# Chromatographic evaluation and anthelmintic activity of *Eucalyptus globulus* oil

## D. J. Taur, V. B. Kulkarni, R. Y. Patil<sup>1</sup>

Department of Pharmacognosy, S.V.P.M's College of Pharmacy, Malegaon (BK), <sup>1</sup>Department of Pharmacognosy, S.U. College of Pharmacy, Kharadi, Pune, Maharashtra, India

Submitted: 27-03-2010

Revised: 28-03-2010

Published: 19-07-2010

# ABSTRACT

In world Helminthes infections are the most widespread of all the infections in humans. The morbidity due to parasitic diseases has been increasing in our population. The gastrointestinal helminthes become resistant to the currently available anthelmintic drugs. Anthelmintic substances having considerable toxicity to human beings are present in foods derived from livestock, posing a serious threat to human health. Due to this, there is a need to derive new chemical substances from natural sources, for helminthes control. In this study, volatile oil isolated from *Eucalyptus globulus* Labill was evaluated for its anthelmintic activity on adult Indian earthworms, *Pheretima posthuma*, which have anatomical and physiological resemblance with the intestinal roundworm parasites of human beings. In concentrations of 0.05, 0.01 and 0.15 ml/ml, respectively, all the oil samples showed potent anthelmintic activity as compared to that of the standard drug albendazole at a concentration of 10 mg/ml.

Key words: Anthelmintic, Eucalyptus globulus, eucalyptus oil, Pheretima posthumag

## **INTRODUCTION**

In world Helminthes infections are the most widespread of all the infections affecting humans. The morbidity due to parasitic diseases has been increasing in our population.<sup>[1]</sup> The gastrointestinal helminthes become resistant to the currently available anthelmintic drugs. Therefore, treatment of helminthes diseases has become difficult.<sup>[2]</sup> Anthelmintic substances having considerable toxicity to human beings are present in foods derived from livestock, posing a serious threat to human health.<sup>[3]</sup> A new lead for helminth control is greatly needed and has promoted studies of traditionally used anthelmintic plants which are generally considered to be very important sources of bioactive substances.<sup>[4]</sup>

*Eucalyptus globulus* Labill (family Myrtaceae) is a lofty tree of about 90 m in height. Leaves are febrifuge, carminative, stimulant, expectorant, antiseptic, antimalarial and anthelmintic. Eucalyptus oil has powerful antiseptic and

Address for correspondence: Dnyaneshwar Jija Taur, Department of Pharmacognosy, S.V.P.M's College of Pharmacy, Malegaon (BK), Tal. Baramati, Dist. Pune, Maharashtra, India. E-mail: dnyaneshtaur@gmail.com

DOI: 10.4103/0974-8490.65504

disinfectant properties. It increases the flow of saliva, gastric and intestinal juices and increases the digestion and appetite.<sup>[5,6]</sup>

Bark of E. globulus contains ellagitannins, methyl and glycosyl derivatives of ellagic acid and free ellagic and gallic acids, gallotannins and cathechin.<sup>[7-10]</sup> Essential oil from this species has a therapeutic application in the treatment of pulmonary infections by inhalation<sup>[11]</sup> and the monoterpene extracted from Eucalyptus citriodora, E. globulus and Eucalyptus teretcorni exhibits antibacterial activity.[12] Besides antibacterial activity, the essential oil from eucalyptus shows analgesic and anti-inflammatory effects.<sup>[13]</sup> Phytochemical analysis has established that the genus Eucalyptus contains monoterpenes.<sup>[14]</sup> Two monoterpene glycosides, conjugated with gallic acid globulusin A and B, together with four known compounds, cypellocarpin A, eucaglobulin, cuniloside and (1S, 2S, 4R)-trans-2-hydroxy-1,8-cineole β-D-glucopyranoside, were isolated from hot water extracts of the leaves of E. globulus.<sup>[15]</sup> The aim of this research was to study chemical composition and anthelmintic property of the volatile oil from the leaves of *E. globulus*.

## **MATERIAL AND METHODS**

#### **Plant material**

The leaves were collected from Baramati region of Dist.

Pune and were authenticated by Prof. R. B. Deshmukh, Head, Department of Botany, Shardabai Pawar Mahila Mahavidyalaya, Shardanagar.

#### Isolation of Essential Oil

The essential oil was isolated from the fresh leaves by hydrodistillation in a clevenger-type apparatus, yielding essential oil 0.53% w/v.

