Study on the assessment of drug resistance on *Trypanosoma vivax* in Tselemti woreda , Tigray, Ethiopia

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Abstract

The study was conducted at Tselemti Woreda, Tigray region, Northern Ethiopia, some 1200 kms away from the capital, from November 2002 to April 2003. In the first phase of this study, questionnaire survey was carried out to assess livestock production constraints. Subsequently, a study on the occurrence of drug resistance to diminazene aceturate and isomethamidum chloride of *Trypanosoma vivax* isolate in artificially infected goats were conducted. Results of the questionnaire survey revealed that 95.7% of the interviewees replied that trypanosomosis is a series problem in their areas. Apart from this, under dosing of trypanocidal drugs appeared a common practice in the areas surveyed. Drug sensitivity test conducted on 18 artificially infected goats revealed that *T. vivax* developed resistance to the curative doses of diminazene aceturate (3.5 mg/kg b.wt) and isomethamidum chloride (0.25 mg/kg b.wt). No relapses were seen for those animals treated with diminazen aceturate at a dose of 7 mg/kg b.wt and 0.5 mg/kg b.wt of isomethamidum chloride. Sanative pair between diminazene aceturate and isometamidum chloride was also confirmed by the occurrence of no relapses after first treatment with diminazene aceturate (at 3.5 mg/kg) and isomethamidum chloride (0.25 mg/kg) and a second treatment with 0.25 mg/kg isomethamidum chloride and 3.5 mg/kg diminazene aceturate, respectively. During the 90 days of trial period there was no statistical significant difference ($P>0.05$) in mean PCV of the five groups before and after treatment: (26 ± 1.23), (27.5 ± 1.33), (26.5 ± 2.03), (29.8 ± 2.12) and (25.5 ± 3.06) for groups I, II, III, IV and V respectively. Generally diminazene aceturate at dose of 7 mg/kg and isomethamidum chloride at dose of 0.5 mg/kg b.wt were most effective in terms of curing infections as well as improving PCV and body weight. It was concluded that where there are indications for drug resistance against isomethamidum and diminazene aceturate the use of trypanocidal drugs should be supervised, the principle of sanative pairs has to be applied and chemotherapy needs to be integrated with other methods like vector (tsetse and biting flies) control.

Keywords: Diminazene-aceturate, drug-resistance, goats, isomethamidum-chloride, trypanosomosis, Tselemti.

Introduction

One of the major constraints of livestock production in Africa is poor animal health. Trypanosomosis is a widespread and economically important disease of man and his farm animals (ILRAD, 1987) exerting adverse effect on the whole pattern of agricultural activities in the continent.Trypanosomosis is a parasitic disease caused by unicellular protozoan parasites of the genus Trypanosoma and family Trypanosomatdae (Itard, 1989). It is transmitted by the bite of a Tsetse fly (Glossina Spp). Getachew Abebe and Yilma Jobre (1996) have also reported that in addition to tsetse transmitted trypanosomes, mechanically transmitted trypanosomes due to *T. vivax* is a potential treat to the huge highland livestock in Ethiopia.

Trypanosomosis is one of the major diseases that cause direct and indirect economic losses (ILRI, 1996). The control programme of the disease with effects on various aspects of the epidemiological cycles of the infection is essential and can be achieved by taking action on the parasite, on the vectors and on the host. In the absence of an effective vaccine and lacking a cohort strategy for controlling the insect vector, in many African countries the dependence on trypanocidal drugs for control of animal trypanosomosis has become alarming (Jordan, 1986). Following the prolonged use of these trypanocides emergence of strains of trypanosomes resistant to the drug has been reported from different parts of the continent (Ainanshe *et al*., 1992).

In spite of all the efforts so far, the emergence of drug resistance has also seriously hampered the control of animal trypanosomosis in Ethiopia. Experimental studies have shown that multiple trypanocidal drug resistant to both dimenazene and isomentamidium reported for T.congolense in Abay/ Didessa Tsetse belt in Metekel district, North west Ethiopia (Yohannes Afework, *et al*., 2000) and in the Ghibe/ Omo Tsetse belt which is adjacent to the upper Didessa river valley (Wubet Mulugeta *et al*., 1997), in Kindo Koysha wereda, Southern Ethiopia (Mesfine Ademe and Getachew Abebe, 2000). Therefore, the distribution and degree of drug resistance has to be carefully monitored so as to work out the best possible therapeutic strategies and /or alternative control measures.

