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Research Article

Pharmacognostical and Phytochemical Study on the Leaves of Methanolic Extract of *Blumea membranacea*

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Abstract

An oxidative stress causes large number of diseases which may affect tissue in cells. Antioxidants neutralized the oxidative stress and help in controlling diseases. However, they may enhance the immune systems' so that cells defend themselves against the diseases caused by free radicals. There are numerous sources of antioxidants such as Vitamins A, C, and E. moreover, selenium protect against cancer, heart diseases as well. The substance having antioxidant property may obtain from both natural and synthetic sources. Synthetic antioxidants are useful and effective but they are harmful for us on the other hand natural sources are available in huge amount. *Blumea membranacea* is one of the medicinal plants which are widely grown in Asian counties including India is a major source of antioxidant. However, all parts (stem, roots and leave) of plant are used as a medicine leaves are more commonly used and they cures several diseases such as cancer, gynecological etc. in addition a recent study described that, it is a strong anti-viral and anti-fungal agent. The essential oils found in leaves of *B. membranacea* has being used in several industries such as pharmaceutical, food, and cosmetic. By this way, essential oil of this medicinal plant contributes an important part in growth in economy of developing countries like India. Hence use of *B. membranacea* based herbal medicine could be effective technology for treatment of several diseases and enhancement of antioxidant in humans.

Keywords: Antioxidant, *Blumea membranacea*, Herbal medicine, eco-friendly technique

Article Info

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1. Introduction

History of use of traditional herbal drugs

Natural products and traditional medicines are of great importance. Such forms of medicine as traditional Chinese medicine, Ayurveda, Kampo, traditional Korean medicine, and Unani have been practiced in some areas of the world and have blossomed into orderly-regulated systems of medicine. This study aims to review the literature on the relationship among natural products, traditional medicines, and modern medicine, and to explore the possible concepts and methodologies from natural products and traditional medicines to further develop drug discovery.

The unique characteristics of theory, application, current role or status, and modern research of eight kinds of traditional medicine systems are summarized in this study. Although only a tiny fraction of the existing plant species have been scientifically researched for bioactivities since 1805, when the first pharmacologically-active compound morphine was isolated from opium, natural products and traditional medicines have already made fruitful contributions for modern medicine. When used to develop new drugs, natural products and traditional medicines have their incomparable advantages, such as abundant clinical experiences, and their unique diversity of chemical structures and biological activities. Since prehistoric times, humans have used natural products, such as plants, animals, microorganisms, and marine organisms, in medicines to alleviate and treat diseases. According to fossil records, the human use of plants as medicines may be traced back at least 60,000 years. The use of natural products as medicines must, of course, have presented a tremendous challenge to early humans. It is highly probable that when seeking food, early humans often consumed poisonous plants, which led to vomiting, diarrhea, coma, or other toxic reactions—perhaps even death. However, in this way, early humans were able to develop knowledge about edible materials and natural medicine.

Traditional medicines (TMs) make use of natural products and are of great importance. Such forms of medicine as traditional Chinese medicine (TCM), Ayurveda, Kampo, traditional Korean medicine (TKM), and Unani employ natural products and have been practiced all over the world for hundreds or even thousands of years, and they have blossomed into orderly-regulated systems of medicine. In their various forms, they may have certain defects, but they are still a valuable repository of human knowledge. In the case of China, Western medicine was introduced in the sixteenth century, but it did not undergo any development until the nineteenth century. Before that, TCM was the dominant form of medical care in the country. Now TCM still plays an important role in China, and it is constantly being developed. TCM is based on 5000

years of medical practice and experience, and is rich in data from “clinical experiments” which guarantee its effectiveness and efficacy. It has developed techniques with respect to such areas as correct dosage, methods of preparing and processing materials, and the appropriate time to collect the various medicinal parts of plants. It is notable that there is increasing convergence between TCM and modern medicine. With the development of modern technology, it has become possible to determine the pharmacology and mechanisms of action of many Chinese herbs, and TCM has become comprehensible in terms of modern medicine. With advances in the theoretical background, therapeutic principles, associated technologies, and understanding of the life sciences, a clearer understanding of the active compounds of TCM has become possible. At the beginning of the nineteenth century, the era of “modern” drugs began. In 1805, the first pharmacologically-active compound morphine was isolated by a young German pharmacist, Friedrich Sertürner, from the opium plant. Subsequently, countless active compounds have been separated from natural products. Among them, some follow their traditional uses and the others do not. Later, the development of synthetic techniques led to a significant reduction in the importance of natural products, and there were concerns that the use of some natural products for medicinal purposes might be completely banned. However, natural products are important for the development of new drugs, and these products have been in constant use. Some type of medicines, such as anticancer, antihypertensive, and antimigraine medication, have benefited greatly from natural products.

