Applied Journal of Hygiene 8 (2): 48-57, 2019
ISSN 2309-8910

DOI: 10.5829/idosi.ajh.2019.48.57

Experimental Study on Insecticide Treated Net Effect on Cattle in Mirab Abaya, Southern Ethiopia

[1]Behailu Negash, [2]Biruk Alemu, [3]Beredu Yohannes, [4]Alamne Hunegnaw and [5]Aweke Yalew

[1]Livestock and fisheries office, Gamo zone, Ethiopia
[2]Veterinary Drug and Animal Feed administration and control authority, Hawassa, Ethiopia
[3]Livestock and fisheries office, Kambata Tambaro Zone, Ethiopia
[4]Hawassa Poultry Multiplication and Breeding Center, Hawassa, Ethiopia
[5]Sodo Farm Service center, Sodo, Ethiopia

Abstract: This study was conducted to evaluate the effect of insecticide-treated nets technology on-farm animals from November 2017 to March 2018 in Mirab Abaya district, Gamo Zone, Southern Ethiopia. For this study in the treated group; insecticide-treated net was fenced around the pen of thirty selected farmers with thirty animals and unfenced for five farmers with ten animals in the control group throughout the study. The pen was 4m × 4m in size, the net placed 10cm above the ground and covers the 1m height of the pen to see the effect of insecticide-treated net fenced around cattle pen protecting animals from tsetse and biting flies. The method of study was done by entomological, parasitological, hematological and defensive movements' data taking. Results from a total of 50 and 25 traps deployed, 161 and 326 biting flies were caught and their number was decreasing from the baseline but remained almost the same in treated and control group, respectively. Also, the overall apparent density biting flies was 0.214 and 0.87 in treatment and control group, respectively. Furthermore, all animals in both groups were found negative to trypanosomiasis and no tsetse flies were caught from both sites. However, the overall mean packed cell volume (PCV) value was higher in treated (28.10) than the control group (24.34). There was the significant statistical difference ($p<0.05$) between treatment groups for PCV, biting flies and defensive movements. The overall mean defensive movements were almost reduced in the treated (8.20) group but increased in the control group (22.36). This showed that the effect of insecticide net on biting flies and animal was significant and it can be transferred to the communities in nearby and help the country utilize from the livestock resource. Conclusions the study indicates that anaemia is caused by biting flies other than trypanosomes. Hence, further study has to be taken to determine factors causing anemia that are caused by biting flies.

Key words: Cattle · Insecticide · Treated Net · Pcv · Pen · Mirab Abaya

INTRODUCTION

Livestock is the mainstay of the vast majority of African people. It contributes a large proportion of the continent's gross domestic product (GDP) and constitutes a major source of foreign currency earning for a number of countries [1]. Domestic livestock in Africa is important as a source of protein (Milk and meat) to humans, animal traction, income (Hides), investment (Social security) and manure for enhancing agricultural /crop production [2]. Ethiopia stands first in Africa and tenth in the world in livestock population. So that it is believed to have the largest cattle population and has the highest livestock number in Africa [3].

Approximately 30% of the total cattle population in the African continent and about 50 million people are exposed to animal trypanosomiasis and human sleeping sickness, respectively [1]. Therefore, trypanosomiasis is a serious disease in domestic livestock that causes a significant negative impact on food production and economic growth in many parts of the world, particularly in sub-Saharan Africa [4, 5]. In southern Africa, the

Corresponding Author: Biruk Alemu, Veterinary Drug and Animal Feed Administration and Control Authority, Hawassa, Ethiopia.

disease is widely known as nagana, which is derived from a Zulu term meaning "To be in low or depressed spirits", a very appropriate description of the disease [6].

Ethiopia is located in the Horn of Africa between latitude from 3°N to 15°N of the equator and longitude from 33°E to 48°E [7]. It is an agrarian country with an estimated human population of above 80 million and a total land of 1,101,000 km^2 [8]. The proportion of total population in the agricultural sector is 82.4%. Livestock are an integral part of the farming system in Ethiopia. Although the country has a huge number of livestock populations, the full utilization of the resource is hindered due to tsetse-transmitted trypanosomiasis and other animal diseases [7].

Trypanosomiasis is one of the most economically important vectors born disease and is mainly transmitted by flies belonging to the *Glossina* species / tsetse flies [9]. The genus *Glossina* consists of three distinct sub-genera, Morsitans (Glossina), Palpalis (Nemorhina) and Fusca (Austenina). These subgenera are differentiated on anatomical features and on their restriction to a relatively specific habitat [10] and thus, the Moriston's group are classed as savannah-inhabiting species, Palpalis group as riverine or lacustrine species and Fusca group as forest-inhabiting species [11]. Their distribution and abundance being determined by the vegetation type along riverbanks [12]. Tsetse and trypanosomiasis lie at the heart of Africa's struggle against poverty affecting 37 countries in sub-Saharan Africa where the disease is endemic [13]. It is recognized as a neglected disease, principally because it is rural and its impact on the socio-economic development isdevastating [14]. Trypanosomosis is mainly restricted to areas in which the vector, tsetse fly can survive. The disease is also found outside the tsetse belt areas being transmitted mechanically by biting flies of the genus *Tabanus*, *Hematopota*, *Chrysops* and *Stomxys*. A number of trypanosome species are responsible for bovine trypanosomiasis (*Trypanosoma brucei*, *T. congolense* and *T. vivax*) that differ from those causing the human form of the disease, sleeping sickness (*T. b. gambiense*, *T. b. rhodesiense*). Economically, tsetse-transmitted trypanosomes *(T. congolense*, *T. vivax* and *T. brucei)* are most important in cattle with 14 million heads are at risk in Ethiopia [15].

