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Research Article

PHYTOCHEMICAL ANALYSIS OF *AERVA LANATA, ADATHODA VASICA, PISONIA ALBA, SESBANIA GRANDIFLORA* AND *INDIGOFERA ASPALATHOIDES*

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ABSTRACT

Objective: The objective of this study was to investigate the presence of various phytochemicals obtained from ethanol (E), ethyl acetate (ET), methanol (M), and chloroform (C) extracts of the whole plant of *Aerva lanata* and leaves of *Adathoda vasica*, *Pisonia alba*, *Sesbania grandiflora,* and *Indigofera aspalathoides*.

Methods: The freshly collected plant materials were subjected to successive extraction separately using E, ET, M and C with soxhlet apparatus. Using the standard protocols, the leaf extracts obtained were subjected to preliminary phytochemical analysis to detect the presence of carbohydrates, proteins, steroids, flavonoids, tannins, and alkaloids.

Results: The phytochemical analysis showed the presence of tannins, flavonoids, alkaloids, terpenoids, glycosides, saponins, resins, carbohydrates and proteins. The whole plant of *A. lanata* showed the presence of flavonoids, terpenoids, glycosides, carbohydrates and proteins. Flavonoids, alkaloids, terpenoids, glycosides, resins, carbohydrates and proteins are found in the leaves of *A. vasica*. Flavonoids, glycosides, and resins were found in the leaves of *P. alba*. No phytochemical constituents were found in the chloroform extract of *P. alba* leaves. Ethanol and methanol leaf extracts from *S. grandiflora* and methanol leaf extract from *I. aspalathoides* indicated that they contained most of the phytochemical compounds.

Conclusion: The different extracts of plants have clearly indicated the presence of all the major phytochemicals; hence, these plants can be used for the extraction of bioactive compounds.

Keywords: Phytochemicals, *Aerva lanata, Adathoda vasica, Pisonia alba, Sesbania grandiflora, Indigofera aspalathoides.*

INTRODUCTION

The plant realm is considered an asset for various kinds of potential drugs. In ancient days, many of the diseases were cured using plant products, and now again, there is an increasing awareness among people about the importance of plants and their medicinal values [1]. An indigenous part of ancestral medicine is herbal medicine. In 2008, according to the World Health Organization, more than 80% of the world's population went back to traditional medicines [2]. The crude forms of plants that are usually used as food supplements are known as herbal therapy in today's world [3]. In ethnomedicine, many of the plants used contain useful chemotherapeutants, which, in turn, are used in orthodox medical practice [4]. The whole plants in crude forms (containing both the active and non-active components) show higher efficacy than the plant products in semi-crude or pure form [5]. The phytochemical constituents in medicinal plants heal and cure human diseases, and these constituents [6] are non-phytotoxic and hence readily biodegradable. Both primary and secondary compounds form phytochemicals, wherein the primary constituents include chlorophyll, proteins and common sugars and the secondary compounds are terpenoid, flavonoids, alkaloids, phenolic compounds, glycosides, gums, tannins and essential oils among others [7]. The most of the active components are found concentrated in the storage organs of the plants [8]. These active secondary compounds determine the medicinal properties of plants. Therefore, there is a need for treasuring these medicinal plants not only to determine the scientific basis for their usage but also to discover fresh or lead compounds for treating various diseases in humans [9]. Added advantage is that these readily available plant medicines are less expensive, safe to use, and biodegradable and rarely have side effects [10]. In this work, five plants were considered for the qualitative phytochemical analysis, namely, *Aerva lanata*, *Adathoda vasica*, *Pisonia alba*, *Sesbania grandiflora*, and *Indigofera aspalathoides*.

METHODS

Collection of the plant materials

The whole plants of *A. lanata* and fresh leaves of *A. vasica, P. alba,* and *S. grandiflora* were collected from Stella Maris College (Autonomous), Chennai, Tamil Nadu, India. Fresh leaves of *I. aspalathoides* were collected from Thakalai, Kanyakumari District, Tamil Nadu.