In view of this the present study was designed with the following objectives:

- To assess the trypanocidal activities of diminazene aceturate and isomethamidum chloride in goats artificially infected with trypanosome isolate (*T. vivax)* from naturally infected cattle.

- To suggest appropriate measures for controlling drug resistance in the field.

Material and Methods

Study Area

The study was carried out at Mekelle Regional Laboratory from November 2002 to April 2003. *T. vivax* isolate was collected from Tselemti Woreda, Northwest part of the region. Mekelle, the capital of Tigray region, is situated at 39°29' E and 13°30' N at an altitude of 2070 meter above sea level. It is located 783 km north of Addis Ababa. Tselemti, which is found in north west part of the region, located 1200km from the capital is found at 38°15' E and 13°48' N, at an altitude of 1400 meter above sea level. The region has a marked variation in rainfall from East to West. The mean annual rainfall varies from less than 200 mm in the eastern extreme on the boarder of Danakil depression to over 1200 mm in the southwestern part of the region. Temperature also shows variation according to altitude. During summer the lowlands of eastern Tigray have a mean temperature of 27.5°c. The extreme western part with altitude below 1000m above sea-level and the slopes of the great escapement running North-South in central Tigray have mean temperatures of 25 °c and 20°c respectively (SAERT, 1993).

Questionnaire Survey

For a questionnaire survey a total of 73 people were interviewed individually. The main questions included were about herd composition, management, extent and type of trypanocides used, importance of trypanosomosis in the area, and relate this to the result of drug sensitivity trial in the field. The survey was carried out in the trypanosomosis infested areas of the Tselemti Wereda include: Mai Anbesa, Feyel Wuha and Mezekere .

Drug Sensitivity Trial

Experimental animals

Goats weighing about 34.2 ± 5.1 kg at the start of the experiment were used. They were obtained from the Mekelle Regional Veterinary Laboratory and their blood was examined and found to be free from trypanosomosis prier to challenge. We were also checked them for other heamoparasites and other blood sucking GIT parasites.

The goats grazed during the day and were confined in sheds at night. As the experiment was carried out during the dry season (November 2002 to April 2003) when grazing was poor, the animals were fed supplementary dry hay.

Trypanosome stock

Trypanosoma. vivax, isolated from a natural infected Zebu cattle in goat at Tselemti wereda, north west part of the Tigray region was used. The goat used for isolation was syringe infected by intravenous (i.v) injection of blood collected from naturally infected cattle, as described by Miwambu, (1969). Then the goat was brought to the Mekelle Veterinary Regional laboratory and stayed for 8 days. The goats used in the experiment were syringe infected by intravenous injection of blood collected from the donor goat. Each infective dose was given in 2ml of blood. The blood was collected using di-sodium salt of ethylene diamine tetra - acetic acid (EDTA) as anticoagulant. Sub-inoculation from infected goat and treatment of experimental goats were performed when parasitemia was of the order of 5-10 trypanosomes per high power microscope field (40X).

Trypanocidal Drugs

Dimenazene aceturate /VeribenTM, Lot No. 33501, Exp. 05-2004, SANOFI SANTE ANIMALE Product., Liborne, France) and Isomethamidum chloride (Trypamidium®, Lot No. A 9809, Exp. 07-2006, MERIAL–17, rue Bourgelat Lyon – France) were drugs used in the experiment. Both drugs where administered by deep intra - muscular injection on the thigh muscle with maximum care to avoid leakage to the sub cutaneous tissues by completing the injection before withdrawing the needle.

Dimenazene aceturate was injected as a 7% solution at doses of 3.5 and 7 mg/kg of body weight while isomethamidium chloride was injected as 1% solution at doses of 0.25 and 0.5 mg/kg of body weight. Both drugs were administered

to animals on the basis of accurate body wt. measurement taken immediately before treatment using a weighing scale.

Experimental Design

Eighteen experimental animals were divided into 5 treatment groups. All the experimental animals were randomly distributed as a case and control in the experiment. Goats weighing about 34.2 ± 5.1 kg at the start of the experiment and all the goats were male.

