

Effect of lupine (*Lupinus termis*) seeds or their water extract on alloxan diabetic rats

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Abstract

Diabetes mellitus represents a major public health problem. Much of the increased mortality and morbidity seen in diabetic patients is the result of various complications. Free radicals play an important role in the cause of complications of diabetes mellitus such as retinopathy, nephropathy, hypertension, arteriosclerosis and ischemic heart diseases. Many secondary plant metabolites have been shown to possess antioxidant activities, improving the effects of oxidative stress in diabetic patient. Thus the present study tries to evaluate the role of different preparations from *Lupinus termis* as a hypoglycemic agent. Alloxan diabetic rats were orally treated with either 5 ml/Kg b.wt/day aqueous lupine seeds extract or 20% w/w in diet edible boiled lupine seeds powder or 20% w/w dry lupine seeds powder for 30 days. The results recorded high levels of glucose, total cholesterol, triglycerides and LDL-cholesterol as well as low levels of total protein, HDL-cholesterol, liver glycogen and serum insulin in diabetic rats. Phospholipids content was increased in the serum but decreased in the liver and pancreas of diabetic rats. In addition, the results confirmed that the alloxan diabetic rats were subjected to oxidative stress as indicated by the extent of lipid peroxidation (high malondialdehyde levels) present in the liver and pancreas and significantly alter activities of some scavenging enzymes (low glutathione reductase, glucose-6-phosphate dehydrogenase, superoxide dismutase and catalase). On the other hand, after administration of any one of the used preparations of lupine seeds, the diabetic rats revealed an improvement on various metabolic abnormalities as well as oxidative stress recorded with diabetes but the best improvement occurred in the animal group treated with dry seeds powder. These results give a good evidence for the amelioration effect of lupine against the alloxan diabetic effects.

Key words: Antidiabetic plants - seed extract - metabolic abnormalities – free radical - alloxan diabetic rats.

Introduction

Diabetes is one of the most common diseases especially in tropical area and the third world. Diabetes mellitus is a multifactorial disease, which is characterized by hyperglycemia and lipoprotein abnormalities (Scoppola *et al.*, 2001). These trails are hypothesized to damage cell membranes which results in elevated production of reactive oxygen species (ROS) (Wolff *et al.*, 1991). This generation of oxygen-free-radicals during cellular metabolism, and by certain environmental factors, including lifestyle, appears to play a critical role in the pathogenesis of diabetes (Hartnett *et al.*, 2000). Hyperglycemia, the main symptom of diabetes, not only increases the production

of ROS but also affects antioxidant reactions catalyzed by ROS scavenging enzymes (Uchimura *et al.*, 1999). Diabetes mellitus has also been associated with an increased risk for developing premature atherosclerosis due to increase in triglycerides and low-density lipoprotein levels and decrease in high-density lipoprotein level (Bierman, 1992). All organisms possess antioxidant enzymes [superoxide dismutase (SOD), catalase (CAT) and glutathione peroxidase (GSHPx)] responsible for scavenging ROS. A defect in ROS scavenging enzyme system has been reported in diabetes mellitus (Kesavulu *et al.*, 2000 and Jung *et al.*, 2004).

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If diabetes is not successfully treated and carefully controlled, many complications may be developed. Treatment of all forms of diabetes depends on normalizing the amount of insulin released from the β -cell or to adjust the requirements of tissue for nutrients to maintain metabolic homeostasis (Flier, 1992). This metabolic imbalance is responsible for the acute metabolic derangements of the chronic complications consequently, the major objective of all forms of treatments is the normalization of nutrients homeostasis and the attainment of this goal may prevent or delay the onset of complications (Mohen *et al.*, 1991).

Nowadays, there is uprising awareness for going back to nature and getting away as much as possible from synthetic drugs. That is because many of the synthetic drugs have shown undesirable side effects, especially when used for long terms. Moreover, the hazardous side effects are even augmented when more than one synthetic drug are used simultaneously due to drug interaction (Sheela and Augusti, 1992). Many minor compounds of foods such as secondary metabolites, have been shown to alter biological processes which may reduce risk of chronic disease in humans. Recent studies showed that aqueous extracts of different antidiabetic plants enhanced insulin secretion and mimicked the effect of insulin on glucose metabolism in vitro (Gray and Flatt, 1998). Such dual pancreatic and extrapancreatic actions would prove to be an important advance on existing therapies used to treat and control diabetes, such as oral hypoglycemic drugs (which act either by enhancing insulin secretion or by improving the action of insulin or producing insulin resistance) (Gray and Flatt, 1997). These combined findings illustrate the enormous potential of plants for use as possible dietary adjuncts and the discovery of natural products for diabetes therapy (Gray *et al.*, 2000).

