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Antidiabetic Properties of *Physalis alkekengi* Extract in Alloxan-Induced Diabetic Rats

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ABSTRACT

The present study investigates the antidiabetic potential of Physalis alkekengi on biochemical profile in alloxan-induced diabetic rats. The effects of an ethanolic extract of Physalis alkekengi on serum glucose, total cholesterol, triglycerides, plasma insulin and liver glycogen were examined in control and experimental groups. Physalis alkekengi extract reduced the serum glucose concentration at 24, 48 and 72 hours. To verify the activity sub-chronically, the extract administered orally in the doses of 25, 50 and 100 mg/kg to diabetic rats for 30 days, that significantly reduced the level of glucose, total cholesterol and triglycerides with an increase in insulin and glycogen concentration to near normal levels in a dose-dependent manner. The results indicate that Physalis alkekengi extract possess antidiabetic potential in alloxan-induced diabetic rats. The activity might be due to high concentration of chemical compounds specially physalin, citric acid and vit C in Physalis alkekengi.

Keywords: Antidiabetic, Physalis alkekengi.

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INTRODUCTION

Diabetes mellitus describes a metabolic disorder of multiple etiologies. It is characterized by chronic hyperglycemia, with perturbations of carbohydrate, lipid and protein metabolism that result from defects in insulin secretion, insulin action or both [1].

The use of herbal medicine has become increasingly popular worldwide and plant therapy's literature is related to that of humanity, because in most cultures man has always been depended on the curative values of medicinal herbs to cure some illnesses. Efforts are ongoing to evaluate botanical drugs for the management of diabetes mellitus [2]. Physalis alkekengi, belongs to the family Solanaceae. It is distributed in Asia (Iran, India, Japan and China) and Europe (Spain, Italy and Turkey) has a large history of herbal use, and an interesting chemistry but it is seldom used in modern practices [3].Chemical studies have demonstrated the presence of physalin, citric acid and vit C as the major components of P. alkekengi extract. Physalin is the most chemical compound with various pharmacological characteristics including, anti bacterial, anti leishmanial and anti tumor and anti-spermatogenesis and anti-conception [4-9].The whole plant is anti phlogistic, anti pyretic, anti tussiveand expectorant [10-12]. It is used in treatment of urinary and skin diseases [13]. Its extract has been used for treatment of wide range of diseases, including kidney and bladder stone, febrile diseases, inflammation, general edema, and arthritis [14].

Despite numerous preventive strategies and treatments, more than 300 million people worldwide are expected to develop diabetes by 2025 [15,16]. Hence researches are on full swing to develop effective medicines from plant sources, so Physalis alkekengi were subjected to antidiabetic screening in experimentally induced hyperglycemic rats.

MATERIALS AND METHODS

Plant material

Physalis alkekengi was collected from Guilan province, and then was identified by a botanist. Its leaves and fruits were dried under shade and powdered. The extract was prepared by the maceration method (80% ethanol in 300 g/lit for 48 hours), filtered with filter paper. After filtration ethanol was removed by rotary evaporator. The extract was dissolved in normal saline and administrated orally into rats.

Adult male albino rats of Wistar strain weighing 150 - 200 g used for the study were obtained from Razi Institute, (Karaj, Iran) and maintained according to the guidelines of Committee for the Purpose of Control and Supervision of Experiments on Animals, Razi Institute, Karaj, Iran. In the present study, thirty rats were used. Group I: contained six animals that served as control. The remaining 24 animals were given alloxan intra-peritoneally (120 mg/kg body weight) to induce hyperglycemia. Alloxan monohydrate was purchased from Sigma

Chemical Company, St Louis, MO, USA. After 72 hours, the hyperglycemic conditions in these animals were ensured from their blood glucose values which were above 250 mg/dl. Further they were segregated into four groups containing six animals each and were treated as follows.

Group II – alloxan treated (120 mg/kg i.p)
Group III - Diabetic + P. alkekengi (25 mg/kg of Body wt)
Group IV - Diabetic + P. alkekengi (50 mg/kg of Body wt)
Group V - Diabetic + P. alkekengi (100 mg/kg of Body wt)

Serum glucose concentration was measured at 24, 48 and 72 hours from the blood samples drawn by the glucose oxidase method, using One Touch Ultra 2 Glucometer (Lifescan, CA, USA).The doses was continued for 30 days and on day 31, the animals were sacrificed by cervical decapitation under mild anesthesia and the blood were collected in tubes with clot activators and heparin to get serum and plasma while the liver was removed immediately, washed with ice-cold saline and some of them stored in deep freezer at -20°C for glycogen estimation and some other fixed for tissue processing.

Plasma insulin was estimated by ELISA method using Biotech-ELX-50, (U.S.), liver glycogen using UV visible spectrophotometer total cholesterol and triglycerides were estimated using Randox-*daytona* fully automated random axis analyzer (U.K.).

