An Evaluation of Anti-Diabetic and Anti-Lipidemic Properties of *Momordica charantia* (Bitter Melon) Fruit Extract in Experimentally Induced Diabetes

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Abstract: Aim: *Momordica charantia* is reported to possess hypoglycemic activity. This study aims at investigating the effect of *Momordica charantia* extract on glucose tolerance and some biochemical parameters in alloxan induced diabetes, comparing it to the effect of rosiglitazone maleate, an oral hypoglycemic drug, and to suggest the possible mechanisms of its action. Main methods: Rats were divided into 5groups: normal control, rats received bitter melon, diabetic control, diabetic treated with rosiglitazone (4mg/kg BW), and diabetic received *Momordica charantia* (300 mg/kg BW). After 4weeks, OGTT, serum insulin, lipid profiles, glycohemoglobin% (HbA1c%), liver enzymes activity and glycogen content, intestinal absorption and diaphragm uptake of glucose and histopathological studies on the pancreas were evaluated. Key findings: Bitter melon (BM) induced a significant improvement of OGTT and induced a significant decrease in HbA1c% (p<0.05), significantly increased insulin release from the pancreas and serum insulin level, increased glucose uptake by rat diaphragm and decreased intestinal glucose absorption (p<0.05). BM improved lipid profile. In addition, BM significantly increased liver glycogen content and reduced liver enzyme activity compared to the diabetic control. BM treatment of diabetic rats resulted in significant hypoglycemic and hypolipidemic effects as compared to rosiglitazone (p<0.05). Significance: Results demonstrated anti-diabetic effects of bitter melon may be through increasing insulin release and serum insulin, increasing glucose uptake by muscles and decreasing intestinal glucose absorption and a hypolipidemic effect and this recommend its therapeutic use in diabetes.
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Key words: *Momordica charantia*, diabetes, glucose absorption, rat diaphragm glucose uptake, rosiglitazone maleate

1. Introduction

Diabetes mellitus is the most common endocrine disease. The prevalence of diabetes for all age-groups worldwide was estimated to be 2.8% in 2000 and 4.4% in 2030 (1). Diabetes mellitus leads to metabolic abnormalities and is characterized by hyperglycemia resulting from defects in insulin secretion, insulin action or both (2).

Although, oral hypoglycemic agents and insulin are the mainstay of treatment of diabetes, they have prominent side effects and fail to significantly alter the course of diabetic complications (3), The common side effects associated with oral hypoglycemic agents are hypoglycemia, weight gain, gastrointestinal disorders, peripheral edema and impaired liver function, in addition to the cost of treatment (4).

Since natural remedies are somehow safer and more efficacious than pharmaceutically derived remedies, herbalism has become mainstream worldwide (5).

Momordica charantia, also known as bitter melon, bitter gourd, or balsam pear, is a plant widely cultivated in many tropical and subtropical regions of the world and is frequently used in South Asia and the Orient as a food stuff and medicinal plant. Extracts from various components of this plant have been reported to possess hypoglycaemic activity (6). Thus, bitter melon can be an alternative therapy used for lowering glucose level in diabetic patients (7).

The hypoglycemic activity of *Momordica charantia* fruit juice is demonstrated in animals with experimental diabetes and also in humans in both type 1 and type 2 diabetes mellitus (8).

Scientists have identified 3 groups of constituents thought to be responsible for blood sugar lowering action of bitter melon; one of these, a compound called charantin which is composed of sitosteryl glucoside & stigmasteryl glucoside and can potentially replace treatment by insulin (9). Another compound, polypeptide p (plant insulin) found in seeds and fruits of bitter melon is similar to insulin in composition, so it can be of a great benefit in therapy of type 1 diabetes (10). Third compound is alkaloids which have also been noted to have a blood sugar lowering effect. Compounds known as oleanolic acid glycosides have been found to improve glucose tolerance in type 2 diabetes (11).

Aim of work:

The present study aims at investigating the effect of *Momordica charantia* (bitter melon) fruit extract on body weight, oral glucose tolerance test, serum insulin, blood glycohemoglobin percentage [HbA1c%], liver glycogen content, serum ALT and

AST and lipid profile (triglycerides, total cholesterol, LDL-cholesterol, HDL-cholesterol) in alloxan induced diabetes, comparing it to the effect of rosiglitazone maleate. The possible mechanisms of the hypoglycemic action of such agents was investigated by studying peripheral glucose uptake by rat diaphragm in vitro, insulin release from the isolated islets of Langerhans *in vitro* and intestinal glucose absorption in situ. Histopathological examination of the rat pancreas was examined.

2. Materials and Methods

Experimental animals

Fifty adult male albino rats weighing about 120-160g were divided into five groups (ten rats in each group) as follow:

Group I: rats of this group served as control group and were fed standard rat chow and pure water (NC).