## Evaluation of Anthelmintic Activity

Indian adult earthworms (Pheretima posthuma) of 3-5 cm length and 0.1-0.2 cm width were used for anthelmintic activity because of their anatomical and physiological resemblance with the intestinal roundworm parasites of human beings.<sup>[16,17]</sup> The earthworms were divided into four groups containing five earthworms in each group. Oil was dissolved in water using tween 80. All the test samples and the standard drug were freshly prepared before starting the experiment. The solution of oils and standard solution were poured in petridishes. All the earthworms were washed in normal saline solution before they were released into petridish containing 10 ml solution of E. globulus oil in concentrations of 0.05, 0.1 and 0.15 ml/ml, respectively, and albendazole (10 mg/ ml). The time taken for the worms to get paralyzed and killed was noted. All readings were expressed as mean and standard error of mean (SEM) of three animals in each group.<sup>[18]</sup>

#### Evaluation of Volatile Oil

Volatile oil was evaluated for optical rotation and refractive index as per the standard procedure.<sup>[19]</sup>

### Thin layer chromatography

Thin layer chromatography was performed using silica gel G as the stationary phase, toluene-ethylacetate (97:3) as the mobile phase and vanillin-sulfuric acid as the spraying reagent.<sup>[20]</sup>

## RESULTS

Essential oil from *E. globulus* contains 1,8-cineole as the major component<sup>[21,22]</sup> and is used in the treatment of pulmonary infections<sup>[11]</sup> and also exhibits antibacterial activity.<sup>[12]</sup> The result of the present study reveals that *E. globulus* oil in concentrations of 0.05, 0.1 and 0.15 ml/ml showed significant anthelmintic activity as compared to the standard drug albendazole at a concentration of 10 mg/ml, as shown in Figure 1. *E. globulus* oil showed anthelmintic activity in a concentration-dependent manner. Evaluation of *E. globulus* oil showed an optical rotation of +0.8° and a refractive index of 1.4554. Chromatographic study confirmed the presence of borneol, linalool, cineol, geranyl acetate, anethol, saffrol as the phytoconstituents

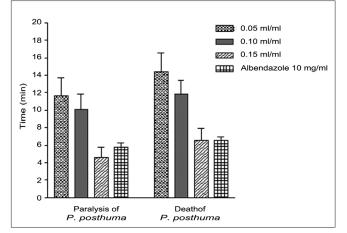


Figure 1: Anthelmintic activity of Eucalyptus globulus oil

of eucalyptus oil, as shown in Table 1.

# DISCUSSION

An Indian adult earthworm (*P. posthuma*) has anatomical and physiological resemblance with the intestinal roundworm parasites of human beings.<sup>[16,17]</sup> Result indicates that time taken for albendazole for causing paralysis and death of *P. posthuma* was  $5.82 \pm 0.466$ and  $6.54 \pm 0.429$ , respectively, whereas *eucalyptus* oil at a concentration of 0.15 ml/ml causes paralysis and death at  $4.598 \pm 1.151$  and  $6.57 \pm 1.374$ , respectively. So, the present investigation concludes that *E. globulus* oil has anthelmintic potential due to the presence of borneol, linalool, cineol, geranyl acetate, anethol, saffrol as phytoconstituents.

## ACKNOWLEDGMENTS

The authors are thankful to the Management and Prof. R. N. Patil, Principal, of S.V.P.M's College of Pharmacy, Malegaon (Bk), Baramati, for providing necessary facilities and also to Prof. R. B. Deshmukh, Head, Department of Botany, Shardabai Pawar Mahila Mahavidyalaya, Shardanagar, for the authentication of the plant.

Table 1: Thin layer chromatography of <i>E.</i> globulus oil		
Spots from bottom	R <sub>F</sub> values	Phytochemical constituents
1	0.266	Borneol
2	0.312	Linalool
3	0.4666	Cineol
4	0.632	Geranyl acetate
5	0.785	Anethol
6	0.814	Saffrol

