



Phytochemical Profiling Of Crude Extracts of Some Medicinal Plants Found In Gangapur Region, District Aurangabad, Maharashtra

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ABSTRACT

The objective of the present study was to find out the presence of phytochemicals in the crude extracts of four traditional medicinal plants such as *Adathoda vasika*, *Anacyclus pyrethrum*, *Butea monosperma* and *Pongamia pinnata* by qualitative screening methods. In qualitative analysis, the phytochemical compounds such as flavonoids, carbohydrates, proteins, phenols, saponins, tannins, terpenoids, quinones, alkaloids and glycosides were screened in the crude extract of six medicinal plants by using standard methods. When compared with other plants ethyl acetate extract more active compounds will be isolated from the selected medicinal plants. Phytochemicals play an important role when used in cosmetic preparations as antimicrobial agents as well as antioxidants. Herbal based cosmetics have gained popularity due to technological advances in manufacturing processes. The application of investigated plant species in various cosmetics was based on their phytochemical content and their pharmacological activities.

KEYWORD:- Phytochemical, Medicinal Plant, Secondary Metabolides etc.

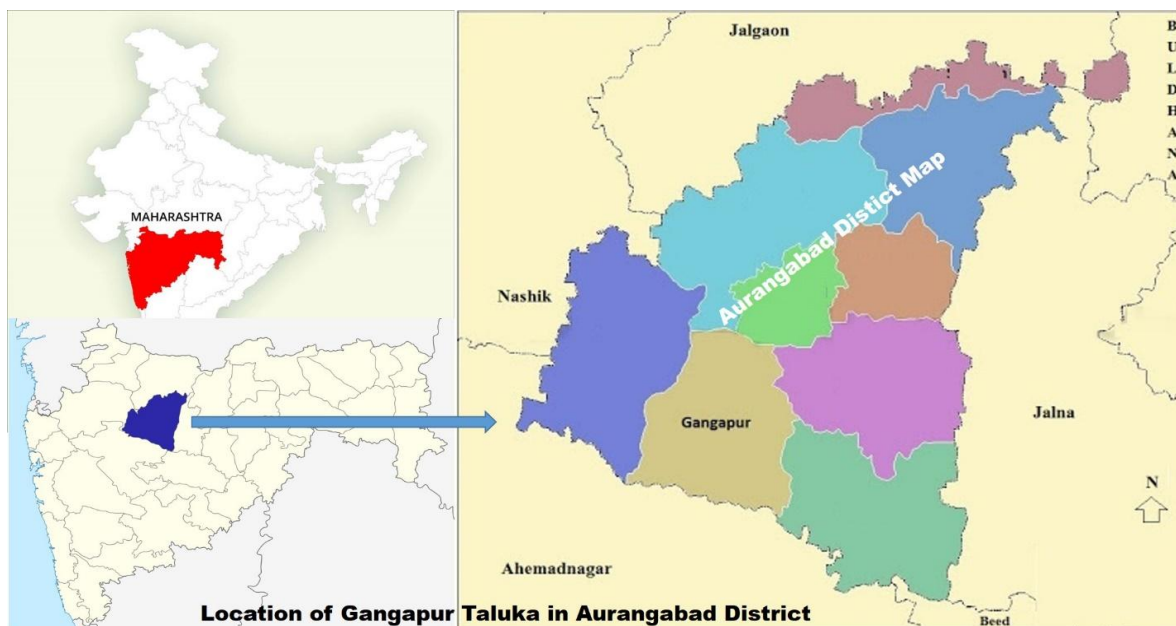
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I. INTRODUCTION

Phytochemicals are bioactive chemicals of plant origin. They are regarded as secondary metabolites because the plants that manufacture them may have little need for them. They are naturally synthesized in all parts of the plant body; bark, leaves, stem, root, flower, fruits, seeds, etc. i.e. any part of the plant body may contain active components (Maruthupandian A. et.al 2011). Medicinal plant products are considered to be the most important components of diet for a good health. A plant as the source of medicine plays an important role in the health services around the globe (Thomson G E et.al 2010). The plants are consumed by both animals and human beings as food. This mineral becomes part of the food chain. The plants absorb much of the essential elements from the soil in which they grow and serve as indicators of the materialization and are in fact used for this purpose (Navneet P 2008).

