

Full Length Research Paper

The effects of Persian shallot extract on the levels of some blood biochemical parameters in streptozotocin-induced diabetic rats

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Diabetes mellitus is a multifarious group of symptoms characterized by hyperglycemia, abnormal lipid and protein metabolism. The purpose of the present study was to investigate the effects of daily oral administration (for 30 days) of Persian shallot extract on some biochemical parameters in streptozotocin-induced diabetic rats. Rats (weighting 210 to 230 g) were divided into 4 groups of 8; normal control, diabetic control, diabetic rats treated with Persian shallot extract in a 100 mg/kg concentration (100 g of dry powder of Persian shallot solved in 900 ml of 0.9% saline) and diabetic rats treated with Persian shallot extract in a 200 mg/kg concentration (200 g of dry powder of Persian shallot solved in 800 ml of 0.9% saline). The plasma glucose level, HbA1c, uric acid and creatinine were reduced while insulin was slightly increased in Persian shallot extract treated groups significantly. Treatment with Persian shallot extract reduced plasma LDL-C and triglyceride but increased HDL-C (these changes were not significant). The plasma total cholesterol, urea, protein and albumin were significantly increased in Persian shallot extract treated groups. Therefore, according to the finding obtained in present study, it could possibly be concluded that the consumption of Persian shallot extract produces a significant hypoglycemic effect in diabetic rats. In addition, Persian shallot extract is almost capable of improving lipid profile but it can not ignore its negative effect on total cholesterol and urea levels, as unwanted side-effect.

Key word: Persian shallot extract, diabetes, glucose, streptozotocin.

INTRODUCTION

Diabetes mellitus is a metabolic disorder with multiple etiologies, characterized by chronic hyperglycemia with disturbances of carbohydrate, fat and protein metabolism resulting from defects in insulin secretion, insulin function or both (Baquer et al., 1998). Hyperlipidemia is a common feature of diabetic patients and is responsible for vascular

complications. In fact, high level of triglyceride and low level of HDL-C are independently related to morbidity and death in diabetic subjects by the induction of coronary heart diseases (Temme et al., 2002). Investigations reported that deficiency of lipoprotein lipase activity may significantly contribute to the elevation of triglycerides in diabetes. In addition to hyperlipidemia and hyperglycemia, albuminemia and proteinemia are significant in diabetes (may be due to microproteinuria and albuminuria, which are important clinical markers of diabetic nephropathy) (Mauer et al., 1981). Therapy of

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diabetes mellitus without any side effects is still a challenge of medicine. These days there is an increasing demand by patients to use natural products with antidiabetic activity, because insulin and oral hypoglycaemic medications have undesirable side effects (Kameswara and Appa, 2001; Li et al., 2004). It is widely believed that the antioxidant micronutrients obtained from fruits and vegetables afford significant protection against diabetes. Onions are rich in two chemical groups that have perceived health benefits to man. These are the flavonoids and the alk(en)yl cysteine sulphoxides (ACSOs).

The ACSOs are the flavour precursors and following cleavage by the enzyme alliinase generate the characteristic odour and taste of onion (Leighton et al., 1992). Flavonoids are potential antioxidants found in onions. There are two major groups of flavonoids in onion are the flavonols and the anthocyanins which intake of them has been reported to be inversely associated with risk of cardiovascular disease in some epidemiological studies (Yochum et al., 1999). Persian shallot (*Allium hirtifolium* Boiss.) called as "Mooseer" in Iran, belongs to Liliaceae family and is one of the important edible onions in Iran. It is a native Iranian plant and grows wildly in the Zagross Mountains (Rechinger, 1984). It is different from common shallot (*Allium ascalonicum* L.) in many characteristics. Persian shallot is yellow, oval, white skinned and usually consists of a single main bulb or rarely two bulbs, each bulb weighting roughly 8 to 15 times of garlic clove. While bulbs of common shallot are pear-shaped, reddish-brown skinned and clustered at the base of the plant and its clusters may contain as many as 15 bulbs (Rubatzky and Yamaguchi, 1997). Furthermore, Persian shallot bulb is darker than garlic and has a stronger odor that correlates with its sulfide content (Mubarak and Kulatilleke, 1990).

A number of intervention studies have similarly shown that garlic significantly reduced plasma glucose in diabetes. Moreover, in animal experiments, garlic extracts have been shown to decrease plasma lipid and cholesterol in rats (Thomson et al., 2006; Gorinstein et al., 2006), but there is not much study about Persian shallot effects on diabetes and biochemical parameters. Though, there are several reports that emphasized shallot medicinal affects as antioxidant (Leelarungrayub et al., 2006), immune system regulating (Jafarian et al., 2003) and anticancer (Ghodrati et al., 2008). Since Persian shallot grows as a wild plant only in some mountains of Iran, limited information is available regarding different aspects of this species. Therefore, the present study was aimed to examine the possible positive effects of hydroalcoholic extract of Persian shallot on some biochemical parameters in streptozotocin-induced diabetes mellitus rats.

