Research Article

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EVALUATION OF MAJOR PHYTOCHEMICALS IN THE LEAVES AND FRUITS OF *SOLANUM MAURITIANUM* SCOP.: A POTENTIAL HERBAL DRUG

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ABSTRACT

Phytochemicals isolation and identification is utmost important to know the active principles of medicinal plants. *Solanum* L., is used in folk and traditional medicine to cure many disorders. Bug weed (*Solanum mauritianum* Scop.) is evergreen woody shrub of South America. Local people use this exotic species for curing many ailments. A preliminary evaluation was attempted to know the major phytochemicals in this species to validate its usage. The leaves and fruits were screened for quantification of major phytochemicals such as alkaloids, flavonoids, saponins, tannins and phenolic compounds. Significant levels of alkaloid (0.59–0.34 mg/g), saponin (4.3–6.4 mg/g), phenolic content (7.2–11.7 mg/g), tannin (4.59–7.4 mg/g) has been detected in the leaves and fruits of this plant. It is fascinating to note that the polyphenols and alkaloids displayed remarkable levels. Thus, the present study reveals medicinal potentiality of the plant. These phytochemicals possess various bioactive properties and may be used as external therapeutic supplement. Further studies are warranted to isolate, fractionate the alkaloids from the leaves and to elucidate its structure.

Key words: *Solanum*, quantitative estimation, therapeutic value, alkaloid, polyphenols.

INTRODUCTION

Plant-based secondary metabolites are potential as defense molecules or as agents of pollination. Secondary metabolites are generally utilized in food, pharmaceutical, chemical, cosmetic industries and also in agriculture. Herbal natural products are abundant in nature and many of them exhibit unique biological activities and some can be used as food additives. Synthetic antioxidants used in the food industry show many health-related issues and most developed countries are tending to give preferences to natural sources. Therefore, investigation of natural molecules has been a changing trend in pharmaceutical research. Antioxidants mitigate oxidative deterioration of food and indirectly scavenging free radicals from it. Butylated hydroxyanisole, butylated hydroxytoluene, propyl gallate and tertiary butylhydroquinone are synthetic antioxidants used in foods to prevent oxidative degradation. No single phytocomponent is responsible for its medicinal properties of many plant-based drugs, and usually their synergic action enhances its efficacy. Therefore, the determination of the total amount of diverse groups of components is essential for the standardization of the plants. Polyphenols, saponins and alkaloids are proven phytochemicals in many plants and display with characteristic toxicity and pharmacological activity. These features are traditionally being exploited by humans for hunting gathering stage in executing drugs.

The genus *Solanum* L., belongs to Solanaceae and is considered as one of the largest genus among the angiosperms with great potential for food security in the developing world. It comprises of about 1500 species and well represented all over the globe. It is rich in alkaloids which are distributed in all parts of the plants. The active principles such as solinidine and other steroids extracted from the roots and leaves of some species are of potent and effective pharmaceuticals. Majority of the *Solanum* species are widely used in folk medicine. The presence of the steroidal alkaloid solasodine, a precursor for the synthesis of steroid hormones and is characteristic active principle of *Solanum* species. This has tremendous impact on utilization of this genus economically and medicinally all over the world. The lack of immediate known use for certain members of this group has lead to their neglect and subsequent genetic erosion. Depletion of such potentially useful plant resources should be taken care of by paying attention to germ plasm exploration and conservation studies. The present study was focused on phytochemical analysis of *Solanum mauritianum* Scop. used locally in traditional and folk medicine to cure many disorders.

MATERIALS AND METHODS

Plant material

Fresh leaves and ripened fruits of *Solanum mauritianum* Scop. were collected from Munnar hills of Idukki district, Kerala. The plant was identified, authenticated by referring manuals and confirmed by referring the herbarium of Jawaharlal Nehru Tropical Botanical Garden and Research Institute (JNTBGRI). The herbarium specimen was deposited in the herbarium of University College, Trivandrum (UCT 1279). The plant was collected during September- October.

Experimental

The samples were shade dried and made in to fine powder for quantitative analysis.

