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## **RESEARCH ARTICLE**

#### Repellent effect of eucalyptus oil applied as paint against house rat, Rattus rattus

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#### Abstract

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..... Increasing community awareness of the moral and animal welfare issues associated with conventional pest animal control has focused interest on nonlethal alternatives such as the use of repellents. Rodent repellents are chemicals which by taste or odour or possibly by both will prevent animal from feeding or gnawing. Mature and healthy house rat, Rattus rattus of both sexes were exposed to 5, 10 and 20% eucalyptus oil applied as paint in laboratory pens in bi-choice tests. Each concentration was applied through three different modes of application i.e. daily, once a week, and alternatively per week. Repellent effect of the oil was assessed based on food consumption from treated and untreated sides for four days. Food consumption was found to be significantly (P  $\leq 0.05$ ) low from treatment side compared to the untreated side at all the concentrations tested in both the sexes. Repellent effect of the oil was found to differ significantly between the two sexes. Percent repellency in both male and female rats was apparently more with daily application of the eucalyptus oil indicating repellent effect of the eucalyptus oil against R. rattus. Further studies may, however, be conducted to enhance the persistence of the repellent effect for longer period of time.

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#### Introduction

Rodents are significant economic pests belonging to the largest, and one of the most interesting groups of mammals. Analysis of the information available on damage and economic losses caused by rodents in various crop fields, horticulture and forestry, poultry farms, rural and urban dwellings and storage facilities in India showed that chronic damages ranging from 2 to 15% persist throughout the country (Parshad, 1999). Rodents destroy food by contaminating with their urine, faecal droppings and hair. They also act as reservoirs of numerous diseases that infect humans, domestic animals and other wildlife species (Singla *et al.*, 2008, 2012). The excessive use of rodenticides has led to increased environmental pollution, direct and indirect poisoning of non-target organisms (Brakes and Smith, 2005) and development of resistance among commensal rodents and the associated impact on pest-control programmes world-wide (Cowan *et al.*, 1995). Increasing community awareness of the moral and animal welfare issues associated with conventional pest animal control has focused interest on non-lethal alternatives (Bomford and O'Brie, 1992).

Natural products are an excellent alternative to synthetic pesticides (Isman and Machial, 2006) as a means to reduce negative impacts to human health and the environment. Many secondary plant metabolites are known for their insecticidal properties, and in many cases, plants have a history of use as home remedies to kill or repel insects (Broussalis *et al.*, 1999; Pavela, 2004). Essential oils from plants belonging to several species have been tested to assess their repellent properties as a valuable natural resource. Among the plant families with promising essential oils used as repellents include *Cymbopogon* spp., *Ocimum* spp., *Thymus* spp., *Eucalyptus* spp. etc (Koul *et al.*, 2008). Plant based non-lethal repellents are most suitable for rodent control (Meehan, 1988, Nolte *et al.*, 1993). These are easy to extract, biodegradable and do not persist in soil and water. These can be useful for the prevention of rodent damage to grains in stores and seeds and seedlings in crop fields and nurseries. Eucalyptus is particularly

useful as it possesses a wide range of desirable properties for pest management and is regarded as non-toxic to humans. The essential oils obtained from the eucalyptus have many medicinal and commercial uses. The oils possess many bioactivities such as antimicrobial, antiviral, fungicidal, insecticidal and herbicidal (Zhang *et al.*, 2010).

Eucalyptus oil is the generic name for distilled oil obtained from the leaf of *Eucalyptus*, a genus of the plant family Myrtaceae native to Australia and cultivated worldwide. Its chief constituent is eucalyptol (cineole), a colourless liquid with camphor like odour and cooling taste. Essential oils of eucalyptus appear particularly potent as mosquito repellents (Choi *et al.*, 2002). Eucalyptus oil can also protect plants against rice weevils, pine processionary moths, mushroom flies (Batish *et al.*, 2008) and mites (Han *et al.*, 2011). Relatively little work has been carried out on plant derived repellents compared to other aspects of rodent control. No study has yet been made on evaluating the potential of eucalyptus oil as repellent against rodent pests. The aim of present study was to evaluate the potential of eucalyptus oil applied as paint as repellent against house rat, *Rattus rattus*, the predominant commensal rodent pest species.

#### **Material and Methods**

The present work was carried out in Animal House Laboratory, Department of Zoology, Punjab Agricultural University (PAU), Ludhiana, India. Commercially available pure eucalyptus oil was used for present study.

#### **Collection and Maintenance of Animals**

For present studies, the house rat, *R. rattus* of both sexes were trapped with the help of single catch and multi catch rat traps from store houses and poultry farms in and around Ludhiana. In the laboratory, rats were acclimatized individually in cages of size 36 x 23 x 23cm for 15-20 days before the commencement of experiment with food and water provided *ad libitum*. Food was prepared by mixing cracked wheat, powdered sugar and groundnut oil (WSO bait) in ratio 96: 2: 2. Animals were used and maintained as per the guidelines of Institutional Animal Ethics Committee. After acclimatization, healthy and mature rats of both sexes were weighed and selected for experimentation.

#### **Experimental Design**

Three different concentrations of eucalyptus oil i.e. 5, 10 and 20% were tested. Different concentrations were prepared by diluting the oils in isopropyl alcohol. For each concentration, a total of twelve rats (six of each sex) were taken. Each concentration of the oil was applied as paint (by using cotton swab dipped in the oil). Rats were exposed to each concentration of the oil for 3 weeks using three different modes of application i.e. oil applied daily, once a week and alternatively. The effect of treatment was recorded after every 24 h for 4 days in a week.

