

ORIGINAL RESEARCH ARTICLE

Preliminary Phytochemical Screening and Blood Glucose Lowering Activity of Methanol Extract of *Salvia tilifolia* Vahl aerial part**A. K.GEBRU¹, G.G. SIBHAT*¹, M.G. HIBEN¹**¹Department of Pharmacy, Pharmacognosy Course and Research Unit, College of Health Sciences, Mekelle University, Mekelle, Ethiopia; P.O.Box: 1871

Received 21 Jun 2014; Revised 04 Oct 2014; Accepted 15 Oct 2014

ABSTRACT

Diabetes mellitus is one of the most common chronic diseases nearly in all countries and the burden continue to increase especially in developing countries. Medicinal plants are being used as alternative medicine for diabetic patients especially in developing countries where cost of conventional drugs is a burden for the population. *Salvia tilifolia* Vahl is traditionally used to relief various disorders in north central Ethiopia. The present study was focused on phytochemical screening and *in vivo* hypoglycemic activity of *Salvia tilifolia* Vahl. Preliminary phytochemical screening was done using common chemical test procedures; and acute toxicity study was done as per Organization for Economic Co-operation and Development (OECD 425) guidelines. The experimental animals were divided into 3 groups comprising of 5 animals in each group: G-I were treated with *S.tilifolia* at 400mg/kg, G-II received standard drug Glibenclamide at 5mg/kg, and G-III were negative control and given normal saline. Results of preliminary phytochemical screening indicated that alkaloid, flavonoids, saponins, phytosterols and carbohydrates were detected in the methanolic extract of *S. tilifolia* aerial part. There were no signs of adverse toxicity and mortality in mice within the first 24hrs as well as during 14 days after oral administration of the test substance up to a dose of 2 g/kg. Reduction in blood glucose level was observed in the mice that received 400 mg/kg extract when compared to those that received normal saline (negative control). Thus, it can be concluded that secondary metabolites such as alkaloids, flavonoids, saponins, and phytosterols may be present in *S. tilifolia* aerial parts and the plant could serve as potential herbal hypoglycemic agent with good safety profile.

Key words: Blood glucose lowering, phytochemical screening, *Salvia tilifolia*.**INTRODUCTION**

Diabetes mellitus is a group of metabolic diseases characterized by hyperglycemia resulting from defects in insulin secretion, insulin action, or both.

^[1] The world prevalence of diabetes among adults (aged 20-79 years) was estimated to be about 6.4%, affecting 285 million adults, in 2010, and it is expected that the prevalence will increase to 7.7% and 439 million adults by 2030. Between 2010 and 2030, there will be a 69% increase in diabetes prevalence among adults in developing countries and around 20% in developed countries.

^[2] Despite the introduction of hypoglycemic agents from natural and synthetic sources, diabetes and its secondary complications continue to be a major health problem. ^[3] Medicinal plants have always been a very good source of drugs and

many of the currently available drugs have been derived directly or indirectly from them and are used for treatment of diabetes especially in developing countries where the cost of conventional medicines is a burden to the population. ^[4] *Salvia* is the largest genus of plants in the family Lamiaceae, with approximately 1000 species of shrubs, herbaceous perennials, and annuals with high diversity in growth forms, secondary compounds, floral morphology and pollination biology. ^[5] Many plants from the genus *salvia* such as *Salvia lavandulifolia* ^[6], *Salvia officinalis* ^[7], *Salvia fruticosa* Mill ^[8], *Salvia splendens* ^[9] have been shown to demonstrate hypoglycemic activity. *Salvia tillifolia* was introduced to Ethiopia in the 1980's. Since then

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the plant has gradually established itself and at present it seems to be spreading vigorously, replacing native herbs in some sites^[10] and traditionally it is used to relief headache, stomach trouble and as memory enhancing herb. In the present study, preliminary phytochemical screening and *in vivo* hypoglycemic activity of methanolic aerial extract of *Salvia tiliifolia* were carried out.

MATERIALS AND METHODS

Plant Material:

The aerial parts of *Salvia tiliifolia* Vahl were collected from and around the town of Harar in the Harari People Regional state, eastern Ethiopia in September 2013. The plant was authenticated by Ato Melaku Wondafrash at Addis Ababa University National Herbarium, Biology Department.

Extraction:

Aerial parts of *Salvia tiliifolia* were air-dried at room temperature and grinded using commercial Hemisphere Control blender machine (Breville BBL605XL, UK). The powdered plant material (500 g) was extracted exhaustively at room temperature by maceration using 80% methanol for 72 h with occasional shaking and extraction was repeated two times. The extracts were filtered, combined and methanol was recovered using Rota vapor (BIBBY Steriling, UK) at a temperature not exceeding 40°C and the remaining aqueous part was dried by hot oven.

Experimental animals:

Swiss albino mice of both sexes weighing between 30-40g were obtained from the animal house of pharmacy department, Mekelle University and acclimatize for one week at room temperature in hygienic condition under natural light and dark schedule and were fed on standard laboratory diet and water before commencement of the activity. Ethical approval was obtained from Mekelle University, college of health sciences research ethics review committee.