## REFERENCES

- Tagboto S, Townson S. Antiparasitic properties of medicinal and other naturally occurring products. Adv Parasitol 2001;50:199-205.
- 2. Coles GC. Nematode control practices and anthelmintic resistance on British sheep farms. Vet Rec 1997;141:91-3.
- Turnipseed SB, Roybal JE, Rupp HS, Gonzales SA, Pfenning AP, Hurlbut JA. Confirmation of avermectin residues in food matrices with negative-ion atmospheric pressure chemical ionization liquid chromatography/mass spectrometry. Rapid Commun Mass Spectrom 1999;13:493-9.
- Hammond JA, Fielding D, Bishop SC. Prospects for plant anthelmintics in tropical veterinary medicine. Vet Res Commun 1997;21:213-28.
- Kirtikar KR, Basu BD. Indian Medicinal Plants. 2<sup>nd</sup> ed, Vol 1. Dehradun: International book Distributor; 1985. p. 1044-5.
- Nadkarni AK. Dr. K.M. Nadkarni's Indian Materia Madica. 3<sup>rd</sup> ed, Vol 1. Bombay: Popular Prakashan; 1992. p. 512-6.
- 7. Yazaki Y, Hillis WE. Polyphenol of *Eucalyptus globulus*, *E. regnans* and *E. deglupta*. Phytochemistry 1976;15:1180-1.
- Fechtal PM, Riedl B. Analyse de extraits tannants des 'ecorces des eucalyptus apr'es hydrolyse acide par la chromatographie en phase gaseuse coupl'ee avec la spectrometrie de masse (GC–MS). Holzforschung 1991;45:269-73.
- Conde E, Cadahia E, Diez-Barra R, Garcia-Vallejo MC. Polyphenolic composition of bark extracts from *Eucalyptus camaldulensis*, *E. globulus* and *E. rudis*. Holz Roh Werkstoff 1996;54:175-81.
- Cadahıa E, Conde E, Fernandez de Simon B, Garcia-Vallejo MC. Tannin composition of *Eucalyptus camaldulensis*, *E. globulus* and *E. rudis*. Part II. Bark Holzforschung 1997;51:125-9.
- Low D, Rawal BD, Griffin WJ. Antibacterial action of the essential oils of some Australian Myrtaceae with special references to the activity of chromatographic fractions of oil of *Eucalyptus citriodora*. Planta Med 1974;26:184-9.
- 12. Ramezani H, Singh HP, Batish DR, Kohli RK. Antifungal

activity of the volatile oil of *Eucalyptus citriodora*. Fitoterapia 2002;73:261-2.

- Silva J, Abebe W, Sousa SM, Duarte VG, Machado MI, Matos FJ. Analgestic and anti-inflammatory effects of essential oils of Eucalyptus. J Ethnopharmacol 2003;89:277-83.
- Foudil-Cherif Y, Meklati BY, Verzera A, Mondello L, Dugo G. Chemical examination of essential oils from the leaves of nine Eucalyptus species growing in Algeria. J Essent Oil Res 2000;12:186-91.
- Hasegawa T, Takano F, Takata T, Niiyama M, Ohta T. Bioactive monoterpene glycosides conjugated with gallic acid from the leaves of *Eucalyptus globules*. Phytochemistry 2008;69:747-53.
- Thorn GW, Adams R, Braunwald E, Isselbacher K, Petersdorf R. Harrison's Principle of Internal Medicine. New York: Mcgraw Hill Co; 1977. p. 1088.
- 17. Vigar Z. Atlas of Medical Parasitology. 2<sup>nd</sup> ed. Singapore: P.G. Publishing House; 1984. p. 216.
- Tambe V, Nirmal S, Jadhav R, Ghogare P, Bhalke R, Girme A, Bhambar R. Anthelmintic activity of *Wedelia trilobata* leaves. Indian J Nat Prod 2006;22:27-9.
- Anonymous, Indian Pharmacopoeia. Vol 2, New Delhi: The Controller of Publications; 1996. p. A-137.
- Wagner H, Bladt S. Plant drug analysiws: A thin layer chromatography atlas. 2<sup>nd</sup> ed, New Delhi, India: Springer; 1996. p. 149.
- Silvestre AJ, Cavaleiro JA, Delmond B, Filliatre C, Bourgeois G. Analysis of the variation of the essential oil composition of *Eucalyptus globules* Labill. from Portugal using multivariate statistical analysis. Ind Crops Prod 1997;6:27-33.
- Viturro CI, Molina AC, Heit CI. Volatile components of *Eucalyptus* globules Labill. ssp. bicostata from Jujuy, Argentina. J Essent Oil Res 2003;15:206-8.

Source of Support: Nil, Conflict of Interest: None declared.