A total of four laboratory pens (each of size  $252 \times 100 \times 72$  cm), were used for each concentration. Each pen consisted of three chambers of equal size. One rat was released in each chamber. Each chamber in a laboratory pen, on its opposite facing sides was connected with holes (each of diameter 6 cm) to two small nest boxes (each of size  $20 \times 15 \times 15$  cm). Rats had free access to these nest boxes. Treatment was carried out in the nest box of one side of each chamber. Oil as paint was applied on all the interior sides of the nest box.

## **Repellent effect**

Repellent effect of the oil was assessed based on the consumption of WSO bait by the rat from the bowls kept in two nest boxes of a chamber in a laboratory pen. Bait consumption was recorded daily after every 24 h from both treated and untreated sides for 4 days in a week to determine mean daily bait consumption (g/100g bw). Based on mean daily bait consumption data, percent repellency was determined using the formula given below:

Percent repellency = 
$$\frac{BUT - BT}{BUT} \times 100$$

Where,

BUT is the mean daily bait consumption from untreated side and BT is the mean daily bait consumption from treated side.

#### **Statistical Analysis**

Values were determined as mean  $\pm$  SD. Significance of differences was determined at 5% level of significance. Significance of differences in bait consumption and percent repellency among three concentrations of oil, three modes of application, four days of application and between the two sexes and treatment and untreated sides of a chamber in a laboratory pen provided to each rat were determined by student's t-test and analysis of variance. The statistical software used was SAS version 9.3.

## **Results and Discussion**

Statistical analysis of the data revealed significantly ( $P \le 0.05$ ) low consumption of bait from treatment side compared to untreated side at all three concentrations and modes of application (Tables 1-3) indicating repellency of eucalyptus oil when applied as paint.

Table. 1- Bait consumption	by <i>Rattus rattus</i> in res	sponse to application of a	5% eucalyptus oil as paint
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			( <b>9</b> , _ 0, <b>9</b> , )			
Mode of application	Days of application	Female rats (n = 6) (Body wt =148.33±30.23g)		Male rats (n = 6) (Body wt = 158.33±25.44g)		
		Treatment side	Untreated side	Treatment side	Untreated side	
	Day 1	1.82±0.82 <sup>a</sup>	3.35±0.37 <sup>в</sup>	0.69±0.52 <sup>a</sup>	8.31±3.97 <sup>b</sup>	
	Day 2	1.83±1.26 <sup>a</sup>	2.88±1.13 <sup>b</sup>	0.49±0.57 <sup>a</sup>	8.89±2.53 <sup>b</sup>	
Ι	Day 3	1.26±1.04 <sup>a</sup>	3.04±3.11 <sup>b</sup>	0.75±1.39 <sup>a</sup>	9.82±2.63 <sup>b</sup>	
	Day 4	0.86±0.78 <sup>a</sup>	4.65±1.25 <sup>b</sup>	3.84±3.25 <sup>a</sup>	7.27±3.98 <sup>b</sup>	
	Average	$1.44 \pm 0.40^{\text{A}}$	3.48±0.69 <sup>B</sup>	1.44±1.38 <sup>A</sup>	8.57±0.92 <sup>B</sup>	
	Day 1	1.45±1.73 <sup>a</sup>	4.50±1.65 <sup>b</sup>	0.85±1.09 <sup>a</sup>	9.84±1.49 <sup>b</sup>	
	Day 2	1.61±1.26 <sup>a</sup>	4.33±2.66 <sup>b</sup>	4.06±4.29 <sup>a</sup>	7.52±3.44 <sup>b</sup>	
II	Day 3	1.51±1.49 <sup>a</sup>	2.71±1.90 <sup>b</sup>	5.02±3.92 <sup>a</sup>	2.72±3.85 <sup>b</sup>	
	Day 4	2.42±1.49 <sup>a</sup>	1.49±1.47 <sup>b</sup>	4.93±3.34 <sup>a</sup>	4.63±4.70 <sup>b</sup>	
	Average	1.74±0.39 <sup>A</sup>	3.25±1.23 AB	3.71±1.69 <sup>A</sup>	6.17±2.71 AB	
	Day 1	1.56±1.10 <sup>a</sup>	3.17±1.58 <sup>b</sup>	3.47±2.32 <sup>a</sup>	6.98±3.60 <sup>b</sup>	
	Day 2	1.68±0.74 <sup>a</sup>	3.30±1.23 <sup>b</sup>	3.04±2.63 <sup>a</sup>	4.60±4.67 <sup>b</sup>	
III	Day 3	1.93±0.78 <sup>a</sup>	3.38±1.16 <sup>b</sup>	2.29±2.43 <sup>a</sup>	6.46±5.06 <sup>b</sup>	
	Day 4	1.27±0.88 <sup>a</sup>	$4.04\pm0.79^{b}$	3.84±3.25 <sup>a</sup>	7.27±3.98 <sup>b</sup>	
	Average	1.61±0.28 <sup>A</sup>	3.47±0.33 <sup>B</sup>	3.16±0.57 <sup>A</sup>	6.32±1.03 <sup>B</sup>	

Mean daily bait consumption (g/100g bw) (Mean±SD)

- I = Daily, II = Once a week, III = Alternatively

- Values with similar superscripts in a column for four days of application (a or b) and for average values (A or B) at each mode of application indicate no significant difference.