The life cycle of trypanosome is complex in both tsetse fly vector and the mammalian host which undergoes a series of transformations into different forms. Most tsetse-transmission is cyclical and begins when blood from trypanosome infected animals are ingested by the fly. The trypanosome losses its surface coat, multiplies in the fly, then reacquire a surface coat and becomes infective [16]. Tsetse transmits trypanosomes in two ways; mechanically and biologically. Mechanical transmission involves the direct transmission of the same individual trypanosomes taken from an infected host into an uninfected host whereas the biological transmission requires a period of morphological development of the trypanosomes within the tsetse host [17].

In Ethiopia, trypanosomosis is widespread in domestic livestock in the Western, South and Southwestern lowland regions and the associated river systems (i.e. Abay, Ghibe, Omo and Baro-Akobo) [18]. About 220,000 Km^2 areas of the above-mentioned regions are infested with five species of tsetse flies namely *Glossina pallidipes*, *G. morsitans*, *G. fuscipes, G. tachinoides* and *G. longipennis* [19]. Farmers strongly recognized trypanosomosis as the primary problem for livestock productivity and agricultural development in the northwestern and western parts of Ethiopia [20]. Trypanosomosis in cattle locally referred, as "Gendi" is a serious constraint to livestock production in areas of the north and southwest Ethiopia at an altitude of below 2000 meters above sea level [21].

Several control approaches are available to eradicate trypanosomosis and its biological vector, the tsetse fly [22]. Trypanosomosis control is a long-term fight and therefore requires the involvement of decision-makers, researchers and farmers. Until now, the use of trypanocidal drugs to treat or to prevent susceptible livestock against trypanosomosis remains the only control measure for most of the farmers [23].

Recently, due to a shortage of grazing land, farmers in Ethiopia are forced to use other alternative methods to free grazing. People have started dairying and fattening in many parts of the country as a source of income as the economic importance of raising livestock especially cattle has improved. All these together led to the development of the habit of keeping cattle, especially fattening and milking animals, in enclosures for indoor feeding. Hence, insecticide-treated nets (ITN's) can be used as a control mechanism for vectors transmitting trypanosomosis and Anaplasmosis causing anemia. These nets are most commonly used as bed-nets. Placing deltamethrin-treated net or screen (Zero Fly livestock) around cattle pen was proposed as an option to reduce challenges from tsetse and biting flies. Besides killing the flies, the technology was designed to protect individual animals from fly attack. The technique would eliminate tsetse flies, biting flies and other nuisance flies and create a sustainable condition for livestock rearing under indoor feeding or Zero grazing system to be considered as an alternative and cost-effective vector control technique in the rural areas [24, 25]. Therefore, this study was conducted on cattle owned by 35 farmers from Mirab Abaya district of Gamo Gofa zone, to evaluate the effect of insecticide-treated nets technology on-farm animals.

MATERIALS AND METHODS

Description of the Study Area: An experimental study was conducted from November 2017 to March 2018 in Mirab Abaya district of Gamo Gofa Zone (5°57'N latitude and 37°32'E longitude), in SNNPR state of Southern Ethiopia (Fig. 1). The area has a sub-humid climate with a moderately hot temperature. Arbaminch, the capital of the zone, is located 505 kilometres south of Addis Ababa and 280 kilometres from the regional town, Hawassa. Its geographical location is 37° 5' east longitude and 6° north latitude with an altitude of 1001-2500 meters above sea level [26]. Mirab Abaya is one of the districts of Gamo Gofa Zone which is located at 455 kilometers from Addis Ababa and 56 km from the zonal city, Arbaminch. The area of the district is about 121,150 hectares of which only 38,342 hectares are used for livestock grazing. The district consists of 24 kebeles with 11% highland, 27% midland and 62% lowland. The area receives an annual rainfall of 800-1600mm and the average annual temperature is 27.5°C. The altitude of the district ranges from 1100 to 2900 masl. The district has a total population of 95,251 and cattle population is estimated to be 46, 417 [27].