The value of medicinal plants depends upon the use of plants as raw materials in the pharmaceutical industry. People living in rural areas from their personal experience know that these traditional remedies are a valuable source of natural products to maintain human health, but they may not understand the science behind these medicines, but knew that some medicinal plants are highly effective only when used at therapeutic doses. Several medicinal compounds have been successfully isolated from plants and presently being use for the treatment and management of different diseases.



The discovery of new drugs through natural products is the single most successful strategy, which led the invention of several modern drugs. With the plants, human beings have taken advantage of the medicinal compound known as secondary metabolites present in the leaves, stems, roots and sap of the plants. The secondary metabolites such as saponins, alkaloids, flavonoids, glycosides, anthraquinone, steroids and tannins from plants has also been used in the modern system of medicines for their extensive therapeutic value. Gangapur is located on west side of Aurangabad-Ahmednagar Highway, 38 km from Aurangabad. (Coordinates 19.6991°N 75.0086°E) Gangapur is Taluka place in Historical District Aurangabad. It is situated near backwater of Jayakwadi dam so most of the area is irrigated.

Collection of samples was done from pre-selected Site as Gangapur, Antapur, gaidapur, Bhendala, Ganeshwadi, Amalner, Lakhmapur, Ager wadgoan, Wazar, Mahalaxmi kheda. this places located near Godavari river sites.

Adathoda vasica-

Adhatoda vasica is an evergreen dense and bushy shrub with long opposite ascending branches, it showed broadly elliptic leaves. The flowers showed off white or purple colour. It was in dense axillary pedunculate and bracteates spikes on it. It grows widely in Bangladesh, Myanmar, and Pakistan and also in India. The Leaves of the plant showing expectorant, bronchodilator, antispasmodic, hypotensive, cardiac depressant, respiratory stimulant, uterotonic, antimicrobial and hypoglycemic properties. The roots and barks are expectorant, antispasmodic and antiseptic. Its vernacular names are in Bengali name-Basakpata, English name is Malabar nut, having Family- Acanthaceae.

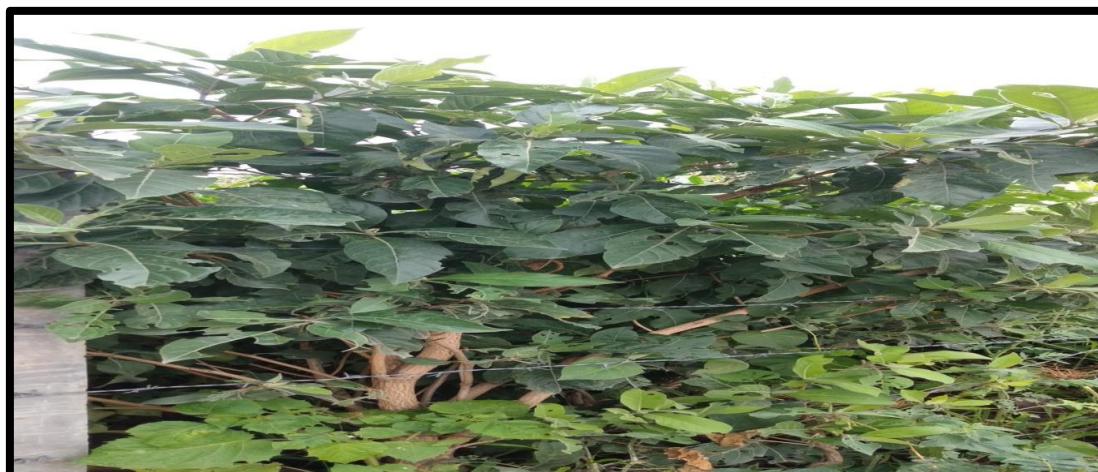


Figure- 1) Showing entire plant of *Adathoda vasica*.

