

Antidiabetic and Antioxidant Property of *Wattakaka volubilis*

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ABSTRACT

Leaf samples of *Wattakaka volubilis* were used to examine their antidiabetic effect on Swiss Albino rat with different concentrations of this selection. Diabetes was induced in Swiss Albino rats by administration of alloxan monohydrate (150 mg/kg, i.p). The ethanol extract of *W.volubilis* at a dosage of 150 mg/kg of body weight was administered in individual dose per day to diabetes induced rats for a period of 14 days. The issue of ethanol extract of *W.volubilis* leaf extract on blood glucose, serum enzymes (serum glutamate pyruvate transaminases (SGPT), serum glutamate oxaloacetate transaminases (SGOT) and alkaline phosphatase (ALP)) and antioxidant enzymes (catalase (CAT) and glutathione peroxidase (GPx)) were appraised from the diabetic rats. The ethanol extract of *W. volubilis* leaf elicited significant reductions of blood glucose ($p < 0.01$), the extracts also caused a significant growth in plasma insulin and antioxidant enzymes in the diabetic rats. The survey confirmed that *W.volubilis* effective in scavenging free radicals and has potential to be powerful antioxidant ability / antidiabetic effects in alloxan induced diabetic rats.

Keywords: *Wattakaka volubilis*, antidiabetic, antioxidant property

Received 22 Feb 2014

Received in revised form 07 March 2014

Accepted 09 March 2014

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INTRODUCTION

Ethnobotanical methods provide important leads in the hunt for new rules and guides for more potent and safe drugs and cosmetics discovery. Medicinal plants have run close to key roles in the health care needs of rural and urban settlements in human and farm animal/animals. Diabetes mellitus is a metabolic disorder featured by hyperglycemia and, alterations in carbohydrate, fat and protein metabolism associated with rank or relative insufficiency of insulin secretion and/or insulin action [1]. *W. volubilis* belongs to the family *Asclepiadaceae*. The plants are distributed along subtropical of Malaysia and India, South China, Taiwan and Srilanka. The flora is important in the Indian traditional system of medicine and is utilized to treat several diseases [2]. The flora is employed particularly for antidiabetic and antiinflammatory action. It

is one of the oldest diseases affecting millions of people all over the world [3]. According to recent estimates the prevalence of diabetes mellitus is 4% worldwide and that indicates 143 million souls are regarded which will increase to 300 million by the year 2025 [4]. Although numerous oral hypoglycemic drugs exist alongside insulin, however there is no promising therapy to cure diabetes [5]. The root is applied to snake bites and given to women to cure headache after child birth. The leaves are applied to boils and abscesses promote suppuration. It is emetic diaphoretic and diuretic [6]. In the present investigation, *W. volubilis* leaves were examined for their antidiabetic efficacy. *W. volubilis* widely practiced in Indian traditional medicines and the leaf paste to treat rheumatic pain, cough, fever and wicked cold [7]. Leaf paste is removed along

with pepper to treat dyspepsia [8]. Bark paste, mixed with hot milk is utilized internally for treating urinary troubles [9], and leaf powder is taken orally along with cow's milk have antidiabetic activity [10]. The present survey was planned to look into the antidiabetic efficacy of ethanolic extract of *W.volubilis* leaf in alloxan induced diabetic rats.

MATERIALS AND METHODS

Plant material

W. volubilis leaves were freshly collected from the Jamal Mohamed College, Tiruchirappalli district in Tamil Nadu, South India. The plants were shade-dried at ambient temperature (31°C) and the dried materials were broken down into fine powder using an electric blender.

Solvent extraction

Fifty grams of dried, powdered materials (Leaves) were soaked separately in 300 ml each of the solvent *vials* (ethanol and aqueous) in a Soxhlet apparatus for 72 hrs. The extracts were evaporated under vacuum and the residues were separately dissolved in the same solvent. These extracts were concentrated to dryness in a flash evaporator under reduced pressure and controlled temperature (40-50°C).