Preparation of the extract

The freshly collected plant materials were thoroughly washed thrice in distilled water, shade dried, cut into fine pieces and powdered using a mechanical blender. The shade dried plant materials were pulverized and subjected to successive extraction separately using ethanol (E), ethyl acetate (ET), methanol (M), and chloroform (C) with Soxhlet apparatus.

Phytochemical analysis of leaf extract

The leaf extracts obtained from E, ET, M, and C were subjected to preliminary qualitative tests to detect the presence of carbohydrates, proteins, steroids, flavonoids, tannins and alkaloids.

Test for carbohydrates

Molisch's test

Few drops of Molisch's reagent and concentrated sulphuric acid (H_2SO_4) were added to 2 ml of methanol extract. Reddish violet ring was observed at the junction of two layers indicating the presence of carbohydrates [11].

Reduction of Fehling's solution

About 10 ml of Fehling's solution (copper sulphate in alkaline condition) were added to the concentrated extracts and heated on a steam bath. Brick-red precipitates indicated the presence of carbohydrates [11].

Test for proteins

Biuret test

To 3 ml of the extract, 4% NaOH and few drops of 1% $CuSO_4$ solution were added. Violet or pink indicates the presence of proteins [12].

Ninhydrin test

To 1 ml of the extract 1% reagent was added and heated on a steam bath. Violet indicates the presence of proteins [12].

Test for glycosides

Keller-Killani test

About 1 ml of glacial acetic acid containing traces of $FeCl_3$ and 1 ml of concentrated H_2SO_4 were added carefully to the extracts. A reddish-brown is formed at the junction of the two layers and the upper layer turns bluish-green indicating the presence of glycosides [12].

Test for tannins

To 1 ml of the extract, 2 ml of 5% $FeCl_3$ was added. A dark blue or green-black indicates the presence of tannins [13].

Test for alkaloids

To 2 ml of the extract, 2 ml concentrated HCl and few drops of Mayer's reagent were added. A green or white indicates the presence of alkaloids [13].

Test for flavonoids

To 2 ml of the extract, 1 ml of 2N NaOH was added. The appearance of yellow indicates the presence of flavonoids [12].

Test for terpenoids

About 2 ml of each extract is mixed with 5 ml of chloroform, and few drops of concentrated H_2SO_4 is carefully added to form a layer. A reddish-brown coloration formed in the interface shows the presence of terpenoids [14,15].

Test for saponins

Foam test

The crude extract is mixed with 5 ml of distilled water and shaken vigorously, resulting in the formation of a stable foam which is a positive indication for saponins [14,15].

Froth test

About 2 g of the powdered sample is boiled with 10 ml of distilled water and then filtered which is mixed with 5 ml of distilled water and few drops of olive oil, shaken vigorously, and then observed for the formation of emulsion [14,15].

Test for resins

Acetone - water test

Extracts are to be treated with acetone followed by the addition of 500 µl water added and mixed well. Turbid appearance of the extract indicates the presence of resins [14,15].

RESULTS AND DISCUSSION

The phytochemical constituents of the five plants tested are summarized in Table 1. In the present study, it was observed that the ethanol extract of the whole plant of *A. lanata* has flavonoids, carbohydrates and proteins but lacks tannins, alkaloids, glycosides, terpenoids, saponins and resins. Similarly, other studies Ragavendran *et al*. and Koperuncholan *et al*. [16,17] reported the presence of tannins, flavonoids, saponins, terpenoids, alkaloids, carbohydrates, proteins and the absence of resins and glycosides. In the current study, ethyl acetate extract of whole plant of *A. lanata* contained tannins, flavonoids, saponins, terpenoids, carbohydrates, and alkaloids but resins, proteins and glycosides were absent. Similarly, the results of Ragavendran *et al*. [16] also showed that the ethyl acetate extract of the whole plant extract had tannins, flavonoids, saponins, terpenoid, carbohydrates and alkaloids but resins, proteins and glycosides were absent. In the current study, the methanol extract of the whole plant of *A. lanata* possessed tannins, alkaloids,

Table 1: Phytochemical constituents of *A. lanata*, *A. vasica*, *P. alba*, *S. grandiflora*, *I. aspalathoides*

