ABSTRACT

Introduction: Essential oils (EOs) are aromatic and are characteristic feature of many angiosperm families. Currently, studies on the biological activities of essential oils have become increasingly important in the search for natural and safe alternative medicines. Helminthiasis, a parasitic infection was considered as the major cause of ill health of peoples throughout the world. Species of Blumea belongs to the family Asteraceae and has been reported for its anti-helminthic use in ancient literature. Methods: Essential oil from leaves of Blumea mollis was extracted by Clevenger apparatus and analysed for its anti-helminthic potentiality against Pheretima posthuma, the Indian earth worm. Results: The essential oil yielded from Blumea mollis by hydro distillation was yellow in colour, transparent and clear with characteristic medicinal aroma and the yield was 0.1ml/100g. The essential oil of B. mollis exhibited significant anti-helminthic potentiality against the earth worms. The time required for paralysis and death of worms against essential oil was less than that of standard Albendazole. Conclusion: Evaluation techniques used in the present study was preliminary and qualitative. The observed anti-helminthic activity of the essential oil against worms substantiate the use of B. mollis as an anti-helminthic in the Indian system of medicine.

KEYWORDS: Essential oil, Blumea, anti-helminthic, Asteraceae, Helminthiasis.

INTRODUCTION

Essential oils (EOs) are unique natural plant product displays various biological properties. Plant essential oils are usually the complex mixture of natural compounds include polar and non polar compounds. Essential oils are used by the plants as defense molecules against infection, function as hormone-like compounds, initiate cellular regeneration and microbialicidal against fungal, viral and animal foes. They are used in perfumes and other products such as creams, soaps, sanitary products, dentistry, agriculture, preservatives, flavor additives for foods, as fragrances for household cleaning products, industrial solvents, as natural remedies (as mixtures with vegetable oil in massages or in baths and in aromatherapy).[1]

Blumea is a shrubs or small tree which comprises of about 80 species distributed along the tropical and subtropical parts of Asia, Africa, and Oceania. This genus includes some important medicinal plants largely used in traditional medicine. The essential oil from Blumea mollis,[2] and Blumea perrottetiana[3] has shown notable insecticidal activities. Antibacterial, antioxidant and antifungal potentials were also reported from many species of Blumea.

Helminthiasis a parasitic infection still considered as the major cause of ill health of peoples throughout the world especially peoples from deprived communities of undeveloped countries with poorer sanitary and health facilities. The worms which are associated with problem of ill health include the trematodes (flukes), cestodes (tapeworms) and intestinal nematodes (roundworms). Many plants have been reported as anti-helminthic but they are still remaining as underutilized due to lack of scientific proofs or records. In this scenario, the present study aims to extract the essential oil from the leaves of Blumea mollis, an aromatic herb of Asteraceae and study their bio-activity in terms of anti-helminthic potentiality.

MATERIALS AND METHODS

Plant material
The plant material selected for the present study was Blumea mollis (D. Don) Merr. (Fig.1). It is an aromatic herb, growing up to 80 cm tall. Leaves are obovate, 2.5 to 7 cm long and 1-3.5 cm broad. Rose to pink flowers is borne in dense flat topped clusters. Flower heads are bell shaped, Florets are bisexual. The plants are collected from Varkala village of Thiruvananthapuram District, Kerala.
**Phreatima posthuma**

*Phreatima posthuma* is the Indian earthworm and was used for the anti-helminthic studies (Fig. 2). It has long, elongated, cylindrical narrow body which is bilaterally symmetrical. The anterior end is tapering, while the posterior end is more or less blunt. A mature earthworm may attain the size up to 150 mm in length and 3-5 mm in width. They are found in the soil rich in decaying organic matters usually in gardens, pastures, lawns, irrigated farm lands, near the banks of the ponds, lakes and rivers. They generally inhabit an upper layer of earth to a depth of about 30 to 45 cm and even they go down up to 3 meters or more during summer. They were collected from moist soil and washed with water to remove all debris.

**Extraction of essential oil**

The fresh plant material including aerial part, tender stem and leaves of *B. mollis* were collected from moist soil and washed with water to remove all debris. The mixture was heated to a depth of about 30 to 45 cm and even they go down up to 3 meters or more during summer. They were collected from moist soil and washed with water to remove all debris.

The plant material including aerial part, tender stem and leaves of *B. mollis* were chopped into small pieces. 500 g of fresh plant material was subjected to hydrodistillation using clevenger type apparatus. 3L of water was added to the material. The mixture was heated on a heating mantle. The distillation was continued for 3 h. The essential oil obtained was collected and stored at 4°C in sealed vials until for further analysis. The percentage of yield was calculated using the equation:

\[ \text{EO} = \frac{\text{M}}{\text{B}_m} \times 100 \]

where M - Mass of extracted oil and B_m - Initial plant biomass.