The development of new drugs relying purely on modern technology appears to be reaching something of a limit. In developing new drugs, the pharmaceutical industry has tended to adopt high-throughput synthesis and combinatorial chemistry-based drug development since the 1980s; however, the considerable efforts made in this direction have not resulted in the expected drug productivity. Some large pharmaceutical companies are facing great challenges to develop new products. Over the past dozen years, increasing attention has accordingly been paid to natural products in the search for novel drugs in combination with new technology, such as high-throughput selection. Natural products, which have evolved over millions of years, have a unique chemical diversity, which results in diversity in their biological activities and drug-like properties. Those products have become one of the most important resources for developing new lead compounds and scaffolds. Natural products will undergo continual use toward meeting the urgent need to develop effective drugs, and they will play a leading role in the discovery of drugs for treating human diseases, especially critical diseases.

2. Natural Products

Natural products have a wide range of diversity of multi-dimensional chemical structures; in the meantime, the utility of natural products as biological function modifiers has also on considerable attention. Subsequently, they have been successfully employed in the discovery of new drugs and have exerted a far-reaching impact on chemobiology. From the past century, the high structural diversity of natural products have been realized from the perspective of physical chemistry. Their efficacy is related to the complexity of their well-organized three-dimensional chemical and steric properties, which offer many advantages in terms of efficiency and selectivity of molecular targets. As a successful example of drug development from natural products, artemisinin and its analogs are presently in wide use for the anti-malaria treatment. This shows how research using natural products has made a significant contribution in drug development. Among anticancer drugs approved in the time frame of about 1940–2002, approximately 54% were derived natural products or drugs inspired from knowledge related to such. For instance, the Vinca alkaloids from *Catharanthus roseus*, and the terpene paclitaxel from *Taxus baccata*, are among successful anticancer drugs originally derived from plants. During the period between 1981 and 2002, the application of natural products in the development of new drugs—especially in the search for novel chemical structures—showed conspicuous success. In that 22-year time frame, drugs derived from natural products have been significant. That is especially true in the case of antihypertensives, where about 64% of newly-synthesized drugs have their origins in natural product structures. Considering their incomparable chemical diversity and novel mechanisms of action, natural products have continued to play a pivotal role in many drug development and research programs. With time, those natural products have undergone interesting and meaningful developments in their ability to interact with numerous, varied biological targets, and some have become the most important drugs in health care system. For example, plants, microorganisms, and animals manufacture small molecules, which have played a major role in drug discovery. Among 69 small-molecule new drugs approved from 2005 to 2007 worldwide, 13 were natural products or originated from natural products, which underlines the importance of such products in drug research and development. Over the past 50 years, there has been a great diversity of new drugs developed using high-throughput screening methods and combinatorial chemistry; however, natural products and their derived compounds have continued to be highly-important components in pharmacopoeias. Of the reckoned 250,000–500,000 existing plant species, only a tiny proportion has been scientifically researched for bioactivities. Therefore, there is great potential for future discoveries from plants and other natural products which,

thus, offer huge potential in deriving useful information about novel chemical structures and their new types of action related to new drug development.