Previous phytochemical investigations of *Adhatoda vasica* led to the isolation of steroid, alkaloids, saponin, phenols. The vasicinone, vasicolinone, vasicoline, adhatodine, vasicolinine, vasicinine, vasicinol, anisotine, 3- hydroxyanisotine, betaine, viciine, adhatodic acid, essential oil, fats, resins, sitosterol, vasicine, vasicionol, essential oil, indole alkaloid, galactoside, D-galactose, deoxyvasicinone, vasicinine, kaempferol, quercetin, tritriacontane, fatty oil consisting of arachidic, behenic, lignoceric, cerotic, oleic and linoleic acids (Ghani A.2003). The plant AV and its component vasicine and its derivatives are extensively being used for bronchodilatory/mucolytic cough syrup preparations since ancient history of Ayurveda and till date.

Ethnobotanical information approximately estimates that more than 6000 higher plant species forming about 60 % of the higher plant diversity are used in the traditional and folk healthcare traditions. It was also found in some part of the Tamil Nadu. The plant is used extensively in the treatment of asthma, cough, bronchitis and tuberculosis. The crude extract of *A. vasica* also proved to be effective in joint pain, lumbar pain, sprains, eczema, malaria, swellings, it is as an anti-hyperglycemic, venereal diseases, rheumatism, anti-diarrhoeal, anti-convulsant, The alkaloid Quinazoline present in the leaves of *A. vasica* were established as active principles. In the indigenous food preparations, The *A. vasica* showed active ingredients and found out that vasicine, oxyvasicine and vasicinone weresome alkaloids present in vasaka. The active ingredient is medicinally active. The alkaloid named vasicine found the market in the treatment of respiratory disorders.

The plant is used in preparation of cough syrup in pharmaceutical industry in various popular formulations. The vasaka as an ingredient is used to clear the airways by decreasing the mucus secretions and opening the air passages, one of the drug Bisolvon (Iyengar et al.,1994; Shete AB1993; Doshi MM,2003). Some reports are available on antimicrobial efficiency of *A. vasica* plant, but more research is required to explore more about its antibacterial potential of this plant. The present paper discusses the antibacterial activity of *A. vasica* leaves extract extracted in different solvents as per polarity nature of solvents.As itshowed no side effect on human but slow and long lasting effect on human pathogens.

***Anacyclus pyrethrum* :-**

Anacyclus pyrethrum (Linn) is commonly known as Akarkara. It is widely known in ayurvedic system of Indian medicine as tonic and rejuvenator. It showed fusiform root which were hard and compact in nature. They are externally brownish, deeply fissured and longitudinal. The different phytochemical and biological evaluations have been reported in the literature for proving importance in medicine of *A. pyrethrum*. So, it have been used in ethno medicine to exploit its medicinal properties including antimicrobial effect, insecticidal, local anesthetic effect, mollusidal effect also showed anti-inflammatory activity.



Figure- 2) Showing entire plant of *Anacyclus pyrethrum*

The root is a pungent in taste which stimulates the salivary glands and irritates the tissues, thereby increasing blood flow to the area. It is used externally to treat toothache, facial neuralgia and chronic catarrh. The root collected in rainy season and dried for stored for later use. In ayurveda the root is considered as tonic and also used in the treatment of paralysis and epilepsy. The root was also used in mouthwashes and in treatment of toothaches. This oil should not be used internally, except under professional supervision. *Anacyclus pyrethrum* DC. Known in different languages as Sanskrit: Agragrahi, Akarakarabha English: Pellitory Hindi:

Akarakara Malayalam: Akkikaruka, Akkrav. Tamil: Akkalkara, Akkirakkaram, Akirakaram, Akkarakaram, Akrakaram Telugu: Akarakaram. (Badhe *et al.*,2010). The less research was studied on antibacterial activity of *A. pyrethrum* so the same plant was selected for study.

***Butea monosperma* (Lam.) Kuntze-**

The *B. monosperma* plants belongs to the family Fabaceae. It is medium size woody plant and is commonly known as *Dhak*, *Tesu* or *Palash*. It was more popularly known as *Flame of the Forest*. It is mainly native from India but it is spread all over countries like Indonesia, Thailand, Cambodia, Nepal Japan, Sri Lanka, China also.



Figure -3) Showing entire plant of *Butea monosperma*

The flowers of *B.monosperma* used as natural dyes are known for their uses in the colouring of food leather, silk, wool, silk and cotton. The Natural dyes may have a vast and wide range of shades and it getting from various plant parts like root, bark, leaves, fruit and flowers. These dyes were did not showed any side effect, in fact it is used for natural colour for Holy festival to enhance the skin glow.

