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In vitro Ectoparasiticidal Efficacy of Commonly Used Acaricides on Selected Herds in and Around Chancho Town, Ethiopia

Dejene Getachew, Aboma Regassa, Tesfaye Birhanu and Kusa Mole

Wolmera District Livestock and Fishery Resource Development Office, Holeta, Ethiopia

Abstract: The study was carried out from October 2019 to April 2020 on selected kebeles of Sululta woreda in and around Chancho town. The therapeutic efficacy of two different acaricides: Amitraz and Diazinon were evaluated in *in vitro* and herders' communities were assessed the on acaricide usage, delivery and methods of tick control practice. Acaricide treatment is the only method for tick control in the community, where Amitraz was drug of choice in the area. Adult immersion technique (AIT) was used to determine the efficacy of acaricides at different concentration on randomized control experiment. Majority of respondents (93.5%) use the acaricides when they face maximum of tick infestation using hand dressing and few of them uses knap sack hand sprayer. The AIT result indicates that most of the engorged females of *Rhipicephalus* (*Boophilus*) *decoloratus* immersed in Diazinon laid more eggs than of Amitraz. According to the finding of these study the antiparstic efficacy of amitraz at half, recommended and double recommended dose was 88.9%, 98.2%,98.86% respectively On the other hand, the antiparasitic efficacy of Diazinon at half, recommended, and double recommended concentrations were 38.8%, 72.6% and 87.6% respectively.

Key words: Acaricide · Amitraz · Concentration · Diazinon · Efficacy · *In vitro* Test

INTRODUCTION

Ethiopia is reported to be endowed with the largest livestock population in Africa. According to the 2010 report of the Central Statistical Agency (CSA) the cattle population was estimated at about 50.9 million. The productivity of these animals is affected by many factors, among which animal diseases, inadequate nutrition, poor management, poor genetic makeup and recurrent drought are major causes [1].

Poor health and productivity of animal due to disease has considerably become the major stumbling block to the potential of livestock industry [2]. A wide range of internal and external parasitic diseases are found in domestic animals. Among external parasites, ticks are undoubtedly the most economically important ectoparasites of livestock on global scale [3].

In tropical Africa, tick and tick borne diseases (TBDs) are economically very important diseases next to trypanosomosis [4]. Among 60 tick species found infesting both domestic and wild animal of Ethiopia, 30 species have been wide spread and are important parasites of livestock and causes significant economic losses to the livestock industry [5]. In Ethiopia ectoparasites in ruminant causes serious economic losses to small holder farmers, the tanning industry and country as a while through mortality of animals, decreased production, downgrading and rejection of skin and hide [6].

Ticks harm the hosts both directly and indirectly: Direct harm results from blood loss, tick burden as well as toxicoses. The bites can be injurious and cause severe hide damage including abscessation and can provide a route for secondary infection. Blood loss and reduction in weight gain resulting from tick feeding are among major factors that affect ruminant production in different parts of the world. Indirectly, ticks can cause economic loss because they play an important role as vectors of a wide range of pathogens to humans and domestic animals [7].

Tick acaricide resistance is reported in various parts of the countries where tick and tick borne diseases are of major problem. Since tick infestation is one of the major reported problems in the area, repeated use of acaricides is the only option in high tick seasons [8]. The major constraint of chemical treatment is selection for acaricide resistant tick strains. Inappropriate acaricide use with

Corresponding Author: Dejene Getachew, Wolmera District Livestock and Fishery Resource Development Office, Holeta, Ethiopia. Tel: +25113490974/+21516858009.

incorrect concentrations probably contributes to the development of resistance, which leads to tick-control program failures [9].

The application of chemicals is still the most effective method of tick control. However, uncontrolled applications of commercial acaricides may have accelerated the emergence of tick resistance to several active ingredients available. Since acaricide introduction in Africa around 1890, tick treatment relying on different application methods have been the main method of tick control in Africa, leading to numerous problems; environmental pollution, development of resistant tick strains and escalating costs [10].

Therefore, this study was contemplated with the aim of assessing the in vitro efficacy of two, commonly used ectoparasitidicidal agents, amitraz and diazinon, against the most abundant and important tick infesting cattle.