3. Traditional Medicines(TM)-

TM is the oldest form of health care in the world and is used in the prevention, and treatment of physical and mental illnesses. Different societies historically developed various useful healing methods to combat a variety of health- and life-threatening diseases. TM is also variously known as complementary and alternative, or ethnic medicine, and it still plays a key role in many countries today. The medicaments used in TM are mostly derived from natural products. In TM, “clinical trials” have been conducted since ancient times. In the case of TCM, considerable experience and advances have been accumulated and developed over the past thousands of years with respect to methods of preparation, selection of herbs, identification of medicinal materials, and the best time for obtaining various different plants. Appropriate processing and dose regulation are urgently needed in TCM to improve drug efficacy and reduce drug toxicity. Considerable amounts of data have been acquired through clinical experiments, and in this way TM has assisted in the development of modern drugs. Through its use of natural products, TM offers merits over other forms of medicine in such areas as the following: discovery of lead compounds and drug candidates; examining drug-like activity; and exploring physicochemical, biochemical, pharmacokinetic, and toxicological characteristics. If any form of TM is applied successfully, it may surprisingly assist in the development of new drugs, thereby resulting in many benefits, such as significant cost reductions. TCM is now an inseparable part of the Chinese public health system. In recent years, TCM has gradually gained considerable approval as a complementary or alternative medicine in Western countries. Chinese herbal medicine, which is the most important component of TCM, is currently used in the health care of an estimated 1.5 billion people worldwide. It should be noted that in TCM, several herbs and ingredients are combined according to strict rules to form prescriptions, which are referred to as formulas (fang ji in Chinese). Commonly, a classic formula is composed of four elements—the “monarch”, “minister”, “assistant”, and “servant”—according to their different roles in the formula, each of which consists of one to several drugs. Ideally, these drugs constitute an organic group to produce the desired therapeutic effect and reduce adverse reactions.

Kampo is the TM of Japan. Between the fifth and sixth centuries, TCM was introduced to Japan from China; since then, TCM has been significantly altered and adapted by Japanese practitioner to meet their particular circumstances and gradually evolved into Kampo. A recent study has found that some physicians in Japan use Kampo

medicines in their daily practice—sometimes as the preferred medication. Together with radiotherapy or chemotherapy, some Japanese physicians frequently utilize Kampo medicines in treating cancer patients. This indicates how modern Western medicine can be well integrated with TM. As the use of Kampo continues to rise in conjunction with Western medicine, there is growing realization of the urgent need to study the interactions between these two types of medicines. Unani is an ancient Greek holistic medical system with a history that can be traced back 2500 years. Since the mid-1970s, when the WHO began to place a greater focus on TM, Unani has attracted considerable attention all over the world, especially in India, where it has been integrated into the national health care system. It was reckoned by WHO that a large quantity of people in the world still depend on TMs for health care. The current status of TM differs in different countries. In 2012, the total value of the TCM industry was equivalent to around one-third of the total for China's pharmaceutical industry. It has been determined that 80% of the population in Africa makes use of TM—either alone or in conjunction with conventional medicine. By contrast, traditional Aboriginal medicine in Australia is in danger of vanishing owing to the prevalence of conventional medicine. In the case of Israel with its ethnic diversity, modern medicine is prevailing, and TM is declining. Many practitioners of Western medical science think such TM systems as being short of reliability; however, they are adopted by the majority of people in the world. It is possible to produce remarkable synergy and yield great benefits in developing reformed medicines and new drugs by connecting powerful modern scientific techniques and methods with the reasonable ethnobotanical and ethnomedical experiences of Traditional medicine.

4. Drugs Developed from Traditional Medicines that Follow the Traditional Uses

Traditional medicines is too valuable to be ignored in the research and development of modern drugs. Though it has an enigmatic character, there are also wide contexts for its use in terms of non-Western medical technology or activities. In TM, a single herb or formula may contain many phytochemical constituents, such as alkaloids, terpenoids, flavonoids, etc. Generally speaking, these chemicals function alone or in conjunction with one another to produce the desired pharmacological effect. It is notable that a lot of plant-originated drugs in clinical medicine today were derived from TM. In addition, it has been demonstrated that the many valuable drugs derived from plants were discovered through their application in TM. Almost 20 years ago, a thorough investigation of the pharmacopoeias of developed and developing nations and the associated world scientific literature was conducted as part of the WHO's TM Program. The aim of that study was to determine whether TM really had inspired modern drug