Study Animals: An experimental study was conducted in the dry season from November 2017 to March 2018 on 67 cattle owned by 35 farming households. Of the 67 animals, 57 owned by 30 households were assigned to the treatment group while 10 cattle owned by 5 households were allocated to the control group. Selected farmers for the experiment constructed cattle protective pens of size 4 meters by 4 meters and covered the roof with a polyethylene sheet. The walls of the pens for the treatment group were covered with deltamethrin-impregnated mosquito nets for 1 meter starting 10 centimetres above the ground. The number of cattle kept in the pens for the treatment group

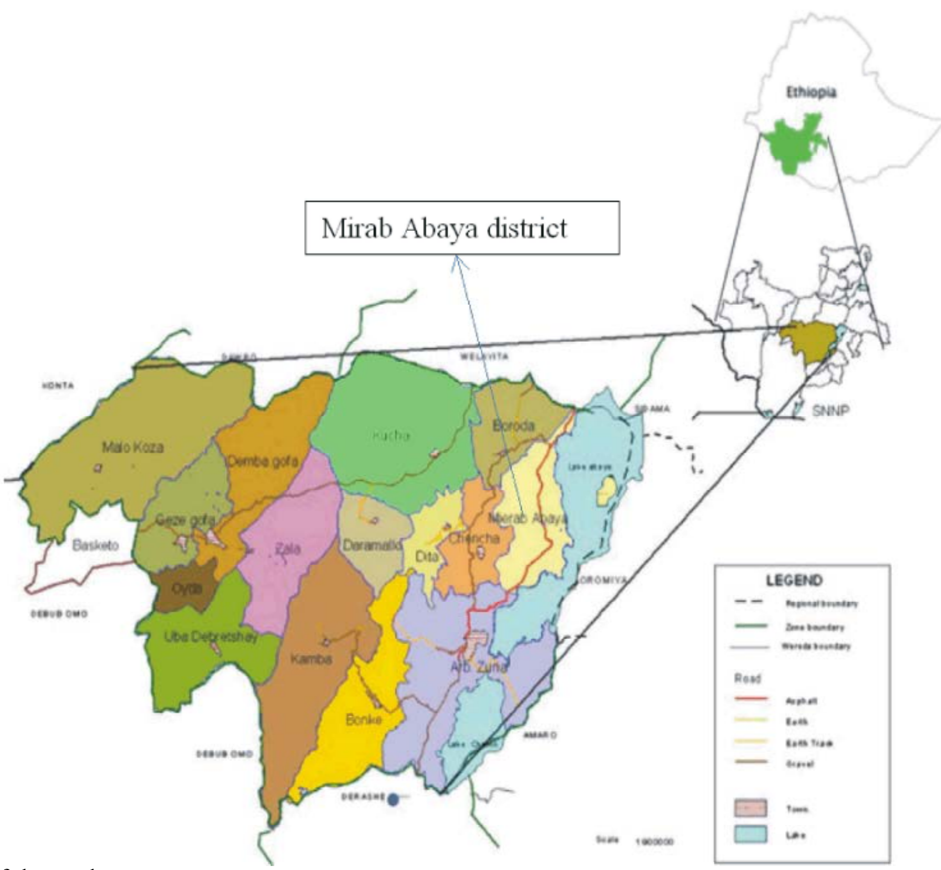

Fig. 1: Map of the study area

varied from 1 to 2 based on the farmers' capacity to use zero grazing and to prepare adequate feed for the cattle in the pen and on the number of cattle an individual owns. On the other hand, 5 farmers with 10 animals were selected for the control group. The number of animals per household varied from 1 to 3 in the control group. The control animals were also kept in pens in a similar fashion to the treatment groups except that the walls of the pens were not covered with insecticide-treated nets/screens.

Experimental Pens and Nets: A total of 35 identical cattle pen of 4 meter x 4 meter size and with a height of net 1 meter and 10 centimeter space gap between the ground and the net with two animals holding capacities were constructed using different types of woods, nails and the roof was made of a corrugated iron sheet. The floor of the pen was not made of concrete but leveled soil. The treated net consisted of a black 150 denier polyester fiber with 2 x 2 millimetres mesh width and impregnated with 80-120 milligram of deltamethrin per square meter [24]

Study Design and Data Collection: The households were selected according to their willingness to participate in the study which involves the construction of pens for the study animals and feeding them housed until completion of the study. At the beginning of the study, all the study animals were screened for trypanosomosis, their PCV was measured, their body condition scored and in milking cows daily milk was measured and recorded to serve as baseline data. Subsequently, these parameters were recorded every month for 4 months. Defensive movements of the body against flies: tail switch, head and ear movement, leg kicks and skin ripples were also counted for 5 minutes and recorded. Traps were also deployed near the treated and control groups to measure densities of tsetse flies, *Tabanus*, *Stomoxys* and other nuisance flies around the pens.

Entomological Monitoring: The entomological Monitoring was carried out to assess the apparent density and species composition of tsetse and other biting flies in the study area. Every trap was odor baited with acetone and flies attractants. The underneath of each trap pole was smeared with grease in order to prevent the ants from climbing up the pole towards the collecting cage that could damage the tsetse flies. The trap deployment time was 72 hours. The flies captured in the collecting cage were then sorted by sex and species and recorded. The species of tsetse was identified based on the characteristic morphology. Other biting flies were also separated according to their morphological characteristics such as size, color, proboscis and wing venation structures at the genus level [28]. For both experimental and control groups everything including feeding system, watering and other management systems and the area, altitude and climate were the same except an insecticide-treated net. During the course of the monitoring cycle, traps were deployed at the beginning of the study and monthly thereafter for 4 months. Each month 15 NGU traps were deployed; 10 traps for the treated groups and 5 traps for the control approximately 5 meters from the pens with 100-200 meters between the traps. Therefore, a total of 75 NGU traps were deployed throughout the study [29]. Generally, the deployment of traps was to check the efficacy of the insecticide-treated net on tsetse and other biting flies throughout the time of the study.