***Pongamia pinnata* (L.)-**

Pongamia pinnata L. is a species of family *fabaceae*, it is a deciduous legume with soft and shiny green leaves. The leaves were used for aliments. The plant extract contains flavonoids, carbohydrates, glycosides, tannins, steroids etc. *Pongamia pinnata* L. contains many alkaloids such as glabrin, pinnatin, pongamal, fatty acids, sterol and disaccharides (Samuel L.A2006; Yadav P.P., 2004; Parekh J. and Chand S., 2007). *Karanja* (*Pongamia pinnata* (Linn) Pierre.) is an important drug which has been used since in Vedic period. It is also known by the name Naktamala in Sanskrit. In Malayalam, It is called as Pongu or Ungu and Hongemara in Kannada.It belongs to Papilionaceae family.

It is a medium sized tree, upto 25m high. *Karanja* is a preferred species for controlling soil erosion and binding sand dunes because of its dense network of lateral roots. The literary review of the drug proved to be having *Vata kapha hara karma*. Root, bark, leaves, flower and seeds of this plant also have different medicinal values and traditionally used as medicinal plants. All the parts of the plant have been used as crude drug for the treatment of tumors, piles, skin diseases, wounds and ulcers (Ahmad S. *et al.*, 2003). It is also well known for its application as animal fodder, green manure, timber and fish poison (Brijesh S, *et al.*,2006).



Figure -4) Showing entire plant of *Pongamia pinnata*

In traditional medicine, the fruits, seeds and even bark of *Karanja* were useful. Seed oil is used in scabies, leprosy, piles, ulcers, chronic fever and lumbago (Ingredient guide, 2006). Powdered form of seeds are used in bronchitis, chronic fever, whooping cough and chronic skin diseases and painful rheumatic joints. The Seed extracts having anti hypotensive activity and produce uterine contractions (Srinivasan K. *et al.*,2001) .The leaves are proved to be possessed with Anti-inflammatory activity, Antioxidant, Anti-diarrhoeal Activity and Anti-hyperammonemic Activity.

II. METHOD AND MATERIAL

Collection of samples-

Collection of samples was done from different pre-selected site of Gangapur taluka region, viz. Antapur, Saidapur, Bhendala, Ganeshwadi, Amalner, Lakhmapur, Ager wadgoan, Wazar, Mahalaxmi kheda. This places located near Godavari river sites. The leaves was collected by an eco-friendly. They were identified by taxonomist. All the parts of the plants were washed with tap water and sterilized by HgCl₂.They get dried and crushed in mixer grinder and the grinding was performed in a hygienic condition. The following plant part was used for phytochemical testing.

Sr. No.	Scientific Name of Plants	Common Name of plants	Family	Parts used
1	<i>Adathoda vasica</i>	Adulsa	Acanthaceae	stem
2	<i>Anacyclus pyrethrum</i>	Akkalkara	Asteraceae	leaves
3	<i>Butea monosperma</i>	Palas	Fabaceae/Papilionaceae	leaves
4	<i>Pongamia pinnata</i>	Karanja	Fabaceae	seeds

Table- 1) Showing list of plants selected for phytochemical screening.

Preparation of Crude Extract-

The coarsely powdered parts were soaked in methanol, ethanol and Aqueous and ethyl acetate solvents in a conical flask and left for 24 hours. The extracts were taken out and filtered using sterile filter paper and concentrated using water bath.

Working Crude Extract solution-

Working concentration of 25mg/ ml were prepared by dissolving respective amount of extract in one ml of methanol, ethanol and aqueous and ethyl acetate solvents in separate test tubes. Then extract was filtered through Whatmann paper No. 1 and solvent was removed by rotary vacuum evaporator (Buchi type-Superfit, Bangalore) under reduced pressure so as to get the crude extract. The concentrated extract was used for further study. (Mangesh S. Kharate.2017and 2018).

Phytochemical screening-

It is the process to know the presence or absence of number of chemical. Plant material is subject to preliminary phytochemical screening for the detection of various plant constituents.

Tests for detection of Alkaloids:-

Mayer reagent (potassium Mercuric iodide Solution)- Test solution produce cream colour precipitate with Mayer reagent which indicates the presence of alkaloids.

