Full Length Research Paper

Preliminary phytochemical screening and ethnomedicinal uses of *Teucrium stocksianum* from Malakand Division

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The extracts of *Teucrium stocksianum* were screened for secondary metabolites by using methanol, chloroform and n-hexane solvents. The methanolic extracts gave positive results for alkaloids, tannins, flavonoids, saponins, steroid, reducing sugar, terpenoid, anthraquinone, phlobatannin and glycoside. In the case of chloroform, alkaloids, tannins, reducing sugar, saponins, flavonoids, terpenoid were present and anthraquinone, glycoside, phlobatannin, steroid were found to be absent, while n- hexane extracts contained tannins, reducing sugar, flavonoids and failed to detect alkaloids, anthraquinone, glycoside, phlobatannin and terpenoid. This paper highlights the significance of plant in traditional medicine and distribution of various chemical constituents with respect to Malakand Division, Pakistan.

Key words: Teucrium stocksianum, secondary metabolites, traditional medicine, phytochemical constituents.

INTRODUCTION

For the existence of life on the earth, plants played very importance role from the time immemorial. Human being is directly relayed on plants to fulfill their daily life requirement. They are used as food, fuel, ornamentals, flavor and medicine. Throughout the world higher plants are being employed to treat various infectious diseases. They provide number of natural products including medicaments to fight against diseases.

To maintain quality of drug, there is need to evaluate active constituents of raw material to ascertain their therapeutic effects. It enables the pharmacist to prescribe numerical values and ensure uniformity of standards (Amen, 1996). Medical plant can be defined as any part of the plant which contains substances that can be used for therapeutic purposes and its precursor for the synthesis of useful drugs. They contain nutrients and phytochemicals that can heal ailments of the body (Trease and Evans, 1978). Plant material that has cellular structure is referred as organized drug in pharmacy, whereas, non-cellular structure as unorganized or a cellular drug (Akinpelu and Onakoya, 2006).

Medicinal and aromatic plants (MAP) are a large group of economically important plants that provide raw materials for pharmaceuticals, perfumery, flavor and cosmetic industries. Teucrium stocksianum Boiss. (Lamiaceae) locally known as Speer botay is a perennial, woody, aromatic herb which is native to the mountainous regions of the United Arab Emirates (UAE), northern Oman (Western 1989; Nadaf et al., 2003), Pakistan (Ahmad et al., 2002) and Iran (Mojab et al., 2003). This plant attains a height of 10 to 25 cm having densely packed stem and grey-green leaves. The leaves and young shoots of T. stocksianum are commonly used for the preparation of traditional medicines to treat several ailments including diabetes, gastro-intestinal ailments and inflammatory conditions (Radhakrishnan et al., 2001). In addition to the gastro protective effect, decoction of T. stocksianum has been used for the treatment of diabetes and burning feet syndrome (Barkatullah and Hussain, 2009).

Secondary metabolites are chemicals produced

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through secondary reactions resulting into primary carbohydrates, amino acids and lipids (Ting, 1982). Their direct function in plant metabolism is not well recognized till date. However, their role in ecosystems (Dey and Harborne, 1997), particularly in plant herbivore interaction (Feeny, 1976; Swain, 1979) and chemotaxonomy (Gibbs, 1974) has been well known. Plants contain secondary metabolites such as alkaloids, saponins and tannins are generally avoided by grazing animals and leaf feeding insects. Their presence in plants and intake at high level reduces the nutrient utilization, feed efficiency, animal productivity and in some cases may cause death (Makkar and Goodchild, 1996).

The extract of *T. stocksianum* has been found to have anti-ulcerogenic and cytoprotective properties when applied on experimentally-induced gastric lesions (Islam et al., 2002). In addition, the plant extract has also been analyzed for analgesic and anti-inflammatory activities, which supports the traditional use of the plant in the treatment of painful inflammatory conditions (Radhakrishnan et al., 2001). This plant is also used as a blood purifier, a remedy for the treatment of hypertension and epilepsy (Ahmad et al., 2002) and sore throat (Iqbal and Hamayun, 2004).