Parasitological Study: Sample collection and parasitological examination were carried out by using two laboratory procedures. First, blood samples were collected after properly securing the animal and aseptically by puncturing with the tip of the lancet on the margin of ear vein followed by filling heparinized capillary tubes. Pair of the capillary tubes was filled ¾th of their length. The capillary tubes were sealed by using a crystal seal at one end. The blood in the capillary was centrifuged at 12,000 rounds per minute for five minutes and consequently, the packed cell volume (PCV) of each sample was determined by a Hawksley micro-hematocrit reader. Second, the capillary tube was cut 1 millimetre below the buffy coat intersection to include the top layer of red blood cells. The content of the capillary tube was then expressed on to a clean microscope slide mixed and covered with 22 x 22 millimetres coverslip. Then the slide was examined using a microscope with 25x objective lens to determine the prevalence of trypanosomosis using the Buffy coat technique (BCT) as described by Murray *et al*. [30]. To determine PCV, the tubes were placed in hematocrit and readings were expressed as a percentage of packed red cells to the total volume of whole blood. Animals with PCV $\leq$ 25% were considered to be anemic [31].

Defensive Movements: The defensive movements were closely monitored for both treatment and control group animals. Five minutes for the fly and host contact landing on the parts of animals especially tail switch, head and ear movements, leg kicks and skin ripples in responsive to flies throughout all the five cycles of the study period were considered according to Maia *et al*. [32].

Data Management and Analysis: Raw data generated from this study was entered into Ms-excel database and was analyzed using STATA software version 11.0 and t-test was applied to compare the effect of insecticide-treated net on tsetse and biting flies. A 95% confidence interval and 5% absolute precision were used to determine whether there was a significant difference between measured parameters. Mean PCV values, defensive movements and fly catches between the treatment and control group were compared by using t-test. F/T/D analysis used to calculate to the flies (*Tabanus* and *Stomoxys*) densities.

RESULTS

Parasitological Examination: Parasitological examination of blood samples for trypanosomosis showed that all animals, in both groups (Treated and control), were negative to the infection throughout all of the five monitoring cycles (At the start of the study and monthly for 4 months afterward) of the study period. There was also no tsetse fly catch.

PCV Determination: Mean PCV (±SE) of cattle kept in the treated and control group were determined in the current study. Hence, the PCV of the animals was found significantly different between the treatment groups ($P<0.05$). The mean PCV of treated animals was 28.10 (0.27%) and control animals 24.34 (0.73%) which indicates higher mean in treated group than in control group animals (Table 1). However, the experiment was originally designed to see the effect of insecticide-treated screen around the wall of cattle pen on trypanosomosis and its vectors, it was not possible to detect parasitemia and catch the main vector of the parasite. The hematological results of all the five monitoring cycles for both (Treatment) and control groups were analyzed in the current study as indicated in the Table 2. Statistical analysis was made using a t-test to compare the mean PCV value of treatment and control group. The mean value of PCV of treatment groups was increasing from 25.37% to 29.30% in treatment group whereas in control groups the mean value of PCV was decreasing from 26.40% to 22.50% throughout the study period. The hematological results indicated that in the first (Baseline) and second monitoring cycle, there was little difference between the mean PCV values of both groups and also there was no significant statistical difference ($p>0.05$) between treatment and control groups. But in the third, fourth and fifth monitoring cycles of both groups there was quite a different in mean PCV values and there was also a significant statistical difference ($p<0.05$) between the treatment and control group (Table 2).

Entomological Result: There was no tsetse fly among the fly catches of this study. However, biting flies (Tabanus and Stomoxys) were cached. The overall mean fly count was higher near control pens compared

Table 1: Overall mean PCV (±SE) of cattle kept in the treated and control group

Treatment group	No. of observation	Mean (SE)	P-value
Treated	285	28.10 (0.27)	0
Control	50	24.34 (0.73)	
Total	335	27.50 (0.26)	

Table 2: Mean PCV result of each cycle for treated and control groups

			(95% Confidence Interval)		
Monitoring cycle	Observation	Mean PCV	Minimum	Maximum	P-value
1st					
Control	10	26.40	22.67	30.13	0.4687
Treatment	57	25.37	24.32	26.41	
2nd					
Control	10	25.80	21.40	30.19	0.2499
Treatment	57	27.77	26.51	29.02	
3rd					
Control	10	24.30	20.83	27.77	0.0083
Treatment	57	28.49	27.31	29.66	
4th					
Control	10	22.70	19.05	26.35	<0.001
Treatment	57	29.30	28.11	30.48	
5th					
Control	10	22.50	19.40	25.60	<0.001
Treatment	57	29.50	28.55	30.47	

Table 3: Overall mean fly catch (±SE) per trap near treated and control cattle pens

Treatment Group	Number of traps deployed	*Tabanus* Mean (SE)	*Tabanus* P value	*Stomoxys* Mean (SE)	*Stomoxys* P value
Treated	50	1.04 (0.29)	<0.001	2.18 (0.64)	<0.001
Control	25	4.24 (0.76)		8.80 (1.20)	
Overall	75	2.12 (0.36)		4.40 (0.68)	