discoveries and whether there was any correlation between the current use of various compounds and their application in TM. The study focused on various compounds used in drugs derived from plants in different countries, and it established that TM had indeed played a significant role in developing effective new drugs. That study focused on 122 compounds, 80% of which were found to be related to pharmaceutical effects in folk medicine, and it was determined that these compounds originated from 94 plant species. Early in China's Jin Dynasty, Doctor Hong Ge (AD 284–384) recorded the efficacy and related details of *Artemisia annua* L. in treating malaria in his book *Zhou Hou Bei Ji Fang*. That is the earliest record anywhere of treating malaria with *Artemisia annua* L., and it shows that Chinese physicians 1700 years ago had reached a sophisticated level of medical treatment. Artemisinin is known as qinghaosu in Chinese, and its study has made significant progress, including the synthesis of new artemisinin analogs and derivatives, and research efforts into the biological activities and related mechanisms. As a result, artemisinin, as well as its effective derivatives, are extensively applied throughout the world as new-type anti-malarial drugs. The discovery of artemisinin can be traced back to the 1960s, when tropical malaria was a serious problem during the Vietnam War. North Vietnam requested China to help tackle the malaria problem. The Chinese government approved a project for malaria control and drug research in 1967. The research group made its investigations and carried out a large-scale search of the literature on the subject. As part of the phytochemical and pharmacological research effort, a lot of Chinese herbal medicines were screened and investigated with respect to their toxicity or efficacy. Eventually artemisinin was derived from *Artemisia annua* L. in 1972. Artemisinin is quite different from previously-used antimalarial drugs, such as chloroquine, in that it has a novel structure, with a sesquiterpene lactone bearing a peroxy group, and it does not contain nitrogen heterocycles. Compared with previous antimalarial drugs, artemisinin has the merit of high efficiency, quick effect, and low toxicity. Artemisinin is effective in treating various forms of malaria, such as falciparum and cerebral malaria, which are resistant to chloroquine, and its mechanism of action is different from traditional antimalarial drugs. The discovery of artemisinin was a great success for TCM at a special period in China's history, and it was achieved through a well-organized team of hundreds of researchers. Since that breakthrough, scientists have conducted comprehensive research in such areas as pharmaceutical chemistry, organic synthetic chemistry, and chemical biology. Through etherification and esterification, they have produced a series of well-known new drugs, such as artemether and artesunate. Those drugs have improved efficacy and solubility, which are of benefit for patients receiving oral or intravenous administration and have overcome the high parasite recrudescence rate and low

solubility of artemisinin. Most importantly, one of these scientists, Youyou Tu, was just awarded the 2015 Nobel Medicine Prize for her significant devotion in discovering artemisinin. The discovery of artemisinin illustrates how TCM constitutes a great store of knowledge about natural products, such as Chinese herbs, and holds much future promise. The discovery of successful new drugs can proceed by profiting from this knowledge.

5. Drugs Developed from Natural Products

In clinical practice in China in the 1960s, it was found that *Schisandra chinensis* (Turcz.) Baill.—a traditional Chinese herb—had obvious enzyme-reducing and hepatoprotective effects. Chinese scientists then began isolating the chemical constituents of *S. chinensis*. In the subsequent total chemical synthesis and pharmacodynamic study of schisandrin C (which is one of the compounds of *S. chinensis*), researchers found that the intermediate compound bifendate had a stronger pharmacological activity and that the cost of preparation was low. They discovered that it may be used to lower the enzyme content in the treatment of hepatitis B virus.



LITERATURE SURVEY OF THE PLANT

Blumea membranacea DC (Local name Almish) grows in waste places, both in shady and sunny situations, sometimes in pure stands. The ability to utilize oxygen has provided humans with the benefit of metabolizing carbohydrates, fats and proteins for energy; however, it does not come without a cost. A paradox in metabolism is that, while the vast majority of complex life on Earth requires oxygen for its existence, oxygen is a highly reactive molecule that can damage living organisms by producing reactive oxygen species. Oxygen is a highly reactive atom that is capable of becoming part of potentially damaging molecules commonly called “free radicals”. The antioxidant activity of essential oils is another a biological property of great interest because they may preserve foods from the toxic effects of oxidants. Plant essential oils as antioxidants were researched in detail with the view to investigating their protective role for highly unsaturated lipids in animal tissues. Moreover, essential oils being also able to scavenge free radicals may play an important role in some disease prevention such as

brain dysfunction, cancer, heart disease and immune system decline

Scientific Classification

Kingdom:	Plantae
<i>Clade:</i>	Angiosperms
<i>Clade:</i>	Eudicots
<i>Clade:</i>	Asterids
Order:	Asterales
Family:	Asteraceae
Supertribe:	Helianthodae
Tribe:	Inuleae
Genus:	<i>Blumea</i>
Type species	
<i>Blumea membranacea</i>	
Synonyms	
<ul style="list-style-type: none"> • <i>Placus</i> Lour. • <i>Bilevillea</i> Vaniot • <i>Leveillea</i> Vaniot • <i>Blumea</i> section <i>Apterae</i> DC. • <i>Conyza</i> subgenus <i>Blumea</i> (DC.) 	