Group II: this group included normal rats received bitter melon (*Momordica charantia*), at a daily dose of 300 mg/kg BW, dissolved in distilled water and given by gavage for 4weeks (NBM).

Group III: this group included diabetic control rats those were given pure distilled water (DC).

Group IV: this group included diabetic rats treated with Avandia® (rosiglitazone). The drug was purchased from Smith Kline Beecham Pharmaceuticals (U.S.A). The tablets were crushed, suspended in distilled water and was administered by gavage daily in a dose of 4 mg/kg BW (12) (DAV).

Group V: this group comprised of diabetic rats received, bitter melon (*Momordica charantia*) at a daily dose of 300 mg/kg BW, dissolved in distilled water and given by gavage for 4weeks (DBM).

Forty alloxan- induced diabetic rats were added to the diabetic group for the in vitro and in situ studies.

The rats were obtained from the animal house of Faculty of Medicine, Cairo University, Egypt. Rats were housed in separate cages temperature 25 ± 5C◦ and were given free access to water and food.

Experimental protocol

In the current study diabetes was induced experimentally in fasted rats by intra-peritoneal injection of a single dose of 100 mg/kg BW alloxan monohydrate (Sigma Company) dissolved in citrate buffer at pH 4.5 (13). Animals were given 5% glucose solution to drink instead of tap water for a few days until sustained hyperglycemia was established. Rats having serum glucose ranging from 180-300 mg/dl after 2 hours of glucose intake were only included in the experiment.

Preliminary testing of hypoglycemic activity of different doses of bitter melon was done for a week using diabetic rats to select the most potent dose which was used in the subsequent studies.

The effect of alloxan induced diabetes, as well as rosiglitazone maleate and *Momordica charantia* treatments, were investigated on: body weight, oral glucose tolerance test, serum insulin, blood glycohemoglobin [HbA1c] percentage, liver glycogen content, and serum ALT and AST activity, and lipid profiles. Peripheral glucose uptake by rat diaphragm, insulin release from isolated islets of Langerhans *in vitro* were performed and intestinal glucose absorption in situ was estimated. The present study also includes the histopathological changes in the pancreata of normal, diabetic control and diabetic treated rats.

At the end of the experimental period (4 weeks), OGTT was done to the fasted rats in the five groups. Twenty four hours later, fasted rats were sacrificed under diethyl ether anesthesia, and blood samples were collected from the rats. Pancreata and livers were excised quickly after dissection of the sacrificed animals. Fresh liver samples were used for determination of glycogen content. Pancreas was fixed in 10% neutral buffered formalin for paraffin section preparation.

Preparation of freeze-dried bitter melon BM juice

According to the methods of **Chen and Li**, (14) unripe BM fresh fruit was cut open and the seeds were removed. The extracted juice from the edible portion was frozen and completely lyophilized by continuous freeze-drying operation for 72hrs. The powder was kept in airtight containers at -70C^0 until used.

Biochemical analysis

1-Serum glucose levels and oral glucose tolerance test were performed according to the method described by **Leatherdale *et al.*** (15), using reagent kits purchased from Bio Merieux Chemicals (France).

2-ALT and AST activity in serum were determined according to the method of **Moss and Henderson** (16) using reagent kits purchased from Randox Company (United Kingdom).

3-Serum triglycerides concentration was determined according to the method of **Nauk *et al.*** (17), using reagent kits obtained from Reactivos Spinreact (Spain).

4-Serum LDL-cholesterol concentration was determined according to **Friendewald *et al.*** (18).

5-Liver glycogen content was determined according to the method of **Seifter *et al.*** (19)

6-Blood HbA1c% was estimated according to the method of **Abraham and Rao**(20), using reagent kits purchased from Stanbio Company (Texas).

Peripheral glucose uptake

Peripheral glucose consumption was studied in preparations from diabetic, 24 hrs fasted rats prior to sacrifice and exsanguinations according to **Zarzuelo *et al.*** (21). Diaphragms were incubated in a nutrient solution at 37°C with constant oxygenation for 1 hr. The preparation was used to compare between the

effect of rosiglitazone maleate and *Momordica charantia* on glucose uptake by the muscle, at their low concentrations (0.45mg/ml&0.2mg/ml respectively) and high concentrations (0.9mg/ml &0.4mg/ml respectively), in absence and presence of 50 μIU/ml insulin.

Intestinal glucose absorption

An intestinal perfusion in situ technique (21) was used to study the effects of rosiglitazone and *Momordica charantia* at their low & high concentrations on intestinal glucose absorption in diabetic 24 hrs fasted rats. First 10 cm of jejunum was perfused by a Kreb`s solution. Results were expressed as percentage glucose absorption calculated from the amount of glucose in solution before and after perfusion with rosiglitazone and *Momordica charantia* compared with a control study.

Histopathological study

The pancreas was immediately removed from each animal after sacrificing, fixed in 10% neutral buffered formalin and transferred to the National Cancer Institute, Cairo, Egypt for preparation. Pancreata were stained with modified aldehyde fuchsine stain method (22).

