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## Phytochemical and Pharmacological Studies on *Achillea Millefolium* (L) Leaves

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### ABSTRACT

The plant *Achillea millefolium* was selected to explore the scientific information on pharmacognostical, preliminary phytochemical and pharmacological screening carried out. This extract was subjected to OECD-Acute Toxicity Studies and found that it is Safe drug category according to the Global Harmonized Classification System quoted in OECD guidelines 1996 (Three doses level 125,250 and 500mg/kg selected). This plant showed good antibacterial activity against gram+ve bacteria strains than gram -ve strains which may be due to the presence of saponin and flavanoids.

**Keywords:** *Achillea millefolium*, pharmacognostical, pharmacological

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## INTRODUCTION

Herbal Medicine sometimes referred to as Herbalism or Botanical medicine is the use of herbs for their therapeutic or medicinal value. Many familiar medications of the twenty century were developed from ancient healing traditions that treated health problems with specific plants. Folk medicine is significant source of Ayurvedic, Unani, Traditional Chinese Medicine and Medical herbalism. It incorporates crude medicinal herbs, decoctions and infusions and syrups. Folk medicine is still practiced by some vendors, hakims and vaidis in remote areas and some folk preparations are of surprising high curative value.

The WHO estimates that up to 80% of the world's population use traditional medicines as their primary form of healthcare. The use of herbal medicine, the dominant form of medicinal treatment in developing countries, has been increasing in developed countries in recent years. WHO notes that of 119 plants derived pharmaceuticals medicines, about 74% are used in modern medicine in ways that correlated directly with their traditional uses as plant medicines by native cultures. Major pharmaceutical companies are currently conducting extensive research on plant materials gathered from the rain forests and other places for their potential medicinal value [1-11].

For most herbs, the specific ingredient that causes a therapeutic effect is not known. Whole herbs contain many ingredients, and it is likely that they work together to produce the desired medicinal effect. Many factors determine how effective an herb will be. For example, the type of environment (climate, bugs, soil quality) in which a plant grew will affect its components, as will how and when it was harvested and processed.

Herblists, chiropractors, naturopathic physicians, pharmacists, medical doctors and practitioners of Traditional Chinese Medicine all may use herbs to treat illness. Naturopathic physicians believe that the body is continuously striving for balance and that natural therapies can be used to support this process. They are trained in 4 year, postgraduate institutions that combine courses in conventional medical science (such as pathology, microbiology, pharmacology and surgery) with clinical training in herbal medicines, homeopathy, nutrition, and lifestyle counseling. Early Greeks and North Americans used this herb as to stop bleeding and promote the healing of wounds. It is used as uterine and gastric tonic for the disorders. Arrow is valuable medicinal herb, with much scientific evidence of use in alternative medicine as an antiseptic, antispasmodic, astringent, carminative, diaphoretic, digestive emmenagogue, stimulant and tonics, vasodilator and vulnerary,. also, used against colds, cramps, fevers, kidney disorders, toothaches, skin.



## AIM AND OBJECTIVE

From time immemorial, plants have been an indispensable source of both preventive and curative medicinal preparation for human being. Many indigenous plants have been scientifically tested and found to have medicinal properties that can be used in western style medicine. Such cures are particularly suitable for use in rural communities as inexpensive, sustainable and culturally appropriate alternative to more expensive conventional treatments. As a result, ethnomedicine is being promoted and supported as a way of providing effective medicines for people in less developed areas.

## MATERIALS AND METHODS

In the present study, investigation of *Achillea. Millifolium* was studied under two parts

### Pharmacognostical work

1. Collection and authentication of the plant
2. Extraction of active constituents with 90% alcohol.
3. Pharmacognostical studies i) Macroscopical studies

### Physio chemical constants

1. Phytochemical analysis on extract [18].
2. Estimation of total flavanoidal content in the extract [20].
3. Estimation of total phenolic content in the extract [19]

### Pharmacological studies

- i) Evaluation of toxicity profile by 14 days single dose acute toxicity study and assessment of gross behavioural changes
  - a) Mortality
  - b) Clinical signs and symptoms
- ii) In – vitro antimicrobial studies by cup plate method using different bacterial strains
- iii) Anti-inflammatory activities using in vivo model

## EXPERIMENTAL PART

### Plant material

The plant material was collected from Nilgiris district in Tamilnadu and authenticated and Voucher specimen deposited in college herbarium library. The plant was identified and confirmed as *Achillea millifolium* belong to family Asteraceae by Dr Rajan, (Botanist), Ooty.

### Detection of glycosides

About 50 mg of the extract was hydrolysed with Conc. HCl for 2 hours on a water bath and filtered.