2. Material and Methods

Botanical Collection

Blumea membranacea plant leaves was collected from Itaura, Chandeshwar, Azamgarh, Uttar Pradesh, India during 2018.

Morphology study

Different parameter were studied in macroscopic evaluation of the leaves of *Blumea membranacea*, which are colour, odour, size and shape.

Microscopy study

Transverse section taken from the middle part of the leaves was observed. Microscopic studies were done by preparing a thin section of leaves of *Blumea membranacea*. The section was cleared with chloral hydrate solution and then stained with iodine solution.

Powder Microscopy

The microscopic powdered materials of leaves of the plant were also further treated iodine solution

Florescence Study

Coumarin, flavonoids and other different compounds produce specific florescence characteristics which helpful for preliminary chemical study as well as for standardization of specific plant materials. For florescence study, the powdered leaves were extracted with different solvents like petroleum ether (40-60°C), cyclohexane, ethyl acetate, acetone, methanol, water in cold maceration technique for 24 h; the sample were filtered and filtrate

was seen under ultra-violet rays at 366 nm for observing any specific fluorescence

Analytical parameters

In this regard, powdered material of and *Blumea membranacea* was mixed with different chemical agents like dilute hydrochloric acid, concentrated hydrochloric acid, dilute sulphuric acid, concentrated sulphuric acid, 10% potassium hydroxide, ferric chloride, distilled water, ammonia, iodine and observed under normal light and ultraviolet light (254 nm and 365 nm) for the effect of different chemical agents on powdered crude drug.

A small quantity of dried and finely powdered leaves sample was placed on a grease free microscopic slide and added 1-2 drops of freshly prepared solution, mixed by gentle tilting the slide and waited for 1-2 minutes. Then the slide was placed inside the UV viewer chamber and viewed in day light, short (254 nm) and long (365 nm) ultraviolet radiations. The colors observed by application of different reagents in various radiations were recorded.

Physico-chemical evaluation

Physico-chemical parameters such as the percentage of loss on drying (LOD), total ash, acid soluble ash, water soluble ash, foaming index, swelling index and water content in the powdered raw materials were determined by following the WHO guidelines (WHO 2002).

Phyto-chemical study

The dried and powdered leaves were subjected to preliminary phyto-chemical screening for qualitative detection of phytoconstituents. Air dried leaves were grinded and kept in air-tight container. The powdered leaves were macerated with methanol for seven days for three times. All solvents were mixed together and concentrated at vacuum to get solid extract.

Different solvent extractive value

This method determines the amount of active constituents in a given amount of medicinal Plant material when extracted with solvents. Different solvent extractive value like as a Chloroform, Acetone, Methanol, Petroleum ether (60-80°C) and Water. For determination of solvent extractive values 1gm of the air dried, coarsely powdered macerated with 25 ml of water close flask for 24 hours, shaking frequently during first 6 hours and allowing stand for 18 hours. Thereafter, filter rapidly taking precautions against loss of solvent; evaporate 25 ml of the filtrate to dryness in a tarred flat bottomed shallow dish, at 105°C and weight. The percentage of solvent soluble extractive with reference to air dried drug has to be calculated

Determined Ash Value

Total ash value

About 2gm of powdered drug was weighted accurately into a tarred silica crucible and incinerated at 450°C in muffle furnace until free from carbon. The crucible was cooled to room temperature and weighted. Percentage of ash was calculated with reference to air dried substance.

Acid soluble ash:

Ash obtained from total ash was boiled with 25ml of 2N HCl for few minutes and filtered through an ashless filter Paper. The filter paper was transferred into a tarred silica crucible and incinerated at 650°C in muffle furnace until free from carbon. The crucible was cooled and weighted. Percentage of acid insoluble ash was calculated with reference to air dried substance.

Water soluble ash

Ash obtained from total ash was boiled with 25 ml of distilled water for few minutes and filtered through an ashless filter paper. The filter paper was transferred into a tarred silica crucible and incinerated at 450°C in muffle furnace until free from carbon. The crucible was cooled and weighted. Percentage of water soluble ash was calculated with reference to air dried substance.

