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Abstract

A number of herbs are traditionally used in Sudan to treat different diseases. In the present study the therapeutic effects of *Trigonella foenum graecum (*Fenugreek), and *Lupinus termis* aqueous extracts in glucose-induced diabetic albino rats and the sub chronic toxicity of these plants were investigated. Forty two albino rats of either sex (weighing 135-250 g and aged 60 days) were used. Animals were assigned to seven equal groups (N=6); intact control and six diabetic groups. Diabetes was induced by 5% glucose (2mg/kg b.w). Group (1) was administered with drinking water and assigned as control, groups 2, 3 and 4 were administered with *Trigonella foenum* aqueous extract (200, 400 and 800 mg/kg b.w, respectively) and groups 5, 6 and 7 were administered with *Lupinus termis* aqueous extract (200, 400 and 800 mg/kg b.w, respectively). Blood samples were obtained to assess blood glucose level, α-amylase, cholesterol, high density lipoprotein (HDL) and triglycerides concentrations. In treated diabetic rats, blood glucose level markedly decreased to the normal range. Reduction in cholesterol, triglycerides and HDL was also observed. Reduction of α-amylase concentration was shown with a low dose (200 mg/kg b.w.) of the extracts of both plants. The present studied plants clearly demonstrate the non-toxic nature and safety profile. The combined history human use of these plants and the data from the current study support the safe use of these plants. It can be concluded that *Trigonella foenum graecum* seed and *Lupinus termis* fruit aqueous extract have anti-hyperglycemic effect by affecting both blood glucose level and α-amylase. *Trigonella foenum graecum* has higher hypoglycemic than *Lupinus termis.*

Key words:Diabetes mellitus, hypoglycemia, hypolipidemia, *Trigonella foenum graecum*, *Lupinus termis*.

دراسة مقارنة للتأثيرات البايولوجية لنباتي الحلبة والترمس الابيض في الجرذان المصابة بداء السكري

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الخلاصة

العديد من النباتات الطبية تستخدم شعبيا في السودان لمعالجة الأمراض المختلفة. في الدراسة الحالية ، تم التحري عن التأثيرات الدوائية للمستخلص المائي لبذور نبات الحلبة (*Trigonella foenum*) و ثمار نبات الترمس الابيض (*Lupinus termis*) في الجرذان المستحث فيها داء السكري تجريبيا باستخدام الكلوكوز علاوة على دراسة التأثيرات تحت السمية. استحدث داء السكري تجريبيا باستخدام التحميل بالكلوكوز 5% بجرعة 2 ملغم/كغم من وزن الجسم. تم توزيع 42 جرذا من كلا الجنسين ، بأعمار 60 يوما وأوزان تراوحت ما بين 135-150 غم ، على 7 مجموعات متساوية العدد ، ضمت الأولى جرذان سليمة وعدت مجموعة سيطرة وجرعت الثانية والثالثة والرابعة بخلاصة نبات الحلبة بالجرعات 200 و400 و800 ملغم/كغم من وزن الجسم ، على التوالي ، وجرعت الخامسة والسادسة والسابعة بخلاصة نبات الترمس

الابيض بالجرعات 200 و 400 و 800 ملغم/كغم من وزن الجسم ، على التوالي . تم سحب نماذج الدم لتقدير تركيز الكلوكوز والأميليز ألفا و الكوليستيرول و البروتينات الدهنية عالية الكثافة والكليسيريدات الثلاثية. أشارت النتائج الى انخفاض تركيز الكلوكوز و الكوليستيرول و البروتينات الدهنية عالية الكثافة والكليسيريدات الثلاثية في دم الجرذان المعاملة الى المستوى الطبيعي في جميع التراكيز المستخدمة ، أما الأميليز ألفا فقد انخفض عند استخدام جرعة 200 ملغم/كغم لكلا النباتين . أوضحت الدراسة الحالية أن النباتات المدروسة غير سامة وأمينة عند استخدامها بالجرعات الحالية.

الكلمات المفتاحيه: الحلبة ، الترمس الابيض ، داء السكري ، انخفاض السكر ، انخفاض الدهون .