### **Detection of Saponins**

#### **Foam or broth test**

A small quantity of extract was diluted with distilled water to 20 ml, the suspension was shaken in a graduated cylinder for 15 min. A 2 cm layer of foam or froth which is stable for 10 min. indicates the presence of saponins.

### **Physio Chemical constants**

#### **Ash values**

Ash values are helpful in determining the quality and purity of crude drug, especially in the powdered form.

The ash content of a crude drug is generally taken to be the residue remaining after incineration. It usually represents the inorganic salts naturally occurring in the drug and adhering to it, but it may also include inorganic matter added for the purpose of adulteration. Hence, an ash determination furnishes a basis of judging the identity and cleanliness of a drug and gives information relative to its adulteration with inorganic matter.

The total ash of a crude drug reflects the care taken in its preparation. The Acid Insoluble Ash is a part of the Total Ash which is insoluble in dil.HCl acid. A higher limit of acid insoluble ash is imposed, especially in cases where silica may be present or when the calcium oxalate content of the drug is very high some analysts favor mixing of sulphuric acid with the powdered crude drug before ashing and this Sulphated Ash Value is normally less fusible than ordinary ash.

#### **Fluorescence analysis**

For fluorescence studies, the powdered drug and various extracts were studied under both the day light and UV light, at wave length of 254 nm and 365 nm. The observations were recorded

#### **Total phenolic content (TPC) by Folin-Ciocalteu (FC) method [20]**

#### **Chemicals and reagents**

Folin-Ciocalteu Reagent: Folin-Ciocalteu was diluted (1:10) with distilled water and used

Sodium carbonate: 202.5 g of Sodium carbonate ( $\text{Na}_2\text{CO}_3 \cdot 10\text{H}_2\text{O}$ ) was dissolved in 1 litre of distilled water and used.

Methanol: Distilled

## Antibacterial Studies

### *In-Vitro* Antimicrobial Screening

The following gram positive and gram negative strains have been used for the antibacterial study.

#### Gram Positive strains

- i) *Staphylococcus aureus*
- ii) *Bacillus subtilis*

#### Gram Negative strains

- i) *Esherichia coli*
- ii) *Aspergillus niger*

## ANTI INFLAMMATORY STUDIES: *IN-VIVO* METHOD

### Carragenin induced rat paw edema

Group I	Solvent control 1ml/kg 0.3%CMC orally
Group II	Animals received Indomethacin 10mg/kg in 0.3CMC orally
Group III	Animals received Achillea millifolium 125mg/kg in 0.3%CMC orally
Group IV	Animals received Achillea millifolium 250mg/kg in 0.3% CMC orally
GroupV	Animals received Achillea millifolium 500mg /kg in 0.3%CMC orally

Acute edema was produced by injecting carragenin 1%w/w (0.1ml) into the sub plantar region of the left hind paw in the rats. The drug As. Millifolium 125, 250 and 500mg/kg and Indomethacin 10mg/kg administered orally one hour before testing. The control group received vehicle 0.1 ml/100gm. The paw volume was measured by using digital plethysmometer(UGO Basile. Italy) at 0,1,2,3,4,and 6 hrs after carragenin challenge. The percentage increase in the edema (paw volume) was calculated by comparing it with zero minute reading. The percentage inhibition of edema was calculated at 4<sup>th</sup> hour assuming 100% inflammation in vehicle group.

## RESULTS AND DISCUSSION

The present project work was performed under two heading namely Phytochemical and pharmacological activities screening.The Pharmacognostical features such as macroscopical

characters, preliminary phytochemical studies of *Aschillea millifolium* was carried out and presented.

The results reveals that the organoleptic characters showed characteristic or specific odour with taste of this plant, may authenticate the plant some extent but required additional informations such as the parameters like physio-chemical constants to be helpful in determining the quality and purity of crude drugs in powder form according to the standard procedures.

### CONCLUSION

The plant *Achillea millefolium* was selected to explore the scientific information on pharmacognostical, preliminary phytochemical and pharmacological screening carried out. This extract was subjected to OECD-Acute Toxicity Studies and found that it is "Safe drug category according to the Global Harmonized Classification System" quoted in OECD guidelines 1996 (Three doses level 125, 250 and 500mg/kg selected) *Achillea millefolium* (L) leaves ethanolic extract was subjected to antibacterial and anti-inflammatory screening by Cup plate method and Carragenin induced rat paw edema respectively.

This plant showed good antibacterial activity against gram+ve bacter strains than gram -ve strains which may be due to the presence of saponin and flavanoids. *Achillea millefolium* ethanolic extract 500mg/kg was more active than 250mg/kg and equipotent as that of Indomethacin 10mg/kg treated group ( $p < 0.001$ ).

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