METERIALS AND METHODS

Study Area: The study was conducted from October 2019 to April 2020 on selected kebeles of Sululta Woreda in and around Chancho town. Sululta is located between 9°4'30"N to 9°30'59"N and 38°31'26"E to 38°58'49"E and the Woreda town Chancho, is situated 40 km northwest of Addis Ababa [11].

Study Design: An experimental randomized controlled in vitro trial was used to assess the effect of commercially available acaricides namely Amitraz and Diazinon against Boophilid tick species randomly collected from herds in and around Chancho town and herds owners who use acaricides for tick control were approached to respond structured questionnaire, which helps to assess the perception of farmers and herd owners towards delivery system of acaricide, as well as the different tick control options and methods of applications practiced in the area.

Study Animals: Animals with high tick infestation and which did not receive any acaricide treatment at least within one month were purposely selected and ticks were collected from those animals for the commencement of the in vitro experimental trail. From those purposively selected animals all visible engorged adult female ticks were collected.

Tick Collection Methods and Species Identification: In each herd, all visible engorged adult female ticks were collected from cattle. These ticks were placed individually in different plastic flasks pre labeled with time, date, place of collection and code number. Then ticks were transported to Addis Ababa University, College of Veterinary Medicine and Agriculture Laboratory within 48 hours of collection for the in-vitro acaricidal efficacy evaluation using AIT. All collected ticks were examined under stereomicroscope and identified using the taxonomic key described by Kaiser [12].

***In-vitro* Acaricide Efficacy Evaluation:** The ticks which are collected from the field were placed individually in different plastic flasks pre-labeled with time, date, place of collection and code number. Afterwards the ticks were transported to Addis Ababa University, College of Veterinary Medicine and Agriculture Laboratory within 48 hours of collection for the in-vitro acaricidal efficacy evaluation using AIT. The AIT was conducted according to the method described by Drummond [13] and then modified by FAO [14].

A total of two hundred seventy (n=270) engorged adult female *Rhipicephalus* (*Boophilus*) *decoloratus* of uniform size were collected from cattle and tick randomly allocated into three groups: Group-1 (n = 10 ticks) and Group-2 (n = 10 ticks) were subjected to each tested acaricide treatment and Group-3 (n=10) were untreated, ticks serve as control. Commercial formulations of Amitraz and Diazinon were diluted in distilled water according to the label recommendations at three different concentrations. The recommended (1:1000 for diazinon, 1.6:1000 for amitraz), double (2:1000 for diazinon, 3.2: 1000 for amitraz) and half doses were prepared. Each group of 10 females were immersed for 10 minutes in 20 mL of the acaricide solutions of different concentration. After 10 minutes of immersion all ticks were cleaned and air dried on absorbent paper. All treatment and control groups ticks were later stuck (ventral side up) with double sided sticky tape in a Petri dish. The plates were then placed in larger, plastic boxes containing a moistened sponge for 7 days at temperature of 27°C and incubated. To estimate the efficacy of each acaricide, both groups (treated and control) were then tested using the egg laying test (ELT) method which involves the comparison of the egg mass of ticks treated with acaricide and the egg mass of untreated ticks and finally estimates the percentage control value, using the following formula according to the method described by Drummond [13] and then modified by FAO [14].

$$Percent\ control = \frac{MEC - MET}{MEC} \times 100$$

where, MEC and MET are mass of eggs laid by control ticks and treated ticks, respectively. During the study period, three successive replicates of the above trails for each acaricide treatment and control group were established.

Data Management and Analysis: Data were organized, edited and analyzed using statistical package for social sciences (SPSS) Version 20. Results generated from the investigation was expressed using descriptive statistics (mean ± standard error of mean, percentage and graphs); and statistically significant was taken if $p \leq 0.05$ at 95% confidence intervals.

RESULTS

Questionnaire Survey: The result of the questionnaire survey indicates that almost none of respondents were using acaricides by schedule for tick controlling. A majority of the interviewed respondents (80%) declared that they used Diazinon 60% EC for long period of time in the area due to the availability and cost of Diazinon 60% EC was fair compared to other acaricides.

According to 69% of the respondents, Amitraz was effective than Diazinon whereas 31% of herd owners arguing that Diazinon was more effective. The farmers and herd owners were also interviewed from which source they get the acaricides and about 48.4% of respondents complained that the source of acaricides are private pharmacies, 35.5% of herders are from government veterinary clinics and 3.2% from the local markets.