lupine (*Lupinus termis*), an edible plant native to the North Africa, especially in Egypt, has been widely used in folk medicine for maintaining tolerance blood glucose levels (Riyad *et al.*, 1988 and Zaahkouf, 2001). Shani *et al.* (1974)

have shown that lupine seeds extract produced a mild and transient hypoglycemic effect in diabetic rats. This was somewhat contradictory to an earlier report by Helmi *et al.* (1969), where lupine seeds powder produced a pronounced hypoglycemic effect in both normal and diabetic rabbits. Moreover, the termis seed extracts used in treatment of chronic eczema of hands and act as anti-histaminic drug (Lemus *et al.*, 1999). The authors also noted that *lupinus termis* extracts showed a blood pressure reduction in hypertensive rats. Studies are available with conflicting results on the effects of termis on lowering the levels of cholesterol and triglycerides in diabetes (Rodriquez *et al.*, 1998).

In view of these information, it was therefore of interest for us to evaluate the effect of different preparations of lupine seeds supplementation diets on the induction of experimental diabetes and its management as judged by blood glucose, lipid profile and oxidative stress in alloxan diabetic rats.

Materials and Methods

Plant material preparations:

Dried Lupine seeds were purchased from the local market. Water extract of lupine were obtained by boiling lupine seeds (300 g/L) for one hour and the extract solution was stored in the refrigerator to avoid possible deterioration of active ingredients. The extract was given orally to rats daily by gastric intubations at a dose of 5 ml/Kg b.wt. at a concentration of 1500 mg/Kg (Zaahkouf, 2001). Two different diets supplemented with lupine seeds were also used for treatment, the first one was prepared by adding 20% (w/w in diet) edible boiled lupine seeds and the other one was prepared by adding 20% (w/w in diet) dry seeds powder (Riyad *et al.*, 1988).

Animals and treatment:

Twenty-five male albino rats weighing 150-170g. were used in this work. Animals were kept under good ventilation and received a balanced diet and water *ad libitum* throughout the experiment. Diabetes was induced by interaperitoneal injection of a single dose

of 120 mg alloxan/Kg b.wt., dissolved in acetate buffer pH 5.5, to overnight fasted rats (Othman, *et al.*, 1989). Successful induction of diabetes was assessed 3 days after alloxan injection by performing glucose urine analysis Glukotest which was obtained from Bohringer, Mannheim Company, Germany.

The experimental animals were then divided into four groups, in addition to a control group that received a single interperitoneal (IP) injection of acetate buffer pH (5.5). The first one left as diabetic group. In the second group diabetic rats were treated daily with aqueous lupine seeds extract by gavage at a dosage of 5 ml/Kg b.wt. While, in the Third and fourth groups, the diabetic rats of each groups fed diets supplemented with 20% edible boiled lupine seeds powder or dry lupine seeds powder respectively. After 30 days from treatment, the rats were sacrificed and the blood was collected in non heparinized tubes and centrifuged at 4000 g for 10 min to remove the serum for biochemical determinations. Liver and pancreas specimens were surgically removed, immediately washed with ice cold saline and prepared for biochemical analysis.

Biochemical analysis:

The level of glucose and high density lipoprotein cholesterol (HDL-c) in the serum, in addition to the concentration of total lipids, total cholesterol, and triglycerides in serum, liver and pancreas were estimated using assay Kits (Purchased from Sigma Chemical Co, USA). Total protein (TP) was assayed by Biuret reaction according to Dumas (1975). The method of Baginiski *et al.* (1972) was adopted to determine the level of total phospholipids. Serum insulin levels was measured using immuno-enzymatic assay Kits for the quantitative measurement of insulin in serum (Flier *et al.*, 1979). Liver glycogen was estimated according to the technique of Van Handle (1965). Glucose-6-phosphate dehydrogenase (G6PD) activity was estimated depending on the method of Chan *et al.* (1965). Moreover, thiobarbituric acid reactive substances (TBARS) was

determined as described by Esterbauer and Cheeseman (1990). The content of Glutathione (GSH) was estimated by the method of Prins and Loose (1969). Glutathione reductase (GSH-Rx) activity was determined by the method of Beutler (1975). The activity of superoxide dismutase (SOD) was assayed according to the method of Niskikimi *et al.* (1972). Catalase activity was detected as described by Aebi (1983).