- Values with different superscripts in a row for each sex for four days of application (a-b) and for average values (A-B) at each mode of application indicate significant difference at  $P \le 0.05$ .

Tab	e 2. Bait consumption by	<b>Rattus rattus</b>	in response t	to application	on of 10% e	eucalyptus o	il as paint	

		Mean daily bait consumption (g/100g bw) (Mean±SD)					
Mode of	Days of		rats (n = 6)	Male rats (n = 6)			
application	application	(Body wt =156.66±24.94g)		(Body wt =143.33±22.85g)			
application	аррисацон	Treatment side	Untreated side	Treatment side	Untreated side		
	Day 1	1.98±1.53 <sup>a</sup>	5.87±2.70 <sup>b</sup>	0.28±0.62 <sup>a</sup>	9.99±3.98 <sup>b</sup>		
	Day 2	2.40±2.19 <sup>a</sup>	6.19±1.60 <sup>b</sup>	0.98±1.94 <sup>a</sup>	9.30±4.61 <sup>в</sup>		
Ι	Day 3	1.36±0.79 <sup>a</sup>	3.67±1.93 <sup>b</sup>	6.25±4.11 <sup>a</sup>	2.93±2.60 <sup>b</sup>		
	Day 4	2.25±0.61 <sup>a</sup>	4.04±0.96 <sup>b</sup>	6.86±7.03 <sup>a</sup>	7.39±4.61 <sup>b</sup>		
	Average	1.99±0.39 <sup>A</sup>	4.94±1.10 <sup>B</sup>	3.59±2.98 <sup>A</sup>	$7.40\pm2.75^{\text{AB}}$		
	Day 1	1.16±1.16 <sup>a</sup>	3.26±1.26 <sup>b</sup>	6.36±4.88 <sup>a</sup>	7.43±3.71 <sup>b</sup>		
	Day 2	1.90±1.20 <sup>a</sup>	3.66±1.08 <sup>b</sup>	6.15±4.20 <sup>a</sup>	6.06±5.14 <sup>b</sup>		
II	Day 3	2.70±0.96 <sup>a</sup>	3.86±0.83 <sup>b</sup>	8.70±5.65 <sup>a</sup>	7.75±4.85 <sup>b</sup>		
	Day 4	2.30±1.21 <sup>a</sup>	5.33±1.95 <sup>b</sup>	9.83±5.94 <sup>a</sup>	3.26±2.93 <sup>b</sup>		
	Average	2.01±0.56 <sup>A</sup>	4.02±0.78 <sup>B</sup>	7.76±1.55 <sup>A</sup>	$6.12 \pm 1.77^{AB}$		
	Day 1	1.18±1.34 <sup>a</sup>	2.59±1.37 <sup>b</sup>	3.66±4.79 <sup>a</sup>	9.59±4.66 <sup>b</sup>		
	Day 2	2.33±0.55 <sup>a</sup>	3.65±1.21 <sup>b</sup>	0.90±1.10 <sup>a</sup>	8.13±4.39 <sup>b</sup>		
III	Day 3	1.85±1.54 <sup>a</sup>	4.57±2.37 <sup>b</sup>	4.42±0.96 <sup>a</sup>	3.52±0.74 <sup>b</sup>		
	Day 4	2.92±1.83 <sup>a</sup>	3.42±1.23 <sup>b</sup>	6.07±4.46 <sup>a</sup>	11.02±5.99 <sup>b</sup>		
	Average	2.07±0.63 <sup>A</sup>	3.55±0.70 <sup>B</sup>	3.76±1.86 <sup>A</sup>	8.06±2.81 <sup>B</sup>		

- I = Daily, II = Once a week, III = Alternatively

- Values with similar superscripts in a column for four days of application (a or b) and for average values (A or B) at each mode of application indicate no significant difference.
- Values with different superscripts in a row for each sex for four days of application (a-b) and for average values (A-B) at each mode of application indicate significant difference at  $P \le 0.05$ .

