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Preliminary phytochemical screening of foliar extract of *Saraca asoca* (Roxb.)

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Abstract

Saraca asoca (Roxb) De. Wild is a small evergreen plant belongs to the family Ceasalpiniaceae commonly known as Ashoka. The plant has many pharmacogonestic activities such as anti-diabetes, uterine toxicity, anti-oxidant and heart disease. Traditionally it was used as medicine against uterine fibroids, leucorrhoea, dysentery and wound healing. The present study was conducted for the phytochemical screening of different extracts such as petroleum ether, chloroform, methanol, ethanol and distilled water of leaves of *Saraca asoca*. Shed dried leaves were grinded and used for the extraction by using soxhlet method. Results indicated the presence of Tri-terpenoids in petroleum ether, chloroform, methanol and ethanol extract. Phenols were present in only chloroform and methanol extract whereas tannins in petroleum ether and chloroform extract. Saponins were found to be present in chloroform and distilled water extract. Distilled water and methanol extract indicated the presence of flavonoids in it. Alkaloids and carbohydrates were present in only methanol and ethanol extract respectively. Steroids were present only in methanol extract. Amino acid and proteins were not found to be present in leaves of *Saraca asoca*. The leaves of this plant posses various potent phytochemicals and is recommended for further phytopharmaceutical importance.

Keywords: *Saraca asoca*, phytochemical, soxhlet, phytopharmaceuticals

Introduction

Ayurvedic science is deeply rooted in India and its neighbouring countries. A variety of plants is used for medicinal treatments either whole or in specific parts i.e bark, root, leaves, flowers and fruits in dried state. Most of the natural products are secondary metabolites which are serves as plant defence mechanisms against predation by micro-organism, insects and herbivores [5].

Saraca asoca (Roxb.) is a evergreen plant belongs to family Caesalpiniaceae, commonly known as Ashoka or Sita Ashoka. This plant is spread throughout in India, especially in Himalaya and whole south region. It is well known for their medicinal value [7.]. Traditionally it was used as medicine against uterine fibroids, leucorrhoea, piles, dysentery, wound healing

and many others [15]. All the parts of *Saraca asoca* such as bark, leaves, flower, fruits, seeds and roots have medicinal value. Bark of this plant shows antimicrobial activity against wild range of pathogenic bacteria whereas flowers and leaves used to treat fever, dysentery, diabetes, constipation, stomach pain and pimples [6,18,10].

Leaves of the *Saraca asoca* are 15-20 cm long and have 6-12 leaflets. The plant contains peripinnates, obolong and lanceolate leaves which are arranged opposite to each other. Young leaves are red in colour which turns into green after maturity [3]. Several studies reported that leaves of this plant contains alkaloids, steroids, flavonoids, tannins, saponins, terpenoids, polyphenolic, glycosides and many carbohydrates [3,14,8,2]. Flowers of this plant are fragmented, polygamous apetalous, deciduous and calyx petaloids [3]. Tannins, flavonoids, saracasin, saracadin, waxy substance, carbohydrates, protein and steroids are reported from the flower [3,14,8]. Bark of Saraca asoca is dark brown and black in colour with water surface and reported to contains flavonoids, tannins, steroids, volatile oil, glycosides and many other compounds [3,14,11]. Several studies revealed the presence of fatty acid, steroids, flavonoids and saracin in seeds [3,14,8]. Reports revealed the presence of fatty acid, palmitic acid, steroids and flavonoids in fruits whereas roots of *Saraca asoca* contains gum, sugar, colouring matter and salts of lime [3,14,8]. The present investigation is carried to screen the presence of different phytochemicals in leaves of Ashoka plant.

Materials and methods

The study was conducted in January - July 2017 in ITM University Gwalior Madhya Pradesh. Fresh and disease free leaves of *Saraca acoka* were collected from the botanical garden and used for extraction. Collected leaves were washed with distilled water and shed dried at room temperature. The shed dried leaves were grinded and stored in air-tight container for the further use. Five different solvents such as petroleum ether, chloroform, ethanol, methanol and distilled water were used for the extraction using soxhlet method. Foliar extract of *Saraca asoca* in all the five extracts were tested for the presence of various bioactive compounds by following standard methods [7,17].

Alkaloids

Alkaloids were tested by Mayer's test. Few drops of Mayer's reagent (1.36gm of Mercuric chloride and 5gm of Potassium iodide in 100ml distilled water) was added in 2-3 ml of test extract. Appearance of cream colour is observed in the sample. This change in colour of extract indicated the presence of alkaloids.