Introduction

Diabetes mellitus is defined as state in which homeostasis of carbohydrate and lipid metabolism is improperly regulated by the pancreatic hormone, insulin; resulting in an increased blood glucose (1). A number of herbs are traditionally used in different countries as drug or toxin induced hepatic, renal and cardiac disorders (2). The term medicinal plant is used to determine the plant or plant products used by human beings in the protection against, or treatment of illness (3).In Sudan, different plants are widely used for the treatment of some diseases for example: *Trigonella Foenum-Graecum* (Fenugreek) seeds, *Lupine termis* seeds, *Solenstomma argel (Gomphocarpus fruticosus),* bark and leaves and *Cinnanomum zeylenicum* are used in treatment of some diseases such as liver and kidney and allergies. Some have an effective remedy for bronchitis and are used for treatment of neuralgia and sciatica. (4). They also used for colds, diabetes, stomach pain, and urinary infection. (5).The present study aimed to investigate the hypoglycemic effect of *Trigonella foenum graecum* (Fenugreek) seeds and *Lupine termis* seeds extract in 5% glucose induced diabetic rats. The study also aimed to evaluate the toxicity effect of these plants.

Material and methods

Study area: Medicinal and Aromatic Plants Research Institute, National Center for Research, Khartoum, Sudan.

Plant materials: Plants selected according to questionnaire which resulted 44.6% of Sudanese diabetic patients used *Trigonella foenum graecum* (Fenugreek) seed and 26.7% used *Lupinus termis* fruit. The plants seed and fruits were obtained from local market at Omdurman, purified, ground to powder using mechanical grinder and preserved in air tight container and kept in clean dry bottles (6).

Preparation of aqueous extraction: Aqueous extracts were prepared according to the method described by Harborne (7), 300g of each powder sample was soaked in 2000 ml of hot distilled water, and left till cooled down with continuous stirring at room temperature. Extract was then filtered and freezed in a deep. Freezed extract was dried using Freeze drying apparatus till powdered extract obtain. The powder residue was re-dissolved in drinking water before experiment.

Animals and induction of diabetes: Wistar albino rats of either sex (weighing 135-250 g and aged 60 days) were kept on a fixed diet so as to stabilize the fasting plasma glucose level at 70-110 mg/dl for 3 days, as an adaptation period. All groups were fasting for 18 hours then loaded with 5% glucose (2mg/kg b.w.) (8) to induce diabetes millitus. Rats with a FBG level higher than 120 mg/dl were included in the study as diabetic animals.

Experimental groups: Forty two (6 intact and 36 diabetic) albino rats were assigned to 7 equal groups (6 each). Intact control (G1) was administered with drinking water. Treated groups (G2, G3 and G4) were administered with *T. foenum* aqueous extract (200, 400 and 800 mg/kg b.w., respectively) and treated groups (G5, G6 and G7) were administered with *Lupinus termis* aqueous extract (200, 400 and 800 mg/kg b.w., respectively). Blood samples (2 ml) were drawn out by capillary tubes in fluorinated test tubes

from the orbital plexus of rats according to Khana *et al*, (9) and centrifuged at 3000 r.p.m for 5 minutes to separate plasma. The plasma prepared was used to assess: blood glucose, α-amylase, cholesterol, high density lipoprotein (HDL) and triglycerides concentrations. Data were statistically analyzed by SPSS using ANOVA-1 and LSD (10).

Results

Figure (1) shows the levels of blood glucose (mg/dl) in control and *T. Foenum* treated diabetic rats (G1, G2, G3 and G4). At zero time, fasting rats revealed low blood glucose level. After loading with glucose, the level increased significantly after 2 hrs. in treated groups compared with control (P=0.016) and then decreased to the normal range after 4hrs. Results of *L. termis* treated diabetic rats (G5, G6 and G7) shown in figure (2) revealed same as that resulted by *T. Foenum* administration. Despite the two plant extracts exhibit same pattern in reducing blood glucose, but *T. foenum* showed higher hypoglycemic effect (35.88% vs 10.08%) compared with *L. termis* aqueous extract. Concerning the effect *T. foenum* seed aqueous extract administration on lipid profile, figures (3, 5 and 7) shows decreased cholesterol, triglycerides, and high density lipoprotein concentrations at 0, 2, and

