pectin)

content (Wolfram *et al.*, 2002). The effect of pectin on serum cholesterol levels could be due to a number of factors. Of prime significance is the possibility that this effect could be mediated through its shifting the bile acids pools away from cholic acid and toward chenodeoxycholic acid. The chenodeoxycholic acid inhibits 3-hydroxy-3-methylglutary (HMG) CoA reductase (a regulatory enzyme necessary for cholesterol biosynthesis). Finally, decreased HMG CoA reductase activity results in reduced hepatic cholesterol synthesis and theoretically lower blood cholesterol concentrations (Groff and Gropper, 2000). On the other hand, Soluble fibers are usually fermented by colonic microflora producing short chain fatty acids (SCFA), which reduce serum and liver cholesterol concentrations. SCFA inhibit the synthesis of hepatic triacylglycerols and therefore reduce serum lipids (Suzuki and Kajuu, 1983 and Hara *et al.*, 1999).

The histopathological investigation of the pancreatic tissue of STZ- diabetic rats represented the presence of necrosis, edema and congested blood vessels in the islets of Langerhans cells. Oral administration of *O. dillenii* fruit juice to STZ-diabetic rats improved the previous changes, the majority of the cells consisting the islets of Langerhans tended to be normal. STZ is frequently used to induce diabetes mellitus in experimental animals through its toxic effects on pancreatic β-cells (Kim *et al.*, (2003). The cytotoxic action of STZ is associated with the generation of reactive oxygen species causing oxidative damage (Szkudelski, 2001). The present results were in coincidence with the observations of Kanter *et al.* (2006) in STZ diabetic rats who found degenerative and necrotic changes and shrinking of the islets of Langerhans, the nucleus of necrotic cells indicated pyknosis. Most of the tissue damage is considered to be mediated by free radicals which attack membranes through peroxidation of unsaturated fatty acids (Stringer *et al.*, 1989). Gul *et al.* (2002) reported that STZ produced oxidative stress and depletion of antioxidant systems in both blood and tissues. The findings of the fruit juice treated diabetic group indicated that *O. dillenii* fruit juice could provide protection against oxidative pancreatic tissue damage.

Conclusion

From the present results, it could be concluded that the edible portion of *O. dillenii* Haw fruit is very nutritious, being a rich source of vitamins B_1 , B_2 and C in addition to the minerals, Fe, Zn, Cu, Cr, Mn, Ca, and Mg. On the other hand, the fruit juice revealed significant hypoglycemic activity in STZ induced-diabetic rats. This activity may be attributed to its antioxidant activity and its high content of chromium. Therefore, it could be recommended that *O. dillenii* should be ingested as fresh fruit to diabetic and hypercholesterolemic patients beside the usual therapy. Further investigations also should be carried out with different doses and for more prolonged periods to complete the profile of the plant in order to introduce it as a natural antidiabetic agent.

Acknowledgment

I would like to thank Mrs T.L. Youssef, Specialist of Classification and Identification of Ornamental Plants, Orman Botanical Garden, Giza, Egypt, for kindly providing the plant material. Thanks also to Prof. Dr. K.H. Al-Batanony, Professor of Taxonomy, Faculty of Science, Cairo University for kindly verifying the identification of the plant material. Finally, full gratitude is extended to Dr. Kawkab A. Ahmed, Assistant Professor of Pathology, Faculty of Veterinary Medicine, Cairo University, for helping in the histopathological study of this work

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تقييم مدى فاعلية عصير ثمار فاكهة *Opuntia dillenii* (نوع من أنواع التين الشوكي) كخافض للسكر في الجرذان المصابة بالسكر المحدث بواسطة الإستربتوزوتوسين

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الملخص: تستخدم فاكهة *Opuntia dillenii* وهي نوع من أنواع التين الشوكي في الطب الشعبي كعلاج لمرض السكر. تهدف هذه الدراسة إلى تقييم الدور العلاجي المحتمل لعصير ثمار فاكهة هذا النوع من التين الشوكي باستخدام الجرذان المصابة بالسكر المحدث بواسطة الاستربتوزوتوسين. تم أيضاً تحليل القيمة الغذائية لثمار هذه الفاكهة وقد اتضح من النتائج وجود نسبة عالية من الألياف و الكربوهيدرات، وفيتامينات (ب$_1$، ب$_2$،جـ) ومعادن الحديد والزنك والنحاس والكروم والمنجنيز والكالسيوم والماغنسيوم.

أوضحت النتائج البيولوجية أن حقن الجرذان بالاستربتوزوتوسين بالمقارنة مع المجموعة الضابطة أحدث انخفاضاً معنوياً ذو دلالة إحصائية عالية في وزن الجسم، وارتفاعاً معنوياً ذو دلالة إحصائية عالية في مستوى سكر الدم مصحوباً بانخفاض معنوي في محتوى الكبد من الجليكوجين. كذلك أظهرت النتائج أن الجرذان المصابة بالسكر مقارنة مع المجموعة الضابطة حدث لها ارتفاعاً معنوياً في مستوى الدهون فوق المؤكسدة، وارتفاعاً معنوياً ذو دلالة إحصائية عالية في مستوى الكوليستيرول الكلي،والجليسريدات الثلاثية وكوليستيرول الليبوبروتينات ذات الكثافة المنخفضة متزامناً مع حدوث انخفاض معنوي ذو دلالة إحصائية عالية في مستوى كوليستيرول الليبوبروتينات ذات الكثافة العالية.

كما سجلت النتائج أن إعطاء عصير ثمار هذه الفاكهة لم يكن له تأثير على الجرذان غير المصابة بالسكر. بينما أدى إعطاء العصير للجرذان المصابة بالسكر مقارنة مع المجموعة المصابة بالسكر وغير المعالجة بالعصير إلى حدوث تحسن ملحوظ ذو دلالة إحصائية في وزن الجسم وأيض الدهون، وانخفاض ملحوظ ذو دلالة إحصائية في مستوى سكر الدم ومستوى الدهون فوق المؤكسدة.

أوضحت الدراسات الهستوباثولوجية لأنسجة البنكرياس للجرذان المصابة بمرض السكري وجود أنسجة ميتة وفراغات بين الخلايا وتجمعات دموية في خلايا جزر لانجرهانز. وقد أدت المعاملة بعصير ثمار الفاكهة موضوع الدراسة إلي التغلب على هذه الأعراض السابقة حيث أن أغلب الخلايا أصبحت أقرب إلى الحالة السليمة. وهذا التحسن في خلايا جزر لانجرهانز ربما يفسر التأثير المضاد لمرض السكري لعصير الفاكهة (تحت الدراسة). كذلك قد يعزي سبب انخفاض سكر الدم إلى فعل العصير المحسن لمستقبلات الأنسولين في خلايا بيتا.

اتضح من هذه الدراسة أن عصير ثمار فاكهة *O. dillenii* كان له تأثير قوى كمخفض لنسبة السكر المرتفعة بالدم. وقد عُزي هذا التأثير لدوره كمضاد للأكسدة ولاحتوائه على نسبة عالية من الكروم والذي ثبت في هذه الدراسة. لذلك توصي الدراسة بتناول الثمار الطازجة لفاكهة *O. dillenii* للمرضى المصابين بالسكر والمصابين بارتفاع مستوى الكوليستيرول في الدم إلى جانب العلاج الاعتيادي.