Carbohydrates

Carbohydrate was tasted by using Molish test. Few drops of Molish reagent (10gm α-napthol in 100ml 95% alcohol) were added in 2-3 ml of test sample. Then few drops of concentrated sulphuric acid were mixed with it through the wall of the test tube. Formation of purple- violet colour ring at the junction indicated the presence of carbohydrates.

Amino acid

Two millilitre Million's reagent (Mercuric nitrate) was added with 2-3 ml of test sample. Formation of white precipitate indicated that amino acid were present in the sample extract.

Protein

Presence of protein was tested by Warming test. For this2-3 ml of test sample was heated in boiling water bath. Coagulation of extract after heating indicated the presence of protein in the sample.

Saponin

Saponin content was tested by Forth formation test. For this 2 ml of test extract was shaken vigorously with distilled water in a test tube. Persistent foam formed at the surface indicated the presence of saponin in the extract.

Flavonoids

Flavonoids were tested by performing Alkaline reagent test. Few dropes of sodium hydroxides solution was added in 2 ml of test extract. Instance yellow colour formed which turned into colour less solution on addition of few drops of dilute acid (H_2SO_4). This change indicated that extract posses flavonoids in it.

Tannins

Tannins were tested by performing Gelation test. Two millilitre of test extract and 1% gelatine solution containing 10% sodium chloride were mixed in a test tube. Formation of precipitate indicated the presence of tannins.

Steroids and tri-terpenoids

Steroids and tri-terpenoids were tested by Salkowski test. Two millilitre ml of test extract was added with few drops of concentrated sulphuric acid. Formation of red colour at the lower level indicated the presence of steroids whereas yellow colour indicated the presence of tri-terpenoids in the extract.

Phenol

Two millilitre of test extract was treated with 2 ml of 5% ferric chloride solution. Formation of blue colour indicated the presence of phenol.

Results and discussion

The present study is based on phytochemical analysis of the active medicinal and chemical constituents present in foliar extract of *Saraca asoca*. For preliminary phytochmical screening, qualitative analysis for alkaloids, amino acids, carbohydrates, flavonoids, saponins, tannins, protein, steroids, tri-terpenoids and phenol were performed.

The study revealed the presence of maximum phytochemicals in methanol extract. Carbohydrates were present in ethanol [4,17]. Tri-terpenoids were present in most of the solvents i.e.petroleum ether, chloroform, methanol, ethanol except distilled water [17,16]. Alkaloids were present in methanolic and ethanolic extracts [17]. Plants having alkaloids are used in medicines for reducing headache and fever. These are also artibuted in antibacterial and analgesic properties [13]. Phenolics were present in chloroform and methanol extract [4]. The higher activity of the chloroform and methanol extracts as compare to the aqueous extract can be attributed to the presence of higher amounts of polyphenols. It means they are more efficient in cell wall which has non polar character and cause polyphenols to be released from the cell. The reason behind this can be because of the ability of the enzyme polyphenol oxidase, which degrades polyphenols in water extract where as in chloroform and methanol they are inactive [21]. Saponins and tannins both were present in chloroform extract but their occurrence varies in distilled water and petroleum ether respectively. These findings were in concordance with some previous findings [4]. Saponins and tannins are very important bioactive compound in plants, which have strong anti-cancer properties [22]. Methanol and aqueous extract revealed the presence of flavonoids in it [16,12]. Steroids were present only in methanolic extract [1]. Amino acid and proteins were not traced in any of extracts [17]. These findings show that leaves offers a wide array of phytochemicals which can be used for further quantitative estimation.

Table 1: Phytochemical analysis of different solvent extracts in leaves of *Saraca asoca*.

Phytoconstituents	Petroleum Ether	Chloroform	Methanol	Ethanol	Distilled Water
Alkaloids	-	-	+	-	-
Amino acid	-	-	-	-	-
Carbohydrate	-	-	-	+	-
Flavonoids	-	-	+	-	+
Saponins	-	+	-	-	+
Tannins	+	+	-		-
Protein	-	-	-	-	-
Steroids	-	-	+	-	-
Tri-terpenoids	+	+	+	+	-
Phenol	-	+	+	-	-

'+'= positive, '-'= negative

Conclusion

In the Ayurvedic system of medicine, herbals extract instead of purified compounds have been used since centuries, because many constituents with more than one mechanism of action are considered essential for the required holistic therapeutic action. *Saraca asoca* is highly recommended plant in ayurvedic medicine. This plant in enriched with various types of important compounds that has many pharmaceutical activities. The present study revealed that *Saraca asoca* has such bioactive compounds which have ability to overcome various abnormalities such as antimicrobial disease, piles, leucorrhoea and uterine fibroids. Considering the above mentioned phytochemicals, further research is required to reveal its hidden properties which provide great interest to authors.

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