After fixation, the liver was rinsed repeatedly in PBS until the yellowish coloration disappeared. Paraffin embedding was done, and 3.5 µm sections were cut on a Reichert-Jung 2050 rotary microtome (Cambridge Instruments, Germany), followed by Periodic acid-Schiff (PAS) staining. Briefly, sections were de-waxed in xylene, hydrated in descending grades of ethanol and then transferred to 0.8 % periodic for 10 min. Rinsing was done in ordinary water (10 min.), followed by exposure to Schiff reagent (30 min). After rinsing in ordinary water (20 min.), sections were stained in Carazzi's haematoxylin (15 min) and eosin Y (1 min.). Photomicrographs were taken with a Nikon digital camera DXM1200F (Nikon, Japan) coupled to a Nikon Eclipse 80i light microscope (Nikon, Japan).

Data were expressed as mean± standard deviation. Student's t-test was used to compare means. A level of $p< 0.05$ was consideredas statistically significant.

RESULTS

Table 1 shows the effect of physalis alkekengi extract on serum glucose level in hyperglycemic animals. The level of glucose in animals treated with physalis alkekengi extract (25 mg/kg) for 24, 48 and 72 hours showed a decrease in the level of glucose. In 50 and 100 mg/kg dose, the level of glucose further decreased with 24, 48 and 72 hours, the decrease was drastic in 72 hours. On observing the response with 100 mg/kg dose for 72 hours, the level of

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glucose was found near to the control value, thereby indicating the antidiabetic potential of physalis alkekengi.

Table 1: Effect of Physalis alkekengi extract on serum glucose (mg/dl) levels in hyperglycemic rats for 24, 48 and 72 hours.

Treatment (mg/kg)	24 hours	48 hours	72 hours
Group I control	112 ± 6.11	113.00 ± 7.04	114 ± 1.12
Group II Alloxan treated	612.77 ± 16.87a***	626.97 ± 13.62a***	616.43 ± 11.98a***
Group III P. alkekengi 25 mg/kg	596.00 ± 16.87bNS	411.65 ± 8.56b***	367.75 ± 13.76b***
Group IV P. alkekengi 50 mg/kg	432.16 ± 8.43b***	298.63 ± 13.32b***	198.97 ± 12.87b***
Group V P. alkekengi 100 mg/kg	412.15 ± 12.76b***	275.54 ± 8.32b***	123.83 ± 10.39b***

Values represent mean ± S.D. of six animals
*P< 0.05, **P<0.01, ***P<0.001 when compared to control animals; NS -Non-significant

Table 2 indicates the effect of p. alkekngi extract for a sub-chronic period of 30 days. The alloxan treated group (Group II) showed a significant increase in glucose, total cholesterol and Triglycerides levels, while plasma insulin and liver glycogen were reduced drastically when compared to the control animals. The animals treated with p. alkekngi extract in different doses (25, 50 and 100 mg/kg) showed dose-dependent decrease in levels of glucose, total cholesterol and triglycerides while increase in plasma insulin and liver glycogen were obtained when compared to the alloxan treated group.

DISCUSSION

The results of antidiabetic study clearly showed that Physalis alkekengi extract produced a significant hypoglycemic action. At 25 mg/kg dose, the activity of Physalis alkekengi extract in lowering the serum glucose and promoting glycogen storage was found to be higher than the standard drug. The possible mechanism for this action might be due to the inhibition of the enzyme glycogen phosphorylase, an enzyme that catalyzes the process of glycogenolysis thereby inhibiting glucagon which on feedback inhibition favours the production of insulin [17]. It is probable that reduction in serum glucose level and increase in glycogen storage performed by chemical compounds in P. alkekengi specially physalin. Our observations on the antihyperglycemic activity of P. alkekengi in diabetic rats as well as the isolation of high yield of citric acid and vit C from the P. alkekengi extract go along with these findings. Further studies

are necessary to determine the exact nature of the active principles and mechanism of action of p. alkekengi extract.

Table 2: Anti diabetic properties of Pysalis alkekengi extract treated in alloxan-induced rats for 30 days

Treatment (mg/kg)	Serum glucose (mg/dl)	Plasma insulin µU/L	Liver glycogen (mg/gm tissue)	Total cholesterol (mg/dl)	Triglycerides (mg/dl)
Group I control	114.08 ± 8.12	14.10 ± 1.75	57.87 ± 3.13	66.10 ± 4.34	68.14 ± 3.81
Group II Alloxan treated	503.53 ± 27.00a***	6.30 ± 2.45a***	10.53 ± 1.71a***	128.00 ± 9.23a***	123.30 ± 7.13a***
Group III P. alkekengi 25 mg/kg	109 ± 6.81b***	12.13 ± 2.43b**	53.17 ± 3.80b***	69.19 ± 4.65b***	84.11 ± 3.27b***
Group IV P. alkekengi 50 mg/kg	101.64 ± 5.84b***	12.00 ± 1.54b***	57.5.64 ± 4.87b***	63.13 ± 3.62b***	64.64 ± 5.12b***
Group V P. alkekengi 100 mg/kg	90.00 ± 4.09 b***	15.43 ± 3.38 b***	59.67 ± 4.62 b***	56.00 ± 5.14 b***	59.17 ± 5.32 b***

Values represent mean ± S.D. of six animals
*P< 0.05, **P<0.01, ***P<0.001 when compared to control animals; NS -Non-significant

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