Table 3. Bait consumption by <i>Rattus rattus</i> in response to application of 20% eucalyptus oil as paint									
	Mean daily bait consumption (g/100g bw) (Mean±SD)								
Modo of	Derref	Female	rats $(n = 6)$	Male rats (n = 6)					
Mode of application	Days of	(Body wt =	141.66±29.10g)	$(Body wt = 147.50 \pm 18.20g)$					
	application	Treatment side	Untreated side	Treatment side	Untreated side				
	Day 1	2.36±1.12 <sup>a</sup>	5.54±1.51 <sup>b</sup>	1.95±3.10 <sup>a</sup>	10.04±5.22 <sup>b</sup>				
	Day 2	2.64±1.68 <sup>a</sup>	4.82±2.44 <sup>b</sup>	8.02±3.60 <sup>a</sup>	15.33±2.13 <sup>в</sup>				
Ι	Day 3	2.91±1.34 <sup>a</sup>	5.48±2.74 <sup>b</sup>	1.21±1.87 <sup>a</sup>	11.05±1.82 <sup>b</sup>				
	Day 4	4.39±1.79 <sup>a</sup>	7.91±2.65 <sup>b</sup>	3.4±2.77 <sup>a</sup>	8.72±2.24 <sup>b</sup>				
	Average	$3.07\pm0.78^{\text{A}}$	5.93±1.17 <sup>B</sup>	$3.64 \pm 2.64^{\text{A}}$	$11.28 \pm 2.47$ <sup>B</sup>				
	Day 1	2.83±1.78 <sup>a</sup>	4.47±1.56 <sup>b</sup>	2.15±2.63 <sup>a</sup>	13.09±3.24 <sup>b</sup>				
	Day 2	2.90±1.29 <sup>a</sup>	5.88±2.18 <sup>b</sup>	2.43±1.92 <sup>a</sup>	9.29±3.73 <sup>b</sup>				
II	Day 3	2.56±1.67 <sup>a</sup>	3.58±1.46	6.13±3.27 <sup>a</sup>	7.56±5.43 <sup>b</sup>				
	Day 4	3.95±5.05 <sup>a</sup>	5.05±1.93 <sup>b</sup>	7.06±2.71 <sup>a</sup>	7.90±6.33 <sup>b</sup>				
	Average	3.06±4.74 <sup>A</sup>	4.74±0.83 <sup>B</sup>	$4.44\pm2.17^{\text{A}}$	9.46±2.19 <sup>B</sup>				
	Day 1	2.06±1.16 <sup>a</sup>	4.64±2.51 <sup>b</sup>	1.47±1.52 <sup>a</sup>	8.67±6.38 <sup>b</sup>				
	Day 2	1.49±0.91 <sup>a</sup>	5.56±1.51 <sup>b</sup>	1.06±1.13 <sup>a</sup>	7.42±2.73 <sup>b</sup>				
III	Day 3	3.11±1.58	5.19±1.56 <sup>b</sup>	2.73±2.80 <sup>a</sup>	7.96±5.30 <sup>b</sup>				
	Day 4	1.65±1.80 <sup>a</sup>	5.15±1.24 <sup>b</sup>	3.41±2.12 <sup>a</sup>	7.4±2.66 <sup>b</sup>				
	Average	2.07±0.63 <sup>A</sup>	5.13±0.32 <sup>B</sup>	2.16±0.94 <sup>A</sup>	7.86±0.51 <sup>B</sup>				

- I = Daily, II = Once a week, III = Alternatively

- Values with similar superscripts in a column for four days of application (a or b) and for average values (A or B) at each mode of application indicate no significant difference.

- Values with different superscripts in a row for each sex for four days of application (a-b) and for average values (A-B) at each mode of application indicate significant difference at  $P \le 0.05$ .

#### Effect of 5% Eucalyptus Oil

The average mean daily bait consumption (g/100g bw) of four days was found to be significantly ( $P \le 0.05$ ) low from treatment side compared to untreated side in rats of both sexes when 5% eucalyptus oil as paint was applied daily and alternatively, however, no significant difference in average mean daily bait consumption of four days between treatment and untreated sides was found when the oil was applied once a week (Table 1). This may be due to the reduced effect of treatment on subsequent days due to volatile nature of the eucalyptus oil. No significant difference in average mean daily bait consumption from treatment side was observed among the three modes of application in rats of both the sexes.

Analysis of data revealed (Table 4) no significant difference in percent repellency among the three modes of application as well as among the four days of treatment in case of female rats when 5% eucalyptus oil was applied as paint. However, in male rats, significant ( $P \le 0.05$ ) difference was observed in average percent repellency between mode I (when the oil was applied daily) and mode II (when the oil was applied once a week) and similarly between mode I and mode III (when the oil was applied alternatively). There was no significant difference in percent repellency between the modes II and III (Figure 1). Percent repellency in male rats was significantly ( $P \le 0.05$ ) less on day 4 (28.34%) of application as compared to days 1 to 3 (90.75-91.15%), when 5% eucalyptus oil as paint was applied daily (Table 4). This may be due to the habituation of rats towards the smell of the oil. When the oil as paint was applied once a week (i.e. on day 1 of the week), percent repellency was significantly ( $P \le 0.05$ ) more on day 1 (92.78%) of application, compared to days 3 to 4 (18.89-27.67%). This may be due to the reduced effect of the oil due to vaporization with time. At this mode of application, the difference in percent repellency between days 1 and 3 and between days 1 and 4 was found to be significant ( $P \le 0.05$ ). No significant difference in percent repellency was observed among all the four days of treatment when the oil was applied as paint alternatively (Table 4).

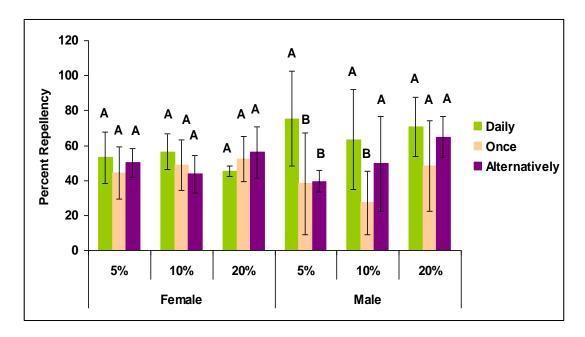


Figure 1: Average percent repellency of eucalyptus oil in *Rattus rattus* among three modes of application at three different concentrations with three different modes of application each. Bars with different superscripts differ significantly at  $P \le 0.05$ .

Significant ( $P \le 0.05$ ) difference in percent repellency was observed between male and female rats at all the four days of treatment when the oil as paint was applied daily (Figure 2). Repellency was significantly ( $P \le 0.05$ ) higher against male rats for first three days (90.75-91.15%), and higher against female rats on day 4 (78.3%) (Table 4). In other two modes of application, no significant difference was observed between rats of two sexes on all the four days of treatment. The average percent repellency among the three modes of application was not found to vary significantly between female rats, however, in male rats, the percent repellency was significantly high when the oil was applied daily (75.31%).