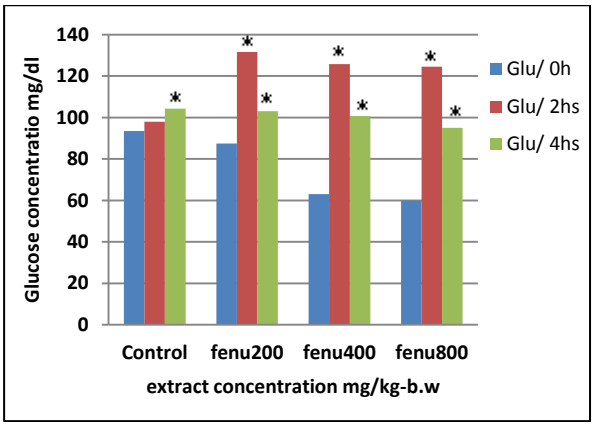

Fig. (1). Effect of administration of different doses of *Trigonella foenum-graecum* seed aqueous extract on blood glucose concentration (mg/dl) in induced-diabetic rats.
***represent significant compare with Glu 0h (p<0.05).**

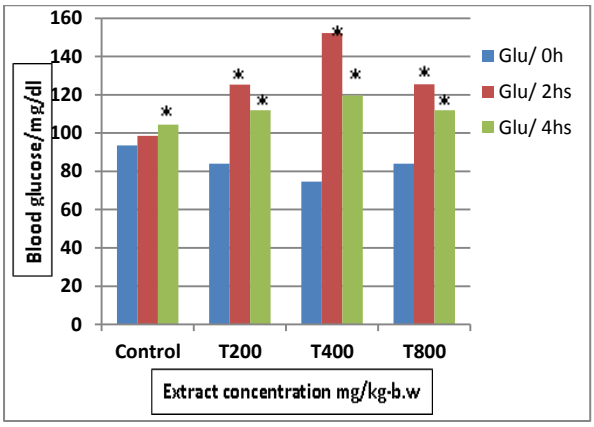

Fig.(2). Effect of administration of different doses of *Lupine termis* fruit aqueous extract on blood glucose concentration (mg/dl) in induced-diabetic rats.
***represent significant compare with Glu 0h (p<0.05).**

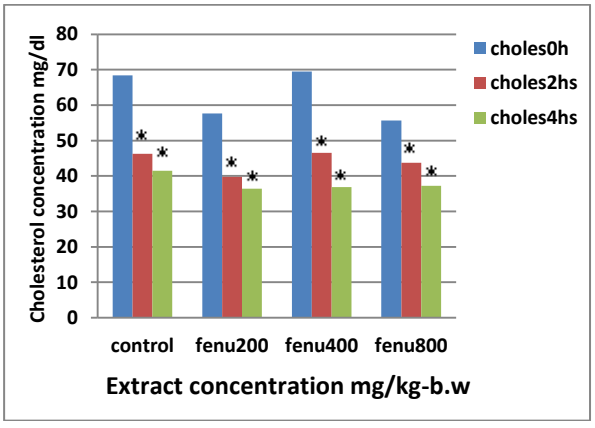

Fig.(3). Effect of administration of different doses of *Trigonella foenum-graecum* seed aqueous extract on blood cholesterol concentration (mg/dl) in induced-diabetic rats.
***represent significant compare with Glu 0h (p<0.05).**

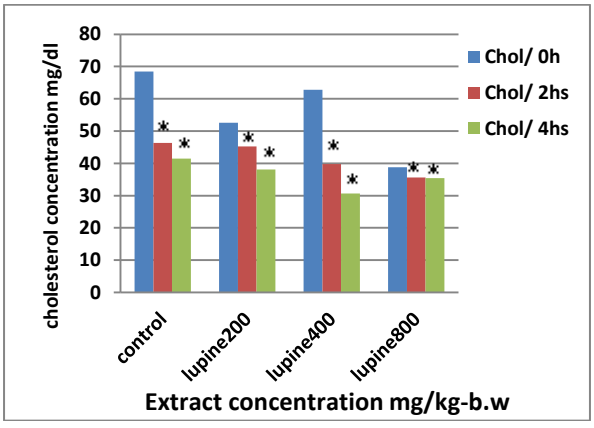

Fig.(4). Effect of administration of different doses of *Lupine termis* fruit aqueous extract on blood cholesterol concentration (mg/dl) in induced-diabetic rats .
***represent significant compare with Glu 0h (p<0.05).**

