

Repellent activity of selected aromatic oils on land leech *Haemadipsa montana*, Moore: 1927.

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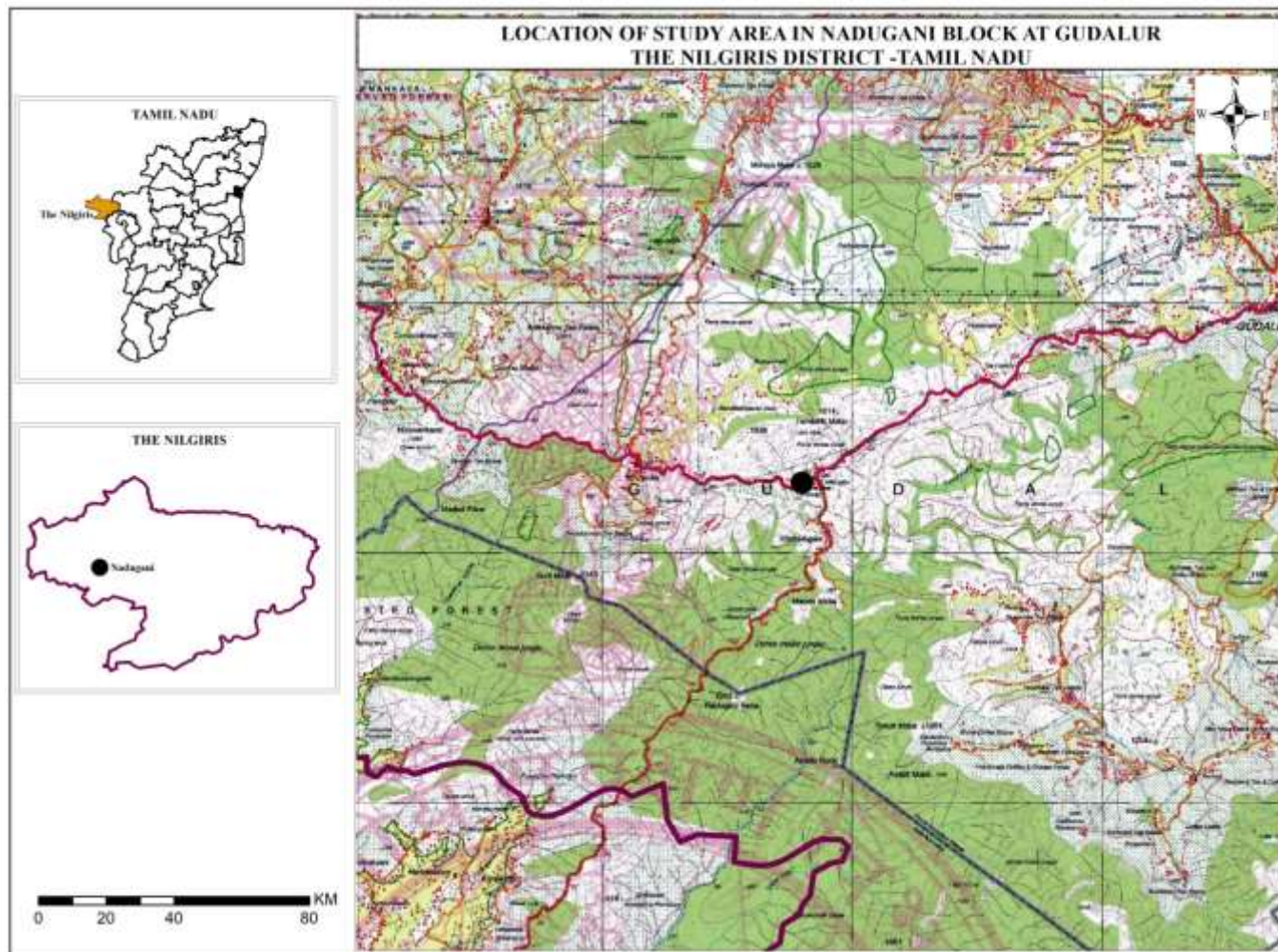
ABSTRACT:

Haematophagus leech species are found distributed in the tropical and subtropical indo-Pacific Regions. Land leech species of *Haemadipsa montana* (Moore, 1927) is the dominant land leech species found in South India. Leeches were collected from Nadugani area of Nilgiris and identified. The leeches were evaluated for repellency against selected aromatic oils viz., tea tree oil, lavender oil, and peppermint oil. Repellency was evaluated in three different concentrations viz., 1%, 3%, and 5% and for each concentration 10 leeches were used. The following indices of repellency activity viz., repellency (%), effectiveness period of oils, and Effectiveness repellency concentration (ECR). ECR50, ECR75, ECR95 in 3 hrs and 6 hrs interval were used. Lavender oil showed high repellency of 83.41% at 5% concentration after 3 hrs exposure and peppermint oil showed least repellency of 28.88% at 1% concentration after 6 hrs exposures. Lavender oil has effective concentration of 50% repellency (ECR-50) as 1.837% at 3 hrs of interval and 95% repellency (ECR-95) of 4.643%. The result revealed that lavender oil shows high activity even at low concentration. Among the three peppermint oil has least activity with effective concentration of 50% repellency (ECR-50) of 3.072 % at 3 hrs of interval and 95% repellency (ECR-95) been 7.520 %. Hence tea tree oil is also considerable for repellent activity against *H. montana*. Even though aromatic oils have repellency against leech bite the oils can be used by Ecotourists and estate workers for avoiding leech bite.

Key words: *Haemadipsa montana*, Tea tree oil, Lavender oil, Peppermint oil, Leech repellent.

INTRODUCTION

Blood feeding terrestrial leeches belong to the family Heamadipsidae. They are also adapted to aquatic and terrestrial habitats and are confined to moist forests that have seasonal rainfall (Sawyer, 1986). Typically leeches are found in fresh water environments, only a few inhabits terrestrial or saltwater environments. Majority of leeches suck blood of other animals which marks them as "Haematophagous." Some species of leeches are not blood sucking, instead they feed other invertebrate tissues or whole (Mooser *et al.*, 2009). Leeches take blood meal from vertebrates including man. The amount of blood ingested is usually 6-10 times the initial fresh weight of the leech (Mann *et al.*, 1962; Fogden and Proctor 1985). In South-East Asia, people are generally aware of the serious inconvenience and blood loss that may be caused by terrestrial leeches. However, after removal of the leech, the bite wound may cause secondary bacterial infection and delay healing (Heukelbach *et al.* 2009). Use of repellents as personal protection measures seems to be the most acceptable solution to prevent leech bite. Repellents of essential oils and extract of plants were also used with promising results. Essential oils are concentrated essences extracted from different parts of plants, containing hundreds of substances, but typically with the prevalence of one, two or three of them that really characterize the fragrance (Mendes *et al.*, 2007). Industrial interest in essential oil is due to their application as fragrances in perfumes and as flavor additives. (Katz *et al.*, 2008; Simic *et al.*, 2008). Numerous plants and derived products, in particular essential oils, have been investigated and described as potential natural sources of insect repellent. However identification of effective repellents against leeches which has no health hazard to human beings is the need of the day. *Haemadipsa montana* is a common terrestrial leech species found in South India and are abundant in a reserve forest areas tea and coffee estates and near human habitat. But still there are no studies available on the repellency effect of aromatic oils against leech species of *Haemadipsa montana*. With these visions in mind the present study was planned and carried out.

Materials and Methods:**Material used:**

Land leech : *Haemadipsa montana*

Systematic Position:

Kingdom : Animalia
 Phylum : Annelida
 Class : Clitellata
 Sub class : Hirudinea
 Order : Arhynchobdellida
 Suborder : Hirudiniformes
 Family : Haemadipsidae
 Genus : *Haemadipsa*
 Species : *Montana*

Collection of samples:

The leeches were collected from Nadugani area of Gudalur Taluk. (11°28'12"N 76°24'30"E) Multiple field visits were conducted, to collect the leeches. The leeches were handpicked and transferred to plastic container with little amount of moist soil, the collected leeches were transported to the laboratory.

Identification:

The identification of this land leeches was done by using "Handbook: Leeches of India" by Mahesh Chandra (1991).

Method for testing Repellency:

The repellency test of (Ribbands, 1946) which was modified by (Ramachandran *et al.*, 1971) was used for the present study. In this method four cloth strips were cut in the proportions of 5cm width and 95.5cm length. These cloth strips were soaked with selected oils for 2 minutes and assembled around a glass panel. The glass panel is used to introduce leech, and the attempts made by the leeches were recorded.