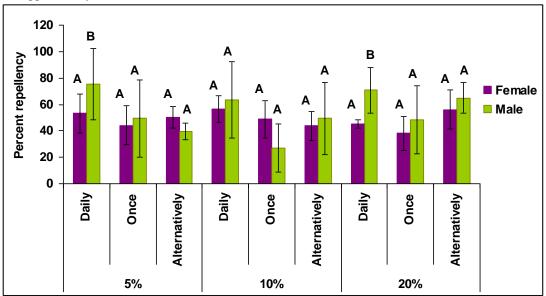


Figure 2: Average percent repellency of eucalyptus oil in *Rattus rattus* between two sexes at three different concentrations in both male and female rats. Bars with differ superscripts differ significantly at  $P \le 0.05$ .

24,5 01							
application	5%	5%		10%		20%	
	Female rats	Male rats	Female rats	Male rats	Female rats	Male rats	
Day 1	47.54±22.82 <sup>a</sup>	91.15±8.70 <sup>b</sup>	63.84±27.40 <sup>a</sup>	97.60±5.35 <sup>b</sup>	50.30±29.78 <sup>a</sup>	87.37±19.08 <sup>b</sup>	
•	43.29±35.84 <sup>a</sup>		61.30±32.44 <sup>ab</sup>	82.54±36.95 <sup>bd</sup>	44.06±31.85 <sup>a</sup>	49.44±20.43 <sup>ab</sup>	
Day 3	43.36±41.70 <sup>a</sup>	90.75±16.11 b	62.03±30.99 ac	23.51±37.35 °	44.43±20.20 <sup>a</sup>	87.73±19.34 <sup>b</sup>	
Day 4	78.30±20.81 <sup>a</sup>	28.34±35.19 <sup>°</sup>	38.90±26.38 <sup>ac</sup>	$50.00 \pm 50.00$ <sup>cd</sup>	42.45±22.74 <sup>a</sup>	58.22±39.53 <sup>ab</sup>	
Average	53.12±14.63 <sup>A</sup>	75.31±27.12 <sup>B</sup>	56.51±10.21 <sup>A</sup>	63.41±28.75 <sup>A</sup>	45.31±2.97 <sup>A</sup>	70.69±17.14 <sup>B</sup>	
Day 1	63.16±44.80 <sup>a</sup>	92.78±8.48 <sup>a</sup>	69.46±16.68 <sup>a</sup>		35.46±37.22 <sup>a</sup>	80.25±26.66 <sup>b</sup>	
Day 2	53.27±39.57 <sup>a</sup>	58.24±44.89 <sup>ab</sup>	45.47±36.02 <sup>a</sup>		49.23±15.23 <sup>ab</sup>	66.83±26.61 <sup>b</sup>	
Day 3			29.35±24.78 <sup>a</sup>		49.74±40.59 <sup>a</sup>	28.00±30.12 <sup>a</sup>	
Day 4					18.29±19.07 <sup>a</sup>	18.51±20.05 <sup>a</sup>	
Average			48.78±14.32 <sup>AB</sup>			48.39±25.80 <sup>AB</sup>	
Day 1	43.32±32.44 <sup>a</sup>	42.49±39.03 <sup>a</sup>	47.34±35.23 <sup>a</sup>	62.60±38.72 <sup>ac</sup>	49.51±28.60 <sup>a</sup>	62.33±45.02 <sup>a</sup>	
Day 2	47.32±19.63 <sup>a</sup>	33.67±37.79 <sup>a</sup>	30.78±26.41 <sup>a</sup>	78.11±35.66 <sup>°</sup>	66.56±30.89 <sup>a</sup>	81.58±19.79 <sup>a</sup>	
Day 3						66.55±36.68 <sup>a</sup>	
Day 4						49.40±33.20 <sup>a</sup>	
Average	50.05±8.23 AB	39.55±6.43 <sup>A</sup>	43.72±10.78 <sup>A</sup>	49.56±27.27 <sup>A</sup>	56.10±14.62 <sup>A</sup>	64.96±11.48 <sup>AB</sup>	
	Day 4 Average Day 1 Day 2 Day 3 Day 4 Average Day 1 Day 2 Day 2 Day 3 Day 4	Female ratsDay 1 $47.54\pm22.82^{a}$ Day 2 $43.29\pm35.84^{a}$ Day 3 $43.36\pm41.70^{a}$ Day 4 $78.30\pm20.81^{a}$ Average $53.12\pm14.63^{A}$ Day 1 $63.16\pm44.80^{a}$ Day 2 $53.27\pm39.57^{a}$ Day 3 $35.98\pm35.64^{ab}$ Day 4 $24.65\pm34.87^{ab}$ Average $44.26\pm14.92^{AB}$ Day 1 $43.32\pm32.44^{a}$ Day 2 $47.32\pm19.63^{a}$ Day 3 $45.47\pm27.25^{a}$ Day 4 $64.10\pm28.79^{a}$	application5%Female ratsMale ratsDay 1 $47.54\pm22.82^{a}$ $91.15\pm8.70^{b}$ Day 2 $43.29\pm35.84^{a}$ $91.03\pm13.64^{b}$ Day 3 $43.36\pm41.70^{a}$ $90.75\pm16.11^{b}$ Day 4 $78.30\pm20.81^{a}$ $28.34\pm35.19^{c}$ Average $53.12\pm14.63^{A}$ $75.31\pm27.12^{B}$ Day 1 $63.16\pm44.80^{a}$ $92.78\pm8.48^{a}$ Day 2 $53.27\pm39.57^{a}$ $58.24\pm44.89^{ab}$ Day 3 $35.98\pm35.64^{ab}$ $18.89\pm30.42^{b}$ Day 4 $24.65\pm34.87^{ab}$ $27.67\pm40.34^{b}$ Average $44.26\pm14.92^{AB}$ $49.39\pm28.99^{A}$ Day 1 $43.32\pm32.44^{a}$ $42.49\pm39.03^{a}$ Day 2 $47.32\pm19.63^{a}$ $33.67\pm37.79^{a}$ Day 3 $45.47\pm27.25^{a}$ $33.33\pm47.14^{a}$ Day 4 $64.10\pm28.79^{a}$ $48.71\pm43.89^{a}$	application $5\%$ 10Female ratsMale ratsFemale ratsDay 1 $47.54\pm22.82^{a}$ $91.15\pm8.70^{b}$ $63.84\pm27.40^{a}$ Day 2 $43.29\pm35.84^{a}$ $91.03\pm13.64^{b}$ $61.30\pm32.44^{ab}$ Day 3 $43.36\pm41.70^{a}$ $90.75\pm16.11^{b}$ $62.03\pm30.99^{ac}$ Day 4 $78.30\pm20.81^{a}$ $28.34\pm35.19^{c}$ $38.90\pm26.38^{ac}$ Average $53.12\pm14.63^{A}$ $75.31\pm27.12^{B}$ $56.51\pm10.21^{A}$ Day 1 $63.16\pm44.80^{a}$ $92.78\pm8.48^{a}$ $69.46\pm16.68^{a}$ Day 2 $53.27\pm39.57^{a}$ $58.24\pm44.89^{ab}$ $45.47\pm36.02^{a}$ Day 3 $35.98\pm35.64^{ab}$ $18.89\pm30.42^{b}$ $29.35\pm24.78^{a}$ Day 4 $24.65\pm34.87^{ab}$ $27.67\pm40.34^{b}$ $50.86\pm31.77^{a}$ Average $44.26\pm14.92^{AB}$ $49.39\pm28.99^{A}$ $48.78\pm14.32^{AB}$ Day 1 $43.32\pm32.44^{a}$ $42.49\pm39.03^{a}$ $47.34\pm35.23^{a}$ Day 2 $47.32\pm19.63^{a}$ $33.67\pm37.79^{a}$ $30.78\pm26.41^{a}$ Day 3 $45.47\pm27.25^{a}$ $33.33\pm47.14^{a}$ $59.37\pm31.25^{a}$ Day 4 $64.10\pm28.79^{a}$ $48.71\pm43.89^{a}$ $37.41\pm33.55^{a}$	$\begin{array}{c c c c c c c c c c c c c c c c c c c $	$\begin{array}{c c c c c c c c c c c c c c c c c c c $	