Isolation of islets of Langerhans and incubation techniques:

Pancreatic islets were isolated from diabetic rats, using the collagenase digestion technique (23). Collagenase (Type V) was purchased from Sigma Company, USA. To study the effect of different treatments on insulin release, 0.35 ml of rosiglitazone and *Momordica charantia*, both at their low (0.45,0.2 mg/ml) and high (0.9,0.4 mg/ml) concentrations respectively, were added to the isolated islets separately and incubated for 1 hr at 37° C. Another preparation was kept without treatments and used as a control study.

Statistical analysis of the results:

The data were analyzed using one way analysis of variance ANOVA, followed by least significant difference LSD analysis to compare various groups with each other. Results were expressed as mean ± standard deviation and values of $P<0.05$ were considered statistically significant.

3. Results

Figure 1 shows that the three doses of bitter melon BM (150, 300, and 600mg/Kg) produced varying significant hypoglycemic effects compared to the control group. However, the most potent dose was 300 mg /kg BW.

Table 1 and figure 2 show that treatment of diabetic rats with BM or Avandia induced a significant increase in BW, decrease in fasting blood glucose levels than those of the diabetic untreated group. Bitter melon and Avandia induced a significant hypoglycemic effect throughout the OGTT, decreased HbA1c% and increase in serum insulin in diabetic rats ($p<0.05$) compared to the diabetic untreated group.

Bitter melon induced a significant hypoglycemic effect, decrease in Hb A1c % and increase in serum insulin in diabetic rats as compared to Avandia (Table 1 and Figure 2).

Both bitter melon and Avandia significantly increased liver glycogen content, decreased liver enzymes of diabetic rats as compared to the diabetic untreated group ($p<0.05$).. Effect of bitter melon treatment was significant when compared to Avandia treated group (Table 1).

Bitter melon induced a significant decrease in serum total cholesterol, triglycerides and LDL but a significant increase in HDL as compared to the diabetic untreated group ($p<0.05$), while its effect on normal rats was insignificant as compared to the normal control group($p>0.05$). Avandia® induced an insignificant decrease in total cholesterol, triglycerides ($p>0.05$), but a significant decrease in LDL($p<0.05$), in addition to an insignificant increase in HDL ($p>0.05$) as compared to the diabetic untreated group. Effect of BM on diabetic rats was significant as compared to Avandia® (Figure 3).

Table 2 shows that, in the absence and presence of insulin, bitter melon caused a significant increase in percentage of glucose uptake by rat diaphragm at low and high concentrations ($p<0.05$), while values obtained with Avandia® were insignificant at low concentration($p>0.05$) and significantly increased at higher concentration ($p<0.05$) as compared to their controls.

Bitter melon induced a significant increase in insulin release from the pancreas of diabetic rats at both low and high concentrations in a dose dependent manner ($p<0.05$) as compared to control values. Avandia® showed no significant effect on insulin release (Table 3). Table 3 shows a significant decrease in % glucose absorption in situ at both low and high concentrations of BM in a dose dependent manner ($p<0.05$), while values obtained with Avandia were insignificant ($p>00.5$)as compared to control values.

Compared to the normal appearance of pancreas shown in figure (4A,B),bitter melon treatment had no effect on normal pancreas (Fig.4C). Intra-peritoneal injection of alloxan, at a dose of 100 mg/kg B. W. resulted in morphological alterations of pancreatic islet cells and showed destructed β cells with decreased number and vacuolated cytoplasm (Fig4D).

Treatments for 4 weeks of diabetic rats with either Avandia® (Fig.4E) or bitter melon (Fig.4F)

stimulated recovery of the islet cells. The islets approximately regained their normal appearance with a marked increase of β cell number and fewer vacuolated cells when compared to the pancreas of untreated diabetic rat.

Table (1): Effect of Avandia® and bitter melon on % change in body weight, serum insulin and HbA1c % and liver glycogen and liver enzymes of normal and alloxan diabetic rats compared to their controls after 4 weeks experimental period.

Groups	% change in body weight	Insulin µIU/ml	HbA1c %	Liver glycogen mg/g. fresh tissue	ALT U/l	AST U/l
NC	10.3±2.07[a]	19.6±2.37[a]	4.5±0.72[c]	10.5±1.70[a]	43.9±4.06[d]	40.2±4.38[d]
NBM	8.4±2.20[a]	19.8±1.75[a]	4.9±0.19[c]	10.7±2.26[a]	43.6±4.74[d]	40.2±4.91[d]
DC	-12.3±2.81[c]	6.3±0.87[d]	13.6±0.53[a]	3.2±1.00[c]	74.6±4.40[a]	63.7±4.5[a]
DAV	5.7±2.11[b]	7.9±0.97[d]	6.2±0.62[b]	5.9±0.93[b]	62.3±3.95[b]	56.9±4.36[b]
DBM	6.4±2.22[b]	12.5±2.12[b]	4.7±0.58[c]	9.8±1.29[a]	55.1±3.86[c]	48.5±5.23[c]