3. Results and Discussion

Morphology

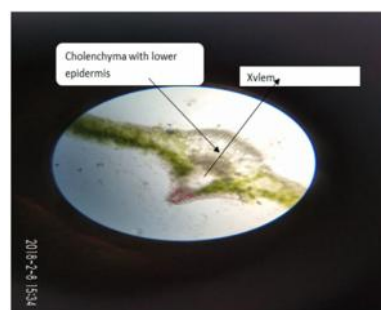
The Fresh leaves characters recorded are described below.

Shape of leaf: Large and palmate shaped.

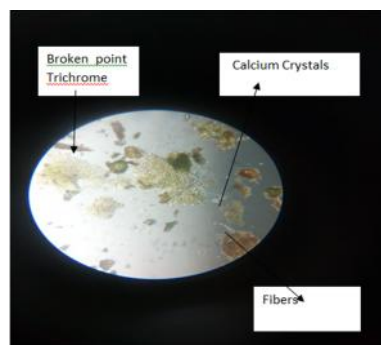
Color: The leaves is green in color

Taste: pungent

Size: Leaves are arranged opposite to one another and are 2.5 to 5cm. It is annual herb grows up to 40-60 cm. the flower color is yellow



Microscopy



Powder Microscopy

Phytochemical screening-

Detection of Alkaloids

Mayer test: On treatment with potassium mercuric iodide solution it gives creamish precipitate which indicates alkaloid is present.

Dragendorff test: Add potassium bismuth iodide solution to sample it gives reddish brown precipitate which indicates alkaloid is present.

Wagner test- Add potassium iodide or aqueous iodide solution to sample it gives reddish brown precipitate which indicates alkaloids is present.

Detection of Proteins-

Precipitation test- 5% HgCl₂ solution gives white colloidal precipitated form.

Detection of Phytosterols

Salkowski's test: Extracts were treated with chloroform and filtered. The filtrates were treated with few drops of conc. sulphuric acid, shaken and allowed to stand.

Appearance of golden yellow colour indicates the presence of triterpenes.

Detection of Flavonoids-

Shinoda test:

To the alcoholic solution of extracts, a few fragments of magnesium ribbon and conc. HCl were added. Appearance of magenta colour after few minutes indicates presence of flavonoids.

Detection of Saponins

Foam test:

Extracts were diluted with distilled water to 20 ml and this was shaken in a graduated cylinder for 15 min. Formation of 1 cm layer of foam indicates the presence of saponins.

Fluorescence Study for various solvents having extract of *Blumea membranacea* leaves

Sr. No.	Solvent	Fluorescence
1	Acetone	Light green
2	Aqueous	Light brown
3	Ethyl acetate	Green
4	Methanol	Light green
5	Petroleum ether	Light brown

Different solvent extractive value

Sr. No.	Solvent	Extractive value %
1	Acetone	4%
2	Chloroform	5%
3	Methanol	7%
4	Petroleum ether (40-60°C)	2%
5	Water	8%

Effect of Different Chemical Agents on *Blumea membranacea* powdered leaves

S.No.	Treatment	Normal Light	Ultra violet Light	
			254nm	366nm
1	Blank	Green	Black	Dark black
2	Powder +Dil. Hydrochloric acid	Greenish brown	Brown	Black
3	Powder+Conc. Hydrochloric acid	Greenish brown	Black	Black
4	Powder+Dil. Sulphuric acid	Greenish	Black	Brown
5	Powder+Conc. Sulphuric acid	Greenish brown	Black brownish	Brown
6	Powder+5%Potassiumhydroxide	Greenish brown	Black	Dark black
7	Powder+10%Potassiumhydroxide	Greenish brown	Brown	Dark black
8	Powder+ 5% Ferric chloride	Greenish brown	Black	Black
9	Powder+ Distill water	Greenish	Black	Dark black
10	Powder+ Ammonia	Greenish	Brown	Black
11	Powder+ Iodine	Greenish brown	Black	Dark black

Physical parameters for *Blumea membranacea* leaves

Sr. No.	Parameter	Result
1	Loss on drying	7%
2	Total ash	13.5%
3	Acid insoluble ash	7.4%
4	Water soluble ash	14.8%
5	Foaming index	+ve
6	Swelling index	1.3 cm