Table 4: Apparent fly catch of treated and control groups

Group				Glossinaspecies		Biting Flies			
Control	Cycle	No. of Traps	Days	*Tsetse*	F/T/D	*Tabanus*	*stomoxys*	Total	F/T/D
	1	5	3	0	0	10	51	61	4.06
	2	5	3	0	0	22	50	72	4.80
	3	5	3	0	0	29	35	64	4.26
	4	5	3	0	0	20	48	68	4.53
	5	5	3	0	0	25	36	61	4.06
	Total	25	15	0	0	106	220	326	0.87
Treatment	1	10	3	0	0	19	86	105	3.50
	2	10	3	0	0	18	4	22	0.73
	3	10	3	0	0	0	18	18	0.60
	4	10	3	0	0	14	0	14	0.47
	5	10	3	0	0	1	1	2	0.067
	Total	50	15	0	0	52	109	161	0.214

Table 5: Overall mean defensive movements within 5 minutes

Treatment group	Number of observations	Mean (SE)	P-value
Treated	285	8.20(0.42)	<0.001
Control	50	22.36 (0.50)	
Overall	335	10.30 (0.46)	

Table 6: Mean (±SE) defensive movements of each body parts observed

Defensive movements	Treated Mean (SE)	Control Mean (SE)	P-value
Tail switch	3.44(0.20)	8.62(0.32)	<0.001
Head and ear	2.30(0.14)	5.74(0.20)	<0.001
Leg kicks	1.55(0.88)	4.60(0.20)	<0.001
Skin ripples	0.89(0.60)	3.4(0.16)	<0.001

to pens covered with insecticide-impregnated screen/net and also there was a significant difference ($p<0.05$) of flies between the treatment groups (Table 3). During the study period, from total of 50 traps for treatment and 25 traps deployed for control, a total of 161 biting flies (52 *Tabanus* and 109 *Stomoxys*) from treated and 326 biting flies (106 *Tabanus* and 220 *Stomoxys*) from control sites were caught and unfortunately, no tsetse flies were caught from both sites of the traps deployed throughout all of the monitoring cycles. Specifically, the number of biting flies (*Tabanus* and *Stomoxys*) was decreasing in the treatment group from time to time (i.e. 105 in first or baseline, 22 in second, 18 in third, 14 in fourth and 2 in fifth cycle) but in control groups, they were remained almost the same i.e. 61, 72, 64, 68 and 61 in first, second, third, fourth and fifth cycle, respectively and also the overall apparent density biting flies was 0.214 and 0.87 in treatment and control group. There was no statistical difference ($p>0.05$) between *Tabanus* and *Stomoxys* in first (Baseline) whereas in the second, third, fourth and fifth cycle there was a significant difference ($p<0.05$) for the mean fly catch in both group sites (Table 4).

Defensive Movements: There was a significant difference ($p<0.05$) between the treatment and control groups for mean defensive movements. In general, the mean defensive movements of body parts observed within five minutes were higher in the treated animals when compared with the control ones. There was also a statistically a significant difference ($p<0.05$) between the groups (Table 5). The most frequent defensive movement of the body parts of animals was observed from tail switch followed by head and ear movements and leg kicks while the skin ripples were infrequent in both groups. The mean defensive movements of all of the body parts observed were lower in the treated than control one. The mean movements or responses of animals to biting flies with their tail, head and ear, legs and skin were statistically associated ($p<0.05$) between the treated and control groups at the current finding (Table 6). The higher rate of defensive movement observed in the animal's body parts was tail swishes followed by head and ear movements and leg kicks while the skin ripples were

Table 7: Defensive movements of each body parts observed at each cycle

Monitoring cycle	Mean Control	Mean Treatment	St. Err. (SE) Control	St. Err. (SE) Treatment	p-value
		1st (baseline)			
Animals examined	10	57	10	57	
Tail movement	8.6	9.12	0.67	0.268	0.4575
Head and ear movement	4.9	6.28	0.433	0.20	0.0102
Leg kicks	4.4	3.8	0.45	0.118	0.0843
Skin ripples	3.1	1.98	0.37	0.13	0.0022
		2nd (after 1 month)			
Tail switch	8.2	2.33	0.78	0.15	<0.001
Head and ear movement	5.9	1.17	0.37	0.16	<0.001
Leg kicks	4.2	1.17	0.44	0.15	<0.001
Skin ripples	3.4	0.86	0.30	0.12	<0.001
		3rd (after 2 months)			
Tail switch	8.5	2.57	0.636	0.185	<0.001
Head and ear movement	6.20	1.05	0.29	0.13	<0.001
Leg kicks	4.70	1.10	0.3	0.13	<0.001
Skin ripples	3.2	0.50	0.38	0.10	<0.001
		4th (after 3 months)			
Tail switch	8.2	1.78	0.72	0.16	<0.001
Head and ear movement	5.5	1.38	0.45	0.11	<0.001
Leg kicks	5.1	0.80	0.31	0.12	<0.001
Skin ripples	4.1	0.649	0.34	0.09	<0.001
		5th (after 4 months)			
Tail switch	9.6	1.36	0.76	0.121	<0.001
Head and ear movement	6.2	1.12	0.51	0.13	<0.001
Leg kicks	4.6	0.87	0.49	0.093	<0.001
Skin ripples	3.2	0.42	0.38	0.065	<0.001

infrequent in treatment and control groups and in first (Baseline) the mean defensive movements were nearly equal whereas for second, third, fourth and fifth cycle there was a significant difference (p<0.05) between the treatment and control groups. Generally, the mean defensive movements of body parts (Tail switch, head and ear movements, leg kicks and skin ripples) were higher in the treated animals when compared with the control ones. In the treatment group the defensive movements were drastically reduced in the second cycle from the baseline and then were decreasing gradually, but in the control, the condition remained almost the same with the baseline throughout the monitoring cycles (Table 7).

