Studies on Prevalence of Ixodid Ticks Infesting Cattle and Their Control by Plant Extracts

Dilpreet kaur, Kamal Jaiswal and Suman Mishra*

Dept. of Applied Animal Sciences, B.B. Ambedkar University Lucknow-226025 *Email: drsumanmishra@gmail.com

Abstract: The present study was conducted to investigate the occurrence of ticks on cattle. The survey was carried out between April, 2014 to March 2015 period at various locations in Lucknow, U.P, India. A total of 2150 cattle were examined on random basis throughout the year, out of which 1262 cattle were found infested with ticks. The overall prevalence of tick infestation in cattle was observed 58.6%. On seasonal investigation, highest tick infestation was found in rainy season (68.08%) followed by summer (59.85%) and winter (48.70%). Overall highest percentage of tick infestations was observed in animals < 1 years (77.1%). A higher prevalence was observed in female cattle (63.63%) than males (50.84%). On the basis of morphological studies, the ticks identified are Rhipicephalus (Boophilus) microplus and Hemaphysalis bispinosa out of which the R.B. microplus was highly abundant. Ticks are commonly controlled by using conventional synthetic acaricides, however it has certain drawbacks like high cost, non biodegradable, toxic to environment, left residuals in animal body and above all development of resistance in ticks. Therefore, the search for herbal alternatives is ongoing process and various researchers are exploring different genera of plants to find extracts with acaricidal properties that can be used in association with or even as an alternative to synthetic compounds. Keeping the importance of plant-based, less-toxic and effective anti-tick agent in view, twenty plants were selected in the present study and evaluated for their anti-tick properties.

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Key words: ixodid ticks, prevalence, livestock, anti-tick etc.

In India, 70% of the rural households own livestock for generating additional employment through milk, meat, and wool and eggs production. According to the latest livestock censes, Uttar Pradesh (U.P.) has got the second highest cattle population and highest buffalo population in the country. The parasitic infection is the major constraint in the livestock sector in which ectoparasitic infestation is among the serious veterinary problems (Faiz, 1972; Hourrigan, 1979; Colebrook and Wall, 2004). Among the arthropod ectoparasites, ticks are important and the most common ectoparasites of mammals, birds and reptiles worldwide (Loomis, 1986; Sonenshine, 1991). They belong to phylum Arthropoda and class Arachnida (Soulsby, 1982). Globally the ticks are second to mosquitoes as vector of infectious pathogens to humans and animals. They transmit diseases like babesiosis, theileriosis, anaplasmosis etc (Norval *et. al.*, 1984; Drummond, 1983; Jongejan and Uilenberg, 2004). In some cases, ticks have been reported to cause lowered productivity, mortality, damages hides (Branscheid and Schroer, 1997; Niyonzema and Kiltz, 1986). Over the past two decades, the incidence of tick borne diseases has increased and poses major public and animal health problems that essentially require the strategic tick control methods.

Introduction

I.

In India, tick and tick borne diseases cause an estimated loss of US \$498.7 million (more than 2000 crores) per annum (Ghosh *et al.*, 2006 and 2007; Minjauw & McLeod, 2003). The study of distribution pattern of ticks in the different agro-climatic area would provide a basis for evolving strategic and tropical control of ticks and diseases caused by them. Although 106 tick species reported from India but a systemic survey of the ticks in different region has not been done yet. Uttar Pradesh is well known for its livestock. The epidemiological conditions of the state aggravate the surplus tick population on them. Systematic survey on ixodid ticks in the area is still lacking, which hampers the tick control management system.