Statistical analysis:

All results were expressed as mean \pm S.E of five animals. The obtained data were compared using Student t-test as described by Snedecor and Cochran (1982). The level of significance was expressed as significant at $P < 0.05$.

Results

The results of the present study as shown in tables 1 and 2 revealed that the injection of alloxan induced a significant increase in glucose level and significant decrease in liver glycogen content of the same animals. In addition, the present data showed that diabetic rats demonstrated significant increase in serum, liver and pancreas total lipids, total cholesterol, triglycerides and LDL-c concentrations. On contrary, serum HDL-c level and total protein content in the same examined tissues was significantly decreased after alloxan injection. In diabetic rats, phospholipid was significantly decrease in liver, while non significantly altered in both serum and pancreas. However, the obtained data regarding diabetes-induced changes in glucose and insulin levels in serum (table 1) as well as in liver glycogen content (table 2) showed occurrence of good treatment by dry seeds powder (group 5) of lupine particularly glucose and insulin which were nearly returned to control values. Moreover, the administration of lupine into diabetic rats showed progressive recovery in total protein content as well as in the level of total lipids, total cholesterol, triglycerides, phospholipids, HDL-c and LDL-c in the examined tissues throughout the experimental duration. Whereas supplemented diets with 20% edible boiled

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lupine seeds powder (group 4) showed non significant change in the levels of the above mentioned serum, hepatic and pancreatic parameters comparing to diabetic rats group except the level of serum total lipids, where the decrease was significant as shown in Table (1&2).

As shown in table 3 it is clear that the TBARS levels in diabetic rats were higher than in normal control group. However, diabetic rats displayed significant decrease in liver and pancreatic glutathione content as well as in the

activities of GSH-Rs and SOD. Also, the data of the activity of G6PD and catalase enzymes recorded significant decrease in diabetic rats compared to control rats. Furthermore, the amelioration noted in the above mentioned parameters after the treatment with water seeds extract or dry seeds powder while addition of edible boiled seeds lupine to the diet of diabetic rats (group 4) did not cause significant changes in the most oxidative stress as compared with diabetic rats (Table 3).

Table 1: Effect of lupine (*Lupinus termis*) on some serum biochemical parameters in diabetic rats

Animal Groups Parameters	Control group	Diabetic group	Diabetic +aqueous seeds extract	Diabetic +edible boiled seeds powder	Diabetic +dry seeds powder
Glucose level (mg/100ml)	112.5±2.4	398.4±3.4 a	230.7±2.3 a&b	388.1±6.6 a	116.4±2.1 b
Insulin level (µU/ml)	4.9±0.61	2.6±0.45 a	3.8±0.12 b	3.1±0.32 a	4.6±0.25 b
Total protein (g/100ml)	8.9±1.2	4.4±1.6 a	6.9±1.3	5.8±2.6 a	8.4±2.4 b
Total lipids (mg/dl)	654.2±3.2	988.8±3.5 a	811.4±4.2 a&b	961.7±2.7a&b	724.3±3.7a &b
Total cholesterol (mg/dl)	198.4±3.2	354.3±3.5 a	280.4±2.3 a&b	334.1±6.3 a	203.6±4.1 b
Triglycerides (mg/dl)	87.5±2.1	140.4±3.2 a	103.6±3.6 a&b	136.2±4.5 a	91.2±2.5 b
Phospholipids (µmol/min/100ml)	6.4±2.3	7.9±0.41	6.02±1.5	7.5±0.85	8.5±0.46 b
HDL-cholesterol (mg/dl)	41.2±2.4	33.1±1.3 a	38.2±1.2 b	36.4±1.7 a	44.2±1.5 b
LDL-cholesterol (mg/dl)	139.4±2.6	293.1±8.1 a	221.5±1.4 a&b	270.5±9.6 a	141.2±2.8 b

Values are presented as means ± SE. (n=5).

a → P<0.05 versus control group.

b → P<0.05 versus diabetic group.

Table 2: Effect of lupine (*Lupinus termis*) on some hepatic and pancreatic biochemical parameters in diabetic rats.