-Data are expressed as mean ±SD. -Number of samples in each group is 10.
-Means with different superscript letters in the same raw differ significantly ($P < 0.05$) and those with same superscript letter do not have a significant difference (P>0.05).
- For % change in B. Wt, LSD at 5% is 2.739 and LSD at 1% is 3.706.
- For insulin, LSD at 5% is 2.057 and LSD at 1% is 2.783.
- For glycol Hb %, LSD at 5% is 0.647 and LSD at 1% is 0.912.
- For liver glycogen, LSD at 5% is 1.808 and LSD at 1% is 2.446.
- For ALT, LSD at 5% is 5.018 and LSD at 1% is 6.789. - For AST, LSD at 5% is 5.586 and LSD at 1% is 7.558.

% change in BW = $W_x - W_o \times 100 / W_o$
W_o: Body weight at the beginning of the experiment. -W_x: Body weight at the end of the experiment.

Table (2): Effect of different concentrations of Avandia® and bitter melon on % glucose uptake by rat diaphragm of diabetic rats in presence and absence of insulin compared to normal control values.

Group / Studies	control	Avandia®		Bitter melon	
		Low 0.45mg/ml	High 0.9mg/ml	Low 0.2mg/ml	High 0.4mg/ml
In absence of insulin	12.0±1.38[c]	12.9±1.57[b,c]	14.1±0.63[b]	14.2±1.26[b]	17.2±1.52[a]
In presence of insulin	12.7±1.57[c]	13.1±1.98[b,c]	14.7±1.44[b]	14.9±1.86[b]	19.7±2.45[a]

-Data are expressed as mean ±SD. -Number of samples in each group is 5.
- Means with different superscript letters in the same raw differ significantly ($P < 0.05$) and those with same superscript letter do not have a significant difference (P>0.05).
- For % glucose absorption values in absence of insulin, LSD at 5% is 1.883 & at 1% is 2.568.and in presence of insulin, LSD at 5% is 2.345 and at 1% is 3.199.

Table (3): Effect of different concentrations of Avandia® and bitter melon on % intestinal glucose absorption in situ and insulin release from isolated islets of diabetic rats compared to control values.

Group / Studies	Control	Avandia®		Bitter melon	
		Low 0.45mg/ml	High 0.9mg/ml	Low 0.2mg/ml	High 0.4mg/ml
% glucose absorption	29.2±1.57[c]	28.7±1.53[b,c]	28.6±2.20[b,c]	26.7±1.89[b]	24.4±1.19[a]
Insulin release (µIu/islet/hour)	7.3±0.86[c]	7.5±1.14[c]	7.3±1.70[c]	9.5±1.85[b]	11.3±1.57[a]

-Data are expressed as mean ±SD.
-Number of samples in each group is 8 for insulin release studies and 10 for % glucose absorption.
-Means with different superscript letters in the same raw differ significantly ($P < 0.05$) and those with same superscript letter do not have a significant difference (P>0.05).
-For % glucose absorption values, LSD at 5% is 2.04 and at 1% is 2.76.
-For insulin release values, LSD at 5 % is 1.754 and at 1% is 2.374.

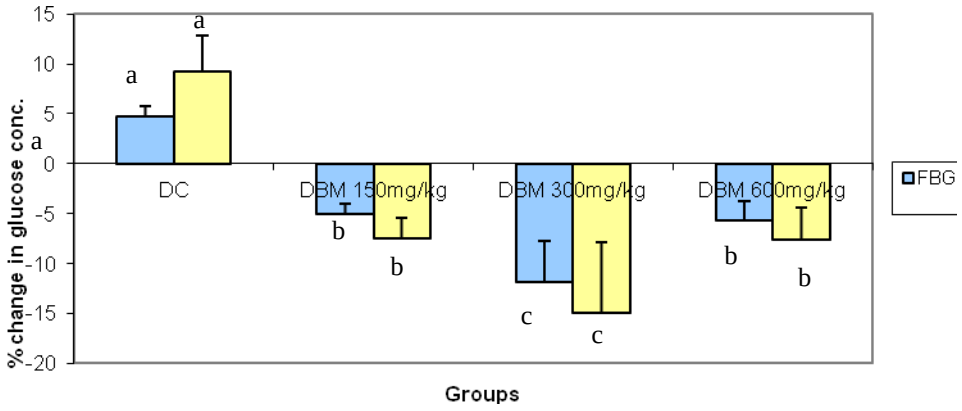

Figure1: Hypoglycemic effect of different doses of bitter melon in diabetic rats treated for one week. Results are expressed as mean±SD. Means with different letters differ §ignificantly.

