Phytochemical analysis of floral extract of Some medicinal plants used by local people of Ranchi district, Jharkhand (India) used to cure skin diseases

[1]Vikram Kumar Sharma and [2]Dr. Rashmi Mishra

[1]Research Scholar, Department of Botany, Dr. Shyama Prasad Mukherjee University Ranchi (Jharkhand)

[2]Assistant Professor, Department of Botany, Dr. Shyama Prasad Mukherjee University Ranchi (Jharkhand)

ABSTRACT : In this present study floral extract of five medicinal plants viz., *Acacia nilotica* (Linn.) Delile, *Butea monosperma* (Lam.) Taub., *Woodfordia fruticosa* Kurz , *Azadirachta indica* and *Millettia pinnata* L. which are used to cure skin disease was qualitative phytochemically analysed with aqueous, ethanol and methanol extract. Qualitative phytochemical analysis was performed for the test of proteins, carbohydrates, phenol-tannins, flavonoides, saponin, glycosides, steroids, phlobatannins, alkaloids and terpenoides with the help of standard protocol. The qualitative phytochemical analysis revealed the presence of protein, carbohydrates, phenol and tannin, saponin, glycosides, steroids, alkaloids and terpenoids in extracts of flowers. On the basis of presence of active phytochemical, the present paper provide justification for the use of medicinal plants by the local inhabitants of Ranchi district, Jharkhand (India) to cure skin diseases.

Keywords: Ethnobotany, local inhabitant, medicinal plants, skin diseases, phytochemical analysis.

I. Introduction

A large number of medicinal plants are claimed to be useful in skin disease in all traditional system of medicine and folklore. While these plant remedies are being used orally and by local application since ancient time.It affects more than 60% of the general population[1].Drugs discovery from medicinal plants led to the isolation of early drugs such as cocaine, codeine, digitoxine, and quinine, in addition to morphine, of which some are still in use [2].

The World Health Organization (WHO) has outlined herbal medicine as culminated labeled medicinal products that incorporate lively ingredients as aerial or underground accessories of plants or other plant fabric[3]. Skin disease refers to disorders of predominantly the superficial layers of the skin. The prevalence of skin disease in any region or country depends on various factors, such as genetics, racial constitution, social and hygienic standards, customs and occupations. Transmissible skin diseases are observed in people who are living under poor socioeconomic and unhygienic conditions[4]. In India there is a significant incidence of infectious disorders in rural communities because of underdeveloped economy and social backwardness[5]. Up to 80% of the population suffering from skin problems may not seek medical help[6]. The common skin problems are Acne, Burn, scars, Psoriasis, Scabies, Skin grafting, Vitiligo, Pediculosis, Herpes simplex infection, Varicella, Herpes Zoster, Erythema, Urticaria etc. They are found in children, young and adults as well as in old persons. Usually for peak level skin disorder, the therapy of skin problems is longer for complete removal of problems. In all over the world use of drug like Benzoyl Peroxides, Proactive, Antibiotics, Retin-A, Oral retinoid, Salicylic acid, Anti-Histaminic, Minerals and Vitamins, Steroids, Analgesic are of more interest for skin specialist for the modern treatment. But the herbal medicine is becoming popular due to toxicity and side effects of allopathic medicines[7]. Most important of such compounds are alkaloids, tannins, flavonoids, terpenoids, saponins and phenolic compounds. Pharmacists are interested in these compounds because of their therapeutic performance and low toxicity[8].

Due to their natural origin and low toxicity, phenolic compounds are a promising tool in eliminating the causes and effects of skin aging, skin diseases, and skin damage, including wounds and burns. Hence, present paper emphasize on evaluation and characterization of five plants viz. *Millettia pinnata* L., *Acacia nilotica* (Linn.) Delile and *Butea monosperma* (Lam.) Taub. belonging to Fabaceae family, *Wood fruticosa* Kurz. belongs to Lythraceae family and Azadirachta indica belonging to *Meliaceae* family[9]. Plant constituents against a number of skin diseases based on their traditional claims of the plants.(Figure 1, 2 ,3,4,and 5 respectively).

Fig 1: *Acacianilotica* (Linn.) Delile figure

Fig 2: *Butea monosperma* (Lam.) Taub

Fig 3: *Woodfordia fruticosa* Kurz

Fig 4 : Azadirachta indica

Fig 5 : Millettia pinnata L.

MATERIALS AND METHODS

The plant materials were shade dried until all the water molecules evaporated and plants became well dried for grinding. After drying, the plant materials were ground well using mechanical blender into fine powder and transferred into airtight containers with proper labeling for future use.

PREPARATION OF PLANT EXTRACTS

Hot water extraction: 5gm of dried finely powdered plant material was taken in a beaker and 200ml of distilled water was added. The mixture was heated on a hot plate with continuous stirring at 30º-40ºC for 20 minutes. Then the water extract was filtered through filter paper and the filtrate was used for the phytochemical analysis. The water extract was kept in refrigerator when not in use.

