



Phytochemical Analysis Of Leaf Of Some Acanthaceous Members From Murshidabad District, West Bengal

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Abstract:

The Present study deals with the phytochemical analysis of leaf and medicinal values of some selected plants belongs to the family Acanthaceae DPPH method is very useful for the detection of antioxidant property of plants. The presence of high amount of phytochemical compounds suggests that the plants have high amount of medicinal compounds and can be extensively used to extract the natural compounds. Phytochemical analysis was carried out to estimate the quantity of phenols, carbohydrates, tannins, flavonoids and proteins. The quantitative analysis showed the presence of alkaloids, tannins, saponins, proteins, carbohydrate and phenols. The high present of scavenging activities of those plants add value to their medicinal properties. Murshidabad district is the historic place from the time of 17th century and flourished with different types of herbaceous, shrubby and tree plants.

Keywords: Phytochemical study, leaf, Acanthaceae, Murshidabad.

Introduction:

India is rich in Medicinal plant diversity which is distributed in different phytogeographical zones and associated with tribal and folk medicinal knowledge. People started to use plant extracts as medicines in many of the traditional practices. It was suggested that ethno-mediated samples were succeeded to identify the drugs in the treatment of different ailments like inflammatory, gastrointestinal and dermatological complaints. Acanthaceae represent about 229 genera and 3450 species, distributed in the tropical regions, Central America, Brazil and Africa. Some plants occur in Mediterranean regions, some are xerophytic species found in steppes. Plants herbs, shrubs, small trees with cystolitus. Leaves opposite, simple and exstipulate. Flowers bisexual, zygomorphic, hypogynous with bracts and bracteoles. Fruit elastically dehiscent bivalve capsule. Seeds often on hook like outgrowths the so called retinacula or jaculators. Traditionally the most important part use in Acanthaceae is the leaves. Leaves are used externally for wounds. Some research has indicated that Acanthaceae posses antifungal, cytotoxic, anti-inflammatory, anti-pyretic, anti-oxidant, insecticidal, hepatoprotective, immunomodulatory, anti-viral potential. The objective of present study was to compare the phytochemical compounds present in the selected members of the family Acanthaceae. We also focused on the quantification of bioactive compounds founds in the plants. The study was also aimed at evaluating the medicinal and antioxidant properties of the selected plants.

Materials and methods:

Fresh and healthy plant parts of *Adhatoda vasica*, *Ruellia tuberosa*, *Rungia repens*, *Andrographis paniculata*, *Barleria cristata*, *Justicia simplex*, *Hemigraphis hirta*, *Peristrophe bicalyculata*, *Hygrophila spinosa*, *Ecbolium viride* were collected in sterile polythene bags from Murshidabad district, West Bengal. Collected plant parts were examined, identified and kept for experimental use in Department of Botany, Berhampore Girls' College, Berhampore, Murshidabad, West Benal.

Preparation of solvent extract:

The fresh, healthy leaf were cut into small pieces and dried under shade for a few days and then dried in a hot air oven. The dried materials were ground into a fine powder. The powder obtained was stored in a desiccators and kept in room temperature for extraction. The methanolic extracts of the samples were prepared by 2gm. Of dried powder in 20 ml. of solvent. The extracts were filtered using filter paper. The extracts were tested for detecting various compounds such as flavonoids, tannins, saponins, protein, phenol, carbohydrates etc. for quantification analysis 5gm. of sample was extracted with methanol using soxhlet apparatus. The extracts were evaporated using a rotary evaporator.

Qualitative analysis of phytochemicals:

Preliminary phytochemical screening was carried out (Harborne, 1980) and (Karthiswaran, 2010).

Test for alkaloids (Mayer's test)

To 1 ml of extract, 1ml of Mayer's reagent (Potassium iodide solution) was added. Formation of whitish yellow or cream coloured precipitate indicates the presence of alkaloids.

Test for steroids (Liebermann Burchard test)

To 1 ml of extract, 2ml of acetic anhydride and 2ml of concentrated sulphuric acid were added. Formation of violet to blue or green colour indicates the presence of steroids.

test for terpenoids (Salkowski test)

To 1 ml of extract, 2ml of chloroform and few drops of sulphuric acid were added. Formation of reddish brown ring indicates the presence of terpenoids.

Test for flavanoids (Alkaline reagent test)

To 1 ml of extract, few drops of dilute ammonium solution and few drops of concentrated hydrochloric acid were added. A yellow colouration indicates the presence of flavanoids.

Test for saponins (Froth test)

To 1 ml of extract, 5ml of distilled water was added and shaken vigorously. Formation of froth indicates the presence of saponins.