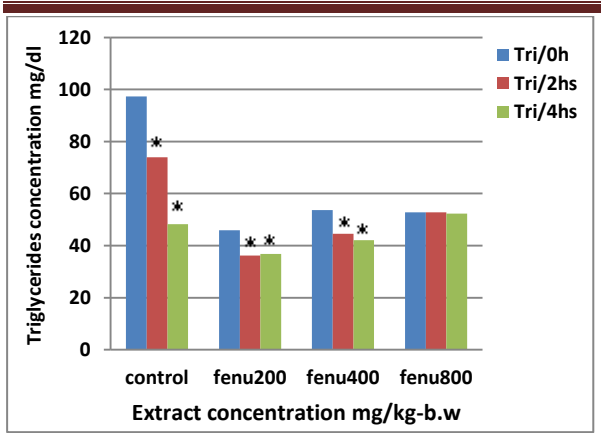

Fig.(5). Effect of administration of different doses of *Trigonella foenum-graecum* seed aqueous extract on blood triglycerides concentration (mg/dl) in induced-diabetic rats .
***represent significant compare with Glu 0h ($p<0.05$).**

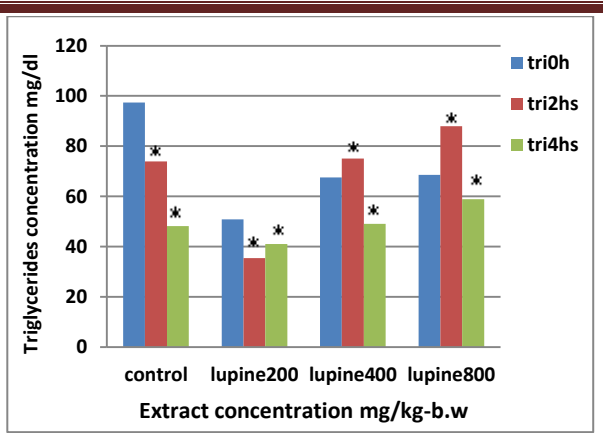

Fig.(6). Effect of administration of different doses of *Lupine termis* fruit aqueous extract on blood triglycerides concentration (mg/dl) in induced-diabetic rats .
***represent significant compare with Glu 0h ($p<0.05$).**

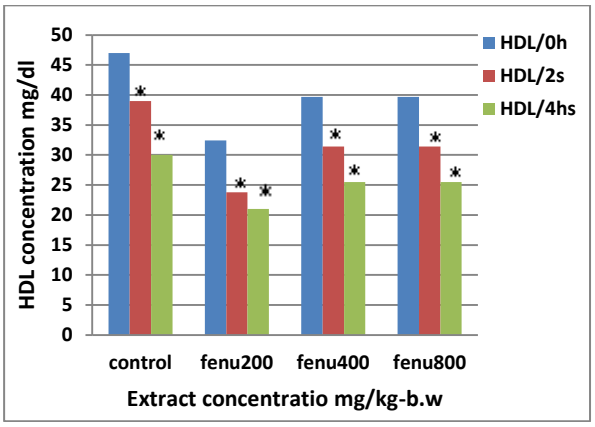

Fig.(7). Effect of administration of different doses of *Trigonella foenum-graecum* seed aqueous extract on blood HDL concentration (mg/dl) in induced-diabetic rats .
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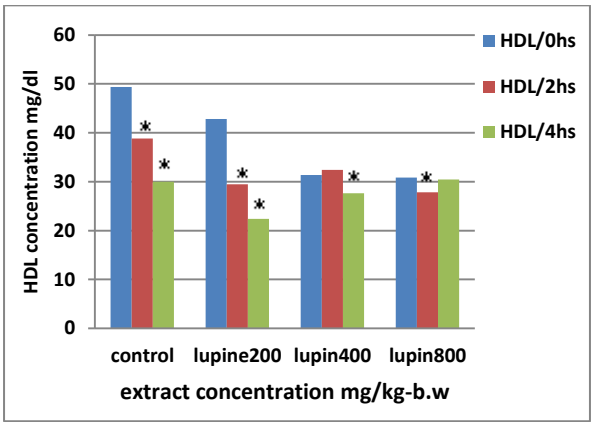

