

**EFFECT OF CITRONELLA OIL AGAINST TICK INFESTATION IN CATTLE**

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**SUMMARY**

The study was carried out to determine the *In-vivo* effect of citronella oil against tick infestation in cattle. Six tick infested cattle and six healthy cattle were selected for the present study. Cattle infested with ticks were treated with citronella oil in sesame oil as carrier applied topically once in a day for 14 days. Therapeutic evaluation of citronella oil was done on the basis of disappearance of clinical manifestations, reduction of tick count and significant variation in haemato-biochemical profiles. The clinical manifestations such as pruritus, erythema, scales, nodules and rough hair coat disappeared at the end of the therapy. The haematological analysis showed Hb, PCV, TEC, TLC, neutrophils and eosinophils to be significantly differ ( $P<0.05$ ) in all the treated cattle at the end of therapy in comparison to pre-treatment values. Non significant alteration ( $P<0.05$ ) in lymphocytes, monocytes and basophils in comparison to pre-treatment values was observed. Biochemical profiles showed significant increase ( $P<0.05$ ) in glucose, total protein, albumin, globulin, A:G ratio, calcium and phosphorus; significant decrease in AST, ALP and CK and there was non-significant change in ALT, BUN and creatinine at the end of therapy as compared to their values before treatment. Tick count started to reduce on day 7 post therapy.

**Keywords:** Cattle, Citronella oil, Essential oil, Ticks

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**INTRODUCTION**

In tropical countries like India ticks and tick-borne diseases, especially bovine theileriosis, babesiosis and anaplasmosis, can cause sudden death of severely infected animals. The cattle tick *Rhipicephalus (Boophilus) microplus* is a significant vector of these deadly diseases (Ghosh *et al.*, 2015). The most common method for controlling tick infestation is to treat the host with synthetic acaricides like Arsenic trioxide, organochlorines, organophosphates, carbamates, amidines, pyrethroids and ivermectins etc. which kill the associated larvae, nymphs, and adults. Although this has limitations due to wide spread environmental pollution, increased risk of insecticide residue, quick development of resistance and parasite reoccurrence (Picinin *et al.*, 2017).

It has been reported that the topical treatment of animals with herbal acaricidal formulations is safe and less toxic as compared to synthetic agents (Chen *et al.*, 2019). In response to the insecticides residue problems, many researchers attempted to develop bioinsecticide products especially from the essential oil of *Cymbopogon winterianus*, the so-called Citronella oil that possesses sufficient biological properties, including repellent, acaricidal, and larvicidal and which in particular acts against *Rhipicephalus microplus* (Martins, 2006). The main objective of the present study was to observe the effect of Citronella oil on tick infested cattle on the basis of improvement in haemato-biochemical attributes, management of clinical manifestations and reduction in tick count.

**MATERIAL AND METHODS**

Tick infestation cases in Instructional Dairy Farm, Pantnagar, Uttarakhand, India were included in the present study. A total of 12 cattle were included in the study, 6 cattle were taken as healthy control with no visible tick infestation and 6 cattle were tick infested. A thorough and ongoing clinical evaluation was carried out on cattle to look for the various clinical manifestations like pruritus, alopecia, rough hair coat, presence of nodules, scales and erythematous lesions before therapy and after therapy. The ticks found on the body surface of cattle were recorded. The estimation of tick count was performed on one half of the body and then doubled as there was no random distribution of ticks on the body surface of infested animals, rather they were limited to few predilection sites (Kivaria *et al.*, 2012). A combination of 10 drops Citronella oil in 10 ml Sesame oil was prepared. The Sesame oil was used as carrier oil. The Citronella essential oil was procured from Medicinal Plants Research and Development Center, G.B.P.U.&T., Pantnagar, Uttarakhand and Sesame oil (manufactured by G.M. Pharmacy) was procured from local market. Cattle infested with ticks were treated with above combination and applied topically once in a day for 14 days.