Preliminary phytochemical screening test:

Preliminary phytochemical screening was performed using common chemical test procedures as described by Debela, (2002).^[11]

Qualitative chemical test was made for alkaloids, flavonoids, coumarins, anthraquinone derivatives, tannins and phenolic compounds.

Acute toxicity test:

Acute toxicity study was carried out on the 80% methanolic aerial extracts (2000mg/kg) *Salvia tiliifolia* according to Organization for Economic Co-operation and Development (OECD425) guidelines for the testing of chemicals.^[12]

Preparation of diabetic animal:

The Study animals were made diabetic by a single administration of intra-peritoneal (i.p.) injection of alloxan monohydrate (150mg/kg) and then kept for an hour with free access to 20 % of glucose solution. After 72 hrs fasting, blood glucose was determined using and mice with blood glucose above 120 mg/dl were taken for the experiment.

Experimental design and animal grouping:

The experimental animals were divided into 3 groups comprising of 5 animals in each group where Group I was treated with methanolic aerial extract of *S.tiliifolia* at 400mg/kg , Group II with standard drug Glibenclamide (5mg/kg) (positive control) and Group III considered as normal control treated with distilled water (negative Control).

Data analysis:

Data obtained were expressed as mean plus standard error of mean (M± SEM) and results were analyzed using SPSS version 16 windows software using one-way analysis of variance (ANOVA) and Tukey's t-test at a 95% confidence interval ($\alpha = 0.05$) to compare results between groups. The results were considered significant when $P < 0.05$.

RESULTS AND DISCUSSIONS

Phytochemical Screening:

As can be seen in (Table 1), results from preliminary phytochemical screening indicated the possible presence of alkaloids, flavonoids, saponins, phytosterols, and carbohydrates; whereas anthraquinone derivatives, cardiac glycosides, coumarins, polyphenols and tannins where not detected.

Table 1: Preliminary phytochemical screening analysis on the methanolic aerial extract of *Salvia tiliifolia*

| Phytochemicals | Result |
|---------------------------|--------|
| Alkaloids | + |
| Antraquinones | - |
| O-antraquinone glycosides | - |
| Cardic glycosides | - |
| Polyphenols | - |
| Flavonoids | + |
| Coumarins | - |
| Saponins | + |
| Tannins | - |
| Phytosterols | + |
| Carbohydrates | + |

Key: + Present, -Absence

The detection of secondary metabolites such as alkaloids, flavonoids, Saponins, phytosterols and carbohydrates in the methanolic aerial extract of *Salvia tilifolia* was in agreement with some

literatures that reported *salvia* species contains mainly terpenoids, and flavonoids and in some extent steroids^[13], alkaloids^[14], and phytosterols.^[15]

Table 2: Blod glucose lowering activity of methanolic aerial extracts *Salvia tilifolia* in mice

| Extract/drug | Dose (mg/kg) | Blood glucose level in (mg/dl) | | | | | | |
|--------------------|--------------|--------------------------------|--------------|--------|-------------|--------|-------------|--------|
| | | 0 h | 2 h | Change | 4 h | Change | 6 h | Change |
| <i>S.tilifolia</i> | 400 | 201±56 | 146 ± 31.03 | 55 | 154.2 ±27.5 | 46.8 | 171.2 ±47.6 | 29.8 |
| Glibenclimed | 5 | 350.2 ± 9 | 271.8 ± 84.0 | 78.4 | 314.2 ± 105 | 36 | 286.8 ±86.2 | 63.4 |
| Vehicle | 0.5ml | 99.2 ±11.3 | 101.2 ±7.9 | -2 | 99 ±13.3 | 0.2 | 99 ± 12.4 | 0.2 |

Values are presented as M ± SEM; n = 5

Acute toxicity test:

No major signs of toxicity such as wieght loss, lacrimation, loss of appetite, comma and death were observed after adiministration of methanolic extract of the plant up to dose of 2g/kg within the first 24 hours and during 14 days of follow up.

Blood Glucose Lowering Activity of methanolic aerial extract of *S.tilifolia* on the blood sugar level of glycemic mice is presented in table2. As can be seen from table 2, oral administration of aerial extract of *Salvia tilifolia* (400mg/kg) showed significantly lower (P <0.001) fasting blood glucose at 2, 4 and 6hrs compared to initial blood glucose level. More over blood glucose lowering effect was significantly higher than normal saline treated diabetic mice which are the negative control group.

The reduction in fasting blood glucose observed in the study animals treated with plant extract could be due to the presence of flavonoids in the plant extract as flavonoids are known to have antioxidant effect^[16] and antioxidants in turn are known to contribute in preventing and therapy of diabetes mellitus.^[17] Another bioactive antidiabetic plant metablites detected in the plant extract were saponins, alkaloids and phytosterols which could be responsible for blood glucose reduction.^[18, 19]

Based on the present finding aerial extract of *Salvia tilifolia* contains alkaloid, flavonoids, saponins, phytosterols and carbohydrates and possesses blood glucose lowering activity with favorable safety profile.

ACKNOWLEDGEMENTS

The authors want to acknowledge Mekelle University, College of Health Science for providing laboratory facility, Mr.Biruk Sintayehu for collecting the plant material and facilitation of its authentication, Abadi Aynalem (Laboratory

technician) for his technical assistance.

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