1. Group I (n=4) dimenazene aceturate (3.5 mg/kg b.w, i.m)
2. Group II (n=4) dimenazene aceturate (7mg/kg b.wt, i.m)
3. Group III (n=4) isomethamidum chloride (0.25 mg/kg b.wt, i.m)
4. Group IV (n=4) isomethamidum chloride (0.5 mg/kg b.wt, i.m)
5. Group V (n=2) control

Relapsed experimental animals from group 1 were retreated with 0.25 mg/kg isomethamidum and from group 3 with 3.5 mg/kg b.wt of diminazene aceturate (Table 1). Animals in the control group were left untreated until day 8 since at day 8 their PCV is markedly decreased; highly parasitemic (4^+ score) and they were clinically sick. Then treated with veriben at a dose of 3.5 mg/kg b.wt.

Table 1. Treatment of the experimental groups

Group	Treatment at day zero (mg/kg b.wt)	Treatment after relapse (mg/kg b.wt)
I	Diminazene aceturate, (3.5)	Isomethamidum chloride, (0.25)
II	Diminazene aceturate, (7)	Isomethamidum chloride, (0.25)
III	Isomethamidum chloride, (0.25)	Diminazene aceturate, (3.5)
IV	Isomethamidum chloride, (0.5)	Diminazene aceturate, (3.5)
V	___	Diminazene aceturate, (3.5)

Recorded Parameters and Experimental Procedure

All the experimental animals were followed on daily basis for the first 7 days for PCV determination and detection of trypanosomes. Then, animals were monitored once a week for the next 11 weeks for PCV and parasite detection, while body weight measurement was conducted monthly. Animals were regarded as cured if following treatment no parasitemia was detected within the period of examination.

Haematology: Blood was taken from ear vein after bleeding using sharp needle from each experimental animals. Then the capillary tube was filled 3/4th of its length and sealed using crystal seal at one end. Then the capillary tube put symmetrically with the sealed part outside and centrifuged at 12000 rpm for 5 minutes using centrifuge and then PCV was determined using a Hawksley microhaematocrit reader (Murry, *et al*., 1983).

Parasitological examination: After PCV was measured, then the capillary tube was broken 1mm below the buffy coat and 1cm above the buffy coat and express the content on the slide and cover with cover slip and examine under 40X objective lens using dark ground buffy coat techniques (Murray, *et al*., 1983) and looking for Trypanosomosis and the level of parasitemia was estimated using the standard method of scoring by Paris, *et al*., (1982).

Body Weight Measurement: Animals are measured just before treatment, one month, two months and three months after treatment to see if there is any improvement in body condition due to treatment using weighing scale.

Data Analysis

The data was analyzed using appropriate statistical method. Students paired t-test was applied to compare the mean PCV before and after treatment. PCV and Body weight improvements were also seen in the five treatment groups, to see if there was any difference (Argawa, 1996)

The mean PCV improvement was calculated using the formula:

$$[(\underline{PCV_2 - PCV_1})/ PCV_1]X100$$

and the mean body weight improvement using

$$\underline{[(BW_2 - BW_1)/ BW_1]X100}$$

Results

Questionnaire Survey

Herd Composition: In the study area, most farmers keep their cattle together with small ruminants, independent of their age and sex. Cattle constituents' major part of the herd. During birth, the dam and the calf will be separated from the rest and would stay together for a certain period of time.

Livestock Management: Animals graze in the field, early in the morning and late in the afternoon. Feed is usually available during the rainy season (June – September) and it is least available during the dry seasons (November – May). During dry periods animals provided with dry hay. Animals and Human share the same roof for their house.

Trypanosomosis Therapy: A total of 73 individuals were interviewed. Almost all (95.7%) of them agree that trypanosomosis is a major problem for there animals and it causes great loss animals every year. There is success for treatment as it is supported by many of the farmers, but sometimes claims of "no change" with treatment probably because of development of resistance or inappropriate diagnosis of the patient. Among the respondents 30% and 25% of the farmers reported that dimenazene aceturate and isomethamidum chloride were used below the recommended dose rates respectively. On the other hand 35% and 30% of the farmers reported that they didn't have any idea on the dose rates of both drugs, respectively.

According to the questionnaire 30% of livestock owners are treating their cattle themselves against Trypanosomosis. It is viewed that various trypanocidals are used in the area, the most common once are ethidium (homidium chloride), diminazene aceturate and isomethamidum chloride. However, most farmers said that they are using one sacket of veriben for one or two cattle and one tablet of ethidium for two cattle.

Drug Sensitivity Test: Goats receiving the different treatments were compared to the control animals. Prier to treatment, heavy parasitemia with *T.vivax* was detected in all the infected goats using HCT as the diagnostic methods.