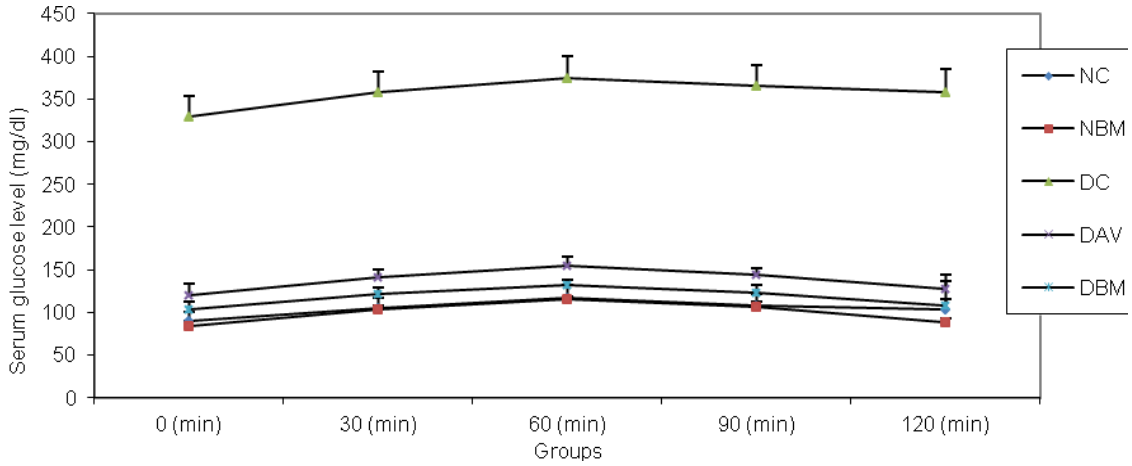

Fig. (2) Effect of Avandia® and bitter melon on OGTT of normal and alloxan diabetic rats compared to their controls after 4 weeks experimental period. Results are expressed as mean±SD.

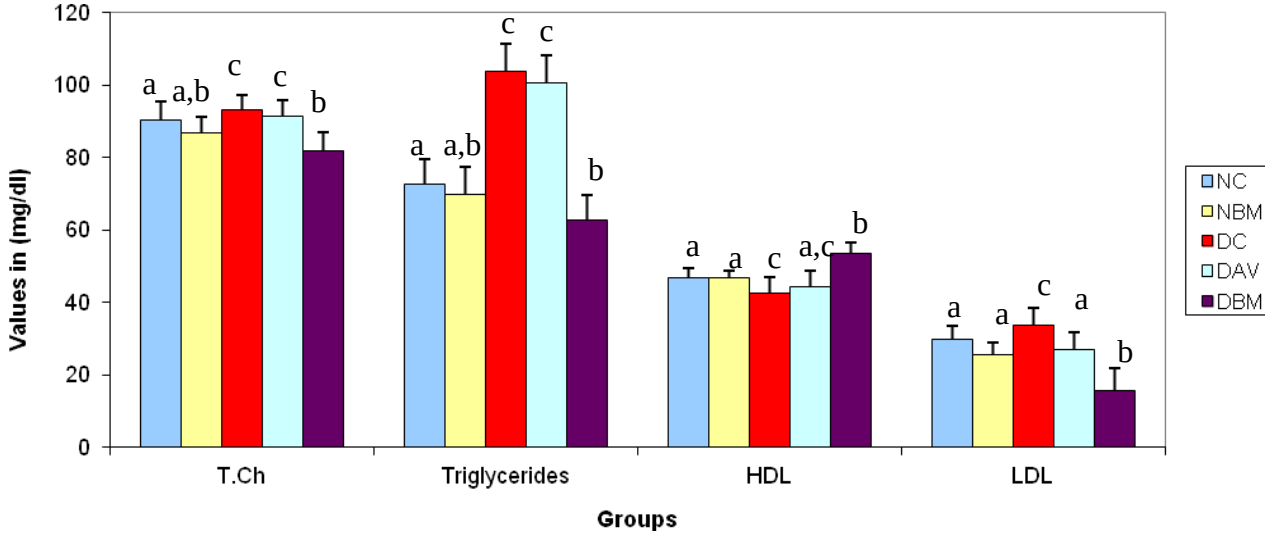

Fig. (3) Effect of avandia and bitter melon on serum total cholesterol, triglycerides, HDL, LDL of normal and alloxan diabetic rats compared to their controls after 4 weeks experimental period. Results are expressed as mean±SD. Means with different letters differ significantly.