According to the respondents, only 54.8% of them dilute acaricides based on the recommendation of manufacturer. The others said that they dilute the acaricide based on burden of the tick and population of their cattle. Majority of respondents use the acaricides when they face maximum of tick infestation using hand dressing and few of them uses knap sack hand sprayer.

In the study area, almost all interviewed herders, responded that they didn't know expiration date of the acaricides and from some of respondents there were reports of intoxication.

***In vitro* Acaricidal Efficacy:** The AIT result indicates that most of the engorged female Boophilus decoloratus ticks immersed in Diazinon at half, recommended and double recommended dose laid eggs with a mean of 0.2133333 gm, 0.1218000gm and 0.045000gm respectively. Whereas those treated with Amitraz at half, recommended and double recommended dose laid eggs with a mean of 0.386667gm, 0.008000gm and 0.0041667gm respectively (Table 1).

Analysis result of this preliminary test indicates that, Amitraz seemed to be superior to Diazinon as measured by acaricide efficacy (%) estimation against Boophilus tick at all concentrations. The antiparasitic efficacy brought about by Amitraz at half recommended, recommended and double recommended dose were 88.9%, 98.2% and 98.86% respectively. On the other hand, the antiparasitic efficacy of Diazinon at half, recommended and double concentrations were 38.8%, 72.6% and 87.6% respectively (Table 1).

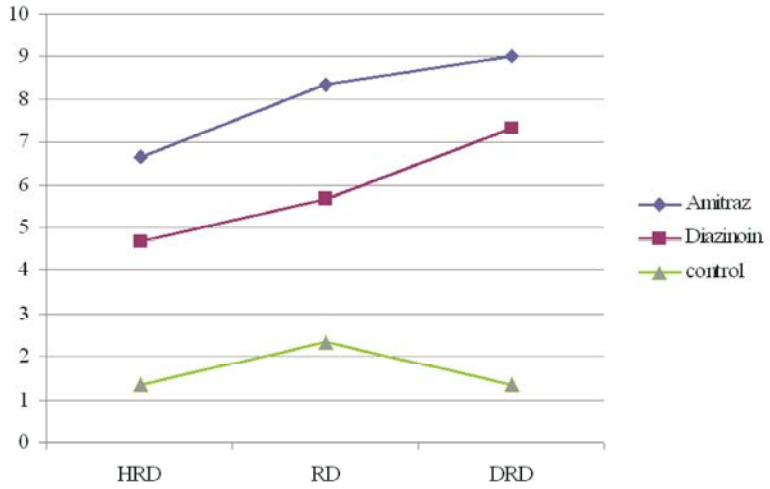

Fig. 1: *In vitro* killing effect of diazinon and amitraz after 7 days incubation at half, recommended and double recommended concentrations. HRD= Half recommended dose; RD= recommended dose; DRD= Double recommended dose

Table 1: *In vitro Rhipicephalus* (*Boophilus*) *decoloratus* ticks killing effect of amitraz and diazinon at double, recommended and half doses after 7 day incubation

Treatment Drug	Mean tick dead	Mean of number of tick lie egg	Mean egg in gram	Anti. parasitic efficacy
Amitriaz HD	6.67±0.577	1.33±1.155	0.0386667±0.04221769	88.9%
Control	1.33±2.309	9.67±0.577	0.3486667±0.06387749	0
Amitraz RD	8.33±0.577	0.33±0.577	0.0080000±0.00346410	98.2%
Control	2.33±2.082	10.00±00	0.4440000±0.04911212	0
Amitraz DRD	9.00±0.00	1.00±00	0.0041667±0.00388373	98.86%
Control	1.33±2.309	9.67±0.577	0.3640000±0.07076722	0
Daizinon HD	4.67±1.155	4.67±5.033	0.2133333±0.10692677	38.8%
Control	1.33±2.309	9.67±0.577	0.0386667±0.04221769	0
Diazinon RD	5.67±0.577	4.33±3.055	0.1218000±0.05487550	72.6%
Control	1.33±2.309	10.00±00	0.4440000±0.04911212	0
DiazinonDRD	7.33±0.577	4.67±3.786	0.0450000±0.00529150	87.6%
Control	1.33±2.309	9.67±0.577	0.3640000±0.07076722	0

According to this study, both Amitraz and Diazinon produced maximum efficacy at their double recommended doses and both of them were less effective at half recommended dose. Amitraz showed evidence of greatest tickicidal effect after exposure to two chemicals at different concentrations after 7 days of incubation (Figure 1).