DISCUSSION

The present study showed that the effect of insecticide-treated net revealed the presence of the net around zero grazing animals in the pens created safe place for cattle from biting flies for treated ones but there were disturbance and panic worry of animals in unfenced pens attributed to the high number of biting flies that were being caught in the trap deployed around the pens. The entomological result in the current study indicated that the total number of biting flies (*Tabanus* and *Stomoxys*) in the treated group was reduced compared to that of the control group in which the total number of fly population was remained almost increased in each monitoring cycles. The overall F/T /D were 0.214 and 0.87 in treated (lower) and control group (higher) respectively and there was also a significant difference (p<0.05) between treatment groups for both of biting flies. This finding agrees with the research done by Mahama *et al*. [33] and Mahama *et al*. [34] in which they reported as the defensive movements of animals in the experimental group were lower than animals in the control group. It was also in agreement with the report of Ghanaian forest Zone at Kumasi by Maia *et al.* [32] and Bauer *et al.* [35]. Similarly, the report by Maia [24] on the impact of insecticide-treated net protecting cattle in zero-grazing units on nuisance flies and biting flies in the forest region of Kumasi Ghana, indicated that the fly catches around treated net was lower throughout the trial when compared to with catches around untreated (Control) pens. This might be due to killing and reducing the flies that come closer or had contact with the impregnated net. As a result, the chance of the flies entering into the fenced pen was insignificant and the animal in the pen remained safe and stable.

However, No tsetse flies were caught in the current findings and this might be attributed to ground and aerial spraying with insecticides and the use of synthetic pyrethroids (Deltamethrin) on cattle had almost eradicated tsetse flies densities in the study area and also using of odor-baited targets impregnated with insecticides was also one of a means of reducing the tsetse flies. In addition to this, all animals of the treatment groups were found negative to trypanosomes in the above result. This result was similar to the report of Kapitano [25] in Arbaminch in which he was reported that all animals from both groups were found negative. In contrast to this result, a report was done by Bauer *et al.* [36] in Kenya shown that trypanosomes is was observed in both groups in which the mean hazard rate for trypanosomosis was significantly lower in protected (Treated) animals than in control animals. The reason for this could be the regular treatment of the animals (Every two weeks for trypanosomosis), low parasitemia andmost of the time the aim the flies were to suck blood to feed itself and also the main vectors were absent around pens of both groups.

The overall mean defensive movements produced by the animals in the treated group (8.20) were lower than control groups (22.36) and there was also a significant difference ($p<0.05$) between the treatment groups during the study period. The findings of this study show that insecticide-treated net (Screen) was highly effective in preventing animals from *Tabanus* and *Stomoxys* and other nuisance flies. This result was also similar tofindings of Maia *et al*. [32] and Bauer *et al*. [35] as they reported that apart from knocking down and killing effect, the treated net provided a physical barrier to the flies protecting animals in the fenced pen from the fly challenge and the animals in the fenced pen remained stable and safe since the chance of flies entering into the fenced pen was insignificant, but the flies have easily accessed into the unfenced pen and caused increased defensive movement to the animals kept inside the pen and caused continuous and worst disturbance to the animals. So fencing insecticide-treated net (Screen) around the cattle pen was observed to significantly reduce fly-animal contact. This was attributed to the presence of insecticide-treated in treated group net and biting fly status and consequently, the animals within the pen protected with a treated net had significantly fewer insects counted on their body regions and performed the least defensive movements. This result was in line with the report of Maia *et al.*[32] in which protection of confined cattle against biting and nuisance flies with insecticide-treated nets in the Ghanaian forest zone at Kumasi, in which the feed uptake and resting of animals in treated group was undisturbed, but relentless disturbance of animals in control group.

Even though there were no tsetse fly catch and trypanosomes in the study, the mean PCV value was increased from 25.37% to 29.30% in treated and reduced from 26.40% to 22.5% in control groups. There was a significant difference ($p<0.05$) betweenthe treatment and control group. Generally, the result indicated that animals in control groups were almost anemic (PCV<25%). This might be due to (in the control group) blood parasites like Anaplasmosis transmitted by biting flies (*Tabanus* and *Stomoxys)* while feeding on blood andas blood feeders, *Stomoxys* flies can consume an average of 11-15 μL of blood per meal [37]. Based on Baldacchino *et al*. [37] in addition to the direct loss of blood from feeding, more blood may pour out from the biting site when the mouthparts are removed from the skin, which may dry on the skin or be promptly absorbed by sucking flies. Often, *Stomoxys*are disturbed by high abundances of sucking flies, which interrupts their blood meal and creates more opportunities to lose blood by pouring from the biting site. *Tabanus* and *Stomoxys* are responsible for the mechanical transmission of *Anaplasma* (Since the outstanding feature of clinical anaplasmosis is anemia associated with phagocytosis of parasitized erythrocytes) through blood-contaminated mouthparts of biting flies. The mechanical transmission by these biting flies is likely the major route of Anaplasma *marginale* in certain areas of Africa when tick vectors are absent. This condition might cause anemia resulting in reduced PCV (PCV<25%) even though there were neither tsetse flies nor trypanosomes.