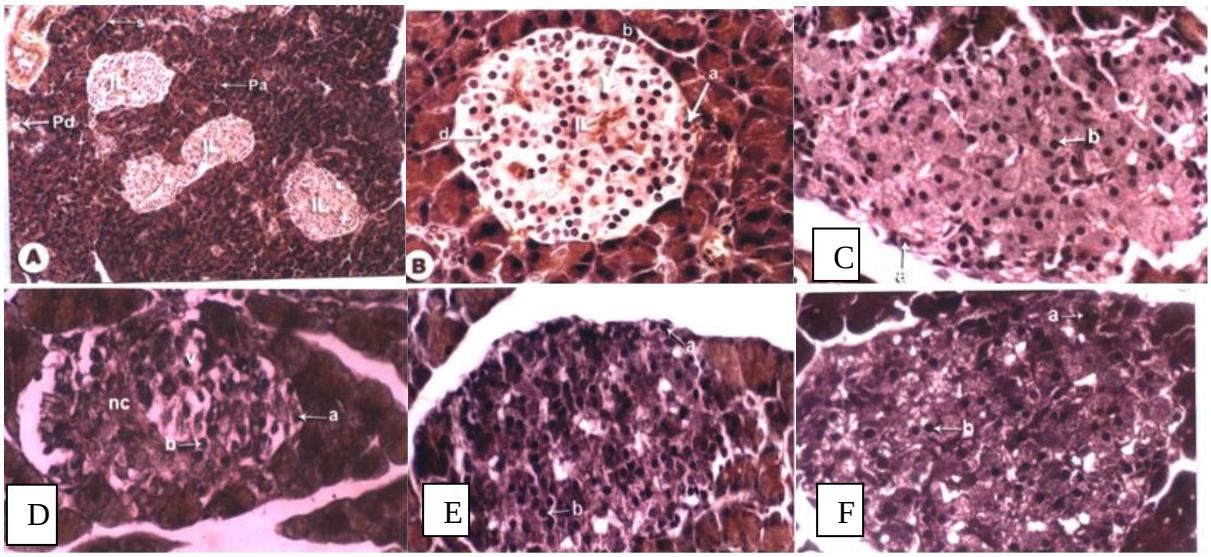

Figure (4)(A): Light micrograph of the pancreas of a normal male albino rat consist of exocrine and endocrine portions. The exocrine portion is subdivided by septa (S) into pancreatic acini (Pa) and ducts (Pd).The endocrine portion consists of the islets of Langerhans (IL). **(B)** Higher magnification of an islet of Langerhans which consists of three types of cells, alpha (a), beta (b) and delta (d). All cell types reveal a normal appearance. **(C)** Pancreas of a normal male albino rat treated with bitter melon for 4 weeks. The islets seemed to have a normal architecture.
(D) Pancreas of untreated diabetic rat after 4 weeks experimental period. The islets showed necrosis (nc) and vaculations (v) **(E)** Pancreas of a diabetic rat treated with rosiglitazone for 4 weeks. The number of alpha and beta cells per islet was increased and there were less vaculations. **(F)** Pancreas of a diabetic rat treated with bitter melon for 4 weeks showing little damage, less vaculations and the number of beta cells per islet was increased.

4. Discussion

The present study revealed that intra-peritoneal injection of a single dose (100 mg/kg B.Wt.) of alloxan to adult male albino rats was suitable to induce histopathological changes in the islets of Langerhans characterized by a marked decrease of β cells and vacuolar appearance, a significant decrease in fasting serum insulin level, decrease in the body weight and a significant increase in serum glucose level in OGTT of diabetic untreated rats, and a significant elevation in blood HbA1c% and an increase in the activity of both transaminases (ALT & AST), with significant decrease in liver glycogen content. The present findings are in agreement with **Lashin and Andrea** (24) and **Umrani *et al*.**(25).

Alloxan decreases body weight due to depressed synthesis of DNA and RNA in diabetic animals (26,27,28).

The hyperglycemia could arise due to destruction of β cells and reduced uptake of glucose to peripheral tissues as evidenced by the decrease rat diaphragm glucose uptake, glycogenolysis (29), and gluconeogenesis (30) as a result of insulin deficiency and may be due to the loss of glycogen synthetase activity, increased activity of glycogen phosphorylase (31) and / or increased activity of glucose-6-phosphatase (32).

This finding is supported by our results that revealed an enormous depletion in hepatic glycogen content and the detected elevation in liver enzymes in diabetic control rats as compared to control rats.

The elevated levels of both transaminases in the serum of diabetic rats of the present study may be ascribed to induced synthesis of these enzymes (33) and or destructive changes in hepatic cells as a result of toxaemia (34).

Treatment of diabetic rats with bitter melon induced a significant increase in body weight as compared to diabetic control rats. These results are in agreement with the findings of **Fernandes *et al***.(35) and **Yuan *et al***.(36), but disagree with **Dans *et al***. (37), who found that bitter melon had no significant effect on body weight of diabetics. This increase in body weight of diabetic rats as a result of bitter melon treatment may be ascribed to the increase in insulin release.

Treatment of diabetic rats with bitter melon produced a significant increase in fasting serum insulin as compared to the diabetic untreated group. The present finding is in agreement with the results of **Fernandes *et al* .** (35), **Yuan *et al*** .(36), **Sundaram and Kumar** (38), **Garau *et al*** .(39), **Yibchok *et al*** .(40), and **Hui *et al*** .(41).