 Table 4. Percent repellency with eucalyptus oil when applied as paint using three different concentrations against *Rattus rattus* 

 Mode of
 Days of

 Percent repellency (Mean±SD) (n = 6 each)

- I = Daily, II = Once a week, III = Alternatively

- Values with similar superscripts in the column for four number of days (a, b or c) and for average values (A or B) at each mode of application indicate no significant difference in percent repellency.

- Values with different superscripts in a row for four number of days (a-c) and for average values (A-B) at each mode of application indicate significant difference in percent repellency between the two sexes at  $P \le 0.05$ .

#### Effect of 10% Eucalyptus Oil

At all the modes of application, no significant difference in bait consumption among the four days of treatment was observed in rats of both the sexes. The average mean daily food consumption at all the three modes of application was found to be significantly ( $P \le 0.05$ ) low from treatment side compared to untreated side in female rats when 10% eucalyptus oil was applied as paint (Table 2). In male rats, no significant difference in average mean daily food consumption between treatment and untreated sides was found when the 10% oil as paint was applied daily and once a week. A significant ( $P \le 0.05$ ) difference in average mean daily food consumption was, however, found between treatment and untreated sides when the oil was applied alternatively (Table 2).

Analysis of data revealed (Table 4) no significant difference in average percent repellency among the three modes of application in case of female rats, however, in male rats significant ( $P \le 0.05$ ) difference was observed between modes I and II and similarly between modes II and III. No significant difference was observed between modes I and III (Figure 1). There was no significant difference observed in percent repellency among four days of treatment at all three modes of application in case of female rats. However in male rats, percent repellency (23.51%) was significantly ( $P \le 0.05$ ) less on day 3 of application as compared to day 1 (97.60%) and 2 (82.54%) and also less on day 4 (50.00%) as compared to day 1 when 10% oil was applied as paint daily. When oil was applied once a week, percent repellency was nil on day 4 of treatment in male rats, however, the difference in percent repellency among the four days of treatment was non-significant statistically.

In male rats, when the oil was applied alternatively, the percent repellency was significantly less (7.42%) on day 3 of treatment, compared to days 1, 2 and 4. Significant ( $P \le 0.05$ ) difference was observed in percent repellency between male and female rats on day 1 of treatment with higher repellency in case of male rats (97.60%) when oil was applied daily and on day 4 of treatment with higher repellency in case of female rats (50.86%) when the oil was applied once a week. However, when the oil was applied as paint alternatively, significant ( $P \le 0.05$ ) difference was observed in percent repellency between male and female rats on day 2 and 3 with higher repellency in case of male rats on day 2 (78.11%) and in case of female rats on day 3 (59.37%). The average percent repellency at all the three modes of application was, however, not found to vary significantly between the rats of two sexes (Figure 2). This may be due to individual variations in response towards the treatment by rats. Similar individual variability in response towards triptolide treatment by *R. rattus* was also observed by Singla *et al.* (2013). Sex specific variation in response of *R. rattus* has also been reported by Kaur *et al.* (2008).