Test for phenols (lead Acetate test)

To 1 ml of extract, 1ml of lead acetate solution was added. Formation of precipitate indicates the presence of phenols.

Test for tannins (lead acetate test)

To 1 ml of extract, 1ml of lead acetate was added. A formation of white precipitate indicates the presence of tannins.

Test for Tannins (Ferric chloride test)

To 1 ml of extract, 1ml of ferric chloride solution was added. Formation of blue, black or brownish green colour indicates the presence of tannins.

Test for cardiac glycosides (Keller killiani test)

To 1 ml of extract, 5ml of distilled water was added and evaporated to dryness. Then to the Sample 2ml of glacial acetic acid containing trace amount of ferric chloride solution was added. Then 1ml of concentrated sulphuric acid was added along the sides of the tube. Formation of brown ring underplayed with blue colour indicates presence of cardiac glycosides.

Test for aminoacids (Ninhydrin test)

To the 1ml of sample, 3 to 4 drops of Ninhydrin solution was added and boiled in water bath for 10 minutes. Formation of purple or blue colour indicates the presence of amino acids.

Test for proteins (Biuret test)

To the 1ml of extract, 1ml of 40% sodium hydroxide solution and 2 drops of 1% copper sulphate solution were added. Formation of violet colour indicates the presence of proteins.

Test for carbohydrates (Barfoed test)

To the 2ml of extract, 1ml of Barfoed's reagent was added and boiled in water bath for few minutes. Formation of reddish brown precipitate indicates the presence of carbohydrates.

Test for reducing sugars (Fehling's test)

To the 1ml of extract, equal quantities of Fehling solution A and B were added and heated. Formation of brick red precipitate indicates the presence of reducing sugars.

Table-1: Qualitative phytochemical analysis of selected plants of the family Acanthaceae:

Sr. No.	Name of the plants	Tests												
		Alkaloid	Steroids	Flavonoi	Terpenoi	Saponins	Phenols	Tannins	Glycoside	Amino Acids	Proteins	Carbohy drates	Reducing Sugars	
1	<i>Adhatoda vasica</i>	*Methanol:	+	+	+	+	+	+	-	+	+	+	+	+
		*Ethanol:	+	+	+	+	-	-	-	-	+	-	-	+
		*Petroleum Ether:	-	+	-	+	-	-	+	+	+	-	+	-
		*Chloroform:	+	-	+	-	-	+	+	-	+	+	-	+
		*Aqueous:												
2	<i>Ruellia tuberosa</i>	*Methanol:	+	+	-	+	+	+	-	+	+	+	-	+
		*Ethanol:	+	+	+	-	+	+	+	+	+	+	+	+
		*Petroleum Ether:	-	+	-	+	-	-	+	-	-	+	+	-
		*Chloroform:	+	+	+	+	-	+	-	+	-	-	-	+
		*Aqueous:												
3	<i>Rungia repens</i>	*Methanol:	+	-	+	+	+	-	+	+	+	+	+	+
		*Ethanol:	+	+	-	-	-	+	+	+	-	-	-	+
		*Petroleum Ether:	+	-	-	+	-	-	+	-	-	-	+	+
		*Aqueous:	-	-	+	+	-	+	+	+	+	-	+	-

