LOWERING CARDIOVASCULAR RISK FACTORS ASSOCIATED WITH DIABETES IS POSSIBLE USING THE EXTRACT OF URTICA PILULIFERA LEAVES

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Abstract

Diabetes mellitus is associated with increased cardiovascular risk factors, and such factors are attributed to disturbances in lipid profile. The present study was conducted to show the direct effect of diabetes on lipid profile in rats with diabetes using alloxan induced diabetic model and to investigate the possible therapeutic effects of using *Urtica pilulifera* . Methodology followed included collection and processing of *Urtica pilulifera* leaves, induction of diabetic rat model and investigating the effects of *Urtica pilulifera* on lipid profile of diabetic group. Study findings showed that in control group, the levels of triglyceride, total cholesterol, and LDL-C increased significantly ($P<0.05$), whereas the level of HDL-C decreased significantly ($P<0.05$) compared with control group. Treatment with the extract of *Urtica pilulifera* leaves using 1.25g/kg/body weight, or 1.88 g/kg body weight decreased the levels of triglyceride, total cholesterol, and LDL-C ($P<0.05$) compared with diabetic group, whereas the level of HDL-C increased significantly compared with diabetic group ($P<0.05$).

Taken together, the present study showed that diabetes is a possible risk factor for cardiovascular disease through induction of disturbances in lipid profile. Treatment using the methanolic extract of *Urtica pilulifera* leaves

reversed the adverse effects of diabetes and restored lipid profile to control level.

Keyword: Diabetes, triglyceride, total cholesterol, LDL-C, HDL-C, *Urtica pilulifera*

Introduction

Diabetes Mellitus (DM) is considered as a metabolic disorder, chronic disease that affects large number of people worldwide. The prevalence of diabetes continues to increase and impacts the quality of life. It has been estimated that 285 million adults in 2010 had DM and it is expected that the number of diabetics to reach 438 million people (Shaw, Sicree, Zimmet, 2010; Ali-Shtayeh and Jamous, 2012).

Diabetes has been associated with many complications among which are the cardiovascular diseases. Lipid disturbances are thought to attribute for this risk (Jaiswal, Schinske, Pop-Busui, 2014).

Both types of diabetes have been associated with dyslipidemia, but the prevalence is higher in DM (Jacobs et al., 2005). It has been indicated that patients, who have poorly controlled type 1 diabetes, have clinical findings including increased concentrations of triglyceride (TG), very low density lipoproteins (VLDL) and chylomicrons, this is thought to due to a reduction in the activity of lipoprotein lipase (LPL) in the muscle and adipocytes. Also, a decrease in the concentration of high-density lipoprotein (HDL) was observed (Chahil and Ginsberg, 2006).

Other studies reported that the deficiency of insulin to be associated with increased levels of LDL-C, LDL particle number, and apolipoprotein B-100 since insulin regulates the expression of LDL receptor (Jacobs et al., 2005).

It has been shown that patients with DM have abnormalities with lipoprotein metabolism such as increased fasting and postprandial TG levels, and lowered HDL-cholesterol levels (Howard et al., 1998).

Urtica pilulifera is known as popular plant by general public and found in various countries including Palestinian and in Sinai areas (Ali-Shtayeh et al., 2000). It belongs to family Urticaceae (Irshaid and Mansi, 2009; Shuwayeb and Khatib, 2013). From a morphological point of view, it is characterized by having the stinging hairs which are carried by its leaves and flowers and usually cause irritation to the skin (Fu etal., 2006). It has been shown that leaves of *Urtica pilulifera* is traditionally used for various medical needs including a stimulating tonic, blood purifier and hemostatic and for enhancement of hemoglobin concentration (Chrubasik et al., 2007). The whole plant has other therapeutic properties including diuretic, antiasthmatic, anti-inflammatory, hypoglycemic, hemostatic, antidandruff

and astringent (Delcourt et al.,1996; Abudoleh et al., 2011). Other studies have pointed o the use of extracts of *Urtica pilulifera* in treating urinary tract infection (Lopatkin etal.,2005). In his study, Kavalali et al (2003) showed that *Urtica pilulifera* has pronounced hypoglycemic activity and b-cell regenerative potency. It is worth to mention that *Urtica pilulifera* is safely used since it is considered non-toxic and does not have any mutagenic and embyrogenic effects (Graf et al.,1994).

Study objectives

The present study was conducted to show the direct effect of diabetes on lipid profile in rats with diabetes using alloxan induced diabetic model and to investigate the possible therapeutic effects of using *Urtica pilulifera*.

Methodology

Collection and preparation of of *Urtica pilulifera* extraction

Urtica pilulifera leaves were collected from various parts that are known by growth of *Urtica pilulifera,* at Jordan. *Urtica pilulifera* leaves were dried by air in shade well-ventilated area and then ground into fine powder. About 350 g of powder was put in a Soxhlet cold extractor using absolute methanol as solvent and remained for three consecutive days (Sadki et al., 2001). The extract was further concentrated and gave a yield of 11.4% viscous greenish-colored extract. The extract was kept at 4°C in a glass container until use. Wister rats were used in this study, in which their average weight was 170 g. The animals were carefully checked and monitored every day for any changes. After determination of lethal dose (LD50), two doses were selected 1.25 g/kg and 1.88 g/kg of body weight. Doses were prepared through dissolving required amount of the viscous extract in 10 mL Tween-20: 0.9% NaCl (1:9, V/V).