Phytochemical Screening of *Blumea membranacea*

S. No.	Chemical Constituent	Chemical Tests	Result
1	Alkaloids	Dragendorff's Mayer's Wagner's	++ ++ ++
2	Protein	Precipitation(Lead acetate solution)	++
3	Amino Acid	Ninhydrin	++
4	Steroid	Salkowski reaction	++
5	Flavonoids	Shinoda	++
6	Tannins and Phenolic	Lead acetate solution Dilute HNO ₃	++ ++
7	Saponin Glycoside	Foam	++

CALCULATION**For *Blumea membranacea*****1. Extractive yield of different extracts:**

$$\% \text{ YIELD OF EXTRACT} = \frac{\text{Weight of solid extract}}{\text{Weight of powder taken for extraction}} \times 100$$

I. Determination of % yield of extract with water:

Weight of powder taken = 1gm

Quantity of water = 25 ml

Weight of empty petry dish = 112.25gm

Weight of petry dish after evaporating the filtrate = 112.gm

Weight of extract = 112.33-112.25gm = 0.08gm

% YIELD OF EXTRACT WITH WATER = 8%**II. Determination of % yield of extract with petroleum ether:**

Weight of powder taken = 1 gm

Quantity of petroleum ether= 25 ml

Weight of empty petry dish = 94.33gm

Weight of perty dish after evaporating the filtrate = 94.35gm

Weight of extract = 94.35-94.33gm = 0.02gm

% YIELD OF EXTRACT WITH PETROLEUM ETHER= 2%**III. Determination of % yield of extract with Methanol:**

Weight of powder taken = 1gm

Quantity of methanol= 25 ml

Weight of empty petry dish = 104.07gm

Weight of petry dish after evaporating the filtrate = 104.14gm

Weight of extract = 104.14-104.07gm = 0.07gm

%YIELD OF EXTRACT WITH METHANOL =7%**IV. Determination of % yield of extract with Chloroform:**

Weight of powder taken = 1gm

Quantity of chloroform= 25 ml

Weight of empty petry dish =113.49gm

Weight of petry dish after evaporating the filtrate =113.54gm

Weight of extract = 113.54-113.49gm = 0.05gm

%YIELD OF EXTRACT WITH CHLOROFORM =5 %**V. Determination of % yield of extract with Acetone:**

Weight of powder taken = 1gm

Quantity of acetone = 25ml

Weight of empty petry dish = 96.12gm

Weight of petry dish after evaporating the filtrate = 96.17gm

Weight of extract = 96.17-96.12gm = 0.04gm

%YIELD OF EXTRACT WITH ACETONE =4%**2. Loss on drying**

Weight of powder taken = 1 gm

Weight of empty bottle = 145.80 gm

Weight of bottle with 1 gm powder = 146.80 gm

When it was placed in hot air oven for 15 minute then the weight of bottle after every 15 minute is taken

Ist reading = 146.78gm

IInd reading = 146.76gm

IIIrd reading = 146.74gm

IVth reading = 146.72gm

Vth reading = 146.71gm

VIth reading = 146.71gm

Loss of drying = 146.78-146.71= 0.07gm

% of loss of drying = 0.07/1×100 =7%

3. Swelling index

Weight of powder taken = 1 gm

Filled in the 25 ml of measuring cylinder and take the initial reading = 1cm

Made up the water level up to mark in measuring cylinder

Measured the length of swelled powder after 24 hours is length =2.3cm

Total swelling of powder = Final reading - Initial reading = 2.3-1= 1.3 cm

4. Ash value

*Total ash value

Weight of the empty dish =28.43gm

Weight of the drug taken =2gm

Weight of the ash =28.70gm

$$\text{Total ash value} = \frac{(\text{Weight of the ash} - \text{Weight of the empty dish})}{\text{Weight of the drug taken}} \times 100$$

$$= \frac{28.70-28.43}{2} \times 100$$

$$= 13.5\%$$

*Acid insoluble ash value-

Weight of the empty dish =28.43gm

Weight of the drug taken = 0.135gm

Weight of the ash =28.44gm

$$\text{Acid insoluble ash value} = \frac{(\text{Weight of the ash} - \text{Weight of the empty dish})}{\text{Weight of the drug taken}} \times 100$$

$$= \frac{28.44-28.43}{0.135} \times 100$$

$$= 7.4\%$$

*Water soluble ash value

Weight of the empty dish =28.43gm

Weight of the drug taken = 0.135gm

Weight of the ash = 28.45gm

$$\text{Water soluble ash value} = \frac{(\text{Weight of the ash} - \text{Weight of the empty dish})}{\text{Weight of the drug taken}} \times 100$$

$$= \frac{28.45-28.43}{0.135} \times 100$$

$$= 14.8\%$$

Phytochemical Analysis of Methanol Extract of *Blumea membranacea* Leaves

Collection of Specimens-

For the phytochemical analysis, the leaves of *Blumea membranacea* were freshly collected from selected healthy plants. Followed by collection the leaves were washed with distilled water and under shade

