Defence Science Journal, Vol. 52, No. 1, January 2002, pp.73-76 © 2002, DESIDOC

#### SHORT COMMUNICATION

# **Bio-repellents for Land Leeches**

# D.R. Nath, N.G. Das and S.C. Das Defence Research Laboratory, Tezpur-784 001

#### ABSTRACT

Oils of Callistemon rigidus (bottle brush), Zanthoxylum armatum, DC. syn. Zanthoxylum alatum Roxb (timur), Azadirachta indica (neem) and synthetic repellent N, N-diethyl-meta-toluamide (DEET) and dimethyl phthalate (DMP) were evaluated for leech repellency properties and persistance on cloth. In statistical analysis, bio-repellents were found at par with DMP. Bottle brush oil afforded better protection against land leeches than *timur* oil, *neem* oil and synthetic repellent DMP at all the three concentrations evaluated. However, DEET was found superior to all the repellents evaluated and remained effective for 19.8 days at 0.93 mg/cm<sup>2</sup> followed by 7 days and 6.3 days at 0.56 mg/cm<sup>2</sup> and 0.37 mg/cm<sup>2</sup> treatment, respectively, while bottle brush oil was found effective for 13 days at 0.93 mg/cm<sup>2</sup>.

Keywords: Bio-repellents, synthetic repellents, dimethyl phthalate (DMP), timur oil, neem oil, land leeches, N, N-diethyl-meta-toluamide (DEET)

### **1. INTRODUCTION**

Land leeches are adaptive terrestrial class of annelids, well known for their biting nuisance to man and domestic animals in sub-Himalayan mountain range. Sanguivorous nature of leeches with hirudin in their saliva causes serious loss of blood and resulting wound takes fairly long time to heal. Infestation of land leeches is very common in sylvan habitats having moist and humid climate. They aggregate in places away from sunshine due to attraction of certain microclimatic factors. Field control of leeches with insecticides and chemicals has been tried with limited scope and success but found not feasible due to involvement of vast forested area, difficult terrain and irreversible damage to ecosystem and non terget organisms. Use of repellents as personal protection measures seems to be the most acceptable solution to prevent leech bite. Repellent-impregnated cloth sleeves for legs found to give protection up to 90 days<sup>1</sup>. Essential oils and extract of plants were also used with promising results<sup>1, 2</sup>. Repellents of plant origin are preferred over synthetic repellents as prolonged use of the later could be harmful due to their toxicity and chemical nature<sup>3-6</sup>.

In the present investigation, repellent properties of the Callistemon rigidus (bottle brush), Zanthoxylum armatum, DC. syn. Zanthoxylum alatum Roxb (timur) and Azadirachta indica (neem) oil-impregnated cloth have been evaluated against land leeches and compared with synthetic repellents N,N-diethyl-meta-toluamide (DEET) and dimethyl phthalate (DMP).

### 2. MATERIALS & METHODS

The area selected for the study was the Tawang valley which is located at 27.33' north latitude and 91.48' east longitude in

Revised 28 February 2001

Arunachal Pradesh at 3200 m on Himalayan range in the northeastern part of India. It is a district headquarter township, mainly inhabitated by Monpas, Adis and Tibetians. The climate of Tawang is fairly cold (21.5 °C to 7.0 °C) and remains snowbound for three months (December to February). Extreme change of climate is observed during May to August due to heavy rainfall (0.2 mm to 48.0 mm) and rise in temperature (19.0 °C to 21.5 °C). Relative humidity ranges between 75 per cent and 100 per cent. The moist and humid climate causes heavy infestation of land leeches in jungles, hill slopes and pathways. Leeches remain inactive during heavy rain and hot weather' but become active during light drizzle. Present trials were carried out during June to July. The temperature and relative humidity recorded were 20.0 °C to 21.5 °C and 70 per cent to 100 per cent, respectively. Rainfall and soil temperature ranged between 0.2 mm and 48.6 mm and 17.1 °C to 17.3 °C, respectively. Infestation of leeches was very high in the study area. It was mainly dominated by Haemadipsa zeylanica, the defender of mountains with Haemadipsa sylvestris and Haemadipsa ornata.

The essential oils of bottle brush and Zanthoxylum armatum were obtained by steam distillation of leaf/seed pericarp. Neem oil was obtained from Unjha Ayurbedic Pharmacy, Unjha, N. Gujarat. The two synthetic repellents DEET and DMP procured from Fluke Buch, Switzerland and High Purity Chemicals, New Delhi, respectively were also evaluated as reference.