The people of Malakand division have good knowledge about this plant for its medicinal use. Based on this indigenous knowledge of *T. stocksianum*, present study was carried out to analyze phytochemicals in order to seek scientific logic for their use in folk medicine.

MATERIALS AND METHODS

Collection and identification of plant materials

The whole plant (leaves, stem, root, and floral head) of *T. stocksianum* was collected in the month of May to June 2011 from the different areas of Talash (Dir lower), Malakand Division, Khyber Pakhtun Khwa (KPK), Pakistan. One set of specimens (No. 2214) was made using conventional method of pressing, drying and mounting on standard herbarium sheet. The plant was described and identified by Dr. Rahmatullah Qureshi in Taxonomy Lab. by employing Flora of Pakistan (Hedge, 1990). The identified specimen was deposited in the department of Botany, Pir Mehr Ali Shah Arid Agriculture University, Rawalpindi for further record.

Preparation of the extracts

The plant material was washed thoroughly 2 to 3 times with running water and once with sterile distilled water immediately after collection to remove dirt prior to the drying process and shade dried. These were then finely powdered (80 mesh) using a laboratory grinding mill and stored in refrigerator. The powdered material was soaked in methanol, chloroform and n-hexane (1:10) by shaken for 24 h at 37°C. The extracts were then filtered using Whatman filter paper No. 1. The filtrates of methanol, chloroform and n-hexane were combined and concentrated under reduced pressure at 40°C using rotary evaporator. Blackish extra cts of methanol, chloroform and n-hexane were obtained for phytochemical screening.

Phytochemical screening

Chemical tests were carried out in methanol, chloroform and nhexane extracts of *T. stocksianum* using standard procedures to identify the constituents as described by Sofowora (1993), Trease and Evans (1978) and Harborne (1973).

Test for alkaloids

About 0.2 g of the extracts was warmed with 2% H₂SO₄ for two minutes. The filtrate was added with few drops of Dragendorff's reagent (solution of potassium bismuth iodide). Presence of orange red precipitate indicated as positiveness for alkaloids.

Test for tannins

Little amount of extract was mixed with water and heated on water bath. The mixture was filtered and ferric chloride was added to the filtrate. The dark green solution indicated the presence of tannins.

Test for anthraquinones

About 0.5 g extract was boiled with 10% HCl for few minutes in water bath. The filtrate was allowed to cool and equal volume of chloroform was added.

It was added with few drops of 10% NH and the mixture was heated. The appearance of rose-pink color indicated the presence of anthraquinone.

Test for glycosides

The extract was hydrolyzed adding with HCI and neutralized with NaOH solution. Afterward few drops of Fehling's solution A and B were added. The occurrence of red precipitate showed the presence of glycosides.

Test for reducing sugars

The extract was shaken with distilled water, filtered and boiled with drops of Fehling's solution A and B for minutes. Showing of an orange red precipitate indicated the occurrence of reducing sugars.

Test for saponins

About 0.2 g of the extract was shaken with 5 ml of distilled water and then heated to boil. Frothing (appearance of creamy miss of small bubbles) shows the presence of saponins.

Test for flavonoids

The 0.2 g extract was dissolved in diluted NaOH and HCI was added. A yellow solution that turned colorless within few minutes indicated the presence of flavonoids.

Test for phlobatanins

The 0.5 g extract was dissolved in distilled water and filtered. It was boiled with 2% HCl solution. The appearance of red precipitation revealed the presence of phlobatanins.

Phytochemicals	Solvent		
	Chloroform	Methanol	n-hexane
Alkaloids	+ve	+ve	-ve
Tannin	+ve	+ve	+ve
Saponins	+ve	+ve	+ve
Anthraquinone	-ve	+ve	-ve
Steroid	-ve	+ve	-ve
Phlobatannin	-ve	+ve	-ve
Terpenoid	+ve	+ve	-ve
Flavonoids	+ve	+ve	+ve
Glycoside	-ve	+ve	-ve
Reducing sugar	+ve	+ve	+ve

Table 1. Phytochemical screening of T. stocksianum using different solvents.