Animals

Normal, healthy male Swiss Albino rats (180-240g) were utilized for the present investigation. Animals were put up under standard environmental conditions at a temperature (25±2 °C), illumination and dark (12:12 h). Rats were fed a standard pellet diet (Goldmohur brand, MS Hindustan lever Ltd., Mumbai, India) and urine.

Acute toxicity study

Acute oral toxicity study was done as per OECD-423 guidelines (acute toxic class method), albino rats (n=6) of either sex selected by random sampling were used for acute toxicity study[11]. The creatures were kept fasting for overnight and offered simply with water, after which the extracts were administered orally at 5mg/kg body weight by gastric intubations and observed for 14 days. If mortality was noted in two out of three animals, then the dosage administered was assigned as toxic dose. If mortality was noted in one creature, then the same dose was doubled again to

support the toxic dosage. If mortality was not mentioned, the process was repeated for higher doses such as 50, 100, and 1000 mg/kg body weight.

Elicitation of experimental diabetes

Rats were induced diabetes by the administration of simple intraperitoneal doses of alloxan monohydrate (150 milligram/kilogram) [12]. Two days after alloxan injection, rats screened for diabetes, having glycosuria and hypoglycemia with a blood glucose level of 200- 260 mg/100 ml were used up for the subject area. All creatures were granted free access to water and pellet diet and kept at room temperature in plastic cages.

Experimental design

During this investigation, a sum of 24 rats (18 diabetic surviving rats and 6 normal rats) was selected and divided into four groups of 6 rats each. Group I - Normal, untreated rats; Group II - Diabetic control rats; Group III - Diabetic rats given an ethanol extract of *Wattakaka volubilis* leaf (150 mg/kilogram of body weight) and Group IV - Diabetic rats given standard drug glibenclamide (600µg/kg of body weight).

Biochemical analysis

The beasts were sacrificed at the final stage of the experimental period of 14 days by decapitation. Blood was collected, sera separated by centrifugation at 3000g for 10 transactions. Serum glucose was measured by the O-toluidine method [13]. The serum glutamate pyruvate transaminase (SGPT) and serum glutamate oxaloacetate transaminase (SGOT) was assessed by a spectrophotometer [14]. Serum alkaline phosphatase (ALP) was assessed by the method of [15], Catalase (CAT) [16], superoxide dismutase (SOD)[17] and glutathione peroxidase (GPx) [18] were analyzed in the normal, diabetic induced and drug treated rats.

Statistical Analysis

The data were analyzed using student's t-test statistical methods to study their significant.

RESULTS

The alloxan induced diabetic rats' elicited significant rise in blood glucose from 69.30 to 333.00 mg/dl (p > 0.01). On the contrary diabetic rats treated with aqueous extract of *W. volubilis* exhibited decreased blood

glucose significantly at a dosage of 100 milligram/kilogram body weight. Serum SGOT and SGPT levels were raised significantly ($p < 0.01$) in alloxan induced diabetic rats (Group-II) when compared to normal control rats (Group-I). In alloxan diabetic rats when treated with the plant leaf extract treated rats (Group-IV). On disposal of aqueous extract of *W.volubilis* leaf to diabetic rats (Group-III), the levels of SGOT, SGPT, ALP, Catalase, Glutathione peroxidase were found to be refurbished up to normal.

Summed up the effect of alloxan on the natural process of the hepatic marker enzymes in serum. The levels of SGOT and SGPT in alloxan induced diabetic rats were raised. The aqueous extract of *W.volubilis* leaf regulated the activity of SGOT and SGPT in liver of rats intoxicated with alloxan. The restorations of SGOT and SGPT to their respective normal levels after treatment with aqueous extract of *W.volubilis*. Further strength the act as indicators of liver function and touch on normal levels of this parameter indicates normal function of livers. Serum ALP increased considerably ($p < 0.05$) in alloxan induced diabetic rats. Treatments with aqueous extract of *W.volubilis* in alloxan induced diabetic rats produced a significant ($p < 0.05$) decline in ALP level. The levels of catalase (CAT) and glutathione peroxidase (GPx) were significantly ($p < 0.05$) reduced in alloxan induced rats. These adverse changes were reversed to near normal values in aqueous extracts of *W.volubilis* leaves treated.