Parasitological findings: Experimental animals were followed daily for the first 7 days and then weekly for 83 days to see if there are relapsed cases after treatment. When the goats were treated with isomethamidum and dimenazene aceturate, the level of parasitemia was significantly reduced beginning 24 hrs later. At day 35 and 63 two animals in group 1 relapsed which were treated with 3.5 mg/kg dimenazene aceturate and at day 56 an animal in group 3 was relapsed, it was treated with 0.25 mg/kg b.wt isomethamidum chloride. But there was senative pair to the stock in the area in which after treatment with 0.25 mg/kg b.wt isomethamidum chloride for group 1 and 3.5 mg/kg b.wt dimenazene aceturate for group 3 there was no relapse for the rest of the experimental period. But all animals in group 2 which had received 7 mg/kg b.wt

dimenzene aceturate and those in group 4 which were treated with 0.5 mg/kg b.wt isomethamidum chloride showed no relapse during the experimental period. That means 3.5 mg/kg b.wt veriben and 0.25 mg/kg b.wt trypamidium do not work as shown in Table 2 and 3.

More relapses occurred in group1 (with relapse percentage of 50%), than the other groups. In general more relapses have been seen in dimenazene aceturate (50%, 0%) than isomethamidum treated groups (25%, 0%) (Table 2).

Table 2. Relapses after the first treatment

Group	Number of animals treated	First treatment (mg/kg b.wt, i.m)	Parasitemia Days 0-5	6-30	31-60	61-90	Total
I	4	3.5 (Diminazene)	4	0	1	1	2=50%
II	4	7 (Diminazene)	4	0	0	0	0=0%
III	4	0.25 (Isomethamidum)	4	0	1	0	1=25%
IV	4	0.5 (Isomethamidum)	4	0	0	0	0=0%

Table 3. Relapsed infections after first treatment with isomethamidum chloride and dimenazene aceturate and subsequent treatment with dimenazene aceturate and isomethamidum chloride respectively.

First treatment with diminazene aceturate (i.m)	Number of Animals	
	2nd treatment with 0.25 mg/kg b.wt isomethamidum chloride	Relapse after second treatment
3.5 mg/kg b.wt	2	-
7 mg/kg b.wt	-	-
First treatment with isomethamidum (i.m)	2nd treatment with 3.5 mg/kg b.wt dimenazene aceturate	Relapse after second treatment
0.25 mg/kg b.wt	1	-
0.50 mg/kg b.wt	-	-

PCV Improvement

Table 4 show the mean PCV improvement of the experimental animals over the entire study period.

Table 4. Mean PCV and percentage improvement during the study period.

Week	Group I		Group II		Group III		Group IV		Group V	
	Mean	Imp.	Mean	Imp.	Mean	Imp.	Mean	Imp.	Mean	Imp.
0		00	19	00	22	00	20.4	00	22	00
1	22.5	3.2	21	10.5	22.8	3.64	23.6	15.7	21	-4.5
4	23.8	9.2		21	24	9	25	22.55	22.4	1.82
8	24.5	12.4	24.5	29	24	9	26.6	30.4	24	9.1
12	26	19.3	27.5	45	26.5	20.45	29.8	46	25.5	15.9

Day 0 to 7 : Statistical analysis using paired t-test on the PCV improvement before and after treatment showed that there is no significant difference ($p > 0.05$) in the means of the five groups. The mean PCV improvement of animals in group IV is higher than that of group I, II, III and V (Table 4).

Day 0 to 90 – using students paired t-test, to see if there is statistically significant difference in the means of the five treatment groups before and after treatments revealed that again there is no significant difference ($p>0.05$). But there is a difference in mean PCV between groups in which group IV has relatively highest PCV than the rest and Group V the lowest. The maximum PCV improvement was 46.0% (Group IV on week 12) and minimum was 4.5% (Group V on week 2).

Body weight change

The mean body weight improvement of the five groups of animals shown in Table 5.

Statistical analysis using students paired t-test revealed that there is no statistical significant difference in body weight improvements in the five study groups. But there is difference in body weight improvements for the study period. Group IV shows the highest improvement (13.8%) and Group V the least (-6.75) at day 90 (Table 5).