Animal Groups	Control group	Diabetic group	Diabetic +aqueous seeds extract	Diabetic +edible boiled seeds powder	Diabetic +dry seeds powder	
Liver	Glycogen content (mg/g wet tissue)	8.8±1.3	4.3±1.4 a	6.7±1.2	4.7±1.4 a	7.5±1.7
	Total protein (g/100g wet tissue)	18.6±2.6	11.6±2.2 a	17.5±3.6	13.8±4.1	17.8±2.8
	Total lipids (mg/g wet tissue)	53.1±0.54	74.3±0.81 a	59.2±0.74 a&b	68.1±0.97 a	51.03±0.54 a&b
	Total cholesterol (mg/100g wet tissue)	135.2±5.6	174.5±4.1 a	142.7±3.8 b	169.7±4.1 a	132.2±3.6 b
	Triglycerides (mg/g wet tissue)	32.4±2.5	54.2±1.3 a	35.7±1.8 b	49.9±2.4 a	34.6±2.9 b
	Phospholipids (µmol/min/g dry tissue)	48.8±1.6	32.9±1.6 a	42.1±2.9 a&b	34.5±2.6 a	47.4±3.6 b
Pancreas	Total protein (g/100g wet tissue)	14.9±1.2	11.8±0.87 a	12.8±1.3	11.6±0.96 a	13.8±1.4
	Total lipids (mg/g wet tissue)	44.2±3.5	54.2±4.8	43.2±6.5	51.3±5.2	39.3±4.1 b
	Total cholesterol (mg/100g wet tissue)	19.5±3.06	30.2±2.4 a	23.1±3.1	28.5±5.2	19.9±4.6 b
	Triglycerides (mg/g wet tissue)	23.8±2.6	38.7±1.7 a	30.6±1.2 a&b	36.1±2.8 a	31.5±2.5 a&b
	Phospholipids (µmol/min/g dry tissue)	34.7±1.3	30.4±2.8	35.4±2.7	31.2±4.1	36.6±1.5 b

Values are presented as means ± SE. (n=5).

a→P<0.05 versus control group.

b→P<0.05 versus diabetic group.

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Table 3: Effect of lupine (*Lupinus termis*) on some hepatic and pancreatic antioxidant parameters in diabetic rats.

Animal Groups	Control group	Diabetic group	Diabetic +aqueous seeds extract	Diabetic +edible boiled seeds powder	Diabetic +dry seeds powder	
Liver	TBARS (n.mol/g wet tissue)	45.4±2.3	67.5±3.5 a	49.2±2.4 b	65.4±3.6 a	46.6±2.7 b
	GSH (mg/g wet tissue)	7.2±0.14	3.5±0.12 a	5.5±0.08 a&b	3.7±0.11 a	5.7±0.16 a&b
	GSH-Rx (U/g wet tissue)	68.7±1.04	50.4±0.58 a	55.4±0.83 a&b	51.4±0.48 a	59.4±0.36 a&b
	G6PD (U/g wet tissue)	3.8±0.10	2.03±0.09 a	3.1±0.12 a&b	2.2±0.08 a	3.6±0.09 b
	SOD (U/g wet tissue)	33.6±1.4	27.4±1.5 a	30.5±1.2	28.2±1.4 a	32.5±1.5 b
	Catalase (KU/sec/g wet tissue)	0.39±0.004	0.20±0.005 a	0.25±0.006 a&b	0.21±0.004 a	0.32±0.004 a&b
Pancreas	TBARS (n.mol/g wet tissue)	21.5±1.5	34.1±2.8 a	26.5±2.6 b	31.2±1.2 a	24.1±2.1 b
	GSH (mg/g wet tissue)	3.4±0.14	1.5±0.11 a	2.4±0.15 a&b	1.8±0.17 a	2.9±0.19 a&b
	GSH-Rx (U/g wet tissue)	18.8±0.12	13.2±0.15 a	17.9±0.14 a&b	13.4±0.19 a	18.5±0.18 b
	G6PD (U/g wet tissue)	1.3±0.003	0.89±0.001 a	1.2±0.002 a&b	0.90±0.006 a	1.29±0.005 b
	SOD (U/g wet tissue)	6.4±0.09	4.8±0.11 a	5.7±0.05 a&b	4.9±0.13 a	6.2±0.12 b
	Catalase (KU/sec/g wet tissue)	0.18±0.02	0.12±0.004 a	0.16±0.003 b	0.13±0.01 a	0.17±0.01 b