Phytochemical tests	*A. lanata*				*A. vasica*				*P. alba*				*S. grandiflora*				*I. aspalathoides*			
Solvents	**E**	**ET**	**M**	**C**	**E**	**ET**	**M**	**C**	**E**	**ET**	**M**	**C**	**E**	**ET**	**M**	**C**	**E**	**ET**	**M**	**C**
Tannins	-	-	-	-	-	-	-	-	-	-	-	-	+	+	+	-	+	-	+	-
Flavonoids																				
Alkaline reagent test	+	-	+	-	-	-	-	+	+	-	+	-	+	-	+	+	+	+	+	+
Lead acetate test	+	-	+	-	-	-	-	+	+	-	+	-	+	-	+	+	+	+	+	+
Alkaloids																				
Mayer' test	-	-	-	-	-	-	+	-	-	-	-	-	-	+	+	-	-	-	-	-
Terpenoids																				
Salkowski test	-	+	-	-	-	+	-	-	-	-	-	-	-	-	+	+	+	+	+	+
Glycoside																				
Liebermann's test	-	+	+	-	-	+	-	-	-	+	-	-	+	+	+	-	-	-	+	-
Salkowski test	-	+	+	-	-	+	-	-	-	+	-	-	+	+	+	-	-	-	+	-
Keller-Kiliani test	-	+	+	-	-	+	-	-	-	+	-	-	+	+	+	-	-	-	+	-
Saponins																				
Foam test	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	+	+	+	+
Froth test	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	+	+	+	+
Resin																				
Acetone-water test	-	-	-	-	-	-	+	+	-	+	+	-	+	+	+	+	+	+	+	+
Carbohydrates																				
Molish's test	+	-	-	+	+	-	-	+	-	-	-	-	+	+	+	-	+	-	+	-
Fehling's test	+	-	-	+	+	-	-	+	-	-	-	-	+	+	+	-	+	-	+	-
Proteins																				
Ninhydrin test	+	-	-	+	+	-	-	+	-	-	-	-	+	+	+	-	+	+	+	+
Biuret test	+	-	-	+	+	-	-	+	-	-	-	-	+	+	+	-	+	+	+	+

E: Ethanol, ET: Ethyl acetate, M: Methanol, C: Chloroform, *A. lanata: Aerva lanata, A. vasica: Adathoda vasica, P. alba: Pisonia alba, S. grandiflora: Sesbania grandiflora, I. aspalathoides: Indigofera aspalathoides*

terpenoids, carbohydrates, saponins and resins whereas proteins were absent in concurrence with the Yamunadevi *et al*. [18] that also have shown the absence of tannins, carbohydrates, saponins, proteins, and resins. Although the study Yamunadevi *et al*. [18] has reported the presence of flavonoids and glycosides as in the current study, they have also shown the presence of terpenoids and alkaloids. The chloroform extract of the whole plant of *A. lanata* possessed only carbohydrates and proteins in contrast to another study Battu and Kumar [19] that showed the presence of tannins, alkaloids and flavonoids apart from carbohydrates and has shown the absence of alkaloids, proteins, saponins and resins but tannins, flavonoids, glycosides, saponins, resins, terpenoids, and alkaloids were absent in the present study.

In this study, it was observed that the ethanol extract of the leaves of *A. vasica*, contained only carbohydrates and proteins but other compounds - such as tannins, alkaloids, flavonoids, glycosides, saponins, terpenoids and resins - were absent. In contrast, one study Emimal [20] has reported that the ethanol extract of leaves showed the presence of alkaloids, flavonoids, glycosides, carbohydrates and saponins and absence of tannins, proteins, resins and terpenoids. The ethyl acetate extract of the leaves of *A. vasica* contained terpenoids and glycosides but other components - such as alkaloids, tannins, saponins, carbohydrates, flavonoids, proteins and resins - were absent. In contrast, another study Bharathi *et al*. [21] has reported the presence of alkaloids, tannins, saponins, flavanoids, proteins, and carbohydrates. In the current investigation, the methanol extract of the leaves of *A. vasica* possessed alkaloids, whereas tannins, flavonoids, terpenoids, carbohydrates, glycosides, saponins, proteins and resins were absent. A study by Subhashini *et al*. [22] has shown that the methanol extract of leaves showed the presence of alkaloids, carbohydrates, flavonoids, glycosides, proteins and terpenoids and the absence of tannins and saponins. In the current study, the chloroform extract of the leaves of *A. vasica* showed only flavonoids, tannins, carbohydrates, resins, terpenoids, glycosides and proteins and alkaloids were absent, but Emimal [20] has reported the presence of alkaloids and flavonoids and absence of resins, carbohydrates, glycosides, tannins, proteins, and saponins.