SOLVENT EXTRACTION

Crude plant extract was prepared by Soxhlet extraction method. About 20gm of powdered plant material was uniformly packed into a thimble and extracted with 250ml of different solvents separately. Solvents used were methanol and ethanol. The process of extraction continues for 24 hours or till the solvent in siphon tube of an extractor become colorless. After that the extract was taken in a beaker and kept on hot plate and heated at 30-40ºC till all the solvent got evaporated. Dried extract was kept in refrigerator at 4ºC for their future use in qualitative phytochemical analysis. The extract was tested for the presence of bioactive compounds by using following standard methods[10,11.12]

PROTEINS MILLON'S TEST

Crude extract when mixed with 2ml of Millon's reagent, white precipitate appeared which turned red upon gentle heating that confirmed the presence of protein.

NINHYDRIN TEST

Crude extract when boiled with 2ml of 0.2% solution of Ninhydrin, violet colour appeared suggesting the presence of amino acids and proteins.

Test for carbohydrates Fehling's test

Equal volume of Fehling A and Fehling B reagents were mixed together and 2ml of it was added to crude extract and gently boiled. A brick red precipitate appeared at the bottom of the test tube indicated the presence of reducing sugars.

BENEDICT'S TEST

Crude extract when mixed with 2ml of Benedict's reagent and boiled, a reddish brown precipitate formed which indicated the presence of the carbohydrates.

IODINE TEST

Crude extract was mixed with 2ml of iodine solution. A dark blue or purple coloration indicated the presence of the carbohydrate.

TEST FOR PHENOLS AND TANNINS

Crude extract was mixed with 2ml of 2% solution of FeCl3. A blue-green or black coloration indicated the presence of phenols and tannins.

TEST FOR FLAVONOIDS SHINODA TEST

Crude extract was mixed with few fragments of magnesium ribbon and concentrated HCl was added drop wise. Pink scarlet colour appeared after few minutes which indicated the presence of flavonoids.

ALKALINE REAGENT TEST

Crude extract was mixed with 2ml of 2% solution of NaOH. An intense yellow colour was formed which turned colourless on addition of few drops of diluted acid which indicated the presence of flavonoids.

TEST FOR SAPONINS

Crude extract was mixed with 5ml of distilled water in a test tube and it was shaken vigorously. The formation of stable foam was taken as an indication for the presence of saponins.

TEST FOR GLYCOSIDES LIEBERMANN'S TEST

Crude extract was mixed with each of 2ml of chloroform and 2ml of acetic acid. The mixture was cooled in ice. Carefully concentrated H2SO4 was added. A colour change from violet to blue to green indicated the presence of steroidal nucleus, i.e., glycone portion of glycoside.

SALKOWSKI'S TEST

Crude extract was mixed with 2ml of chloroform. Then 2ml of concentrated H_2SO_4 was added carefully and shaken gently. A reddish brown colour indicated the presence of steroidal ring, i.e., glycone portion of the glycoside.

KELLER-KILANI TEST

Crude extract was mixed with 2ml of glacial acetic acid containing 1-2 drops of 2% solution of $FeCl_3$. The mixture was then poured into another test tube containing 2ml of concentrated H_2SO_4. A brown ring at the interphase indicated the presence of cardiac glycosides.

TEST FOR STEROID

Liebermann's Test

2 ml Crude extract was mixed with 2 ml acetic acid (CH_2COOH) and then 1 ml of concentrated H_2SO_4 was added drop wise, the presence of blue green color indicates the presence of steroids

SALKOWSKI TEST

2 ml Crude extract was mixed with 2ml of chloroform and concentrated H_2SO_4 was added sidewise. A red colour produced in the lower chloroform layer indicated the presence of steroids.

LIEBERMANN'S BRUCHARD TEST

2 ml Crude extract is mixed with 2ml of chloroform. Then 2ml of each of concentrated H_2SO_4 and acetic acid were poured into the mixture. The development of a greenish coloration indicated the presence of steroids.

TEST FOR PHLOBATANNIN

2 ml Crude extract is mixed with 2ml of 1% HCl which gives red precipitate on gentle heating confirms the presence of phlobatannin.

TEST FOR ALKALOIDS

2 ml Crude extract was mixed with 2ml of 1% HCl and heated gently. Mayer's And Wagner's reagents were then added to the mixture. Turbidity of the resulting precipitate was taken as evidence for the presence of alkaloids.

TEST FOR TERPENOIDS

Crude extract was dissolved in 2ml of chloroform andevaporated to dryness. To this, 2ml of concentrated H_2SO_4 wasadded and heated for about 2 minutes. A grayish colour indicated thepresence of terpenoids.