		*Chloroform:	-	+	-	+	+	-	-	+	+	+	+	+
		*Aqueous:												
4	<i>Andrographis paniculata</i>	*Methanol:	+	+	+	+	-	+	+	-	+	+	+	+
		*Ethanol:	+	-	+	-	+	+	+	+	-	+	+	+
		*Petroleum	-	-	+	+	+	-	-	+	-	+	-	+
		Ether:	-	+	+	+	-	+	-	+	-	-	+	+
		*Chloroform:	+	+	-	+	+	-	+	+	+	-	-	-
		*Aqueous:												
5	<i>Barleria cristata</i>	*Methanol:	+	-	+	+	+	-	+	+	+	+	+	+
		*Ethanol:	+	+	-	-	-	+	+	+	-	-	-	+
		*Petroleum	+	-	-	+	-	-	+	-	-	-	+	+
		Ether:	-	-	+	+	-	+	+	+	+	-	+	-
		*Chloroform:	-	+	-	+	+	-	-	+	+	+	+	+
		*Aqueous:												
6	<i>Justicia simplex</i>	*Methanol:	+	+	+	+	+	+	-	+	+	+	+	+
		*Ethanol:	+	+	+	+	-	-	-	-	-	+	-	+
		*Petroleum	-	+	-	+	-	-	+	+	+	-	-	-
		Ether:	-	+	-	+	+	-	+	+	+	-	+	-
		*Chloroform:	+	-	+	-	-	+	+	-	+	+	-	+
		*Aqueous:												
7	<i>Hemigraphis hirta</i>	*Methanol:	+	+	-	+	+	+	-	+	+	+	-	+
		*Ethanol:	+	+	+	-	+	+	+	+	+	+	+	+
		*Petroleum	-	+	-	+	-	-	+	-	-	+	+	-
		Ether:	-	-	-	+	+	+	+	-	-	-	-	+
		*Chloroform:	+	+	+	+	-	+	-	+	-	-	-	+
		*Aqueous:												
8	<i>Peristrophe bicalyculata</i>	*Methanol:	+	+	+	+	+	+	-	+	+	+	+	+
		*Ethanol:	+	+	+	+	-	-	-	-	-	+	-	+
		*Petroleum	-	+	-	+	-	-	+	+	+	-	-	-
		Ether:	-	+	-	+	+	-	+	+	+	-	+	-
		*Chloroform:	+	-	+	-	-	+	+	-	+	+	-	+
		*Aqueous:												
9	<i>Hygrophila spinosa</i>	*Methanol:	+	-	+	+	+	-	+	+	+	+	+	+
		*Ethanol:	+	+	-	-	-	+	+	-	-	-	-	+
		*Petroleum	+	-	-	+	-	-	+	-	-	-	+	+
		Ether:	-	-	+	+	-	+	+	+	+	-	+	-
		*Chloroform:	-	+	-	+	+	-	-	+	+	+	+	+
		*Aqueous:												
10	<i>Ecboium viride</i>	*Methanol:	+	+	-	+	+	+	-	+	+	+	-	+
		*Ethanol:	+	+	+	-	+	+	+	+	+	+	+	+
		*Petroleum	-	+	-	+	-	-	+	-	-	+	+	-
		Ether:	-	-	-	+	+	+	+	-	-	-	-	+
		*Chloroform:	+	+	+	+	-	+	-	+	-	-	-	+
		*Aqueous:												

Quantitative estimation of phytochemicals:

Alkaloid determination:

5 gm of sample was added to 200 ml of 10% acetic acid in ethanol in beaker. The beaker was tightly covered and allowed to stand for 4 hours. This was filtered and the extract was concentrated on a water bath to one quarter of the original volume. The entire solution was precipitated by the drop wise addition of concentrated ammonium hydroxide solution. The precipitate was collected and washed with dilute ammonium hydroxide and filtered. The residue is alkaloid, which was dried and weighed (Harborne, 1980).

Flavanoid determination:

10 gm of sample was added to 100 ml of 80% aqueous methanol in a beaker. The whole solution was filtered through Whatman filter paper No. 42 (125mm). the filtrate was then evaporated to dryness and weighed (Harborne, 1980).

Table-2: Quantitative phytochemical analysis of selected plants of the family Acanthaceae:

Sr. No.	Name of the plants	Tests			
		Alkaloid		Flavonoids	Phenols
1	<i>Adhatoda vasica</i>	*Methanol:	8.24	2.90	17.20
		*Ethanol:	7.20	3.42	14.31
		*Petroleum Ether:	1.20	3.93	6.57
		*Chloroform:	1.32	1.20	11.22
		*Aqueous:	7.90	6.92	15.30
2	<i>Ruellia tuberosa</i>	*Methanol:	8.10	2.40	16.21
		*Ethanol:	7.20	3.41	13.39
		*Petroleum Ether:	1.10	3.90	6.51
		*Chloroform:	1.22	1.05	11.20
		*Aqueous:	6.92	6.82	15.19
3	<i>Rungia repens</i>	*Methanol:	7.23	2.65	16.21
		*Ethanol:	7.30	3.33	14.20
		*Petroleum Ether:	1.02	3.23	6.31
		*Chloroform:	1.05	1.10	11.20
		*Aqueous:	6.51	6.20	15.11
4	<i>Andrographis paniculata</i>	*Methanol:	7.59	2.30	16.69
		*Ethanol:	7.24	3.35	14.11
		*Petroleum Ether:	1.21	3.21	6.39
		*Chloroform:	1.13	1.00	11.20
		*Aqueous:	6.70	6.65	15.21
5	<i>Barleria cristata</i>	*Methanol:	7.61	2.51	16.57
		*Ethanol:	7.33	3.32	14.20
		*Petroleum Ether:	1.02	3.20	6.21
		*Chloroform:	1.05	1.12	11.11
		*Aqueous:	6.85	6.51	15.12
6	<i>Justicia simplex</i>	*Methanol:	8.10	2.78	16.90
		*Ethanol:	7.79	3.40	14.10
		*Petroleum Ether:	1.10	3.12	5.90
		*Chloroform:	1.15	1.17	10.92
		*Aqueous:	6.90	6.43	14.29
7	<i>Hemigraphis hirta</i>	*Methanol:	18.11	2.69	16.70
		*Ethanol:	7.90	3.41	13.92
		*Petroleum Ether:	1.12	3.30	5.61
		*Chloroform:	1.14	1.10	10.68
		*Aqueous:	7.20	6.73	14.31
8	<i>Peristrophe bicalyculata</i>	*Methanol:	8.31	2.48	16.80
		*Ethanol:	7.23	3.21	13.81
		*Petroleum Ether:	1.11	3.27	5.90
		*Chloroform:	1.20	1.18	11.10
		*Aqueous:	7.42	6.60	15.20
9	<i>Hygrophila spinosa</i>	*Methanol:	7.90	2.81	17.10
		*Ethanol:	6.23	3.33	13.86
		*Petroleum Ether:	1.12	3.37	6.31
		*Chloroform:	1.20	1.19	11.12
		*Aqueous:	7.61	6.67	15.10
10	<i>Ecboium viride</i>	*Methanol:	7.45	2.32	16.82