Ticks are mainly control by conventional acaricides. But these acaricides have undesirable effects on host organisms and the environment. Problems like environmental contamination, residues in food and feed, high costs, residual in milk and meat, development of acaricide resistance in tick stimulated research on new safe methods for tick control (Kaaya *et al.*, 2000). Tick resistance to chemical acaricides has been on the rise and it is estimated that for chemicals like arsenicals a wide range of organophosphates, carbamates, amidines and synthetic pyrethroids resistance have been developed in ticks (Willadsen &Kemp, 1988). Plant extracts used in ethno-veterinary medicine represent a cheaper and easily accessible medicine for tick control (Nchu *et al.*, 2005). The ingredients of plants and herbs are known to possess insecticidal, growth inhibiting, anti-molting and repellent activities (Habeeb, 2010). However, the seasonal availability, harvesting timing, application time, variable efficacy, uncertainty over dosages and standardization of the herbal preparations may be drawbacks (Sri

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Balaji and Chakravarty, 2010). A number of reports are available on the effect of different extracts of plant material on tick species (A Bagavan *et al.*, 2005). Kalakumar *et al.*, (2000) have evaluated acaricidal activity of custard seed oil(*Annona squamosa*), neem oil (*Azadirachta indica*) and pyrethrins against 3 tick species *Boophilus microplus*, *Hyalomma anatolicum*, *Rhipicephalus sp.* and *Haemaphysalis* both in vitro and in vivo.

Keeping in view the problem of lack of extensive tick survey, development of tick resistance and lack of plant based acaricides this study is designed to find out the prevalence of ixodid ticks in the study area and to evaluate the acaricidal effect of a herbal formulation prepared by 20 plants: Lantana camara, Ricinus communis, Eucalyptus sp., Thevetia peruviana, Chenopodium album, Cannabis sativa, Cassia fistula, Calotropis procera, Achyranthes aspera, Solanum nigrum, Tamarindus indica, Brassica alba, Cuscuta reflexa, Trachyspermum ammi, Trigonella foenum graecum, Nicotiana tabacum, Mentha sp. and Cynodon dactylon.

II. Materials And Methods

Study Area and Design

The study was conducted from April, 2014 to March, 2015 in and around Lucknow district. A cross sectional study was conducted to determine the prevalence of ticks in cattle. All the animals were randomly selected as sampling unit and were checked thoroughly for any tick infestation. The level of tick burden was estimated based on number of ticks infested. The evaluation of plant extract for anti-tick activity was done by using bioassay proposed by FAO (2004)

Methodology:

1. For prevalence Study:

Adult ticks were collected from different body parts of the cattle The half body tick counts was doubled to obtain whole body tick burden (Keiser, 1987). The ticks (un-engorged, semi-engorged, fully engorged) were removed out of the skin of cattle with the help of blunt forcep without damaging their mouth parts. Collected adult ticks from each body region were kept separately for identification in sample vial containing 70% Alcohol. The vials were brought in the Parasitology laboratory of Babasaheb Bhimrao Ambedkar University, Lucknow for identification using stereomicroscope. The identification was done by the standard keys of Walker *et al.*, 2003.

2. Plant Collection and Preparation of Extract:

The leaves/seeds/stem of 20 plants were selected on the basis of extensive literature survey. The plants selected for the study have ethnoveterinary values. These plant materials were collected from BBAU campus and nearby areas. The plants were: Lantana camara, Ricinus communis, Eucalyptus sp., Thevetia peruviana, Chenopodium album, Cannabis sativa, Cassia fistula, Calotropis procera, Achyranthes aspera, Solanum nigrum, Tamarindus indica, Brassica alba, Cuscuta reflexa, Trachyspermum ammi, Trigonella foenum graecum, Nicotiana tabacum, Mentha sp. and Cynodon dactylon. The dried leaves/stem/seeds were crushed, powdered and mixed in distilled water for 3 days. After that the mixture was filtered out and the filtrate was kept in water bath for complete drying. Dried aqueous extract thus obtained was kept in properly sealed vials and stored for further use. A concentration of 2000 ppm and 3000ppm were made from the extract for bioassay.