#### Effect of 20% Eucalyptus Oil

The average mean daily bait consumption at all the three modes of application was found to be significantly ( $P \le 0.05$ ) low from treatment side compared to untreated side in both male and female rats when 20% eucalyptus oil was applied as paint (Table 3). Analysis of data revealed (Table 4) no significant difference in average percent repellency among the three modes of application in case of both female and male rats (Figure 1). There was also no significant difference observed in percent repellency among four days of treatment at all three modes of application in case of female rats. In male rats, also no significant difference was observed in percent repellency among four days of treatment when 20% eucalyptus oil was applied as paint daily and alternatively. However, when the oil was applied once a week, percent repellency was significantly less on day 3 (28.00%) and day 4 (18.51%) of treatment, compared to that observed on days 1 (80.25%) and day 2 (66.83%).

Significant ( $P \le 0.05$ ) difference was observed in percent repellency between male and female rats on days 1 and 3 of treatment with higher repellency in case of male rats when the oil was applied daily. However, when the oil was applied once a week, significant ( $P \le 0.05$ ) difference in percent repellency was observed between male and female rats on day 1 with higher repellency in case of male rats (80.25%). No significant difference in percent repellency was observed between male and female rats when oil was applied as paint alternatively. The average percent repellency was found to vary significantly ( $P \le 0.05$ ) between the rats of two sexes at mode I (Figure 2). The average percent repellency at mode II and III was not found to vary significantly between the rats of two sexes. In overall, statistical analysis revealed significant difference in repellent effects of eucalyptus oil applied as paint between the two sexes and among the three concentrations of the oil tested.

Similar to present studies, consumption of feed treated with chemicals like malathion, carbaryl, captaf, bavistin, blitox, thiram, copper sulphate, sodium fluoride and neem oil by lesser bandicoot rat was very low in laboratory cages when offered in-bichoice with plain bait (Rimple, 2000). The mean daily intake of food differed non-significantly on different days suggesting that the lesser bandicoot rat developed aversion on day 1, after sampling the treated food which persisted on all the subsequent days except for captaf (0.5%) and thiram (1.0%). Siberian pine needle oil produced avoidance behavior in the rodent species in a bi-choice test (Wager-Page *et al.*,

1997). Singla and Parshad (2007) reported repellent/antifeeding potential of neem based formulation against R. *rattus* in choice experiments.

Among the various components of eucalyptus oil, 1, 8-cineole is the most important one and, in fact, a characteristic compound of the genus *Eucalyptus*, and is largely responsible for a variety of its pesticidal properties (Duke, 2004). The presence of essential oil also provides defense advantage to eucalyptus leaves against herbivory and attack by harmful insects (Brooker and Kleinig, 2006). Repellency of eucalyptus oil has also been recorded against the tick, *Ixodes ricinus* in the laboratory and field studies (Jaenson *et al.*, 2006). Application of 1, 8 cineole reduced oviposition rate by 30-50% at 1% concentration as compared to untreated controls (Koshier and Sedy, 2001). Eucalyptus oil (1%) added to sugar syrup, repelled honey bees (Patyal and Kumar, 1989). Eucalyptus oil (2%) on filter paper and wood floor repelled termites (Lin, 1998). Since eucalyptus oils possess a wide spectrum of biological activity and are regarded as safer compounds, there have been attempts to commercialize and market the insecticide/repellent products containing eucalyptus oil as such or based upon them. Quwenling is a eucalyptus-based product that has been successfully marketed as an insect-repellent in China (Trigg, 1996). It provides protection against *Anopheles* mosquitoes parallel to DEET and has, in fact, replaced the widely used synthetic repellent, dimethyl phthalate.

#### Conclusions

Present studies reveal the potential of eucalyptus oil in repelling away house rat, *R. rattus* when applied as paint, however, persistence of this repellent effect of the oil is not for longer period. Further studies may be conducted to enhance its persistence for longer period of time.

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#### References

- 1. Batish, D.R., H.P. Singh and R.K. Kohli. (2008). Eucalyptus essential oil as a natural pesticide. *Forest Ecol. Manage.*, 256: 2166-2174.
- 2. **Bomford, M. and P. O'Brien. (1992).** A role for fertility control wildlife management in Australia. In: *Proc.* 15<sup>th</sup> Vertbr. Pest Conf. (Eds., Borrecco J.E. and R. Marsh), University of California, Davis, pp. 344-348.
- 3. Brakes, C.R. and R.H. Smith. (2005). Exposure of non-target small mammals to rodenticides: short-term effects, recovery and implications for secondary poisoning. *J. Appl. Ecol.*, **42**: 118-128.
- 4. Brooker, M.I.H. and D.A. Kleinig. (2006). *Field Guide to Eucalyptus*. Vol 1, 3rd ed. South-eastern Australia, Bloomings, Melbourne.
- 5. Broussalis, A.M., G.E. Ferraro, V.S. Martino, R. Pinzo'n, J.D. Coussio and J.C. Alvarez. (1999). Argentine plants as potential source of insecticidal compounds. *J. Ethnopharmacol.*, 67: 219-223.
- 6. Choi, W.S., B.S. Park, S.K. Ku and S.E. Lee. (2002). Repellent activities of essential oils and monoterpenes against *Culex pipiens pallens*. J. Am. Mosquito Contr. Assoc., 18(4): 348-351.
- Cowan, D.P., G. Dunsford, J.E. Gill, A. Jones, G.M. Kerins, A.D. MacNicoll and R.J. Quy. (1995). The impact of resistance on the use of second-generation anticoagulants against rats on farms in southern England. *Pestic. Sci.*, 43: 83-93.
- 8. Duke, J.A. (2004). Dr. Duke's Phytochemical and Ethnobotanical databases. http://www.ars-grin.gov/duke.