Statistical analysis of data was performed using software SPSS-16.0. Data pertaining tick count, haematological and biochemical profiles was analysed by paired t-test and one way ANOVA technique to test the significance of means and  $P<0.05$  was accepted as statistically significant.

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## RESULTS AND DISCUSSION

Therapeutic efficacy of Citronella essential oil was evaluated in tick infested cattle at seven days interval for 21 days. The results of clinical observations after treatment with Citronella oil are presented in Table 1. On day 7 of therapy, the tick population started to decrease in all the animals of the group but was not noticeably reduced. On day 14 of therapy, the ticks were significantly reduced on 5 out of 6 animals. The remaining animal was recovered on day 21 after the start of therapy. On day 7 of the start of therapy, pruritus, erythema, nodules, rough hair coat and scaling started to reduce and there was complete disappearance of erythema, pruritus, scaling, nodules (except 1 animal) and rough hair coat on day 14 of the start of therapy and all the animals recovered on day 21 after the start of therapy. Clinical signs like pruritus, scratching, rubbing, alopecia were recorded in tick infested cattle.

Mean tick count in the cattle of the group before treatment was  $57.11 \pm 4.71$  and a significant reduction in total tick count was observed on day 7 i.e.  $30.78 \pm 2.29$  and day 14 i.e.  $10.94 \pm 0.93$  after the treatment. Percent reduction in tick count was 46.10% on 7th day of treatment and 80.84% reduction on 14th day after treatment (Table 2).

Haemoglobin, packed cell volume, total erythrocyte count, total leucocyte count, neutrophils and eosinophils showed significant differences between post treatment and pre-treatment mean values and were similar to healthy control. No significant alteration ( $P < 0.05$ ) in lymphocytes, monocytes and basophils in comparison to pre-treatment values was observed (Table 3). Significant reduction in haemoglobin, total erythrocytes count, packed cell volume and neutrophils and significant increase in total leucocyte count, eosinophils and lymphocytes was seen in tick infested cattle in comparison to non-tick infested cattle (Kaur *et al.*, 2017). Lower haemoglobin is present in tick infested animal due to anaemia because of the blood sucking nature of ticks and the haemorrhage caused by the blood sucking activity of the ticks (Soulsby, 1982). The reason for higher TLC may be due to inflammatory reactions produced because of the tick bite leading to migration of leucocytes as a response against tick bite. Higher value of eosinophils reveals introduction of foreign particle through saliva of tick in the body of affected animal continuously.

Biochemical profile of treated animals showed significant increase ( $P < 0.05$ ) in glucose, total protein, albumin, globulin, calcium and phosphorus; significant decrease ( $P < 0.05$ ) in Aspartate aminotransferase, Alkaline phosphatase and Creatine Kinase and no significant alteration in Alanine aminotransferase, Albumin: Globulin ratio, creatinine and Blood Urea Nitrogen in comparison to

**Table 1**

**Therapeutic assessment of topical Citronella oil on the basis of disappearance of clinical manifestations against tick infestation (n=6)**

Clinical examination	0 <sup>th</sup> day	7 <sup>th</sup> day	14 <sup>th</sup> day	21 <sup>st</sup> day
Presence of ticks	+++ (6)	++(6)	+(1)	-
Pruritus	+++ (6)	++(6)	+(1)	-
Erythematous lesions	++(5)	+(5)	+(1)	-
Scales	++(4)	+(4)	+(1)	-
Rough hair coat	+++ (4)	+(4)	-(0)	-
Nodules	++(4)	+(1)	+(1)	-

(Figures in parenthesis indicate the number of cattle affected out of 6 cattle of the test group)

(- = Absent; + = Mild; ++ = Moderate; +++ = Severe; ++++ = Very severe)