3. Bioassay:

A series of filter paper was treated with each concentration (2,000 ppm and 3,000 ppm) of extracts. Five envelops were impregnated with each tested solution. Each filter paper was folded to form a packet. 10 ticks were placed in each packet with the help of forceps and brush. These packets were closed and incubated for 24 hr, 48 hr and 72 hr, after which alive and dead ticks were counted. Percentage mortality was calculated after each interval of 24 hour for three days.

III. Results

Overall Prevalence of ixodid ticks:

A total of 2150 cattle were observed during the study period in which 1262 cattle were observed to be infested with ticks. Overall prevalence was calculated by dividing the number of positive samples by the total sample size, then multiplied by 100. The prevalence was found to be 58.74 %. A total of 1412 ticks were collected from different body parts of cattle. The collected ticks were subjected to species assignment, accordingly. The ixodid tick species identified were *Rhipicephalus (Boophilus) microplus* and *Haemaphysalis bispinosa*. In the present study the relative infestation rate of tick species on cattle sampled showed that *Rhipicephalus (Boophilus) microplus* is the most abundant tick species (68.69%) with male to female tick (M:F)sex ratio of 1.55:1 followed by *Haemaphysalis bispinosa* (31.30% and M:F sex ratio1.07:1) (Table 1, Figure 1). The number of ticks collected from cattle was dominated by male ticks.

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Cattle Age Factor:

Cattle were categorized in three groups on the basis of age. Group I < 1 year: 350, Group II 1-4 years: 750, Group III > 4 years: 1050. Highest prevalence was recorded in the cattle of age group I(< I years) in which 262 out of 350 calves were observed infested with ticks (77.71%). The second highest prevalence was found in group II (1-4 years) where 485 cattle out of 750 were parasitized by the ticks (64.66%). The least infestation was observed in group III (>4 years) in which 505 adult cattle out of 1050 were found infested with ixodid ticks (48.09%) (Table 2, Figure 2).

Gender factor:

In the present study a total of 830 male cattle and 1320 female cattle were examined, out of which 422 male cattle and 840 female cattle were found to be infested with ixodid ticks. The rate of prevalence of tick infestation was found higher in female cattle (63.63%) than males (50.84%)(Table 3, Figure 3).

Seasonal factor:

In the present study the period of tick survey was categorized into three seasons viz. Summer (March to June), rainy (July to October) and winter (November to February) seasons. The survey revealed that higher rate of tick infestation occurs in rainy season (68.08%) followed by summer (59.85 %) and winter (48.70%) (table 4, Figure 4).

Evaluation of plant extracts for anti-tick activity:

Plant extracts were evaluated for their anti-tick activity at 2000ppm and 3000ppm concentration for 24hr., 48 hr and 72 hr of exposure. The results of bioassay indicated that all the aqueous plant extracts have shown variable toxicity against ticks (table 5).

IV. Discussion

Ticks are the known to have high medical and veterinary importance. The distribution and abundance of ticks species infesting cattle vary greatly from one area to another. The present study deals with the distribution and prevalence of ixodid ticks in cattle in the study area i.e. Lucknow, U.P. During the study period a total of 2150 cattle were examined from different localities of Lucknow district for tick infestation. It was observed that more than half of the cattle examined was found to be infested with different species of ixodid ticks. The prevalence of tick infesting cattle was observed 58.6%. The prevalence of ixodid ticks was also studied in several districts of other states and in Mathura and Azamgarh district of Uttar Pradesh, India (Ali and Singh, 2013; Prakasan and Ramani, 2007; Latha et al, 2004; Kumar and Balakrishnan, 2002; Jaggannath et al., 1988; Miranpuri and Naithani, 1978). Similar findings have been reported by Patel et al. (2013) who in their study examined 2515 cattle in Mathura district and found an infection rate of 60.07%. Other workers (Misra, 1984; Kumar, 1996, Sajeed et al., 2009) have reported more than 50.00% of prevalence in cattle. However some other workers like Manan et al. (2007) and Vatsya et al. (2007) have reported the prevalence rate of 20.40% and 41.78% respectively. Several factors such as lack of improved husbandry practices, inadequate use of veterinary services, poor awareness of the harmful effects of ticks among the cattle owner in rural areas have contributed much to the wide distribution of tick species in the area. Difference in the results of present and previous findings may be subjected to variations in climatic, geographical conditions of the study area and methodology adopted for the study (Kabir et al., 2011).