- Han, J., S. Kim, B.R. Choi, S.G. Lee and Y.J. Ahn. (2011). Fumigant toxicity of lemon eucalyptus oil constituents to acaricide-susceptible and acaricide-resistant <u>*Tetranychus urticae*</u>. Pest Manage. Sci., 67(12): 1583-1588.
- Isman, M.B. and C.M. Machial. (2006). Pesticides based on plant essential oils: from traditional practice to commercialization. In: *Naturally Occurring Bioactive Compounds*. (Eds., Rai, M. and M.C. Carpinella), Elsevier, BV, pp. 29-44.
- 11. Jaenson, T.G.T., S. Garboul and K. Palsson. (2006). Repellency of oils of lemon, eucalyptus, geranium, and lavender and the mosquito repellent MyggA natural to *Ixodes ricinus* (Acari: Ixodidae) in the laboratory and field. *J. Med. Entomol.*, 43: 731-736.
- 12. Kaur, G., D.K. Kocher, H.S. Sandhu and R.S. Brar. (2008). Sex specific value of oral LD<sub>50</sub> of cholecalciferol (vitamin D<sub>3</sub>) against house rat (*Rattus rattus*). *Toxicol. Int.*, **15**: 143-144.
- 13. Koschier, E.L. and K.A. Sedy. (2001). Effects of plant volatiles on the feeding and oviposition of *Thrips tabaci*. In: *Thrips and Tospoviruses* (Eds., Marullo R. and L. Mound), CSIRO, Australia, pp. 185-187.
- 14. Koul, O., S. Walia, and G.S. Dhaliwal. (2008). Essential oils as green pesticides: potential and constraints. *Biopestic. Int.*, 4(1): 63-84.
- 15. Lin, T.S. (1998). Effects of essential oil from the leaves of seven *Eucalyptus* on the control of termite. *Forest Products Industries*, 17: 751-760.
- Meehan, A.P. (1988). Chemical Repellents. In: *Rodent Pest Management* (Ed., Prakash I.), CRC Press, Boca Raton, FL, pp. 399-400.
- 17. Nolte, D.L., J.R. Mason and L. Clark. (1993). Avoidance of bird repellents by mice (*Mus musculus*). J. Chem. Ecol., 19: 427-432.
- 18. Parshad, V.R. (1999). Rodent control in India. Integ. Pest Manage. Rev., 4: 97-126.
- 19. Patyal, S.K. and J. Kumar. (1989). Olfacto-gustatory repellency of some essential oils to the Indian honey bee, *Apis cerana indica* Fabr. *J. Indian Bee* **51**: 5-8.
- 20. Pavela, R. (2004). Insecticidal activity of certain medicinal plants. Fitoterapia 75: 745-749.
- 21. Rimple, R. (2000). Identification and application of repellents for prevention of seed damage from Bandicota bengalensis. M.Sc. Thesis, Punjab Agricultural University, Ludhiana, India.
- 22. Singla, L.D., N. Singla, V.R. Parshad, P.D. Juyal and N.K. Sood. (2008). Rodents as reservoir of parasites in India. *Integr. Zool.*, **3**: 21-26.
- 23. Singla, N. and V.R. Parshad. (2007). Antifeeding effects of a neem (*Azadirachta indica*) based formulation botanical bird repellent against the house rat (*Rattus rattus*). *Indian J. Agric. Sci.*, 77(8): 502-504.
- 24. Singla, N., L.D. Singla, K. Gupta and N.K. Sood. (2012). Pathological alterations in natural cases of *Capillaria hepatica* infection alone and in concurrence with *Cysticercus fasciolaris* in *Bandicota bengalensis. J. Parasit. Diseas.*, 37(1): 16-20.
- 25. Singla, N., G. Kaur, B.K. Babbar and B.S. Sandhu. (2013). Potential of triptolide in reproductive management of house rat, *Rattus rattus* (Linnaeus). *Integr. Zool.*, 8: 260-276.
- 26. Trigg, J.K. (1996). Evaluation of a eucalyptus-based repellent against Anopheles spp. in Tanzania. J. Am. Mosquito Contr. Assoc., 12(2): 243-246.

- 27. Wager-Page, S.S., J.R. Maso, E. Aronovand and G. Epple. (1997). The role of sensory cues and feeding context in the mediation of pine needle oils repellency in prairie voles. *National Wildlife Research Center Repellents Conference 1995*. Paper 38. http:// digitalcommons.unl.edu/ nwrcrepellants/ 38.
- 28. Zhang, J. M. An, H. Wu, R. Stanton and D. Lemerle. (2010). Chemistry and bioactivity of *Eucalyptus* essential oils. *Allelopathy J.*, 25(2): 313-331.