	*Ethanol:	6.43	3.20	13.80
	*Petroleum Ether:	1.00	3.12	5.60
	*Chloroform:	1.15	1.11	11.10
	*Aqueous:	7.45	6.39	15.13

Table-3: Medicinal uses of the selected plants of the family Acanthaceae:

Sr. No.	Name of the plants	Medicinal uses of leaf
1	<i>Adhatoda vasica</i>	*Act as broncho dialator* Act as an anti asthmatic * Reduce inflammation *Act as analgesic * Have diuretic activity * Act as an antioxidant
2	<i>Andrographis paniculata</i>	* Act as an immune system booster * Has anti inflammatory properties * Benefits lever functions * Digestive health * Helps with respiratory disorders
3	<i>Ruellia tuberosa</i>	*Used to treat urinary retention * Used to treat cardiac problem * Used in case of scorpion bites wounds * Used to treat gonorrhoea and syphilis * It has anti piratic properties
4	<i>Barleria cristata</i>	*It has anti bacterial activity against some gram positive bacteria * Has anti fungal activity * Has anti viral activity * Has anthelmintic properties * Used in the treatment of inflammation * Have anti arthritic property
5	<i>Justicia simplex</i>	* It has diuretic property * Remove indigestion * Used in the treatment of burning of body * It strengthens the lungs * Good in dieses of the spleen
6	<i>Hemigraphis hirta</i>	*Has galactogenic activity, it works on mammary glands and induce secretion * It has anti asthmatic activity due to the relaxation effect of bronchial tube * Effect on urine output and electrolytes * It prevents anemia * Used in the treatment of disentry
7	<i>Rungia repens</i>	*Used in the treatment of small pox * Used to reduce swelling * Used in the relevant pain * It has diuretic property
8	<i>Hygrophila spinosa</i>	*Stimulates male genital system * Gives remedy for kidney stone * It helps in evacuation
9	<i>Peristrophe bicalyculata</i>	*It helps against snake poison * Used in the treatment of ear and eye * Prepared medicine for tuberculosis
10	<i>Ecbolium viride</i>	*The leaf is used for the treatment of jaundice and rheumatism * The leaf extract is used for menorrhagia

Result and Discussion:

The qualitative phytochemical analysis of the leaves of selected members of Acanthaceae is summarized in the Table 1. The quantification of important phytochemicals of the leaves of selected members of Acanthaceae is summarized in Table 2. The methanolic extract of leaves shows the presence of high number of phytochemicals when compared with other solvents like ethanol, petroleum ether, chloroform and aqueous. It shows the presence of alkaloids, steroids, terpenoids, phenols, tannins, saponins, proteins and amino acids. Phytochemicals such as saponins, terpenoids, and alkaloids have hypoglycemic activities (Cherian and Augusti, 1995). The leaves show the presence of tannins and they play a major role in the treatment of intestinal disorders like diarrhea and dydentry (Akinpelu and Onakoya, 2006). The leaves also have terpenoids which can act as antioxidants. Phytochemicals have highest therapeutic efficiency in pharmaceutical field (Thilagavathi *et. al.*, 2015). This paves way for further studies on isolation and identification of specific phytochemicals for pharmacological studies.

Conclusion:

The qualitative and quantitative analysis shows that the leaves of selected members of Acanthaceae contains important bioactive components such as alkaloids, steroids, terpenoids, phenols, tannins, proteins, amino acids and saponins. The methanolic extracts are rich in phytoconstituents when compared with other extracts. From this study it is evident that the selected members of Acanthaceae is valued with bioactive components. Further researches are being undertaken to isolate the bioactive components and to identify its properties in the field of medicine.

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