In the present study R. B. *microplus* was found to be the most prevalent tick species with infection rate of 61.69% followed by H. *bispinosa* (31.30%). *R.B. microplus* has also been reported as one of the common ixodid tick of cattle by other workers also (Patel *et al.*, 2013; Kumar and Balakrishnan, 2002; Kumar, 1996). The tick species was reported in buffalo also by Mamun *et al.*, 2010; Prakasan and Ramani, 2007. However, the infestation rate of R.B. *microplus* was higher in cattle than in buffalo probably due to preference of denser hair coat in cattle than buffalo and also due to the fact that buffalo have access to mud for wallowing that might cause tick dropping (Patel *et al.*, 2013). Another tick species H. *bispinosa* has been reported earlier as a common tick species infesting cattle, goat, buffaloes, sheep and monkeys (Mamun *et al.*, 2010; Latha *et al.*, 2004; Saxena, 1997; Trapido *et al.*, 1964). H. *bispinosa* is found as the most common tick infesting a range of domestic animals in Kerala state of India by Prakasan and Ramani (2007). The male to female ratio in both the species found in the area shows that male ticks are found higher than female ticks on cattle's body. The similar findings have been reported by Tadesse *et al.* (2012). Solomen *et al.* (2001) suggested that this difference is due to the fact that fully engorged female tick drops off to the ground for egg laying while male tends to remain on cattle's body to continue feeding and mating, hence the number of male ticks are found higher than the female ticks.

In the present study it was observed that young cattle are very prone to tick infestation. The rate of infection of tick in < 1 years of calves was recorded 77.71%. Similar results have been obtained by Patel *et al.*, 2012 in Mathura district, Uttar Pradesh, where infection rate was much higher (80.20%) in < 1 year of cattle.

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Kabir *et al.* (2011) reported that young cattle were 2.23 times more susceptible to tick infestation when compared with the adults. They also reported that age of the host had a significant (p<0.01) effect on tick and other ectoparasite infestation. It is very difficult to give the exact reason of high infestation rate of ticks in young calves, however, Mamun *et al.* (2010) suggested that less developed immune system of calves might be responsible for high prevalence rate of tick infection in them. However Wasihun & Doda, (2013), Feseha (1997) and Tessema & Gashaw (2010) reported the higher tick infestation in adult cattle.

The prevalence rate of tick infestation was found higher in female cattle (63.63%) than males (50.84%) in the present study. Mamun *et al.* (2010) also reported that female cattle were 2.61 times more susceptible to tick infestation. Bilkis *et al.* (2007) also reported significantly higher prevalence rate in females (33.47%) than in males (14.28%). Bilkis *et al.* (2007), Kabir *et al.* (2011) and Mamun *et al.* (2010) suggested that higher prevalence rate in female cattle may be due to hormonal effects. Lloyd (1983) reported that high levels of prolactin and progesterone hormone make the individual more prone to any infection. Moreover, female cattle bear higher stress than males due to pregnancy, lactation and production which makes them more prone to infection (Mamun *et al.* 2011). The results revealed out by the studies of Wasihun & Doda, (2013) in Ethiopa, Atif *et al.* (2012) in Punjab state of Pakistan and Rony *et al.* (2010) in Bangladesh show a higher prevalence of tick infestation in male cattle.