Wagner reagent (Iodine Potassium Iodide solution)- Test solution produce reddish brown Precipitate with Wagner reagent which indicates the presence of alkaloids.

Hager's reagent (Saturated solution of Picric acid)- Test solution produce yellow precipitate with hager reagent which indicates the presence of alkaloids.

Tests for detection of Carbohydrates

Molish's Test- To prepare this reagent, 10 gm of -naphthol was dissolved in 100 ml of 95% ethanol [18]. The reagent was added to aqueous and alcoholic extract as such as to hydrolyzed extract (heated with dil HCl on a water bath). Purple colour was obtained indicating the presence of carbohydrates.

Test for Proteins and Amino acids

Millon's test- To the test solution add about 2ml of millon reagents white precipitate was not observe it indicates absence of amino acid.

Biuret test- To the alcoholic extract of the powdered drug 1 ml of dilute sodium hydroxide was added. Followed by this one drop of very dilute copper sulphate solution was added. Violet color was not obtained indicating the absence of proteins.

Test for Glycosides

General test- (Test A) 200 mg of the powdered drug was extracted with 5 ml of dilute sulphuric acid by warming on a water bath, filtered and neutralised with 5% sodium hydroxide solution. Then 0.1 ml of Fehlings solution A and B were added, until it becomes alkaline and heated on a water bath for 2 minutes.

(Test B) 200 mg of the powdered drug was extracted with 5 ml of water instead of sulphuric acid. Boiled and equal amount of water was added instead of sodium hydroxide solution. Then 0.1 ml of Fehlings solution A and B were added, until it becomes alkaline and heated on a water bath for 2 minutes. The quantity of red precipitate formed in test A is greater than in test B indicating the presence of glycosides.

Test for Anthraquinones glycosides:

Borntrager's test- The powdered leaf was boiled with dilute sulphuric acid, filtered and to the filtrate benzene was added and shaken well. The inorganic layer was separated and ammonia solution was added slowly. No red color is observe in ammonical layer indicating the absence of anthracene derived glycosides.

Modified Borntrager's test- About 0.1 gm of the powdered leaf was boiled for two minutes with dilute hydrochloric acid and few drops of ferric chloride solution was added, filtered while hot and cooled. The filtrate was then extracted with benzene and the benzene layer was separated. Equal volume of dilute ammonia solution was added to the benzene extract and shaken well. Colour was not observed in ammonical layer indicating the not of anthracene derived glycosides.

Test for cyanogenetic glycosides

Small quantity of the powdered leaf was placed in a stoppered conical flask with just sufficient water to cover it. A sodium picrate paper strip was inserted through the stopper so that it was suspended in the flask and it was set aside for 2 hours in a warm place [19]. No Change in the colour of the sodium picrate paper was observed indicating the absence of cyanogenetic glycosides.

Test for cardiac glycosides

Keller Killiani test- About 1 gm of the powdered leaf was boiled with 10 ml of 70% alcohol for two minutes, cooled and filtered. To the filtrate 10 ml of water and 5 drops of solution of lead sub acetate were added and filtered. The filtrate was then extracted with chloroform and the chloroform layer was separated and evaporated to dryness. The residue was dissolved in 3 ml of glacial acetic acid containing a trace of ferric chloride. To this 3 ml of concentrated sulphuric acid was added to the sides of the test tube carefully. Reddish brown layer acquiring bluish green colour after standing was observed indicating the presence of deoxy sugars of cardiac glycosides.

Raymond Test - To the alcoholic extract of the leaf, hot methanolic alkali was added. Violet color was produced indicating the presence of cardiac glycosides.

Legal's Test -To the alcoholic extract of the powdered drug, pyridine and alkaline sodium nitro prusside solution were added. Red colour was formed indicating the presence of cardiac glycosides.

Coumarin glycosides

A small amount of powdered drug was placed in test tube and covered with a filter paper moistened with dilute sodium hydroxide solution. The covered test tube was placed on water bath for several minutes. Then the paper was removed and exposed to UV light. Green fluorescence was not observed indicating the absence of coumarin glycosides.

Test for Steroid and Triterpenoids

Salkowski Test- Few drops of concentrated sulphuric acid were added to the above solution, shaken well and set aside. The chloroform layer of the solution was not turned red in color indicating the absence of sterols.