CONCLUSION AND RECOMMENDATIONS

The current study revealed that the insecticide-treated net or screen was effective in protecting animals kept in the pen (Under indoor feeding/Zero grazing system) from biting (*Tabanus* and *Stomoxys*) flies by killing and reducing their number and thereby reduced the defensive movements in the treated group whereas the animals in pen without fence was exposed to flies and become aggressive in response to biting resulting increased defensive movements in the control groups. The overall apparent density biting flies was 0.214 and 0.87 in treated and control group. This was shown by a significant difference ($p<0.05$) between the groups for the parameters like PCV, entomology and defensive movements. The PCV was increasing and decreasing in treated and control groups in the absence of tsetse flies and trypanosomes throughout the study time. So, there should be a factor that was responsible to cause anemia in the control but prevented in the treated group which were caused by biting flies (*Tabanus* and *Stomoxys*). In light of the above conclusion the following points are forwarded as a recommendation:

- Further study needs to be carried out to know the causes of anemia which were transmitted by biting flies.
- A year-round (Both dry and wet season) study should be conducted to determine the seasonal impact on tsetse and biting fly densities.
- The use of insecticide-treated net technology should be supported and extended to the community in the country at large.

ACKNOWLEDGEMENT

The authors would like to thank the farm owners for their co-operation up on data collection.

REFERENCES

1. WHO, 2006. World health organization fact sheet. N0259 Http:// www.who.int/ mediacentre/ factsheets/ fs259/ en/ accessed in May, 2018.
2. Alemayehu, B., B. Bogale, T. Fentahun and M. Chanie, 2012. Bovine trypanosomosis: A threat to cattle production in Chena district, Southwest Ethiopia. Open J. Anim. Sci., 2(4): 287-294. http:// dx.doi.org/ 10.4236/ ojas.2012.24039Vol.2.
3. CSA, 2012. FDRE Agricultural Sampling Survey. Vol.II: Reports on Livestock and Livestock Characteristics, Addis Ababa, Ethiopia.
4. Taylor, K.A., 1998. Immune response of cattle to African trypanosomosis. Protective or pathogenic? Int. J. Parasitology, 28: 219-240.
5. Tesfaye, M., 2002. Report of Trypanosome infection rate in Glossina morsitans submorsitans and Glossina tachnoides in Didessa Valley, Bedelle.
6. Mare, C.J., 2004. African Animal Trypanosomiasis (Nagana, Tsetse Disease, Tsetse Fly Disease). Veterinary Science/ Microbiology, University of Arizona Tuson.Pp.1 http:// www.vet.uga.edu/ vpp/ gray_book/ Handheld/ aat.htm (2 of 12)3/ 5/2004, 4:07 AM
7. CSA, 2013. FDRE Agricultural Sample Survey. Vol. II: Report on Livestock and livestock characteristics (Private peasant holdings). Statistical Bulletin 570. Addis Ababa, Ethiopia.
8. Tamire, M., 2003. Comprehensive geography. Mega publishing enterprise, Addis Ababa.
9. Hu, C. and S. Askoy, 2006. Innate immune responses regulate trypanosome parasite infection of the tsetse fly Glossina morsitansmorsitans. Mol. Microbiol, 60: 1194-1204.
10. Leak, S.G.A., 1999. Tsetse Biology and Ecology: Their Role in the Epidemiology and Control of Trypanosomiasis. 1st Edn. ILRI, Wallingford, ISBN., pp: 568.
11. Leak, S.G.A., D. Ejigu and M.J.B. Vreysen, 2008. Collection of Entomological Baseline Data for Tsetse Area-Wide Integrated Pest Management Programmes. 1stedn. Food and Agriculture Organization of the United Nations, Rome, ISBN., pp: 205.
12. Bouyer, J., L. Guerrini, J. Cesar, S.D. Rocque and D. Cuisance, 2005. A phyto-sociological analysis of the distribution of riverine tsetse flies in Burkina Faso. Med. Vet. Entomol., 19: 372-378.
13. PAAT, 2008. Program against African trypanosomiasis, Food Agricultural Organizations of the United Nations, Rome, 31: 2.
14. Oluwafemi, R.A., 2009. The impact of African animal trypanosomiasis and tsetse fly on the livelihood and well-being of cattle and their owners in the BICOT study area of Nigeria. Inter. J. Vet. Med.
15. Getachew, A., 2005. Review article trypanasomiasis in Ethiopia. Journal of Biological Science, 27: 1-8.
16. Teka, W., D. Terefe and A. Wondimu, 2012. Prevalence study of bovine trypanosomosis and tsetse density in selected villages of Arbaminch, Ethiopia. Journal of Veterinary Medicine and Animal Health, 4(3): 36-41. Available online at http:// www.academicjournals.org/ JVMAH.
17. Urquhart, G.M., J. Armour, J.L. Duncan, A.M. Dunn and F.W. Jenning, 1995. Veterinary parasitology. The University of Glasgow, pp: 203-212.
18. MoA, 1995. Ethiopian ruminant livestock development strategy. Addis Ababa, Ethiopia, Ministry of Agricultural, pp: 112-113.
19. NTTICC, 2004. Annual report, National Tsetse and Trypanosomosis Investigation and Control Centre, NTTICC. Bedele, Ethiopia, pp: 30.