In the present study a relatively higher tick infestation was found in rainy season (68.08%) followed by summer (59.85%) and winter (48.70%). Almost similar findings were reported by Patel *et al.* (2013) in Mathura district of Uttar Pradesh where highest prevalence was recorded in rainy season (69.46%) followed by summer (62.55%) and winter (47.96%). Similar observations have been reported by Vatsya *et al.* (2008), Kumar (1996), Sanjay *et al.* (2007), Atif *et al.*(2012), Rony *et al.* (2010). The higher prevalence of ticks in rainy season suggests that humidity seems to be macroclimatic factor influencing infestation rate of ticks (Vatsya *et al.*, 2007). Some researchers like Kabir et al. (2011), Islam *et al.* (2006) reported higher rate of infection in summer season. However, some workers like Mamun *et al.* (2010), Islam (1989) reported high tick prevalence in winter season. The contrast in between the present and earlier findings may be due to variations in geographical locations, topography and composition of soil type, temperature and humidity of the experimental area.

In the present study toxic effect of aqueous extracts of 20 plants at 2000ppm and 3000ppm were evaluated against ticks and tabulated (Table 5). The percentage mortality obtained for *Azadirachta indica* (86%±0.61) extracts was quite high when compared to those for other plants in 2000 ppm and 3000ppm. Neem (*A. indica*) is one commonly grown indigenous plants in domestic backyard in India. Experiments conducted with neem by other workers (Kalakumar *et al.*, 2000; Benavides *et al.*, 2001; Srivastava *et al.*, 2008) have shown some acaricide properties of its extracts. The leaf extracts of *Azadirachta* inhibit egg production of immersed B. microplus ticks (Borges *et al.*, 2003). Weekly spraying with neem seed extracts decreased the number of ticks on goats (Schwalbach *et al.*, 2003) and cattle (Webb and David, 2002) in Southern Africa. Abdel-Shafy and Zayed (2002) recorded 100% death of unfed larvae of *Hyalomma anatolicum excavatum* with neem seed oil on day 3 post-exposure.

In the present study, beside neem several other plant extracts have also shown higher toxicity against ticks like *Ricinus communis, Nicotiana tabaccum, Calotropis procera, Trachyspermum ammi, Trigonella faenum grecum., Mentha* sp., *Lantana camara, Cannabis sativa.* The other plants which are known to have acaricidal property are *Gyandropsis gynandra* (Lwande *et al.*, 1999), *Cleome hitra* (Ndungu *et al.*,1999), *Pimento dioica* (Brown *et al.*,1998). However extensive studies need to be conducted to also determine the side effects of these extracts on the animals.

V. Conclusion

The tick infestation in cattle and other livestock pose a serious damage to the livestock industry and it is a challenging task for the workers to control them. The results obtain by the present study shows that ticks have seasonal distribution and preferable host selection. The observations from the present study may contribute to the increased understanding of epidemiology of ticks in the area. This may help in adopting tick control strategies. However no statistical significant difference was observed in various indices used in the study, this may be because of less data. Therefore, further studies on tick prevalence are also suggested in other areas of Uttar Pradesh to clear the faded knowledge of tick distribution in cattle and other livestock. Focus should also be given on tick distribution pattern on different attachment sites on host body, tick prevalence in relation to body score and breed of host, management practices adopted by the rearers. The results obtained from the bioassay of plant extracts against ticks open the possibility of further investigations of efficacy on their acaricidal properties of natural product extracts. The isolation and purification of aqueous extracts will be the future prospects of the study.

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Table 1: Distribution of tick species

S.No	Tick species	Male ticks	Female ticks	M:F ratio	Total count of tick species	Prev alenc e%
1	Rhipicephalus (Boophilus) microplus	590	380	1.55:1	970	68.69
3	Hemaphysalis bispinosa	229	213	1.07:1	442	31.30
	Total	809	593		1412	100

Table 2: Table showing the prevalence of ixodid ticks (In relation to cattle age)

S.No	Range of Age of Cattle	Total Cattle surveyed	Infected Cattle	Prevalence%
1	< 1 year	350	262	77.71
2	1-4 years	750	485	64.66
3	> 4 years	1050	505	48.09