RESULT AND DISCUSSION

Table 1: Qualitative phytochemical analysisof floral extract of three medicinal plants

S. No	Phytochemical test	*Acacia nilotica* (Linn.) Delile			*Butea monosperma* (Lam.) Taub.			*Woodfordia fruticosa* Kurz.			,***Azadirachta indica***			*Millettia pinnata* L		
		A.E	E.E	M.E	A.E	E.E	M.E	A.E	E.E	M.E	A.E	E.E	M.E	A.E	E.E	M.E
1	Proteins	+	+	+	+	+	+	_	+	+	+	+	+	+	+	+
2	Carbohydrates	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
3	Phenol&Tannins	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
4	Flavonoids	+	_	+	_	_	+	+	+	+		+		_	_	+
5	Saponin	_	_	+	+	+	+	+	+	+		+		_	_	+
6	Glycosides	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
7	Steroids	+	+	+	+	+	+	_	+	+	+	+	+	+	+	+
8	Phlobatannins	_	_	+	_	_	_	_	_	_		+	-	_	_	+
9	Alkaloids	_	+	+	+	+	+	_	+	+	+	_	-	+	+	+
10	Terpenoids	_	_	_	_	+	+	+	+	+		_	+	+	+	_

A.E= Aqueous extract, E.E = Ethanolic extract, M.E= Methanolic extract, (+) = Present, (-) = Absent

RESULT

- In *Acacia nilotica* (Linn.) Delile It has been observed that proteins, carbohydrate, phenol & tannins, glycosides and steroids were present in all the three extracts whereas saponin and phlobatannins were found only in methanolic extract. Flavonoids are present only in ethanolic plant extract. Alkaloid was found absent in aqueous extract. It was observed that terpenoids were absent in all three plant extract.
- In *Butea monosperma* (Lam.) Taub. Proteins. carbohydrates, phenol & tannins, saponin glycosides, steroids and alkaloids were observed to be present in all the three extract while it was found that flavonoid was only present in methanolic extract. Phlobatannin was absent in all the three exracts and it was observed that terpenoids was present in ethanolic and methanolic plant extracts.
- In *Woodfordia fruticosa* Kurz. Carbohydrate, phenol & tannins, flavonoids, saponins were observed to be

present in all the three extracts whereas protein, steroids and alkaloids were present in aqueous extract and it was found that phlobatannin was absent in all the three extracts.

- In *Azadirachta indica* Proteins. carbohydrates, phenol & tannins, saponin glycosides, steroids and alkaloids were observed to be present in all the three extract whereas saponin and phlobatannins were found only in methanolic extract. Flavonoids are present only in ethanolic plant extract. . Alkaloid was found absent in aqueous extract. It was observed that terpenoids were present in methonolic plant extract
- In *Millettia pinnata* L It has been observed that proteins, carbohydrate, phenol & tannins, glycosides and steroids were present in all the three extracts whereas saponin and phlobatannins were found only in methanolic extract. Flavonoid was only present in methanolic extract .Phlobatannin was absent in all the three exracts and it was observed that terpenoids was present in ethanolic and methanolic plant extracts

DISCUSSION

From the result obtained by the phytochemical analysis of five medicinal plants, it could be seen that carbohydrates, phenol & tannins and glycosides were present in all the three plants. Several reports revealed that medicinal plants are rich in phenolic compounds and have antioxidant properties[13,14]. Phenolic compounds also possess potent antifungal, antiviral and antibacterial activity. It is also mentioned that phytochemical analysis on plants extracts revealed the presence of constituents that are known to exhibit medicinal as well as physiological activities. The plant extracts also revealed to contain phenol & tannins. It was reported that tannins contribute property of astringency i.e. fasten the healing of wounds and inflamed mucous membrane and have received considerable attention in the fields of nutrition, health and medicine, largely due to their physiological activity, such as antioxidant, antimicrobial and anti- inflammatory properties[15]. Flavonoids were found present in methanolic plant extract of all the five plants. Several studies shows that flavonoids are a group of polyphenolic compounds which influence the radical scavenging, inhibition of hydrolytic and oxidative enzymes and also act as anti- inflammatory agent[16]. Saponins was found present in methanolic plant extract of *Acacia nilotica* (Linn.) Delile while it was found in all the five plant extract. According to several studies the plant extracts were also revealed to contain saponins which are known to produce inhibitory effect on inflammation[17]. Steroids have been reported to have antibacterial properties[18]. Terpenoids are active against bacteria[19,20,21].

ACKNOWLEDGEMENT

The first author is highly grateful to Dr. I P Gupta, Head, Department of Botany, Dr DSPMU University,Ranchi. Dr.Rashmi Mishra, Assistant Professor, Department of Botany for providing support at al stage of research work. The first author is also grateful to UGC for financial assistance in the form of fellowship under the CSIR-UGC NET JRF scheme.

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