Quantification of total alkaloids

Alkaloids estimation

Crude alkaloid content was estimated using Dragendorff's reagent following the protocol of Luis et al., 2012[1]. Briefly, 10 ml of each crude ethanolic extract was centrifuged over 10 min (3000 rpm) to remove residual suspended particles. 5 ml of the supernatant were mixed with 1 ml of 0.1 N HCl. Add 2.5 ml of

Dragendorff's reagent to the previous mixture for precipitation and the precipitate was centrifuged for 5 min (3000 rpm). Subsequently, the precipitate was washed with 2.5 ml ethanol. Discard the filtrate and the pellet was treated with 2.5 ml of disodium sulfide solution (1% w/v). The resulted brownish black precipitate was then centrifuged for 5 min (3000 rpm). This residue was redissolved in 2 ml of conc. nitric acid, with slight warming. This solution was diluted with distilled water to 10 ml in a standard flask. Then, pipette out 1 ml and mixed with 5 ml of thiourea solution (3% w/v). Absorbance of this solution was read at 435 nm against blank containing 1 ml of conc. nitric acid + 2.5 ml of thiourea solution (3% w/v). The standard curve was prepared using pilocarpine nitrate (750, 500, 400, 250, 200, 150 and 100 mg/ml solutions) in HCl 0.1 N (y = 0.0012x-0.1044; R2 = 0.9851). Total crude alkaloid contents were expressed as pilocarpine nitrate equivalents (mg PNE/g of dry mass).

Estimation of total flavonoids

Total flavonoid level of leaves and fruits was determined using the aluminium chloride method. 0.5 ml of extracts was taken in different test tubes and adds 2ml of distilled water + 0.15 ml of sodium nitrite (5% w/v) and allowed to stand for 6 min. Later, add aluminium trichloride (10%, 0.15 ml) and incubate for 6 min, followed by the addition of 2 ml of sodium hydroxide (4% w/v). The volume was made up to 5ml with distilled water. After 15 min of incubation the reaction mixture turns into pink and read the absorbance at 510 nm. Distilled water was used as blank. Flavonoid level was expressed in mg of catechin equivalents (CE) per g of extract[2].

Determination of total Phenols

100 mg of the sample was extracted with 5 ml of 80% ethanol and centrifuged at 2000 rpm. 1 ml of folin-ciocalteu reagent was added to 0.5 ml of the extract of the sample. 2 ml of 20% sodium carbonate was added and heated for 1 min. After cooling, the solution was made up to 10 ml with distilled water. The blank contains all the reagents except the sample. The OD was read at 650 nm wavelength[3].

Estimation of saponin

10 g of powdered leaf and fruit samples in 100 ml of dilute ethanol (20%) were taken in two different 250 ml conical flask. The ethanolic mixture was heated at 60°C for 6 h with gentle stirring in a hot water bath. Subsequently, the residue was filtrated by a Whatman filter paper No. 1. The filtrate was kept in new container. Further, the pellet was subjected to the same protocol for elucidating the maximum yield. The resulted fractions were mixed and heated till the volume of the residue was reduced to 20 - 25%. The concentrated crude compound was shaken with 20 ml of diethyl ether in a separating funnel. Separate the aqueous layer and the process was repeated twice. Lastly, mix the residue with n-butanol and washed thrice with 10 ml 5% NaCl solution. The semisolid was dried in an oven and the saponin level was estimated[4].

Determination of Tannins

500 mg powdered sample was transferred to 250 ml conical flask containing 75 ml of distilled water and boiled for 30 min. The residue was centrifuged for 2000 rpm for 20 min. The supernatant was collected in 100 ml volumetric flask and made up to a known volume. 1 ml of the sample extract was transferred to a 100 ml volumetric flask containing 75 ml of distilled water. To this, add 5 ml of Folin-Denis reagent and 10 ml of sodium carbonate solution and diluted to 100 ml. It was shaken thoroughly and left for 30 min. The absorbance was read at 700 nm against blank as water[5].

Statistical analysis

The results were recorded after repeating the experiments six times. The experimental results were expressed as mean ± standard deviation of (6n) measurements. The statistical analysis of the data was carried out using student's t-test and the results were considered significant when $p<0.05$.

RESULTS AND DISCUSSION

Therapeutic use of herbals dates back to the BC and is still utilized to cure many ailments in most part of the world. Developed countries referred this as Complementary and Alternative Medicine. Most of the local ethnic societies inhabitated in the village prone areas depends basically on plant based crude extracts. The efficacy of these extracts is based on the pool of secondary metabolites which act synergistically to cure the disease. Different plant parts contain varying amounts of phytochemicals such as roots, leaves, fruits or seeds. The principle molecule often may cause toxicity in the cells. In this juncture phytochemicals in *S. mauritianum* was analyzed to validate its use as drug by the local people. Leaves and mature fruits were analyzed.