Test for steroids

2 ml acetic anhydride was added to 0.5 g plant extract along with 2 ml of $H_2SO_4.$ The change of color from violet to blue or green indicated the presence of steroids.

Test for terpenoids (Salkowski test)

The 0.2 g plant extract was mixed with 2 ml chloroform and 3 ml concentrated H_2SO_4 . The formation of reddish brown color to the interface was formed that indicated positive sign for terpenoids.

Ethnobotanical use

Indigenous communities of Malakand Division were interviewed to get important recipes and anecdotal uses of *T. stocksianum* following the methodology of Qureshi and Bhatti (2008).

RESULTS

The results of phytochemical screening of *T. stocksianum* by using methanol, chloroform and n-hexane solvents are summarized in Table 1. The methanolic extracts exhibited positive for the presence of all screened phytochemicals such as alkaloids, tannins, flavonoids, saponins, steroid, reducing sugar, terpenoid, anthraquinone, phlobatannin and glycoside. The chloroform extracts detected alkaloids, tannins, reducing sugar, saponins, flavonoids, terpenoid except anthraquinone, glycoside, phlobatannin, steroid. Similarly, n- hexane extracts traced tannins, reducing sugar, flavonoids and failed to detect alkaloids, anthraquinone, glycoside, phlobatannin and terpenoid.

Ethnobotanical use

The people of Malakand Division have good knowledge about this plant for its medicinal use. Ethnobotanically, the juice of T. stocksianum is given for the treatment of

jaundice and as blood purifier as well as cooling agent. The decoction of plant is also prescribed to treat chronic fever. Leaves soaked in water overnight and the juice is given before breakfast to diarrhea and abdominal pain. Young leaves are boiled in water and obtained juice is used for curing cough.

DISCUSSION

The present research work reveals that there are various important phytochemicals such as alkaloids, tannin, saponins, anthraquinone, steroid, phlobatannin, terpenoid, flavonoids, glycoside and reducing sugar were present in *T. stocksianum* (Table 1). Besides, tannin, saponins, flavonoids and reducing sugar were uniformly detected by all solvents (Figure 1). The chemical constituents extracted by various solvents were in the order of methanol> chloroform> n-hexane.

It is an established fact that secondary metabolites present in plants derive therapeutic activities (Rabe, 2000). Particularly, alkaloids are reported the most important therapeutic significant substance (Njoku and Akumefula, 2007). The present study stratified that alkaloids were detected by methanol and chloroform solvents that may signify for further investigation from these solvent base extracts. Tannins and flavonoids are also reported to possess biological activities which are used for the prevention and management of many diseases (James et al., 2007). Our investigation from T. stocksianum reveals that tannins and flavonoids are traced by all solvents and link logically with ethnomedicinal use of this plant in traditional system of medicine. The medicinal properties of the plant could be attributed to the presence of one or more of the detected plant natural products. These findings give credibility to the traditional medicinal relevance of the plant as remedies for abdominal pain, chronic fever, diarrhea, blood purification, jaundice and cough. However, the pharmacological actions of the plant cannot be

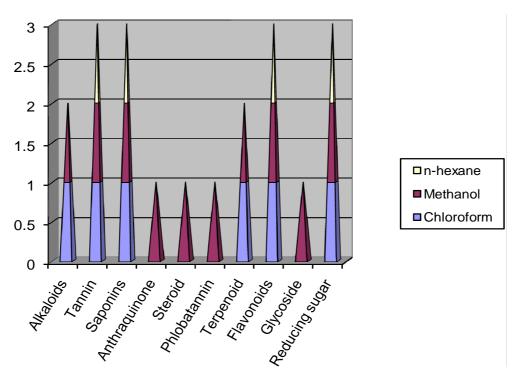


Figure 1. Phytochemical analysis of *Teucrium stocksianum* using various solvents.

ascertained by the result of the phytochemical analysis only. Results of this investigation offer a scientific foundation for carrying out *in vitro* and *in vivo* activities. In conclusion, isolation and purification of the phytochemical followed by a detailed study might result in identification lead compound for curing various diseases.

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