DISCUSSION

The present study indicates CAT and GPX in alloxan induced diabetic rats. The events

disclosed that the protective function of plant extracts in decreasing lipid peroxidation by the normalizing antioxidant system. The hypoglycaemic ethanol effect of *W.volubilis* leaf was found to be induced insulin release from pancreatic cells of diabetic rats [4] in the beginning; many plants have been examined for their hypoglycaemic and insulin release stimulatory effects [7]. Glycosylated haemoglobin determinations are self monitoring of blood glucose; therefore play an important complementary function in the direction of diabetes mellitus [8]. The effect of glibenclamide on the recovery of hepatic enzyme activity in serum was very similar to that of the earlier study [3].

In summation to the assessment of SGPT and SGOT levels during diabetes the measurement of the enzymatic activities of phosphatases such as acid phosphatases (ACP) and alkaline phosphatases (ALP) is of clinical and toxicological importance as changes in their actions are indicative of tissue damage by toxicants [12]. The lipolipidemic effect may be due to suppression of fatty acid synthesis [15]. Earlier surveys have reported that there was an increased lipid peroxidation in liver. Kidney and brain of diabetic rats [6], this could be correlated with previous study with *Cassia auriculata* flower [3] and *Scoparia dulcis* [7]. Few reports stated that the flavonoids, sterols / terpenoids, phenolic acids are recognized to be bioactivities antidiabetic principles [9]. Flavonoids are known to restore the damaged beta cells in the alloxan diabetic rats [16]. Phenolics are found to be effective antihyperglycemic agents [18].

Table 1: Effect of Aqueous leaf extract of *Wattakaka volubilis* on serum blood glucose, SGOT, SGPT, ALP, Cat and Glutathione peroxidase level of normal diabetic and extract treated rats

Groups	Blood glucose (mg/dl)	SGOT (μ l)	SGPT (μ l)	ALP (μ l)	Catalase (mM/mg Hb)	Glutathione Peroxidase (μ mol/ml)
Normal healthy control	69.30 \pm 0.78	69.37 \pm 1.9	40.57 \pm 0.70	137.8 \pm 5.56	3.23 \pm 0.01	2.67 \pm 0.128
Diabetic control	333.00 \pm 11.34	137.9 \pm 2.14	99.49 \pm 1.46	254.1 \pm 18.9	1.98 \pm 0.07	1.92 \pm 0.01
Alloxan + Plant extract	83.20 \pm 0.60	88.86 \pm 8.82	52.46 \pm 1.27	168.6 \pm 3.14	2.50 \pm 0.05	2.7 \pm 0.03
Plant extract	79.05 \pm 1.26	86.28 \pm 2.95	51.30 \pm 1.47	159.5 \pm 6.06	2.85 \pm 0.06	2.66 \pm 0.02

CONCLUSION

The result of aqueous extract of *W. volubilis* leaf extract on blood glucose, SGPT, SGOT, ALP, CAT, GPx were measured in the diabetic rats. The aqueous extract of *W. volubilis* leaf elicited significant reductions of blood glucose in the diabetic rats. The present work suggests that this plant aqueous extract can be employed as an effective antidiabetic drug.

ACKNOWLEDGEMENT

The authors thank the Biospark Biotechnological Research Center (BBRC), Tiruchirapalli, Tamil Nadu, India for antidiabetic and antioxidant studies.