The fruits of *S. mauritianum* contained high level of flavonoids (5.6 mg/g) followed by leaves (4.2 mg/g). Meanwhile, the saponin content was high in the leaves (6.4 mg/g) followed by the leaves (4.3 mg/g). Total phenol content was registered high in the leaves (11.7 mg/g) followed by fruits (7.2 mg/g). Remarkable percentage of alkaloid has been detected in the leaves of the species (0.59 g ± 0.08) compared to fruits (0.34 g ± 0.04). Tannin also showed optimal level i.e., 4.89 to 7.4 mg/g in the leaves and fruits (Table 1).

Alkaloids are class of nitrogen containing natural compound. Diverse 12,000 alkaloids are reported in about 20% of plant species and limited of them are exploited commercially as medicinal such as vinblastine and vincristine (anti-tumor agents), reserpine (anti-hypertensive) and quinine (anti-malarial agent). The bioactive phytochemicals of *S. mauritianum* may play vital role in developing anti-tumor drugs in human being.

Saponins are high molecular weight compound. Sugar molecule present in the saponin combined with triterpene or steroid glycone to form glycosides which has cholesterol binding property. Similarly, it displays antimicrobial properties[6,7]. Therapeutically, they are shown to have hypo lipidemic and anti-cancer activity i.e., they react with cholesterol rich plasma membrane of various cancer cells and inhibits their proliferation[8]. High level of saponin was seen in the leaves of *S. mauritianum*.

Flavonoids are established botanicals with proven records in protecting cells against free radicals and reactive oxygen species, the products of stress in living cells. This may indirectly a boon to many degenerative diseases including myo-cardiac attack and cancer[9]. Further, by inhibiting the estrogen producing enzyme flavonoid blocks the cancer proliferation. The range of total flavonoid in the leaves and fruits were 4.2–5.6 mg/g.

Remarkable amount of total phenolic compound further supports its natural antioxidant potentiality and may be used as nutraceuticals. Polyphenols has ability to combat cancer, prevent heart ailments and inflammation. They are also potent vasodilator[10]. Gnana Sundari *et al*., evaluated phytochemicals in three *Solanum* species *S. torvum*, *S.trilobatum* and *S. xanthocarpum*[11]. The present data is comparable with the above species.

Table 1: Phytochemicals in the leaf and fruits of *S. mauritianum* Significance 5% level ($P < 0.05$)

Phytochemicals	Leaves	Fruits
Saponins	2.1 ±0.04	1.6 ± 0.05
Alkaloids	4.78 ± 0.01	3.5 ± 0.12
Phenols	3.3 ± 0.09	2.7 ± 0.04
Flavonoids	3.5 ± 0.09	2.6 ± 0.04
Tannins	1.56 ± 0.01	1.9 ± 0.03

Quantitative phytochemical evaluation of the seed, leaves, stem and berries of *S. surattense, S. trilobatum* and *S. sisymbriifolium* by Shahiladevi *et al*., also substantiate the present results[12]. Muthumani et al., estimated the total alkaloid from *Solanum* leave with its medicinal potentiality and were lower than that of *S. mauritianum*[13].

Pharmacognostical and phytochemical comparison of roots of *Solanum* species used in Ayurvedic formulations was chiefly based on alkaloid content. In this scenario, the high alkaloid content in *S. mauritianum* demands its further evaluation[13]. Major alkaloids in *Solanum* species are steroidal glycoalkaloids. These contain three portions: a non-polar steroid and a basic portion with either a indolizidine or oxa-azaspirodecane structure which form the aglycone part and a polar, water-soluble sugar moiety with three or four monosaccharides attached to the 3-OH group of the first ring of the aglycone. Medicinal uses of glycoalkaloids have fascinated the pharmacological industries. Solamargine and solasodine exhibit potent cytotoxicity to human hepatoma cells via apoptosis. Solasodine, solamargine, and solasonine from *S. incanum* were proven liver protective agents against CCl_4-induced liver damage. Furthermore, α-chaconine, α-solanine, α–solamargine, α-solasonine, α-tomatine and their hydrolytic products inhibit the growth of colon and liver carcinoma cells. Plasma low-density lipoprotein cholesterol and triglycerides was lowered by α- tomatine by inducing cytokines in immunized animals. Solanine and chaconine separately or jointly reduce cervical liver, lymphoma and stomach cancer[14,15].

CONCLUSION

The results of the present investigation report the phytochemical analysis of phenolics, flavonoids, saponins, tannins and alkaloids present in leaves and fruits of *S. mauritianum.* However further investigations are required to isolate and characterize the active constituents from this plant to evaluate their therapeutic potential. Future studies are warranted to isolate, purify the lead molecules and evaluation of its biological potentialities.

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