20. Tewolde, N., 2001. Study on the occurrence of drug resistant trypanosomes in cattle in the farming in tsetse control areas (FITCA) project in Western Ethiopia. MSc thesis, Addis Ababa University, Faculty of veterinary medicine, Ethiopia, pp: 42-49.
21. FAO, 2002. Program Against African Trypanosomiasis (PAAT). Twenty second Regional Conference for Africa, Cairo, Egypt.
22. Maudlin, I., P.H. Holmes and M.A. Miles, 2004. The Trypanosomiasis. Wallingford, UK: CAB International Publishing, pp: 1-634.
23. Geerts, S., P.H. Holmes, O. Diall and M.C. Eisler, 2001. African bovine trypanosomiasis: the problem of drug resistance. Trends Parasitol, 17: 25-28.
24. Maia, M.F., 2009. Impact Of Insecticide-Treated Nets Protecting Cattle In Zero-Grazing Units On Nuisance And Biting Insects In The Forest Region Of Kumasi, Ghana. Thesis Submitted For The Fulfilment Of A Doctoral Degree In Veterinary Medicine At The Free University Of Berlin, Germany. Journal, pp: 3298.
25. Kapitano, B., 2013. Effect of Deltamethrin-treated Screen (ZeroFly® livestock) fenced around cattle pen in protecting animals from tsetse flies, biting flies and other flies, Arbaminch, STEP, Ethiopia. (Unpublished).
26. GZARDO, 2007. GamoGofa Zone Agricultural and Rural Development Office, Arba Minch, GamoGofa, Ethiopia.
27. MAWDA, 2007. Report from Merab Abaya woreda Agricultural office, GamoGofa, SNNPRs, Ethiopia.
28. Fisher, M.S. and R. Say, 1989. Manual of Tropical Veterinary Parasitology. UK: CAB International Publication, pp: 100-278.
29. Brightwell, R., R.D. Dransfield, C.A. Korku, T.K. Golder, S.A. Tarimo and D. Mugnai, 1992. A new trap for Glossina pallidipes. Trop. Pest Manag, 33: 151-159.
30. Murray, M., P.K. Murrary and W.I.M. McIntrye, 1977. Unimproved parasitology teaching for the diagnosis of African trypanosomosis. Trans. R. Soc. Trop. Med. Hyg., 72: 325-326.
31. Radostitis, O.M., C.C. Gay, K.W. Hinchcliff and P.D. Constable, 2007. Veterinary Medicine. A text book of the disease of cattle, horse, sheep, pigs and goats, 10th edition. Saunders Elsevier, Edinburgh, London, New York, Oxford, Philadelphia, St Louis, Sydney and Toronto., pp: 1533-2047.
32. Maia, M.F., P.H. Clausen, D. Mehlitz, R. Garms and B. Bauer, 2010. Protection of confined cattle against biting and nuisance flies (Muscidae: Diptera) with insecticide-treated screens in the Ghanaian forest zone at Kumasi. Parasitol Res., 106: 1307-1313.
33. Mahama, C.I., M. Desquesnes, M.L. Dia, B. Losson, R. De Deken and S. Geerts, 2004a. A cross-sectional epidemiological survey of bovine trypanosomosis and its vectors in the Savelugu and West Mamprusi districts of northern Ghana. Vet Parasitol, 122(1): 1-13.
34. Mahama, C.I., M. Desquesnes, M.L. Dia, B. Losson, R. De Deken, N. Speybroeck and S. Geerts, 2004b. A longitudinal epidemiological survey of bovine trypanosomiasis and its vectors in the White Volta river basin of Northern Ghana. Vet. Parasitol., 128(3-4): 201-208.
35. Bauer, B., B. Holzgrefe, C.I. Mahama, M.P. Baumann and D. Mehlitz, 2011. Managing tsetse transmitted trypanosomosis by insecticide treated screens-an affordable and sustainable method for resource poor pig farmers in Ghana.
36. Bauer, B., D. Gitau, F.P. Oloo and S.M. Karanja, 2006. Evaluation of a preliminary trial to protect zero-grazed dairy cattle with insecticide treated mosquito netting in Western Kenya. Trop.Anim.Health Prod, 38: 29-34.
37. Baldacchino, F., V. Muenworn, M. Desquesnes, F. Desoli, T. Charoenviriyaphap and G. ADuvallet, 2013. Transmission of pathogens by Stomoxys flies (Diptera, Muscidae): A review. Parasite, 20: 26.