In the current investigation, the ethanol extract of the leaves of *P. alba* contained flavonoids and absence of tannins, alkaloids, terpenoids, glycosides, saponins, resins, carbohydrates and proteins were noted. Poongothai and Shubashini [23] have reported the absence of alkaloids and terpenoids. The ethyl acetate extract of the leaves of *P. alba* contained glycosides and resins, whereas tannins, flavonoids, alkaloids, terpenoids, saponins, carbohydrates and proteins were absent. The methanol extract of the leaves of *P. alba* possessed flavonoids and resins but tannins, alkaloids, terpenoids, glycosides, saponins, carbohydrates and proteins were absent. In this study, no phytochemical constituents were extracted in chloroform in contrast to the study of Poongothai and Shubashini [23] who have reported the presence of flavonoids, tannins, alkaloids, trepenoids, carbohydrates and proteins.

In the current study, the ethanol and methanol extracts of *S. grandiflora* contained tannins, flavonoids, alkaloids, terpenoids, glycosides, resins, carbohydrates and proteins. Saponins were absent. A study Nadia *et al*. [24] reported the presence of tannins, flavonoids, alkaloids, phenols, carbohydrates and proteins and absence of terpenoids in the methanol extract. Another study [25] reported the absence of flavonoids, glycosides, and proteins in the ethanol extract. The ethyl acetate extract contains tannins, alkaloids, glycosides, resins, carbohydrates and proteins, but flavonoids, terpenoids, and saponins were absent. The chloroform extract contained flavonoids, trepenoids and resins whereas the other constituents were absent. In contrast Arun *et al*. [25] reported the presence of steroids and proteins and the absence of tannins, flavonoids, alkaloids, glycosides and carbohydrates.

In the current investigation, ethanol extracts of *I. aspalathoides* contain tannins, flavonoids, terpenoids, saponins, resins, carbohydrate and protein while alkaloids and glycosides were absent. Similarly, Subhashini *et al*. [22] also reported that the ethanol extract of *I. aspalathoides* contains flavonoids, terpenoids and saponins. Ethyl acetate extract contains flavanoids, terpenoids, saponins, resins and protein, whereas tannins, alkaloids, glycosides and carbohydrate were absent. Elangovan *et al*. [26] reported the presence of alkaloid, glycosides, flavonoids and tannins and absence of terpinoids in ethyl acetate extract. The methanolic extracts of *I. aspalathoides* contain tannins, flavonoids, terpenoids, glycosides, saponins, resins, carbohydrate and protein but alkaloids were absent. Subhashini *et al*. and Tamilselvi *et al*. [22,27] also reported similar findings. Apart from alkaloids being absent, the absence of saponins was also reported by them. In this study, the chloroform extract of *I. aspalathoides* contains flavanoids, terpenoids, saponins, resins, and protein. In the chloroform extract, tannins, glycosides, and carbohydrates were absent.

CONCLUSION

The phytochemical analysis of the crude extracts of *A. lanata*, *A. vasica*, *P. alba, S. grandiflora*, *I. aspalathoides* indicates the presence of major phytochemical compounds such as tannins, flavonoids, alkaloids, terpenoids, glycosides, saponins, resins, carbohydrates and proteins which are secondary metabolites. These compounds are associated with antioxidant, antimicrobial activities, antidiuretic, anti-inflammatory, anti-analgesic, anti-cancer, antiviral, anti-malarial and anti-fungal activities. Thus, the traditional system of medicine provides biologically active molecules that are promising sources of potential secondary metabolites which can be used as medicinal compounds. Further studies aim at identifying the antimicrobial and antioxidant compounds which may be exploited in herbal formulations.

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