REFERENCES

1. Kameswara Rao B, Renuka Sudarshan P, Raja Sekar MD, Nagaraju N and Appa Rao CH: Antidiabetic activity of Terminalia pallida fruit in alloxan induced diabetic rats. J Ethnopharmacol, 85, 2003: 169-172.
2. Koperuncholan M and Ahmed John S, Biosynthesis of Silver and Gold Nanoparticles and Antimicrobial Studies of Some Ethno medicinal Plants in South-Eastern Slope of Western Ghats, IJPI's- Journal of Pharmacognosy and Herbal Formulations, 1, 2011: 10-15.
3. Andallu B, Control of hyperglycemic and retardation of Cataract by mulberry (*Morus indica*. L) Leaves in streptozotocin diabetic rats. Indian Journal of Experimental Biology, 40, 2002: 791-795.
4. Mitra A, Bhattacharya D and Roy S: Dietary influence on type2 Diabetes (NIDDM). Journal of Human Ecology, 21, 2007: 139-147.
5. Sumana G and Suryawarshi SA: Effect of Vinca rosea extracts in treatment of alloxan diabetes in male albino rats. Indian Journal of Experimental Biology, 39, 2001: 748-758.
6. Rajadurai M, Vidhya VG, Ramya M and Bhaskar A : Ethno-Medicinal plants used by the Traditional Healers of Pacchamalai Hills, Tamil Nadu, India. Ethnomedicine, 3, 2009: 39-41.
7. Muthu C, Ayyanar M, Raja N and Ignacimuthu S : Medicinal plants used by traditional healers in Kancheepuram District of Tamil Nadu, India. Journal of Ethnobiology and ethnomedicine, 2, 2006: 43-52 doi: 10.1186/1746-4269-2-43.
8. Pandikumar P, Ayyanar M and Ignacimuthu S: Medicinal plants used by Malasar tribes of Coimbatore district, Tamil Nadu. Indian Journal of Traditional Knowledge, 6, 2007: 579-582.
9. Silija VP, Samitha Varma K and Mohanan KV: Ethnomedicinal plant knowledge of the Mullukuruma tribe of Wayanad district, Kerala, Indian Journal of Traditional Knowledge, 7, 2008: 604-612.
10. Ayyanar M, Sankara Sivaraman K and Ignacimuthu S: Traditional Herbal Medicines used for the treatment of Diabetes among two major tribal groups in South Tamil Nadu, India. Ethno Botanical Leaflet, 47, 2008: 389-394.
11. OECD, (Organisation for Economic co-operation and Development). OECD guidelines for the testing of chemicals/Section 4: Health Effects Test No. 423; acute oral Toxicity- Acute Toxic Class method. OECD. Paris. 2002.
12. Nagappa AN, Thakurdesai PA, Venkat Rao N and Sing J: Antidiabetic activity of Terminalia catappa Linn. Fruits. Journal of Ethnopharmacology, 88, 2003: 45-50.
13. Sasaki T, Mastu S and Sonae A: Effect of acetic acid concentration on the colour reaction in the O-toluidine boric acid method for blood glucose estimation. Rinsho Kagaku, 1, 1972: 346-353.
14. King E.J and Armstrong A.R. A convenient method for determining serum and bile phosphatase activity. Can. Med. Assn. J, 31 (4), 1934: 376 - 381.
15. Reitman S and Frankel SA: Colorimetric method for the determination of serum glutamic oxaloacetic and glutamic pyruvic transaminases. American Journal of Clinical Pathology, 28, 1957: 56-63.
16. Bergmayer HU: UV method of catalase assay. In Methods of Enzymatic Analysis, Weidheim Deer field Beach, Florida, Bansal, 3, 1983: 273.
17. Madesh M and Balasubramanian KA: Microtitre plate assay for superoxide dismutase using MTT reduction by superoxide. Indian Journal of Biochemistry and Biophysics, 35, 1998: 184-188.
18. Pagila DE, Valentine WN: Studies on the quantitative and qualitative characterization of erythrocyte glutathione peroxidase. Journal of Laboratory and Clinical medicine, 70, 1967: 158-169.