Values are presented as means ± SE. (n=5).

a→P<0.05 versus control group.

b→P<0.05 versus diabetic group

Discussion

The present data demonstrated that, alloxan-induced diabetic rats showed a marked disturbance as severe hyperglycemia and hypoinsulinemia. Hyperglycemia can be considered as a direct reflex to the marked hypoinsulinemia caused by the selective destructive cytotoxic effect of alloxan on the β -cells of the pancreas (Bolaffi *et al.*, 1986). The biochemical basis of diabetes-induced insulin resistance could involve changes in glucose transporter numbers (Lillioja *et al.*, 1988) or in insulin-induced activation of glycogen synthesis, which reflect primarily depressed glycogen synthesis ((Margolis *et al.*, 1985 and Ortmeyer *et al.*, 1994). This could provide explanation for the decreased ability of the liver to accumulate glycogen in the present study. In association with these reportedly diabetes induced changes in carbohydrate metabolism, various studies have demonstrated a general elevation in almost plasma lipid fractions that often coexists with hyperglycemia (Orchard, 1990 and Betteridge, 1994). Apparently, the most commonly elevated plasma lipids in diabetes is the triglycerides, total cholesterol and LDL-c levels. In our study, alloxan-diabetic rats exhibited marked hyperlipidemia, hypertriglyceridemia and hypercholesterolemia in serum, liver and pancreas, a result which agree with that of Jaiprakash *et al.* (1993) and Ginsberg (1996). Because insulin is known to suppress VLDL secretion, the lack of this suppression by deficient insulin secretion in diabetes may lead to hypertriglyceridemia, which possibly can mediate the mechanism by which diabetes causes hyperlipidemia (Saudek and Eder, 1979). The detected increase in cholesterol may originate from the increase in cholesterol biosynthesis in various tissues and its migration to the blood circulation (Zaahkoug, 2001). In other words, the reduction in the rate of cholesterol removal from circulation appears responsible for hypercholesterolemia recognized in diabetic states (Ebara, 1994). Diabetic dyslipidemia is due mainly to decreased removal of

triglycerides into the fat depots, also the increase in serum concentration of very low density lipoprotein (VLDL) and low density lipoprotein (LDL) may be due to overproduction of VLDL by the liver or decreased removal of VLDL and LDL from the circulation (Tsutsumi *et al.*, 1995).

Furthermore, in the present work, diabetic rats showed a significant decrease in serum liver and pancreas total protein content. Mortimore and Mondon (1970) found that lack of insulin effects on synthesis of hepatic protein. Therefore, the major effect of insulin on hepatic protein metabolism of proteolysis (Nair *et al.*, 1983) and thus it appears that the decreased protein contents observed in the present diabetic study was a result of increased protein breakdown rather than decreased protein synthesis (Devlin *et al.*, 1994).

The present data, on the other hand, revealed that the different preparations from lupine seeds intake to alloxan diabetic rats suppressed the various metabolic disturbances associated with diabetes. This is indicated by an improvement of glucose and insulin levels, concomitantly with tendency of hepatic glycogen content to nearly control value, indicating stimulated glucose utilization. Furthermore, lipid fractions were found to be lowered near to normal values. At the same time, no lipid deposition occurred in both liver and pancreas tissues, indicating that the serum lipid lowering effect of lupine was not mediated by a transport of lipids from serum to the tissues. Along with this, amelioration in the level of total proteins were observed in the serum and in both pancreas and liver following lupine treatment.

The present results run in agreement with the previous studies of Harazti and Vetter (1983) and Zahran (2004). These observed metabolic improvements could contribute to the hypoglycemic effect of *Lupinus termis* that may be due to its alkaloidal contents of sparteine and lupinine and /or its high manganese content, (Abdel-Kader, 1986 and Al-zaid,

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1987). Manganese chloride has been shown to exert a hypoglycemic action in independent diabetic mellitus (IDDM) patients and manganese is now recognized as necessary co-factor for ATP phosphorylation of β -subunit of the insulin receptor (Reddy and King, 1987). Moreover, the treatment with manganese salt resulted in a net increase in acute insulin response and the rate of glucose disappearance after glucose loading (White and Campell, 1993). Also the effect of lupine seeds in lowering blood glucose level in alloxan diabetic rats may be due to the positive action of these plants on the impaired glucoreceptor mechanism in diabetes and indirectly in large part perhaps dependent on insulin release from the pancreas (Kimura *et al.*, 1981).