Diabetic model

Diabetes was induced depending on using alloxan so that rats were injected by alloxan monohydrate "B.O.H chemical LTD England" intraperitoneally at a dose of 150 ml/kg body weight (dissolved in fresh normal saline) to 18 hr fasted rat. Rats were monitored for blood glucose and rats with blood glucose level over 200 mg/ml, were considered diabetic and employed in the study.

Animals were assigned into the following groups:

Group I: control group; Group II: diabetic group; Group III: diabetic treated with 1.25 g/kg of body weight; Group IV: diabetic treated with 1.88 g/kg of body weight.

Statistical analysis

Data analysis was carried out using SPSS 20. Data were presented as mean and standard deviation. T test was used to investigate the difference between kidney function tests in study groups. Significance level was considered at alpha level ≤ 0.05.

Results

As shown in table 1, the concentration of total cholesterol was 126.21 ± 6.26 mg/dl in control group. The concentration of total cholesterol significantly increased in diabetic group to 166.53±10.26 mg/dl ($P<0.05$). Treating diabetic group with methanol extract of *Urtica pilulifera* (1.25g/kg) was able to significantly decrease the concentration of total cholesterol to 142.47±8.36. The same effect was observed when diabetic group was treated with methanol extract of *Urtica pilulifera* (1.88g/kg) and the concentration of total cholesterol significantly decreased to 148.82±6.37 mg/dl ($P<0.05$).

The concentration of triglyceride in control group was 86.45±9.36 mg/dl, and significantly increased in diabetic group 118.36± 14.2 mg/dl ($P<0.05$). When diabetic was treated with methanol extract of *Urtica pilulifera* (1.25g/kg), the concentration of triglyceride significantly decreased to 76.27± 9.34 mg/dl ($P<0.05$). Treatment of diabetic group with methanol extract of *Urtica pilulifera* (1.88g/kg) showed significant reduction of triglyceride concentration to 93.42±11.4iabetic gr6 mg/dl ($P<0.05$).

The concentration of HDL-C was 33.12± 4.56 mg/dl in control group, and it significantly decreased in diabetic group 20.31± 3.18 mg/dl ($p<0.05$). The treatment of diabetic group with methanol extract of *Urtica pilulifera* (1.25g/kg) was able to increase significantly the concentration of HDL-C to 29.44± 3.28 mg/dl ($P<0.05$). The same effect was also observed when diabetic group was treated with methanol extract of *Urtica pilulifera* (1.88g/kg) in which the concentration of HDL-C was significantly increased 28.49 ± 3.66 mg/dl ($P<0.05$).

The data of the present study showed that the concentration of LDL-C was 28.46±3.94 mg/dl in control group, which significantly increased in diabetic group to 54.86± 9.27 mg/dl ($P<0.05$). the treatment of diabetic group with either methanol extract of *Urtica pilulifera* (1.25g/kg, 1.88 g/kg) was able to significantly decrease the concentration of LDL-C (22.48±4.22 mg/dl, 21.25±3.74 mg/dl respectively) ($P<0.05$).

Table 1: Effect of treatment with methanol extract of *Urtica pilulifera* for 4 weeks on lipid profile in alloxan induced diabetic in rats

Group	Treatment	N	Total cholesterol (mg/dl	Triglyceride mg/dl	HDL-C (mg/dl)	LDL-C (mg/dl)
I	Control	6	126.21± 6.26	86.45±9.36	33.12± 4.56	28.46±3.94
II	Diabetic	6	166.53± 10.26*	118.36± 14.2*	20.31± 3.18*	54.86± 9.27*

III	Diabetic +1.25 g/kg	6	142.47±8.36**	76.27± 9.34**	29.44± 3.28**	22.48±4.22**
IV	Diabetic +1.88 g/kg	6	148.82± 6.37 **	93.42±11.46**	28.49 ± 3.66**	21.25±3.74**

* compared with control group
** compared with diabetic group

Discussion

The results of the present study showed that induced diabetes in rats using alloxan model for 4 weeks has lead to disturbances in lipid profile in diabetic group compared with control group. Lipid profile included total cholesterol, triglyceride, HDL-C , and LDL-C.

Diabetes significantly ($P<0.05$) increased the concentration of total cholesterol, triglyceride and LDL-C, while the concentration of HDL-C decreased significantly in diabetic group compared with control group. Our findings are in line with other reported studies in literature. It has been indicated that diabetes is associated with disturbances in lipid profile (Jaiswal, Schinske, Pop-Busui, 2014). Other studies have reported that patients with diabetes either type 1 or type 2 have increased concentrations of triglyceride (TG), very low density lipoproteins (VLDL) and chylomicrons (Howard et al., 1998; Jacobs et al., 2005; Chahil and Ginsberg, 2006).

Diabetic groups that were treated with the methanolic extract of *Urtica pilulifera* leaves showed reversed effects of diabetic impacts. Treatment included two doses of the methanolic extract 1,25 g/kg, and 1.88g/kg/ body weight. Either of the two doses showed significant variations in reducing total cholesterol, triglyceride, and LDL-C, while both treatments were able to increase the levels of HDL-C. The results of our study are in agreement with other studies reported in literature (Irshaid and Mansi, 2009; Singala et al., 2009; Dina et al., 2013).

Conclusion

The present study showed that diabetes is a possible risk factor for cardiovascular disease through induction of disturbances in lipid profile. Treatment using the methanolic extract of *Urtica pilulifera* leaves reversed the adverse effects of diabetes and restored lipid profile to control level.

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