**Table 2**

**Acaricidal potential of Citronella oil on the basis of tick count**

Area of tick infestation	Total tick count		
	Before treatment	After treatment	
	Day 0	Day 7	Day 14
Ear pinna	$30.67 \pm 3.32^a$	$21.67 \pm 2.28^b$	$7.00 \pm 1.62^c$
Neck	$91.67 \pm 13.37^a$	$44.00 \pm 7.48^b$	$10.00 \pm 2.67^b$
Dewlap	$46.33 \pm 6.95^a$	$31.33 \pm 5.35^a$	$11.67 \pm 1.10^b$
Udder	$69.67 \pm 6.47^a$	$33.00 \pm 3.76^b$	$14.00 \pm 3.27^c$
Perineal region	$59.67 \pm 6.37^a$	$29.33 \pm 2.39^b$	$10.00 \pm 1.15^c$
Under Tail	$44.67 \pm 9.58^a$	$25.33 \pm 5.49^{ab}$	$10.00 \pm 1.15^b$
Mean tick count	$57.11 \pm 4.71^a$	$30.78 \pm 2.29^b$ (46.10%)	$10.94 \pm 0.93^c$ (80.84%)

(Figures having different superscripts across the rows are significantly different upto 5% level of significance)

pre-treatment values (Table 3). The values of glucose, total protein, albumin, globulin, calcium and phosphorus were significantly lower and values of creatine kinase was significantly higher in tick infested cattle in comparison to the healthy cattle (Pandey *et al.*, 2007). The explanation for hypoproteinemia may be the negative energy balance caused due to parasitism. The increased serum activity of CK and AST reflect the damaged skin due to the tick bite (Al-Ani and Vestweber, 2004).

Mild recovery started on day 7 post therapy and a noticeable reduction in tick count of all the cattle was observed by day 14 of therapy except one cattle in which mild infestation and clinical manifestations such as pruritus, scales, nodules and erythema were seen and which subsequently disappeared on day 21 after start of therapy. Due to the anti-tick action of the herbal preparation there was reduction in the tick burden and thus the nuisance caused by ticks was reduced thus leading to improvement in the clinico-haematobiochemical findings. *Cymbopogon*



Fig. 1. Effect of topical application of Citronella oil on tick infested cattle

Table 3

**Therapeutic assessment (Mean±SE) of Citronella oil against tick infestation in cattle on the basis of hematological and biochemical attributes of the infested cattle before and after treatment**