MATERIALS AND METHODS

Preparation of hydroalcoholic extract

Fresh Persian shallot (*A. hirtifolium* Boiss) bulbs were obtained from Kangavar (Kermanshah-Iran). The genus and species of the bulbs were confirmed by the botanists (Department of Botany, Valiasr University Rafsanjan-Iran). Then, 100 g of fresh bulbs was well crushed and 400 ml distilled water/ethanol (25/75) was added. After 48 h incubation, the solution was filtered using a filter paper through a Buchner funnel. The filtered resultant solutions obtained from this stage, concentrated by means of a vacuum distillation and decanted to dry powder (5 g), then, needed concentrations prepared; 5 g of Persian shallot dry powder dissolved in 45 ml of 0.9% saline for preparing of 100 mg/kg concentration and 10 g of Persian shallot dry powder dissolved in 40 ml of 0.9% saline for preparing of 200 mg/kg concentration (Momeni, 2000).

Animals and treatments

In this study, 32 male albino Wistar rats weighing 210 to 230 g, twenty four rats were injected with 45 mg/kg body weight of streptozotocin (STZ) (diabetic type-1 rats that had FBS level more than 250 mg/dl) and eight rats considered as normal group. After being matched according to body weight, the rats were divided into 4 groups, with 8 rats in each group:

Group 1

Diabetic rats received daily 200 mg/kg Persian shallot extract (2 ml) for 30 days.

Group 2

Diabetic rats received daily 100 mg/kg Persian shallot extract (2 ml) for 30 days.

Group 3

Diabetic rats received daily 0.9% saline (2 ml) for 30 days (diabetic control).

Group 4

Normal rats received daily 0.9% saline (2 ml) for 30 days (normal control).

The solutions (2 ml) were given to animals by gavage, using a gavage syringe. The animals were then housed in cages and had free access to water and standard food. Animal handling was performed with regard to Iranian animal ethics society and local university rules. Following, 30 days blood samples were collected.

Measurement of insulin and biochemical parameters

The rats' serum insulin level was quantitatively measured using ELISA kits (Mercodia, sweden); and the levels of glucose, HbA1c, urea, creatinine, uric acid, protein, albumin, triglyceride, total

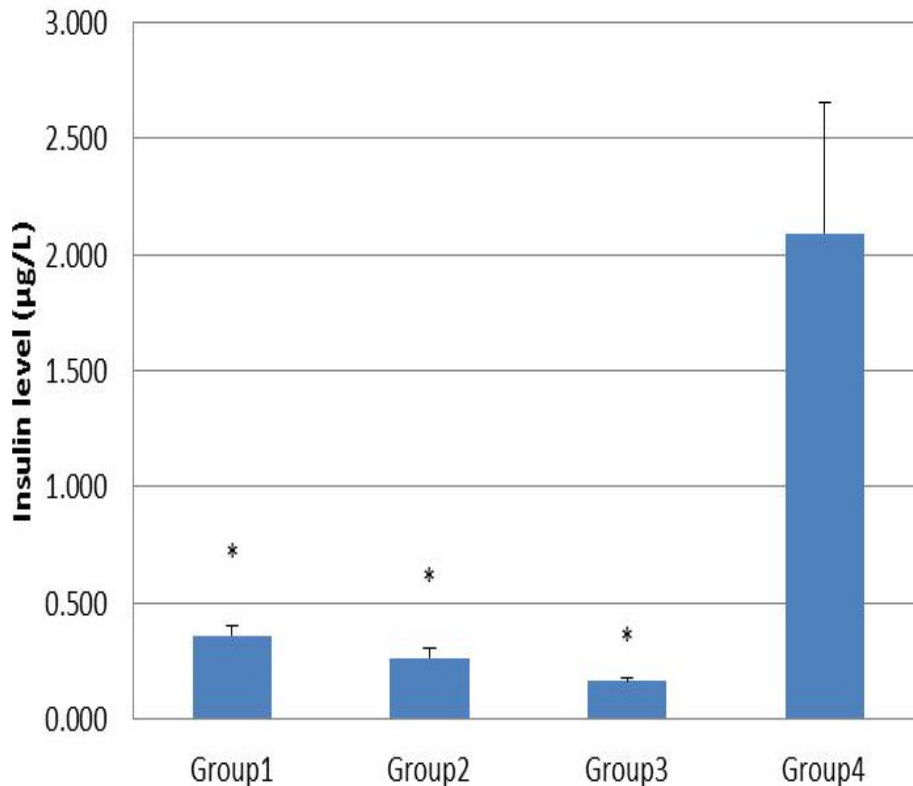


Figure 1. The effect of different concentration of Persian shallot on insulin level ($\mu\text{g/L}$) (Mean \pm SD). Group 1: diabetic rats received 200 mg/kg Persian shallot, Group 2: diabetic rats received 100 mg/kg Persian shallot, Group 3: diabetic rats received 0.9% saline and Group 4: normal rats received 0.9% saline. *Significant differences with Group 4 ($P < 0.05$).

cholesterol, LDL-C and HDL-C were measured in all study groups by BT-3000 autoanalyzer.