Libermann-Burchard's Test- To the chloroform solution few drops of acetic anhydride was added and mixed well. 1 ml of concentrated sulphuric acid was added through the sides of the test tube and set aside for a while. A brown ring was not formed at the junction indicating the absence of sterols.

Test for Saponins-

About 0.5 gm of the powdered drug was boiled gently for 2 minute with 20 ml of water and filtered while hot and allowed to cool. 5 ml of the filtrate was then diluted with water and shaken vigorously. Frothing occurred indicating the presence of saponins.

Test for Tannins-

To the aqueous of the powdered drug, few drops of ferric chloride solution were added. Bluish black color was produced, indicating the presence of tannins.

Test for Flavonoids

Shinoda Test- A little amount of the powdered drug was heated with alcohol and filtered. To the alcoholic solution a few magnesium turnings and few drops of concentrated hydrochloric acid were added and boiled for 5 minutes. Purple color was obtained indicating the presence of flavonoids.

Alkaline reagent test- To the alcoholic extract of the powdered drug, few drop of sodium hydroxide solution was added. Yellow color formed, turning to colorless on addition of few drops of dilute acid indicating the presence of flavonoids.

Zinc Hydrochloride Test- To the alcoholic extract, mixture of zinc dust and concentrated hydrochloric acid was added. Formation of red color indicating the presence of flavonoid. Qualitative phytochemical analysis of total extracts was carried out using standard procedures to detect flavonoids, alkaloids, triterpenoids, glycosides, steroids, saponins, fixed oils and fats, proteins, tannins, phenolic compounds (Khandelwal K.R.1999).

III. RESULTS AND DISCUSSION

Plants contain many novel compounds with medicinal values which need scientific exploration. Phytochemical are very important in medicine and constitute most of the valuable drugs. Several chemicals which are derived from plants acts as a drug that is currently used in more countries in the world. The phytochemical activity of *A.vasica* proved to be presence of variety of phytochemicals.