Fig.(8). Effect of administration of different doses of *Lupine termis* fruit aqueous extract on blood HDL concentration (mg/dl) in induced-diabetic rats .
***represent significant compare with Glu 0h ($p<0.05$).**

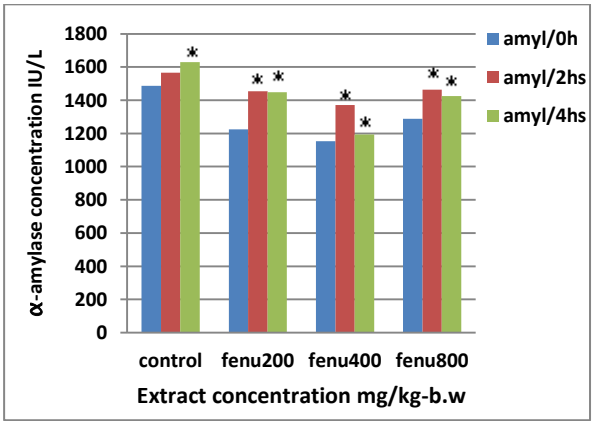

Fig.(9). Effect of administration of different doses of *Trigonella foenum-graecum* seed aqueous extract on blood α-amylase concentration (IU/dl) in induced-diabetic rats .
***represent significant compare with Glu 0h ($p<0.05$).**

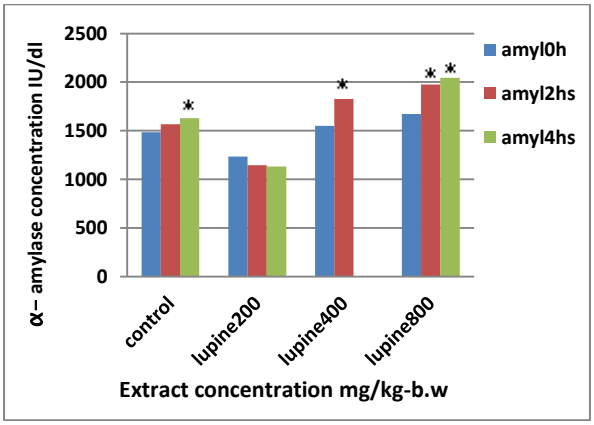

Fig.(10). Effect of administration of different doses of *Lupine termis* fruit aqueous extract on blood α-amylase concentration (IU/dl) in induced-diabetic rats .
***represent significant compare with Glu 0h ($p<0.05$).**

4 hrs. intervals in fasting rats treated with 200 mg/kg b.w. When treatment with 400 mg/kg b.w., the concentration of cholesterol, TG, and HDL registered no significant difference (P>0.05) in comparison with control. But treatment with 800 mg/kg b.w. showed significant decrease (P<0.05) in cholesterol and HDL compared with control, whereas TG registered slightly higher concentration than control. Treatment with *L. termis* (200 mg/kg b.w.) decreased the level of cholesterol, TG, and HDL in comparison to that resulted from *T. foenum* treatment and that of control (figure 4, 6, and 8). At 400 and 800 mg/kg b.w., *L. termis* extract treatment decreased the concentrations of cholesterol, TG, and HDL than control at the three time intervals (0, 2, and 4 hrs.). HDL concentration decrement was dose dependent when diabetic rats treated with *L. termis* fruit extract. Effect of *T. foenum* seed extract administration on α-amylase concentration in diabetic rats shown in figure (9) revealed significant decrease (P<0.05) at zero time in all treated groups compared with control. After 2 hrs., the concentration increased but still lower than that of control, whereas 4 hrs. later, only 400 mg/kg b.w. treatment showed significant decrease (P<0.05) in comparison with that received 200 and 800 mg/kg b.w., however last groups still significantly lower (P<0.05) than that of control. On the other hand, effect of *L. termis* administration on α-amylase concentration in diabetic rats shown in figure (10) revealed significant decrease (P<0.05) in all time periods (0, 2 and 4 hrs.) at the dose of 200 mg/kg b.w. (G5) compared with control and other treated groups (G6 and G7) which showed slight increase than that of control. Whereas the doses of 400 and 800 mg/kg b.w. showed no significant changes in zero time but caused significant increase (P<0.05) after 2 and 4 hrs. interval periods in comparison with control.