Table 5. Mean body weight improvement of the study animals (Kg)

Days post treatment	Group I		Group II		Group III		Group IV		Group V	
	Mean	Imp.	Mean	Imp.	Mean	Imp.	Mean	Imp.	Mean	Imp.
0	32	00	36	00	35.5	00	29	00	40	00
30	31	–3.125	36.7	1.94	34.5	–2.8	29.3	1	36	-10
60	30.5	-4.7	37	2.78	33.7	–5.1	32	10.3	36.5	–8.75
90	30	–6.25	38	5.55	34	–4.2	33	13.8	37.3	–6.75
Change D0 – D90	-6.25		5.55		-4.2		13.8		-6.75	

Discussion

Questionnaire Survey

According to the result of the questionnaire survey 95.7% of the interviewed reported that trypanosomosis is a serious problem to keep livestock in their areas. According to the present findings, all the farmers interviewed reported that trypanosomosis cases occur either in the dry season (November – May), start of the rainy season (May – June) and after the rainy season (September – November). Yohannes Afework (1998) and Nega Tewolde (2001) also reported similar findings.

With regard to the usage of trypanocidal drugs, 30% and 25% of the farmers reported that dimenazene aceturate and isomethamidum chloride were used below the recommended dose rates respectively. On the other hand 35% and 30% of the farmers reported that they didn't have any idea on the dose rates of both drugs, respectively. Yohannes Afework (1998) and Nega Tewolde (2001) also reported that more than 40% of the interviewees responded that they used trypanocidal drugs below the recommended dose rate whilst below 20%, Yohannes Afework (1998) and above 40%, Nega Tewolde, (2001) of the respondents did not have any idea on the doses of trypanocidal drugs.

About 30% of the respondents said, they themselves would treat their sick animals or by other uncertified individuals. Yohannes Afework (1998) and Nega Tewolde (2001) had also reported about 43% and 57% of it to be applied in the same manner in village cattle of Metekel district, Northwest Ethiopia and in FITCA in Western Ethiopia respectively. One of the reasons for trypanocidal drug misuse could be related to the inefficiencies of government services in Ethiopia to discharge their responsibilities in remote areas.

Drug Sensitivity Trial

In the present study relapse occurred to diminazene aceturate and isomethamidum chloride at 3.5 mg/kg body weight and 0.25 mg/kg of body weight respectively. This could be taken as evidence of drug resistance in this stock of *T. vivax* contrary to the view expressed by Fairclough (1963) and Folkers (1966) that it is difficult to induce resistance to isomethamidum even by repeated low dosages (0.25 – 0.5 mg/kg).

From animals treated with dimenzaene aceturate at 3.5 mg/kg, relapse percentage of 50% was found. Similarly, Leeflang *et al*., (1976a) noted, during their studies of the infectivity for mice of Nigerian isolates of *T. vivax*, that in Zebu cattle infections reappeared after treatment with 3.5 mg/kg diminazene aceturate. Rottcher and Schillinger (1985) also claimed that, in the coast province of Kenya, *T. vivax* isolates which caused haemorrhagic disease were resistant to diminazene aceturate (at 3.5 mg/kg). Rowlands *et al*., (1993) in Ghibe, also reported the same result. Unlike Codjia *et al*., (1993) who reported a 100% relapse, no relapses were recorded following treatment with 7 mg/kg dimenazene aceturate. Kupper and Wolters(1983) in northern Ivory Coast and Rottcher and Schillinger (1985) in the coast province of Kenya was also reported similar results. *T. vivax* expecially appear not to develop persistently drug resistant forms (Stephen 1986).

During the drug sensitivity trial, relapse was detected after treatment with isomethamidum chloride at 0.5 mg/kg. Similarly Peregrine *et al*., (1987) in Kenya also found 15 steers treated with 0.5 mg/kg isomethamidum chloride were all cured. Another work conducted with two clones of *T. congolense* (from Uganda and Tanzania) showed them to be sensitive to the therapeutic activity of isomethamidum chloride at 0.5 mg/kg. In contrary to this studies carried out in northern Ivory Coast on cattle trekked from Mali and Burkina Faso showed a heavy preponderance of *T. vivax* infections (Kupper and Wolters, 1983) resistance to doses of 0.5 – 1 mg/kg isomethamidum chloride was demonstrated and the resistant infections also expressed resistance to homidium. In addition to this Codjia *et al*, (1993) conducted drug sensitivity trial on 12 Ghibe isolates by inoclutating Boran calves and also found out that 11 of them relapsed after treatment with isomethamidum chloride at 0.5 mg/kg b.wt.