Preparation of Extract

Air dried leaves were grinded and kept in air-tight container. The powdered leaves were macerated with

methanol for seven days for three times. All solvents was mixed together and concentrated at vacuum to get solid extract.

Phytochemical Analysis

The phytochemical analysis was done according to the standard method (Khandelwal 2006) few of them are as follows.

Test for tannins and phenolic compounds:

With Lead Acetate:

Tannins are precipitated with lead acetate solution.

With Ferric Chloride:

Generally phenols are precipitated with 5% w/v solution of ferric chloride where deep blue-black colour obtained.

With dilute iodine solution:

When methanolic extract treated with dilute iodine solution yellowish red precipitate obtained.

Test for saponins:

Foam Test

1 ml of methanol extract was diluted with distilled water and shaken in kept tube foam observed indications the presence of saponins.

Test for calcium oxalate crystals:

When methanol extract of bark is treated with hydrochloric acid soluble crystals of calcium oxalate formed.

4. Conclusion

The plant *Blumea membranacea* is a small ornamental shrub; it is often found in all parts of India. characterization of different pharmacognostical parameters of *Blumea membranacea* leaves, included botanical identification, macroscopical study, phytochemical study, powder done characteristics, leaf constants, analytical standardization, florescence study etc. was. In the phytochemical analysis amino acids, alkaloids, coumarin glycosides, steroids were found as secondary metabolites in methanol extract of the plant leaves. The work will help for further exploitation of the plant for isolation and biological studies.

5. Reference

- [1] Arora NK, Khare E, Verma A, Sahu RK (2008). In vivo control of *Macrophomina phaseolina* by a chitinase and β -1,3-glucanase-producing *Pseudomonas* NDN1 Symbiosis, 46:129-135
- [2] Garrity GM, Bell JA, Lilburn T (2005). The Revised Road Map to the Manual. In: Brenner, Krieg, Staley and Garrity (ed.), *Bergey's Manual of Systematic Bacteriology, The Proteobacteria, Part A, Introductory Essays*. Springer, New York. 2:159-220
- [3] Jambhulkar PP and Sharma P (2013). Development of bioformulation and delivery system of *Pseudomonas fluorescens* against bacterial leaf blight of rice (*Xanthomonas oryzae* pv. *oryzae* Vol.35 843-849
- [4] A.Chakraborty, B. R.K.Devi, R. Sanjebam, S.Khumbong, and I. S. Thokchom, "Preliminary studies on local anesthetic and antipyretic activities of *Spilanthes acmella* Murr. in experimental animal models," *Indian Journal of Pharmacology*, vol. 42, no. 5, pp. 277-279, 2010.

- [5] K. R. Narayana, M. S. Reddy, M. R. Chaluvadi, and D. R. Krishna, "Bioflavonoids classification, pharmacological, biochemical effects and therapeutic potential," *Indian Journal of Pharmacology*, vol. 33, no. 1, pp. 2-16, 2001.
- [6] A.Chakraborty, R. K. B.Devi, S.Rita, K. Sharatchandra, and T. I. Singh, "Preliminary studies on antiinflammatory and analgesic activities of *Spilanthes acmella* in experimental animal models," *Indian Journal of Pharmacology*, vol. 36, no. 3, pp. 148-150, 2004.
- [7] S. A. Rani and S. U. Murty, "Antifungal potential of flower head extract of *Spilanthes acmella* Linn," *African Journal of Biomedical Research*, vol. 9, pp. 67-69, 2006.
- [8] W.D. Ratnasooriya, K. P. P. Pieris, U. Samaratunga, and J. R. A. C. Jayakody, "Diuretic activity of *Spilanthes acmella* flowers in rats," *Journal of ethnopharmacology*, vol. 91, no. 2-3, pp. 317-320, 2004.