Discussion

Medicinal plants are used in a wide range in order to ameliorate the hyperglycemia by induction of insulin secretion, improvement the utilization of glucose by body cells or by reduction of carbohydrates absorption and gluconeogenesis. The current study was carried out on two Sudanese plants (*Trigonella foenum graecum* seeds and *Lupinus termis* fruits) widely used traditionally for several medical purposes specially treatment of hyperglycemia. Different concentrations (200, 400 and 800 mg/kg b.w.) of *T. foenum* seeds aqueous extract reduced blood glucose level to the normal range. This agreed with Kumar (11) who demonstrated that *in vitro* and *in vivo* studies on herbs used for diabetes, fenugreek showed potent hypoglycemic action among others. Our findings also agreed with previous studies (12) whom demonstrated that *T. foenum-graecum* seed extract has an efficiency as anti-hyperglycemic and antilipidemic agents in diabetic rats. The hypolipidemic effect of *T. foenum-graecum* seed extract has been evaluated in the present study. Our results were in agreement with Samia *et al.* (13) whom represented that Fenugreek seeds lower serum triglycerides, total cholesterol and low-density lipoprotein. When diabetic rats have been administered with three different concentration doses of *Lupine termis* (200, 400 and 800 mg/kg b.w.), a significant decrease in blood glucose has been observed. The hypoglycemic effect of *L. termis* fruit extract may be attributed to the enhancement of insulin secretion and release from pancreatic islets as has been reported in previous studies carried out by Pereira *et al.* (14) whom demonstrated that *Lupinus* aqueous extract enhances insulin release from isolated rat pancreatic islets. In the current study, when comparing the effect of the two plants on blood glucose level, lower value was observed when the diabetic rats treated with 800 mg/kg b.w. of *T. foenum-graecum* seed extract.

Hypocholesterolemic effect of *L. termis* revealed in this study is in agreement with Martins *et al.* (15,16) whom found that feeding raw peas and whole blue *lupine* seeds to pigs exerted a marked hypocholesterolemic effect. This effect has been explained in other studies by the consequence of a marked decrease in the intestinal absorption of cholesterol probably modulated by bile acid reabsorption and a higher content of dietary phytosterols. The present results also agreed with Hall *et al.* (17) Whom suggested that addition of *lupine* kernel fiber to the diet provided favorable changes to some serum lipid total cholesterol, and high-density lipoprotein. Our study showed that *T. foenum* seed aqueous extract reduced the plasma concentration of α-amylase with increasing of the dose. This agreed with Arpita (18) who identified the α-amylase inhibitory effect of *T. foenum* in his study. In present study the effect of *L. termis* on α-amylase activity had been investigated, higher inhibition of the enzyme has been observed when diabetic rats treated with 200 mg/kg b.w. of *L. termis*. The result showed that 200 mg/kg b.w. of each plant inhibit the enzyme activity in spite of activation of enzyme with higher doses. In this study, It can be concluded that *Trigonella foenum-graecum (*Fenugreek) seed and *Lupinus termis* fruit aqueous extract have hypoglycemic and hypolipidemic effects in 5% glucose- induced diabetic albino rats. *T. foenum* has higher hypoglycemic effect than *L. termis*. However, results obtained from questionnaire designed for the purpose of this study, revealed that most Sudanese diabetic people (44.60%) use *T. foenum-graecum* compared to 26.70% using *L. termis*. Further studies need to be carried out to explain the mechanism of the hypoglycemic action of these plants.

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