DISCUSSION

The major tick species identified in the study areas was *Rhipicephalus* (formerly *Boophilus*) *decoloratus*. In the study area, amitraz and diazinon are the most commonly applied chemicals for tick control and treatment in different species of domestic animals.

The result of the questionnaire indicats that tick control in the area mainly accomplished by use of acaricides and according to 69% of interviewed herders Amitraz 12.5% was effective than Diazinon 60% which support also the finding of the expermental trail on these study. At the time of this study, about 48.4% of respondents complained that the source of acaricides are private pharmacies, 35.5% of herders are from government veterinary clinics and 3.2% from the local markets.

According to finding obtained from the questionnaire survey, only 54.8% of the farmers use the acaricides based on the recommendation of manufacturer. Other respondants were diluting based on burden of the tick and population of their cattle. This could be one of the main reason which leads to the decreased efficacy of the acaricides that are used in the study area.This observation was in agreement with the study done by Eshetu [15] in Borena, Ethiopia, found that most pastoralists make concentration and dilution of acaricides based on the extent of tick infestation and number of cattle population.

Most of respondents complained that the source of acaricides are private pharmacies, because of its availability and the governmental clinics are closed when they came to market. Although 3.2% of respondents were only bought acaricides from local markets, only 6.5% of respondents were using acaricides by schedule for tick controlling. Majority of respondents use the acaricides when they face maximum of tick infestation using hand dressing and few of them uses knap sack hand sprayer. Other study done in Ethiopia also agree with this study that ticks on indigenous cattle are treated whenever the farmers bring their animals to the veterinary clinics either for tick control or for other complaints. There is no planned program of tick control except on dairy farms Horak [16].

Analysis result of this preliminary test indicated, Amitraz seemed to be superior to Diazinon as measured by acaricide efficacy estimation against Boophilus tick at recommended dose shows that 98.2% and 72.6% respectively. Sileshi [17], compared the efficacy of amitraz and diazinon in Ethiopia, on R. pulchellus and other ticks and found that amitraz at recommended concentration provides better efficient oviposition inhibition than diazinon with tickicidal efficacy of 90.94% and 71.41% and agrees with the finding of this study. The finding of Furlong *et al.* [18] in Southeast Brazil also obtained mean Amitraz efficacy of 95% and showed the superiority of Amitraz.

In contrast to the present study, Freitas *et al.* [19] found mean acaricidal efficacy of Amitraz as 47.9%. In North eastern Brazil low acaricidal effect of Amitraz (40.5% and 30.95%). These diffence could be due to the fact that amitraz may be used in those araes for a very long period of time.Still we can assume that amitraz was effective than Diazinon.

At present study, both Amitraz and Diazinon were produced maximum efficacy only at their recommended doses with efficacy of 98.86% and 87.6% respectively. Similarly, a closely comparable finding, tickicidal effect of 100% and 92.8% for both amitraz and Diazinon were reported by Silva [20] at Brazil, in their study.

In the present study, the efficacy variation between the two presently tested acaricides might be associated with prolonged exposure of the most prevalent tick species in the area to Diazinon 60% EC. In the present questionnaires survey, majority of the interviewed farmers declared that they used Diazinon 60% EC for long period of time in the area depending on the availability of them and cost of the acaricides. In South Africa, Horak [16] also observed relatively higher level of resistance to organophosphorous chemicals than to Amitraz 12.5% due to its utilization for over 10 years. In addition, the work of Natala [21], highly supports this finding and observed that sequential use of products from the same chemical group for long periods favored the development of resistance.

CONCLUSION AND RECOOMENDATIONS

In the study area respondents were using acaricides for tick controlling. The questioner survey revealed that farmers in the area used Diazinon 60% EC for long period of time in the area depending on the availability of them and cost of the acaricides. The present work demonstrated that amitraz has relatively conserved its tickicidal efficacy than diazinon as the result of in vitro test. A long time usage of the same acaricide type, abnormal concentration, usage of expired acaricides, and usage of acaricides without schedule are the common phenomenon of tick control methods in the area. Therefore, awareness creation for farmers on the ways of proper acaricide usage, application, dilution and systematic ways of substitution is recommended. Moreover, the distribution of acaricides should be under supervision of professionals and authorized body.

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