Methodology described by Nath<sup>1</sup>, et al. was adopted for evaluation of repellents and their persistance on cloth against land leeches. White poplin cloth sleeves (90 cm  $\times$  60 cm) were stitched (for legs) and impregnated with desired quantity of repellents disolved in acetone (260 ml) for one pair of sleeves. Field trials were conducted with leech repellent concentration of 2 per cent (0.37 mg/cm<sup>2</sup>), 3 per cent (0.56 mg/cm<sup>2</sup>) and 5 per cent (0.93 mg/cm<sup>2</sup>). In all instances, cloth sleeves were treated with repellents 24 hr prior to conducting the experiments. Six volunteers (five treated and one control) were taken to a leech-infested area. Two-cloth sleeves treated with two different repellents at the same concentration were given to each subject for wearing on both legs as per Round Robin or incomplete block design test<sup>1,8,9</sup>. Subjects were asked to walk through the leech-infested foottract on the hill slope for 10 min. Simultaneously, a control subject wearing untreated cloth sleeves was also asked to walk through the same path. Leeches attached to untreated cloth sleeves were recorded as control landing. Number of days between impregnation and first landing/ attachment of leeches on cloth sleeves was recorded as the protection period<sup>1</sup>, i.e., persistance of repellents on cloth. Ten observations were made during a day. To minimise the subject preference, if any, subjects were given cloth sleeves with new combination of repellents by rotation on each successive day of experiment. Five sets of experiments were conducted with one control (untreated) in each set. The treated and untreated (control) cloth sleeves were kept in polythene bags seperately for each concentration during the days of experiments. Trials were conducted during early hours of the day when the leeches become active. Data obtained were analysed as per Fisher's F-test.

### 3. RESULTS & DISCUSSION

Results of the field trials revealed that the herbal oil of bottle brush afforded better protection than *timur* oil, *neem* oil and synthetic repellent DMP at all the three concentrations. Bottle brush oil exhibited higher persistancy (13 days) at 5 per cent (0.93 mg/cm<sup>2</sup>) treatment, while *timur* oil, *neem* oil and DMP afforded protection for 11.5 days, 7 days and 11.8 days, respectively. However, synthetic repellent DEET exhibited the highest persistancy (19.8 days) at the same concentration and maintained its supfemacy althrough the experiments (Table 1).

Statistical analysis of the data revealed that synthetic repellent DEET exhibited the highest persistancy than the other three repellents, affording maximum protection time in all the treatments. Bottle brush oil afforded significantly better persistancy than *timur* oil, *neem* oil and DMP at 2 per cent (0.37 mg/cm<sup>2</sup>) treatment. At 3 per cent

74

land leeches.					
Repellent	Conc (mg/cm²)	Adjusted mean (days)	ratio	Tabulated F value at 1% level	
Bottle brush oil	0.37	4.5*	24.623	5.41	1.02
Timur oil		2.3			
<i>Neem</i> oil		2.3			
DEET		6.3*			
DMP	•	3.3			
Bottle brush oil	0.56	<b>4.8</b> *	17.952	5.41	1.12
<i>Timur</i> oil		3.8			
<i>Neem</i> oil		3.3			
DEET		7.0*			
DMP		3.5			
Bottle brush oil	0.93	13.0	78.675	5.41	1.60
<i>Timur</i> oil		11.5			
Neem oil		7.0			
DEET		19.8 <sup>*</sup>			
DMP		11.8			

Table 1.	Results of persistancy of repellents on cloth against
	land leeches.

Control: 26-34 leeches/man hr

\* Least significant difference, P value > 0.01

No. of replecates: 5

concentration, bottle brush oil was found at par with *timur* oil and significantly better than *neem* oil and DMP. However, no significant difference was observed in protection time among bottle brush oil, *timur* oil and DMP at 5 per cent (0.93 mg/cm<sup>2</sup>).