The repellency was calculated by using the following formula

$$\text{Repellency} = \frac{\text{No. Probes attempted} - \text{No. Cross over}}{\text{No. Probes attempted}} \times 100$$

The repellency of aromatic oils at three concentrations (1%, 3%, and 5%) incorporated in the cloth strips were calculated as gms/cm² and presented (table1). Ten leeches were used to evaluate repellency in each concentration

Effectiveness period:

For testing the effectiveness period the experiment was repeated at intervals of 3 hrs and 6 hrs after soaking to the aromatic oil. And the cloth strips were kept at room temperature in between the test periods.

Statistical analysis:

The difference between control and treated groups were analyzed using DMRT with SPSS software the repellency of effect of different concentration was calculated using Curve expert software 1.8 versions and Effective concentration for 50%, 75%, and 95% repellency (ECR) were also calculated.

RESULTS AND DISCUSSION:

The repellency of three aromatic oils on *H. montana*, during different periods of exposure is presented in Table-2

All the oils revealed concentration dependent action with high repellency at higher concentration and declining with the decrease in concentration. After 3 hrs of exposure repellency activity of three aromatic oils differed significantly ($p < 0.01$). The repellency of lavender oil exhibited highest activity of 83.41% and 48.72 % in 3 hrs and 6hrs respectively while tea tree oil 74.05 % in 3 hrs and 48.97 % in 6 hrs and peppermint oil 60.95% in 3hrs and 41.26% in 6hrs were comparatively less repellent than lavender oil. All the oils exhibit significantly higher repellent action at higher concentration, while substantial action could also be prominent in the lowest concentrations as well.

The effect of different treatment on 3 hrs of exposure is presented in (Table-3).

It reveals that least ECR50 rate of 1.837 is found in lavender oil and highest ECR 50 rate of 3.072 it found in Peppermint oil. Similar trend was found ECR 75 and ECR 95. The least value of ECR 95 is found in lavender oil which shows that the lavender oil has high activity even at low concentration 4.643% .

The effect of different treatment on 3 hrs of exposure is presented in (Table-4).

It also reveals that least ECR50 rate of 3.811 is found in lavender oil and highest ECR 50 rate of 5.762 it found in Peppermint oil. Similar trend was found ECR 75 and ECR 95. The least value of ECR 95 is found in lavender oil which shows that the lavender oil has high activity even at low concentration and has high activity even after 6 hrs of exposure.

According to Nath *et al* (2002) synthetic repellent DEET exhibited the highest persistency than the other three repellents, affording maximum protection time in the treatments. Bottle brush oil afforded significantly better persistency than timur oil, neem oil and DMP at 2% (0.37 mg/cm²). According to his studies neem oil and DMP had high repellent activity than timur oil and bottle brush oil at 3% concentration. The highest repellency was (0.56 mg/cm²) at 3% concentration. According to (Nath *et al.*,1986) cinnamon oil was found to be the best followed by oils of pine, Citronella, Eucalyptus and orange oil 25% concentration of Cinnamon oil repelles 100% results. The result of the present study reveals that 95% repellency can be achieved with lavender oil at 4.643 % which is for low than concentration of other aromatic oils used in previous reports.

The efficacy of leech repellents depends, among other properties on the duration of activity on cloth. Nath *et al* (1993) compared the repellency of N,N-diethyl phenyl acetamide (DEPA), N,N-diethyl-m-toluamide (DEET), 3-acetyl 2(2-6-dimethyl-5-heptenyl)-oxazolidine(Citronyl), dimethyl phthalate (DMP), N-benzoyl piperidine (NBP), and the volatile oil of *Zanthoxylum armatum* DC., and found that DEPA and DEET were more long-lasting than *Z. armatum* oil, citronyl, DMP and NBP. (Tawatsin *et al.*,2006) carried out field trials of DEET and the essential oils of *Boesenbergia rotunda* (L.), *Psidium guajava* L., and *Curcuma longa* L., and showed that all afforded 100% protection for at least 8 hour against terrestrial leeches. Our results revealed that lavender oil is the most effective against *Haemadipsa montana.*, the other aromatic oils studied also show noticeable effects against *H. montana*. Hence the selected three aromatic oils can be effectively used as repellent against *H. montana*. The fragrance of lavender oil is also preferred by treatment and as it has high potency to repel *H. montana* it is suggested as a suitable repellent.

Conclusion

The present study revealed that the aromatic oils have appreciable repellent activity against the leech bite include tea tree oil, lavender oil, and peppermint oil. All had leech repellent effect, and therefore, it may be used in the treatment of leech bite. Aromatic oils had an anti leech bite and therefore may be used in the fields both by ecotourists visiting these areas as well as estate workers to protect themselves from leech bite.