Statistical data analysis

Data are expressed in terms of mean \pm standard deviation (SD). Differences in measured parameters between control groups and Persian shallot treated groups were analyzed by the one-way ANOVA. P values less than 0.05 were considered statistically significant.

RESULTS

Insulin levels

In the present study, we demonstrated that diabetic groups showed significantly lower insulin levels compare to normal control. However, there were no significant differences between Persian shallots treated groups (Groups 1 and 2). The fasting plasma Insulin levels of all groups of rats during experimental period are displayed in Figure 1.

Biochemical parameters

As shown in Table 1, induction of diabetes by STZ injection significantly increases glucose, HbA1c, creatinine, urea and uric acid. It also increases cholesterol, triglyceride and LDL-C but it is not significant with control. The administration of both concentrations of Persian shallot extract to the diabetic rats significantly reduced plasma glucose level, HbA1c and uric acid when compared with control groups. These reductions were not enough to reach normal rats, but it was still significantly lower when compared with the normal group as shown in Table 1 ($p < 0.05$). The concentration of 100 mg/kg of Persian shallot extract declined the creatinine level significantly ($p < 0.05$) and the 200 mg/kg concentration of Persian shallot extract reduced it but insignificantly. On the other hand, the levels of plasma triglyceride and LDL-C were decreased in rats treated with Persian shallot extract; it was clear from Table 1 that only reduction in the level of triglycerides reach normal level. Diabetic rats have lower level of protein, albumin and HDL-C.

As illustrated in Table 1, a significant increase ($p < 0.05$)

Table 1. The effects of Persian shallot on the levels of some blood biochemical parameters in diabetic rats; (Mean±SD).

Parameter	Group 1 (diabetic group + 200 mg/kg Persian shallot)	Group 2 (diabetic group + 100 mg/kg Persian shallot)	Group 3 (diabetic control)	Group 4 (normal control)
Glucose (mg/dl)	283.9±14* ^{Δ□}	366±12* ^Δ	390±9*	110±13
HbA1c (%)	10.4±0.4* ^{Δ□}	11.7±0.2* ^Δ	12.5±0.2*	8.6±0.3
Urea (mg/dl)	128±21* ^{Δ□}	79±8*	73±8*	49±9
Creatinine (mg/dl)	0.7	0.75±0.08*	0.78±0.07*	0.62±0.1
Uric acid (mg/dl)	1.65±0.1 ^Δ	2.02±0.6* ^Δ	4.27±0.1*	1.97±0.2
Protein (mg/dl)	7.2±0.17 ^Δ	6.9±0.69	6.4±0.1	6.7±0.4
Albumin (mg/dl)	3.6±0.2* ^{Δ□}	3.1±0.2	2.8±0.05*	3.2±0.2
Triglyceride (mg/dl)	76±26	77±29	124±42	98±41
Total cholesterol (mg/dl)	68±8.7*	58±5.3*	52±5.3 ^Δ	47±6
LDL-C (mg/dl)	17.5±0.5	17.5±1.6	18.6±0.5	18.3±0.5
HDL-C (mg/dl)	37.8±1.1	35.4±1.9	34.1±3.9	35.5±1.7

* Significant differences with Group 4 (P<0.05), ^Δ Significant differences with Group 3 (P<0.05) and [□] Significant differences with Group 2 (P<0.05).

was recorded in the levels of plasma total cholesterol and urea (both concentrations), protein and albumin (200 mg/kg concentrations) when compared with control groups, however, the elevation in HDL-C level was not significant.