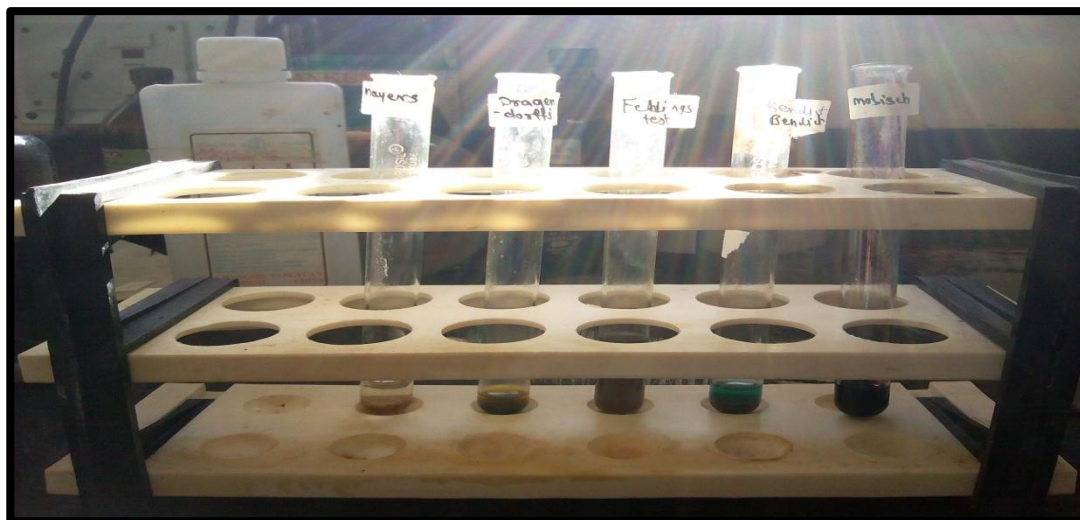


Figure -5) Showing phytochemical testing of *A. vasica*



Figure -6) Showing phytochemical testing of *A. pyrethrum*

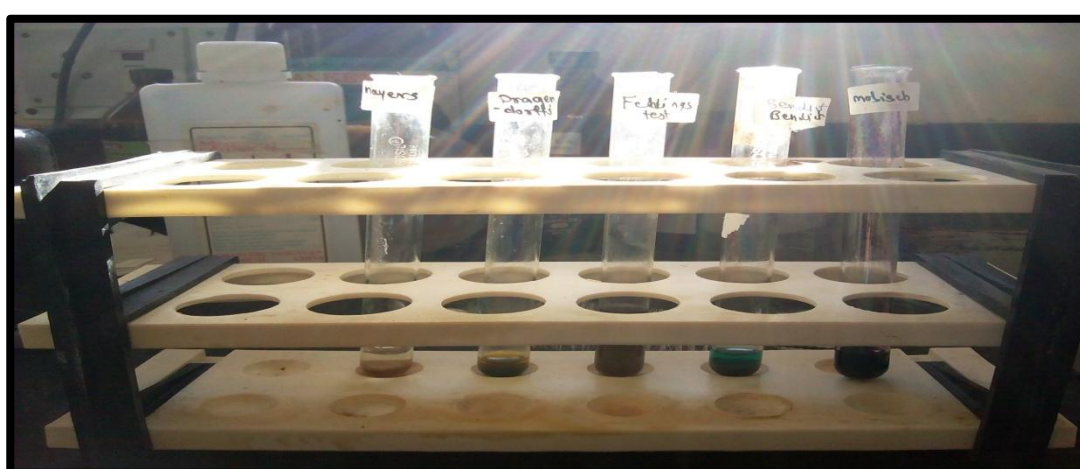


Figure -7) Showing phytochemical testing of *B. monosperma*



Figure -8) Showing phytochemical testing of *P. pinnata*

The alkaloids, proteins, glycosides were present. saponins, triterpenoids were absent. Due to presence of main phytochemicals antibacterial activity might be showed. The phytochemical analysis also showed alkaloids, proteins. But the carbohydrates were absent. These phytochemicals were might be responsible for antibacterial activity. The *B. monosperma* extracts of methanol, ethanol, aqueous and ethyl acetate was tannins, glycosides, proteins and carbohydrates were present. The steroids were absent. The *P. pinnata* extracts showed presence of important phytochemicals such as alkaloids, proteins, glycosides.

Phytochemicals	Presence or absence of phytochemical present in following plant crude extract															
	<i>A. vasica</i>				<i>Anacyclus pyrethrum</i>				<i>Butea monosperma</i>				<i>P. pinnata</i>			
	ME	EE	AQ	EA	ME	EE	AQ	EA	ME	EE	AQ	EA	ME	EE	AQ	EA
Alkaloids																
a. Mayer's test	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
b. Wagner's test	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
c. Hagers test	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Carbohydrates a. Molish test	-	-	-	-	-	-	-	-	+	+	+	+	-	-	-	-
Test for protein and amino acids																
a. Millons test	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
b. Biuret test	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Test for glycosides	+	+	+	+	+	+	+	+	+	+	+	+	-	-	-	-
Test for anthroquinone glycosides.																
a. Bomtragers test	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
b. Modified bomtragers test.	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Cynogenetic glycosides	+	-	-	+	+	-	-	+	+	-	-	+	+	-	-	+
Cardiac glycosides																
a. Raymond test	-	-	-	-	-	-	-	-	+	++	++	+	-	-	-	-
b. Legals test	-	-	-	-	-	-	-	-	-	+	+	+	-	-	-	-
Coumarin glycosides	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Steroids and triterpenoids	-	+	-	-	-	+	-	-	-	+	-	-	-	+	-	-
Liebermann-Burchard test	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Test for saponin	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Test for tannins																
a. Shinoda test	+	+	+	+	+	+	+	+	+	+	+	+	-	-	-	-
b. Alkaline reagent test	+	+	+	+	+	+	+	+	+	+	+	+	-	-	-	-
Zinc hydrochloric test.	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-

Table- 2) Showing Phytochemical analysis of crude extract of selected medicinal plants.

IV. CONCLUSION

Phytochemicals found in six medicinal leaf ethyl acetate extracts of plants indicates their potential as a source of principles that may supply novel medicines. Further studies are therefore suggested to ascertain their antibacterial and antifungal activities. Furthermore, isolation purification and characterization of the phytochemicals found present will make interesting studies.

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