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Tables

Table1: Relative repellency of aromatic oils against *H.montana*

Aromatic oils	Concentration (%)	Quantity of oil used g/cm ²
Tea tree oil	1%	0.001537
	3%	0.004611
	5%	0.007685
Lavender	1%	0.002581
	3%	0.007743
	5%	0.012905
Peppermint	1%	0.0002407
	3%	0.0007221
	5%	0.0012035

Table2: Efficiency of different aromatic oils against land leech *H. montana* during different periods after application

S.no	Time after exposure (hrs)	Concentration	Repellency in treatment %			
			Control	Tea tree oil	Lavender oil	Peppermint oil
1	3	1%	0.00±0.00 ^a	54.30±3.63 ^b	60.01±4.97 ^c	51.70±6.91 ^b
		3%	0.00±0.00 ^a	65.67±6.54 ^c	70.32±4.05 ^d	55.95±4.72 ^b

		5%	0.00±0.00 ^a	74.05±6.13 ^d	83.41±3.00 ^d	60.95±6.07 ^c
2	6	1%	0.00±0.00 ^a	38.66±5.91 ^b	42.75±2.94 ^c	28.88±8.74 ^b
		3%	0.00±0.00 ^a	44.44±4.62 ^b	48.72±4.28 ^b	32.10±6.88 ^b
		5%	0.00±0.00 ^a	48.97±5.81 ^b	53.48±5.54 ^c	41.26±7.08 ^b

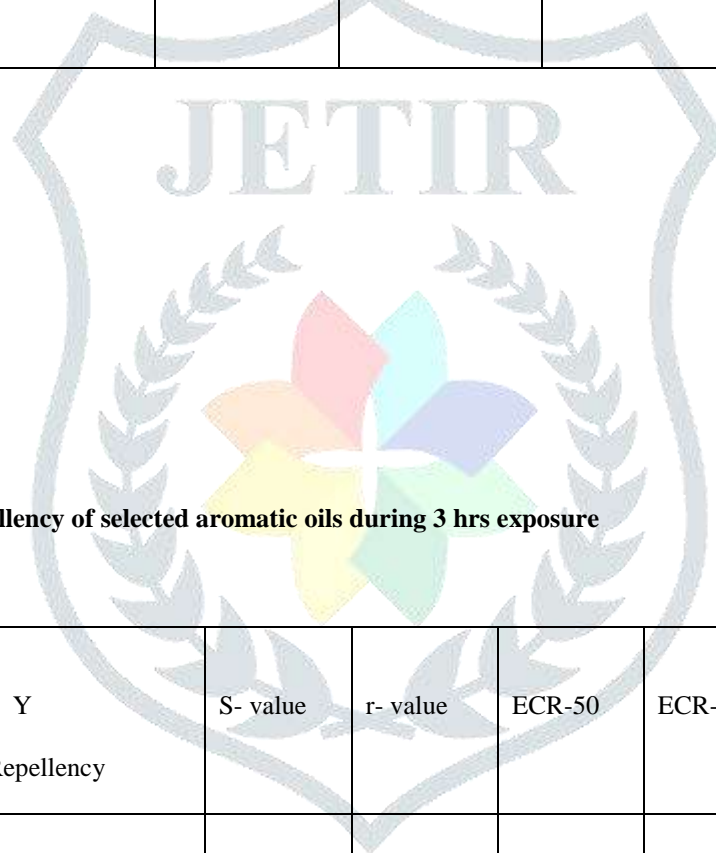


Table3: This table is showing repellency of selected aromatic oils during 3 hrs exposure

Aromatic oils	Y Repellency	S- value	r- value	ECR-50	ECR-75	ECR-95
Tea tree oil	R=1.914+1.283C	17.06	0.829	2.403	4.350	5.908
Lavender	R=2.051+1.604C	18.099	0.868	1.837	3.396	4.643
Pepper mint	R=1.892+1.011	16.960	0.762	3.072	5.543	7.520

Table4: This table is showing repellency of selected aromatic oils during 6 hrs exposure

Aromatic oils	Repellency (Y)	S- value	r- value	ECR-50	ECR-75	ECR-95
Tea tree oil	$R=1.393+8.326$	12.789	0.789	4.330	7.333	9.735
Lavender	$R=1.546+9.061$	13.751	0.793	3.811	6.570	8.777
Pepper mint	$R=9.521+7.024$	10.404	0.800	5.762	9.322	12.169

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