Table 3: Table showing the prevalence of ixodid ticks (In relation to cattle's sex)

S.No	Gender of Cattle	Total Cattle surveyed	Infected Cattle	Prevalence%
1	Male	830	422	50.84
2	Female	1320	840	63.63

Table 4: Table showing the prevalence of ixodid ticks (In relation to different seasons)

S.No	C	Total Cattle graveyed Infected Cattle Providence 9/			
3.N0	Season	Total Cattle surveyed	Infected Cattle	Prevalence%	
1	Summer (March to June)	700	419	59.85	
2	Rainy (July to October)	680	463	68.08	
3	Winter(November to February)	770	375	48.70	

Table 5: Percentage mortality of plant extracts at 2000ppm and 3000ppm

S.No	Plant's name Plant Part Used	Plant Part	Percentage of Mortality at 2000 ppm.			Percentage of Mortality at 3000 ppm.		
		Used	24 hr.	48 hr.	72 hr.	24 hr.	48 hr.	72 hr.
1.	Azadirachta indica	Leaves	45%± 0.12	58%± 0.43	64%±0.71	62%± 0.83	78%±0.89	86%±0.61
2.	Lantana camara	Leaves	52%± 0.73	$54\% \pm 0.23$	$62\% \pm 0.63$	55%± 0.12	$64\% \pm 0.23$	$78\% \pm 0.69$
3.	Ricinus communis	leaves	$38\% \pm 0.21$	$49\% \pm 0.56$	$60\% \pm 0.77$	$40\% \pm 0.95$	$56\% \pm 0.65$	$75\% \pm 0.67$
4.	Eucalyptus sp.	leaves	$25\% \pm 0.63$	$46\% \pm 0.61$	$64\% \pm 0.23$	$30\% \pm 0.63$	$48\% \pm 0.33$	69%± 0.23
5.	Thevetia peruviana	Leaves	$34\% \pm 0.21$	$56\% \pm 0.76$	$66\% \pm 0.65$	$39\% \pm 0.43$	$68\% \pm 0.23$	$78\% \pm 0.12$
6.	Chenopodium album	Whole herb	24%± 0.54	58% ± 0.32	63% ± 0.25	25%± 0.28	60% ± 0.23	69%± 0.28
7.	Cannabis sativa	Whole herb	$28\% \pm 0.65$	$60\% \pm 0.33$	$65\% \pm 0.24$	$30\% \pm 0.46$	$65\% \pm 0.74$	$78\% \pm 0.39$
8.	Cassia fistula	Leaves	20%± 0.66	$24\% \pm 0.23$	$45\% \pm 0.13$	$30\% \pm 0.43$	$35\% \pm 0.83$	$55\% \pm 0.03$
9.	Calotropis procera	Leaves	10%± 0.12	$38\% \pm 0.86$	66% ± 0.53	26%± 0.23	$48\% \pm 0.73$	$74\% \pm 0.43$
10.	Achyranthes aspera	Whole herb	24%± 0.21	42%± 0.87	67% ± 0.54	34%± 0.65	$44\% \pm 0.43$	$72\% \pm 0.53$
11.	Abutilon indicum	Whole herb	20%± 0.14	35%± 0.54	54%± 0.70	30%± 0.16	48%± 0.27	66%± 0.56
12.	Tamarindus indica	Fruit	30%± 0.36	46%± 0.50	52%± 0.71	38%± 0.86	48%± 0.73	62%± 0.73
13.	Solanum nigrum	Whole Herb	22%± 0.48	32%± 0.12	47%± 0.36	$24\% \pm 0.20$	46%± 0.29	$68\% \pm 0.56$
14.	Brassica alba	seed	28%± 0.12	$34\% \pm 0.05$	56%± 0.61	$38\% \pm 0.15$	48%± 0.62	60%± 0.37
15.	Cuscuta reflexa	Whole herb	20%± 0.53	34%± 0.12	52%± 0.86	22%± 0.18	$38\% \pm 0.94$	$66\% \pm 0.96$
16.	Trigonella foenum graecum	seed	16%± 0.55	$42\% \pm 0.33$	50% ± 0.96	28%± 0.38	48%± 0.93	62%± 0.66
17.	Trachyspermum ammi	seed	24%± 0.84	$32\% \pm 0.40$	$54\% \pm 0.34$	30%± 0.24	$64\% \pm 0.44$	$78\% \pm 0.94$
18.	Nicotiana tabacum	leaves	24%± 0.45	44%± 0.24	61%± 0.17	26%± 0.83	43%± 0.25	72%± 0.26
19.	Mentha sp.	leaves	20%± 0.26	42%± 0.27	52%± 0.35	26%± 0.27	52%± 0.26	$76\% \pm 0.66$
20.	Cynodon dactylon	leaves	06%± 0.36	22%± 0.27	28%± 0.37	08%± 0.32	$28\% \pm 0.56$	48%± 0.84