There are several explanations for the hypoglycemic effect of *Lupinus termis*. Helmi *et al.* (1969) suggested that the *Lupinus termis* has a potency effect equal to those of oral hypoglycemic drugs, like tolbutamide or acetohexamide. Moreover, Youness *et al.* (1985) concluded that, this plant can be used as a safe and natural mean to lower blood glucose level. Meanwhile, Abdel-Aal *et al.* (1993) found that *Lupinus* seeds produced a definite hypoglycemic effect in normal and alloxan-diabetic rats. The authors attributed this result to the considerable improvement in the histologic picture of the islets of Langerhans of the pancreas which was noticed by lupine seeds administration. The islets increased in number and size compared to those in the alloxan-diabetic rats. Also, Bwititi *et al.* (2002) observed that the plant has possibly a pancreatic β -cell stimulating action (as an insulin-releasing natural product). This view might run in accordance with earlier study of Collip (1923) who suggested that in the plant kingdom there is a hormone analogous to insulin, which is known as glucokinin. In addition to, the anti-diabetic activity of the *Lupinus termis* may be due to the presence of an active ingredient such as lupanine where lupanine is the main alkaloid which causes hypoglycemic action (Ajabnoor and Tilmisany, 1988). In this study, both water seeds extract or dry seeds lupine powder significantly reduced

the triglyceride, total cholesterol, phospholipids and LDL-c levels as well as increased HDL-c level in treated diabetic rats when compared to untreated diabetic rats. These reductions could be beneficial in preventing diabetic complications as well as improving metabolism in diabetics (Cho *et al.*, 2002). This decrease in the lipid fractions may be due to utilization of circulating free fatty acid by the liver and/or its fairly high contents of fiber and plant protein (Gabrial and Morcos, 1976). The present suggestion is based on the findings of Trowell (1972) who found that dietary fiber decreases the reabsorption of bile salts, increases fecal excretion, reduced hyperlipidemia and may exhibit a hypocholesterolemic effect in rats. Also, treatment with aqueous plant extract or dry seeds powder ameliorated the disturbance of protein in serum, liver and pancreas of diabetic rats. These findings are in accordance with Zaahkouk (2001) who recorded a remarkable increase in serum total protein and globulin in diabetic rats treated with aqueous lupine and fenugreek extracts. This increase in protein content may be due to the high protein content of the *Lupinus termis* (Gabrial and Morcos, 1976).

Furthermore, impaired glucose metabolism leads to oxidative stress (Ceriello *et al.*, 1992) and protein glycation products free radicals (Wolff *et al.*, 1991). Therefore, the presently noted decrease in antioxidant enzyme activity in control diabetic rats when compared to diabetic rats treated with the aqueous seeds extract or seeds powder from *Lupinus termis* could at least in part result from inactivation of the enzymes by H_2O_2 or by glycation, which are known to occur during diabetes (Sozmen *et al.*, 2001). In search of a second factor conditioning such as oxygen radicals accumulation we have measured the level of the principal non-enzymatic antioxidant GSH. The detected decrease in GSH content in this study is concomitant with the lowered GSH-Rx activity observed in diabetic rats. An association between the impairment in GSH-Rx activity and GSH depletion is suggested by the observation of Murakami *et al.* (1989). Moreover, the present finding of decreased G6PD

activity, may relate in part to decreased GSH level observed in this study. A view which supported by Montgomery *et al.* (1980) who showed that the reduced G6PD activity may be in part involved in the mechanism of GSH depletion in diabetes. Lipid peroxidation were also evaluated by measuring hepatic and pancreatic levels of TBARS. In agreement with other studies (Aydin *et al.*, 2001 and Cho *et al.*, 2002), we observed a significant increase in TBARS levels. This increase may be attributed to the increased peroxidative destruction of cellular membrane phospholipids. Moreover, the change in phospholipid fatty acids composition in the cellular membranes in diabetes may lead to increased lipid peroxidation, leading to disturbance of both membrane structure and function (Weber, 1994). The results from the present study showed that diabetes decreases phospholipids content in both pancreas and liver seems to support this suggestion.