Holmes and Jenning (1976) showed that haematological parameter recovers rapidly after treatment with any trypanocidal drug. This was confirmed in the present study in groups of animals, which were treated, with either of the

trypanocidals. But PCV improvements in groups I and III are lower than in group II and IV. This can be due to relatively lower dosages given, the relapse occurred and/or re-emerging of trypanosomes from sites which are inaccessible to the trypanocides. The improvement in PCV readings after treatment may be due to elimination of the sensitive population of trypanosomes from the animal body. However, the overall mean PCV is below the physiological value of 33% (Aiello, 1998). This may be due to the presence of drug resistant populations, re-emerging of trypanosomes from drug inaccessible sites and/or other blood sucking helminthiasis like haemonchosis, bunostomiasis and oesophagostomiasis, protozoal disease like babesiosis, anaplamosis and coccidiosis; and/or reduced response of the bone marrow due to exhaustion when the infection runs a chronic course (Murry *et al.*, 1977). In the present study there is no significant difference in mean PCV improvement in the five experimental groups before and after treatment ($p>0.05$). This finding supports the fact that *T. Congolense* is generally more virulent than *T. vivax* and consequently cattle develop tolerance to *T. Vivax* more readily and easily than to *T. congolense* (Stephen, 1986).

The use of trypanocidal drugs improved the growth rates of animals in groups II and IV for the whole experimental period by 5.5 and 13.8% respectively. Group I and group III animals had growth improvement of - 6.25% and - 4.2% respectively. The non – treated group V animals had growth rate of - 6.75% for the entire experimental period. This emphasizing the stunting effect of trypanosomosis. The growth rates might have been better if the experiment had been conducted during the rainy season instead of during the dry season when the grazing was poor. Body weight improvement in group II and IV could be due to the double dose of each trypanocidals that the animals received. The loss of weight in-group I and III may be attributed to the number of relapses experienced to dimenazene aceturate (3.5 mg/kg b.wt) and isomethamidum chloride (0.25 mg/kg b.wt) respectively. The mean body weight gain of animals in-group IV was higher than the rest of the groups. This may be due to the relative efficacy of isomethamidum chloride at higher dose (0.5 mg/kg b.wt) to cure infection in the study area. Isomethamidum chloride and diminazene aceturate are prescribed in general as "a senative pair" in the control of bovine trypanosomosis (Whiteside 1960). The present work also confirms the same. Since experimental goats were kept in non – tsetse area or in non-biting flies area the risk of reinfection during the study was eliminated. So the relapse is directly owing to the trypanosome population, which was already present in animals before treatment.

The present work disclosed the occurrence of *T. vivax* resistance population due to indiscriminate and frequent use of dimenazene aceturate and isomethamidum chloride in the Tselemti Woreda. There is, therefore, urgent need for detailed experimental work in the field as well as under laboratory conditions to monitor the development of drug resistant pathogenic trypanosome and its impact on livestock productivity in Tselemti Woreda in particular and across trypanosomosis prevalent zones of Ethiopia in general. Furthermore, by taking more number of isolates from different areas of the wereda, drug sensitivity trial should be conducted to know the exact level of drug resistance in the area.

Conclusion and Recommendations

It was concluded that where there are indications for drug resistance in the study area, because it is very unlikely that new trypanodical drugs will be released on to the market in the near future, it is essential to try to maintain the efficacy of the currently available drugs. The most important and most efficient measure is to adopt an integrated disease management strategy which includes control of the agent and vector. Furthermore, better data (instead of case reports) are required on both the true prevalence of trypanocides resistance and its impact on the production of livestock.

Finally, some measures, which may be adopted, to delay the development of drug resistance and to control drug resistance when it occurs are recommended.

- The great potential of livestock to rural farmers, in Northwest Tigray, Northern Ethiopia, can only be exploited if trypanosomosis and the arising appearance of drug resistance are controlled. Therefore, more attention should be given to adopting an integrated disease management strategy involving the vector as well as the parasite. Such strategies should be economically feasible, socially acceptable and sustainable and environmentally sound.
- Legislative reinforcement by way of elaborating a national drug use policy is required to address the indiscriminate drug usage around the study area.
- Further study should be conducted in the Tselemti Woreda in particular and across the tsetse and biting flies infested area of zones of Ethiopia in general to further elaborate the problem of drug resistance.
- Training the livestock owners on the situation of trypanosomosis in the area, the characteristics and consequence of the occurrence of drug resistance

strains of trypanosome as well as on the factors that lead to drug resistance is also recommended.

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