On contrary, **Toshihiro *et al***.(42) and **Subratty *et al.*** (43) reported that treatment of diabetic rats with bitter melon decreased serum insulin. **Dans *et al*** .(37) reported that bitter melon had no effect on serum insulin.

The significant increase in serum insulin concentration of diabetic rats after bitter melon treatment in the present study might be ascribed to the ability of this agent to stimulate the spontaneous recovery of β cells of the islets of Langerhans**.** In vitro studies using isolated islets of Langerhans demonstrated that bitter melon induced a significant increase in insulin release. The work of **Fernandes *et al.***(35), **Garau *et al.*** (39) and **Singh and Gupta** (44) supports this finding. Treatment of diabetic rats with bitter melon showed a significant increase in β cell number. This indicates that bitter melon has a regenerative effect on β cells. On the other hand, **Sundaram and Kumar** (38) reported that treatment of diabetic rats with bitter melon did not restore β cells of islet of Langerhans destroyed by alloxan, however, viable β cells were found to be more active and granulated on bitter melon treatment.

Bitter melon may exert its effect by either preventing the death of beta cells by decreasing the oxidative stress caused by alloxan in diabetic rats since bitter melon contains vitamin C (anti-oxidant). Antioxidants act by neutralizing the free radicals released (45). **Xiang *et al.*** (46) suggested that bitter melon may act as a growth factor for pancreatic beta cells.

Regarding serum glucose level (OGTT), treatment of diabetic rats with bitter melon caused significant decreases in fasting and post- prandial serum glucose levels as compared to the diabetic untreated group. These results are in accordance with the findings of **Jayasuriya *et al.*** (7), **Fernandes *et al.*** (35) **Yuan *et al.*** (36) and **Chatuvedi *et al.*** (47). The present finding disagrees with the finding of **Dans *et al.*** (37) who reported that bitter melon had no significant hypoglycemic effect in alloxan diabetic rats.

In an attempt to gain an insight on the underlying physiological mechanisms of the hypoglycemic effect of bitter melon, we assayed its effect on peripheral glucose uptake by rat diaphragm (*in vitro*) and intestinal glucose absorption in situ.

Regarding peripheral glucose uptake of rat diaphragm, the obtained data indicated that, in both absence and presence of insulin, bitter melon induced a significant increase of glucose uptake as compared to a control study.

The present results are in agreement with the results of **Fernandes *et al.*** (35),**Garau *et al.***(39), **Ahmed *et al.*** (48), and **Shih *et al.*** (49). The mechanism by which bitter melon increases glucose uptake by skeletal muscle and adipose tissue was suggested by **Shih *et al.*** (49) and **Chuang *et al.*** (50) who demonstrated that bitter melon significantly increases mRNA expression and protein of glucose transporter 4 (GLUT4) in skeletal muscle. Bitter melon extract may stimulate GLUT4 translocation on the cell membrane in both myocytes and adipocytes (51).

These results on peripheral glucose uptake give evidence that bitter melon also have insulin-mimetic effects in addition to its insulin secretogogue or insulinotropic effect.

Concerning intestinal glucose absorption, the obtained data revealed that bitter melon produced a significant decrease of intestinal glucose absorption in diabetic rats compared to a control study. These results are in agreement with the findings of **Garau *et al.*** (39), **Ahmed *et al.*** (48) and **Mahmoodally *et al.*** (52).

It is hypothesized that bioactive phytochemicals such as saponins in bitter melon extract inhibit the active transport of d-glucose, l-tyrosine and fluid across rat intestine by inhibiting the ATPase responsible for the active transport of these molecules (52). This positive influence of feeding bitter melon on intestinal glucose absorption may also be through affecting disaccharidase activity (53). Also, it has been shown that oleanolic acid glycosides isolated from bitter melon suppress gastric emptying in alloxan diabetic rats and decrease glucose absorption in small intestine *in vitro* (54).

Based on the above mentioned data, it is worth mentioning that an enhancement of insulin release, increase of peripheral glucose uptake, and suppression of intestinal glucose absorption are involved in the mechanisms of hypoglycemic action of bitter melon in alloxan diabetic rats.

Treatment of alloxan diabetic rats in the present study with bitter melon induced a significant decrease of HbA1c% as compared to the diabetic untreated group. Such decrease may be ascribed to the insulinotropic effect of this agent. This finding is supported by **Fernandes *et al.*** (35) and **Garau *et al.*** (39), but disagrees with **Dans *et al.*** (37) who reported no significant effect of bitter melon on blood glycohemoglobin (%) in alloxan diabetic rats.