Parameters	Healthy group (with no visible tick infestation) (n=6)	Tick infested Animals treated with Citronella oil (n=6)	
		Before treatment	After treatment
Hb (gm/dl)	12.03±0.18 <sup>a</sup>	8.63±0.39 <sup>b</sup>	11.55±0.49 <sup>a</sup>
PCV (%)	41.03±0.25 <sup>a</sup>	36.85±0.69 <sup>b</sup>	40.21±0.34 <sup>a</sup>
TEC (×106/mm <sup>3</sup> )	8.09±0.22 <sup>a</sup>	6.33±0.38 <sup>b</sup>	7.68±0.44 <sup>a</sup>
TLC (×103/mm <sup>3</sup> )	9.76±0.20 <sup>b</sup>	12.26±0.42 <sup>a</sup>	10.81±0.39 <sup>b</sup>
DLC (%)			
Neutrophils	45.17±0.36 <sup>a</sup>	30.33±0.66 <sup>b</sup>	40.88±1.12 <sup>a</sup>
Eosinophils	1.83±0.21 <sup>b</sup>	7.16±0.60 <sup>a</sup>	2.00±0.25 <sup>b</sup>
Lymphocytes	47.16±0.38 <sup>b</sup>	60.33±1.05 <sup>a</sup>	59.16±0.70 <sup>a</sup>
Monocytes	1.66±0.15 <sup>a</sup>	2.00±0.36 <sup>a</sup>	1.50±0.56 <sup>a</sup>
Basophils	1.33±0.19 <sup>a</sup>	1.50±0.34 <sup>a</sup>	1.83±0.60 <sup>a</sup>
Glucose (mg/dl)	62.33±0.68 <sup>a</sup>	46.69±1.42 <sup>b</sup>	57.23±1.57 <sup>a</sup>
Total protein (g/l)	68.43±0.98 <sup>a</sup>	45.01±0.63 <sup>b</sup>	63.52±1.29 <sup>a</sup>
Albumin (g/l)	32.73±0.85 <sup>a</sup>	17.55±0.23 <sup>b</sup>	25.50±1.84 <sup>a</sup>
Globulin (g/l)	35.70±1.43 <sup>a</sup>	27.46±0.43 <sup>b</sup>	38.01±2.6 <sup>a</sup>
A: G	0.93±0.05 <sup>a</sup>	0.64±0.17 <sup>b</sup>	0.71±0.11 <sup>b</sup>
ALT (IU/l)	31.03±1.95 <sup>a</sup>	31.84±2.02 <sup>a</sup>	31.70±1.82 <sup>a</sup>
AST (IU/l)	86.90±6.57 <sup>b</sup>	108.02±4.32 <sup>a</sup>	75.64±5.57 <sup>b</sup>
ALP (IU/l)	135.16±3.67 <sup>b</sup>	152.37±3.79 <sup>a</sup>	140.06±2.97 <sup>b</sup>
Creatinine (mg/dl)	1.07±0.10 <sup>a</sup>	1.30±0.17 <sup>a</sup>	1.12±0.14 <sup>a</sup>
BUN (mg/dl)	21.87±0.93 <sup>a</sup>	20.94±1.36 <sup>a</sup>	19.95±0.85 <sup>a</sup>
Calcium (mmol/l)	2.90±0.11 <sup>a</sup>	1.48±0.14 <sup>b</sup>	2.13±0.21 <sup>a</sup>
Phosphorus (mmol/l)	1.93±0.04 <sup>a</sup>	1.18±0.12 <sup>b</sup>	1.49±0.10 <sup>a</sup>
CK (IU/l)	8.32±0.66 <sup>b</sup>	51.80±1.68 <sup>a</sup>	10.22±0.92 <sup>b</sup>

(Figures having different superscripts across the rows are significantly different upto 5% level of significance)

*genus* has both repellent and acaricidal action against both larvae and adult of *Boophilus (Rhipicephalus) microplus* (Chagas *et al.*, 2012). The essential oil from *Cymbopogon nardus* showed deleterious effects on engorged females of the cattle tick, and the acaricidal activity is due to the presence of citronellal (Olivo *et al.*, 2008). *Cymbopogon winterianus* showed 58.01% inhibition of oviposition by *Rhipicephalus microplus* at the dose of 50 mg/ml (Martins, 2006). *Cymbopogon winterianus* contains citronellal that shows activity against *Rhipicephalus microplus* (Singh *et al.*, 2014). Crude citronella essential oil was found effective

against various insects (ticks, mosquitoes and mites) at concentration of 0.05–15% and its effectiveness can be increased with the use of different binders (Kaur *et al.*, 2021). Many of the essential oils need a carrier oil to avoid evaporation and sesame seed oil is a potent carrier oil to be used along essential oils to increase their duration of action (Kreidel and Jhaveri, 2021). Sesame oil has been shown to be synergistic when combined with essential oils like clove essential oil against the pulse beetle, *Callosobruchus maculatus* adults (Soe *et al.*, 2019) but no reported acaricidal activity. Sesame oil contains vitamin E in abundance along

with vitamin B complex and vitamin A which helps nourish and rejuvenate skin.

### CONCLUSION

The citronella oil preparation was found to be efficacious to a varying degree in tick infestation cases as there was improvement in clinical manifestations and haemato-biochemical attributes at the end of therapy and this preparation may be recommended for the management of tick infestation in cattle.

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