Comparing the experimental data obtained with earlier observations made by Nath<sup>1</sup>, et al. it was observed that much lower concentrations of DEET and DMP were required in the present trials to achieve the identical results. Persistance on cloth of bottle brush oil, *timur* oil and *neem* oil was much longer than the oils of Cinamon and Citronella<sup>10</sup>. Saxena<sup>7</sup>, et al. also used much higher concentration of seed extract. Kumar<sup>11</sup>, et al. obtained 7.25 hr and 2.5 hr of protection with DEET and DMP, respectively when applied at 0.5 mg/cm<sup>2</sup> directly on skin. In the present trial, 3.3 days to 4.8 days protection was obtained with 0.56 mg/cm<sup>2</sup> treatment of bio-repellents on poplin cloth sleeves. Similarly, DEET and DMP also provided protection for 7 days and 3.5 days, respectively. Results of the experiment further conformed the earlier observations of Nath', *et al.* It was also observed during trials that users prefer cloth sleeves impregnated with bio-repellents than the synthetic repellents like DEET and DMP. This may be due to natural fragnance, compatibility and long association of bioproducts with mankind.

Many plants are known to produce chemicals with insecticidal and repellent properties which are environmentally safe than the synthetic ones. Among these, *neem* has created a lot of interest due to its anti-larval and repellent properties<sup>12</sup>. To add, essential oils of *timur* and bottle brush can be good bio-repellent for haematophagous insects and leeches.

Impregnation of cloth with desired concentration of repellents preferably of bio-origin seems to be a practical proposition to get rid of the bites of leeches in the jungle rather than through chemical control, affecting the environment.

## ACKNOWLEDGEMENT

The authors wish to thank Dr M.P. Chacharkar, Director, Defence Research Laboratory, Tezpur, for his valuable suggestions and critically going through this paper.

#### REFERENCES

- Nath, D.R.; Das, N.G. & Das, S.C. Persistance of leech repellents on cloth. *Ind. J. Med. Res.*, 1993, (A)97, 128-31.
- Nath, D.R.; Das, N.G. & Malhotra, P.R. Efficacy of certain essential oils against land leeches. Def. Sci. J., 1986, 36(3), 327-30.
- 3. Skinner, W.A. & Johnson, H.L. The design of insect repellents. *In* Drug design, Vol. 10, edited by E.J. Arients. Academic Press, New York, 1980. pp. 277-02.
- 4. Rabbins, P.J. & Cherniack, M.G. Review of bio-distribution and toxicity of insect repellent N,N, diethyl-m-toluamide (DEET). J. Toxicol. Environ. Health, 1986, 18, 503-25.

- Ansari, M.A.; Sharma, V.P.; Razdan, R.K. & Mittal, P.K. Evaluation of certain mosquito repellents marketed in India. *Ind. J. Malariol.*, 1990, 27, 56-64.
- Lipscomb, J. W.; Kramer, J.E. & Leikin, J.B. Seizure following brief exposure to insect repellent N,N-diethyl-m-toluamide. Ann. Emerg. Med., 1992, 21, 315-17.
- Saxena, B.N.; Dubey, D.N. & Nair, L.N. Studies on the insecticidal and repellent properties of the seed extract of *Tephrosia purpuria* (Linn.) Pers. *Def. Sci. J.*, 1974, 24(2), 43-48.
- Gilbert, I.H.; Gouck, H.K. & Smith, C.N. New insect repellent. Soap Chem. Spec., 1957, 33, 129.

- Das, S.C.; Bhuyan, M.; Das, N.G. & Malhotra, P.R. Field trials on the relative efficacy of five repellents against Simulium himalayense (Diptera: Simuliidae). Ind. J. Med. Res., 1985, 81, 378-81.
- Dixit, R.S.; Saxena, B.N. & Khalsa, H.G. Evaluation of repellents against land leeches. Labdev J. Sci. Technol., 1967, 5(2), 140-45.
- Kumar, S.; Prakash, S.; Sharma, R.K.; Join, S.K.; Kalyanasundaram, M.; Swami, R.V. & Rao, K.M. Field evaluation of three repellents against mosquitoes, black flies and land leeches. *Ind. J. Med. Res.*, 1984, 80, 541-45.
- Sharma, V.P.; Ansari, M. A. & Razdan, R.K. Mosquito repellent action of *neem* oil. J. Amer. Mosq. Contr. Assoc., 1993, 9(3), 359-60.

Contributor



**Mr DR Nath** obtained his MSc Zoology (Entomology). He is working as Technical Officer B at the Defence Research Laboratory, Tezpur. His area of specialisation is medical entomology. He has published 22 papers in national/international journals.