The present study revealed that administration of bitter melon to diabetic rats induced an increase in hepatic glycogen concentration. This finding is in agreement with **Garau *et al.*** (39), **Singh & Gupta** (44), and **Rathi *et al.*** (55) but disagrees with **Fernandes *et al.*** (35) and **Yuan *et al.*** (36) who reported decreased glycogen content of the liver of alloxan diabetic rats treated with bitter melon.

Stimulated insulin release induced by bitter melon treatment, as shown in the current study, may be responsible for increasing glycogen synthetase activity (56).

Regarding liver enzymes, the present study revealed a significant decrease in the activities of both alanine aminotransferase (ALT) and aspartate aminotransferase (AST) in diabetic rats treated with bitter melon as compared to the diabetic untreated group. These findings are in agreement with studies of **Garau *et al.*** (39). Results of **Dans *et al.*** (37) on

diabetic rats treated with alloxan showed no effect on serum ALT and AST.

The present results elucidated a significant increase of total cholesterol, triglycerides and LDL-cholesterol concentrations in the serum of diabetic control rats as compared to normal control group. These results are in agreement with **Newairy *et al*.** (57). On the other hand, HDL-cholesterol level was significantly decreased in serum of diabetic control rats in the present study as compared to the normal control group. This finding parallels that of **Nakura *et al*.** (32), and disagrees with **Wasan *et al*.** (58) who reported a significant increase of HDL-cholesterol in alloxan diabetic rats.

The markedly increased level of triglycerides and LDL-cholesterol in the serum of diabetic rats of the present work may be a consequence of either overproduction by the liver or defective removal from the circulation or both secondary to insulin deficiency (59).

Mechanisms by which HDL decreases in diabetes may be due to the impaired metabolism of triglycerides rich lipoprotein with decreased activity of lipoprotein lipase and impaired transfer of materials to the HDL components, in addition to the high level of hepatic lipase among diabetics (60). Finally, insulin resistance may be a direct cause of decrease of HDL concentration (61).

In a view of the present results, it was found that treatment of diabetic rats with bitter melon produced marked decreases of serum total cholesterol, triglycerides and LDL-cholesterol concentrations and an increase in serum HDL-cholesterol concentration as compared to the diabetic control group. These obtained data are concomitant with the results of **Fernandes *et al*.** (35), **Yuan *et al*.** (36) and **Chatuvedi *et al*.** (47), The present findings disagree with the results of **Dans *et al*.** (37) who found that the addition of bitter melon to hypercholesterolemic diet of rats had no effect on serum lipid profiles.

Bitter melon may affect the break down of specific lipoprotein (e.g LDL) or it may enhance fat oxidation in the body. The saponins and plant sterol in bitter melon also reduce blood triglyceride level and they also reduce the absorption of cholesterol from the intestine. In addition, the insulin like molecule in bitter melon may, like insulin, prevent the increase in triglyceride level due to the movement of fat from body cells into the blood stream (7).

In this study, bitter melon did not have a hypoglycemic or hypolipidemic effect on normal rats treated for 4 weeks. **Toshihiro *et al*.** (42) and **Ouviña *et al*.** (62) supported these findings. On the other hand, these findings disagree with those obtained by **Yibchok *et al*.** (40) and **Ojewole *et al*.** (63) who found that bitter melon fruit extract had significant hypoglycemic and hypolipidemic effects in normal rats.

In the current study, results obtained from diabetic rats treated with rosiglitazone (Avandia), an oral hypoglycemic drug, revealed that rosiglitazone decreased serum glucose, blood glycohemoglobin %, increased liver glycogen, and decreased liver ALT and AST significantly, but had insignificant effects on serum insulin and lipid profiles except LDL which decreased significantly with rosiglitazone treatment. These results except for lipid profiles agree with **Al-Salman *et al*.** (64) and **Leibowitz and Cerasi** (65) who demonstrated that rosigliazone had significant hypoglycemic and hypolipidemic effects in diabetic rats.

Regarding the in vitro and insitu studies, rosiglitazone had no significant effect on insulin release from isolated beta cells of the pancreas or on intestinal glucose absorption, while, it increased glucose uptake significantly by rat diaphragm only at high concentration. In addition, rosiglitazone treatment was found to increase number of beta cells in the pancreas of diabetic rats. These findings are in agreement with findings of **Finegood *et al*.** (66) and **Smith *et al*.** (67).

Bitter melon treatment of diabetic rats resulted in significant hypoglycemic and hypolipidemic effects as compared to rosiglitazone.

Conclusion:

In conclusion, the present study calls attention to the therapeutic use of bitter melon in diabetes mellitus. The results of the current study demonstrated that bitter melon has numerous anti-diabetic effects such as, decreasing serum glucose concentration, increasing serum insulin level, increasing glucose uptake by the peripheral tissues and decreasing intestinal glucose absorption. In addition, it showed hypolipidemic and thus cardiac protective effects. It was shown in this study that bitter melon did not cause hypoglycemia when given for normal rats, this indicates that it is safe if utilized by normoglycemic persons for its other beneficial effects.

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