On the other hand, our results have demonstrated the efficacy of some preparations of lupine seeds to prevent the progression of cellular lipid peroxidation, as indicated by the decreased TBARS level in both liver and pancreas tissues, along with restoring the normal phospholipids content in the examined tissues. The marked improvement in oxidative stress may be attributed to the antioxidant defense function of traditional antidiabetic plants as shown by increased reduced glutathione content as well as glutathione reductase, glucose 6-phosphate dehydrogenase, superoxide dismutase and catalase enzymes activity (Ugochukwu, *et al.*, 2003). Supporting this suggestion, it has been mentioned that termis has a potent to act as free radical scavenger, hypolipidemic and hypoglycemic agents by the effect of lupanine and sparteine. The latter have the ability to bind or chelate the divalent cation which led to decrease in the oxidative stress and oxidation damage in tissue (Helmi *et al.*, 1969 and Shani *et al.*, 1974).

From the present investigation, it could be concluded that diabetes leads to several complications such as hyperglycemia, hyperlipidemia, disturbance in

the lipid peroxidation and elevation in the free radicals. On the other hand, treatment of diabetic rats with *lupinus termis* exerts antidiabetic action against alloxan diabetic effects through modulation of the various metabolic disturbances associated with diabetes, in addition to the improvement in the defense mechanisms in examined tissues. Finally, it is obvious that the used of dry seeds powder of *lupinus* as hypoglycemic agent mean to lessen successfully the complications of diabetes better than seeds extract or boiled seeds of lupine, suggesting the possibility that the addition of antidiabetic plants to the treatment protocols used for diabetic patients may improve diabetic therapy.

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تأثير بذور الترمس ومستخلصه المائي على الجرذان المصابة بمرض السكر بالألوكزان

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يمثل مرض السكر أكبر المشاكل الصحية العامة والتي تسبب الكثير من حالات الوفاة والمرض نتيجة للمضاعفات العديدة. وقد لوحظ أن الشوارد الحرة تلعب دورا هاما في حدوث مضاعفات مرض السكر.

وبناء عليه، أجرى هذا البحث لتوضيح مدى تأثير كل من مستخلص أو مطحون بذور نبات الترمس على بعض المعايير الفسيولوجية للجرذان المصابة بمرض السكر الناتج عن الحقن بعقار الألوكزان بجرعة 120 مليجرام / كيلوجرام من وزن الجسم وقد أوضحت النتائج أن الألوكزان يسبب زيادة معنوية في مستوى السكر في مصل الدم ويصاحب ذلك نقص معنوي في محتوى الجليكوجين في الكبد ومستوى الانسولين في مصل الدم كما لوحظ زيادة محتوى الدهون الكلية والكوليسترول الكلى والجليسيريدات الثلاثية في مصل وكبد وبنكرياس الجرذان المصابة بمرض السكر.

هذا بالإضافة إلى نقص البروتين الكلى في مصل وكبد وبنكرياس نفس الحيوانات. كما أظهرت النتائج زيادة في أكسدة الدهون ونقص في نشاط إنزيم تحويل الأيونات الحرة وإنزيم الجلوتاثيون المرجع والجلوكوز سداسي الفوسفات الهيدروجيني كما لوحظ نقص في محتوى كل من الجلوتاثيون والدهون الفوسفورية في الكبد والبنكرياس.

وعند تعاطى المستخلص المائي لبذور الترمس عن طريق الفم بتركيز 5 سم³/كجم من وزن الجسم لمدة 30 يوما أو إضافة مطحون بذور نبات الترمس الجاف بتركيز 20% في الوجبة الغذائية للجرذان المصابة بالسكر وجد تحسن ملحوظ في جميع المعايير المقاسة بالمقارنة بالجرذان المصابة بمرض السكر. بينما كان تأثير إضافة مطحون بذور الترمس بعد تعرضه للغليان لمدة ساعة بنفس التركيز السابق (20%) على الجرذان المصابة بمرض السكر تأثير غير ذي دلالة إحصائية.

وتخلص هذه الدراسة إلى أن استخدام مطحون بذور الترمس الجاف يعد الأفضل في علاج الجرذان المصابة بالسكري يليه استخدام المستخلص المائي لبذور الترمس حيث أن جرعة الأكثر تثبيطا للجلوكوز أظهرت تحسنا ملحوظا على كل من القياسات البيوكيميائية الوظيفية للجرذان المصابة بمرض السكر ويرجع ذلك إلى وجود المادة الفعالة الشبة قلوية (ليوبينين) و يحتمل أن المادة الفعالة لخفض السكر يثبط مفعولها بالغليان.