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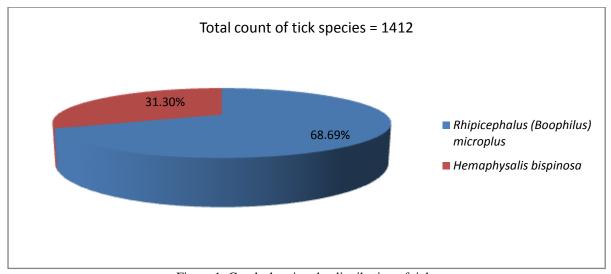


Figure 1: Graph showing the distribution of ticks

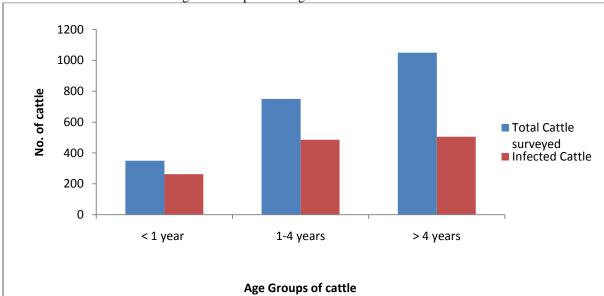


Figure 2: Graph showing the prevalence of ticks (Age Wise)

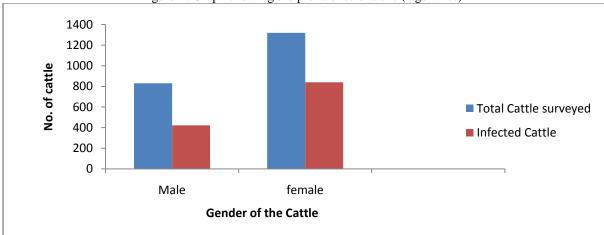


Figure 3: Graph showing the prevalence of ticks (Gender Wise)

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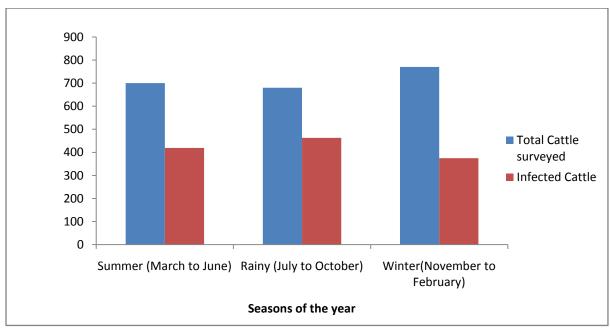


Figure 4: Graph showing the prevalence of ticks (Season Wise)



Figure 6: Heavily infested cattle



Figure 7: Ticks attached on body

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Figure 8: Ticks attached on dewlap





Rhipicephalus Boophilus microplus (anterior view) Rhipicephalus Boophilus microplus (posterior view)

Figure 9: Rhipicephalus Boophilus microplus

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