**Anti-helminthic studies**

The anthelmintic activity was performed according to the method of Madhavulu and Souris on the adult Indian earthworm *Phreatima posthuma*. *Phreatima posthuma* was placed in petridish containing different concentrations (0.1, 0.2, 0.3, 0.4 and 0.5%) of *B. mollis* essential oil dissolved in DMSO (Dimethyl sulphoxide). Each petridish was placed with 2 worms and observed for paralysis or death. Mean time for paralysis was noted as no movement of any sort and the time of death (min) were also recorded after ascertaining that worms neither moved when shaken nor when given external stimuli. The test results were compared with reference compound Albendazole (1%) treated samples.

**Statistical analysis**

All the experiments were conducted in triplicate. Data were presented as mean ±SD. Significance was noted as *p* < 0.05.

**RESULTS AND DISCUSSION**

**Physical properties and yield of essential oil**

The amount of essential oil yielded from *Blumea mollis* was 0.1ml/100g. It is yellow in colour, transparent and clear with characteristic medicinal aroma. The result is comparable with the yield of *Blumea balsamifera* (0.4 - 0.6 ml/100g).\(^5\) Essential oil yields from leaves of *Blepharocalyx salicifolius* and *Myracrodruon urundeuva* were also at par with *B. mollis*.\(^6\) Similarly *Artemisia pallens* also showed yield of 0.33%.\(^7\) The results substantiate that *B. mollis* is a potent source of essential oil.

**Anti-helminthic properties**

The essential oil of *B. mollis* exhibited significant anti-helminthic potentiality against earth worms. The activity of the essential oil has been found to be better than that of Albendazole, the standard. The time required for paralysis and death of worms in the case of essential oil was less than the standard, under same conditions (Table 1). The negative control DMSO not showed any activity against the worms.

**Phreatima posthuma** has been used as test worm in most of anti-helminthic screening, as it shows anatomical and physiological resemblance with intestinal round worm parasite of human beings.\(^8\) The results of the study reveal that essential oil of *B. mollis* was showing significant anthelmintic activity in a dose dependent manner. The study of Aditya et al.\(^9\) on anti-helminthic properties of *Blumea lacerana* was comparable with the present results. In *B. lacerana* the secondary metabolites was responsible for the anti-helminthic activity. The death and paralysis time of *B. mollis* essential oil shows similarities with aqueous, alcoholic and methanolic extracts of *Cycas beddomei* against *Phreatima posthuma*.\(^10\) *Acorus calamus* leaves extracts exhibited...
significant anthelmintic activity at a concentration of 100 mg/ml. Peak activity was exhibited by the MeOH extract at a concentration of 100 mg/ml. Comparing the reported data, B. mollis essential oil is more potent against *Pheretima posthuma*.

Senthilkumar et al.\(^2\) reported the GC–mass spectroscopy (GC–MS) of leaf essential oil of *B. mollis*. The essential oil of *B. mollis* reported with 39 compounds, the major chemical compounds identified were linalool (19.43%), γ- elemene (12.19%), copaene (10.93%), estragole (10.81%), Allo-ocimene (10.03%), γ-terpinene (8.28%) and allo-aromadendrene (7.44%). Linalool is a natural plant-product, which has proven antimicrobial and insect-repellant properties and indicate it might be useful for control of enteropathogens or insect pests in poultry farms. Duman et al.,\(^12\) reported the anti bacterial activity of linalool-rich essential oils from *Ocimum basilicum* and *Coriandrum sativum* varieties.

The anthelmintic potentiality of essential oils may be due to single or a combined effect of the compounds or chemical groups present in them. Different active compounds may have different targets to exert anthelmintic effect. One of the major reported mechanisms is uncoupling of oxidative phosphorylation. This may be like the inhibition of enzyme activity.

Table 1: Anti-helminthic activity of *B. mollis* essential oil against *Pheretima posthuma*

<table>
<thead>
<tr>
<th>Sl. no</th>
<th>Essential oil (%)</th>
<th>Albenzazole (%)</th>
<th>DMSO (%)</th>
<th>Time period for paralysis (min)</th>
<th>Time period for death (min)</th>
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<td>79±1</td>
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</tbody>
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*p* < 0.05.

making complexes with proteins or polysaccharides and block channels of ions. These specific actions may upset the routine biochemical and physiological activities of the earthworm, i.e., depriving of nutrition, abnormality in structure and disturbing neuromuscular aspects.\(^13\)\(^14\)

Essential oil in this study has demonstrated *in vitro* anthelmintic activity. The results obtained in this study should be confirmed by *in vivo* experimentation, so as to standardize doses for developing a drug.

CONCLUSION

Essential oils are good source of several bioactive compounds. *Blumea mollis* showed potent anti-helminthic activity against Indian earth worm *Pheretima posthuma*. The observed anti-helminthic activity of the essential oil against worms substantiate the use of *B. mollis* as an anti-helminthic in the Indian system of medicine and also enhances the credibility of ethno botanical explorations.

REFERENCES