DISCUSSION

In the present study, we found that treatment of STZ-induced diabetic rats with Persian shallot extract reduced the plasma glucose and HbA1c levels and slightly increased insulin level. In this context, a number of other plants have also been reported to have hypoglycemic along with stimulatory effects on insulin release (Pari and Uma, 2000; Prince et al., 1997). Since there is not any other survey on Persian shallot, we have to compare our result with other plants studies. Present results are in agreement with our previous study that found that treatment of garlic extract significantly decreased the blood glucose level and also increase gently insulin level in diabetic group (Mahmoodi et al., 2011). Kar et al. (1999) reported that, the inorganic part of a medicinal plant contains mainly mineral elements which are responsible for the hypoglycemic activity. In support of this notion, a number of essential minerals [for example, Ca (calcium), Zn (zinc), K (potassium), Mn (manganese) and Cr (chromium)], are known to be associated with the mechanisms of insulin release and its activity in different animals and in human beings (Castro, 1998). In an investigation, Ebrahimi et al. (2008) reported that kangavar shallot is an enriched source of Fe (iron) and Cu (copper) compared to 17 different types of Persian shallots obtained from different geographical regions of

Iran. Therefore, probably some of the antioxidant properties of Persian shallot observed in present study are mostly related to these compounds. Also, an investigation demonstrated that shallot extracts had antioxidant properties similarly or slightly higher than garlic extracts.

The relative antioxidant potentials of the garlic and shallot extracts are probably due to their content of phenolic and sulfur compounds (Leelarungrayub et al., 2004). Previous studies reported that the total content of phenolic and diallyl disulfide compounds in shallot extract were higher than garlic (Terrance et al., 1992). Hyperlipidemia is associated with diabetic state (Kumarappan et al., 2007) and this may possibly be due to uninhibited action of lipase (Bangar et al., 2009). In consistence with previous studies by Newairy et al. (2002), we also observed that high levels of serum cholesterol, triglyceride, LDL-C, along with low levels of serum HDL-C in STZ-induced diabetic state are in parallel with the low level of serum insulin. In the present study, the treatment of the induced-diabetic rats with Persian shallot extract led to reduced plasma triglycerides and LDL-C, while in presence of these extracts plasma total cholesterol (significantly) and HDL-C (insignificantly) level were increased. Since insulin inhibits adipose tissue hormone sensitive lipase and reduces lipolysis, the Persian shallot extract may correct the aforementioned disorders via mimicking insulin action. As far as we know, there is no other related data about Persian shallot in literature but, this present report is in agreement with previous studies by Bhandari et al. (2005) which revealed that ethanolic extract of ginger produced significant decrease in serum triglycerides levels and increased HDL-C level in diabetic rats.

In contrast to our findings (that showed elevation in total cholesterol), a study reported declined cholesterol level in garlic treated group (Zamani et al., 2009). In the present study, the decreasing levels of plasma triglycerides and LDL-C following the treatment with Persian shallot extract in our model might be due to the stimulatory effects of Persian shallot extract on insulin secretion. The reduction in plasma total protein and albumin levels that was observed in our model of diabetic rats is consistent with the results presented by Bakris (1997) and Tuvemo et al. (1997). The decrease in protein and albumin may be due to microproteinuria and albuminuria, which are important clinical markers of diabetic nephropathy (Mauer et al., 1981) and/or may be due to increased protein catabolism (Almdal and Vilstrup, 1998). Treatment with Persian shallot increased albumin and protein in present model. It has been established that insulin stimulates the incorporation of amino acids into proteins and this may reflect the albumin and total protein production which happened in present model (Mansour and Newairy, 2000). Such an improvement in serum protein and albumin production was previously observed following oral administration of *Balanites aegyptiaca* to experimentally diabetic rats (Shinde and Goyal, 2003). The STZ-induced diabetic rats exhibited a pattern of significantly higher plasma urea, uric acid and creatinine levels compared to the normal group.

A significant elevation in serum creatinine, uric acid and urea levels is indicative for impaired renal function in diabetic animals. The present study demonstrated that Persian shallot extract improved renal function, which was evident from the lowered serum uric acid and creatinine levels in the rats treated with Persian shallot extract. These results are in agreement with other previous studies on the mesocarp extract of *B. aegyptiaca* (Saeed et al., 1995) and herbal formulation D-400 (Dubey et al., 1994). Analysis of shallot extracts has confirmed the presence of flavonols such as quercetin, and sulfur compounds such as diallyl disulfide (Leelarungrayub et al., 2006). We suggested that the presence of polyphenols and flavonoids in Persian shallot extract might be responsible for the antioxidant nephroprotective activities and the reduction of serum uric acid and creatinine levels (Kaleem et al., 2008); however, Persian shallot extract increase urea level significantly.

The beneficial effect of Persian shallot extracts on altering blood biochemical parameters in diabetic rats may be due to the antioxidant capacity of its content.

Conclusion

It could probably be stated that the extract of Persian shallot may provide a new therapeutic avenue against

diabetes and diabetes-related complications. Overall, from the data obtained, treatment with Persian shallot extract produced a significant antihyperglycemic effect. Furthermore, Persian shallot is capable of improving lipid and protein profiles in diabetic rats. Moreover, further work is necessary to seek the active ingredients present in this extract having antidiabetic efficacy.

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