EFFICACY OF COMMONLY USED ANTHELMINTICS IN SELECTED SHEEP FARMS IN ARUSHA

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ABSTRACT

A study was conducted to determine the efficacy of albendazole, levamizole and oxyclozanide and ivermectin anthelmintics in selected sheep farms in Arusha, northern part of Tanzania. Eight farms under study were screened and in each farm a total of 100 sheep were involved. The sheep were allocated in groups of 25 animals and each group received different anthelmintic. We sampled pre- and post-treatment faecal samples for faecal egg count reduction test. In addition, we incubated the samples for identification of different helminthes. Resistance to anthelmintics was assumed to be present if egg count percent reduction was less than 95% and the lower limit of the 95% confidence interval for percentage reduction was equal to or less than 90%. Our results show that all the farms studied had developed resistance to albendazole, levamisole and oxyclozanide and ivermectin with the egg count reduction ranging from 67-87%; 69-87% and 74-90% respectively. The most predominant nematode species in the resistant population were *Haemonchus* and *Trichostrongylus*. It was concluded that albendazole, levamizole and oxyclozanide and ivermectin anthelmintics are not effective against some nematode strains of sheep in the studied farms.

INTRODUCTION

Livestock production is an essential industry in many parts of the world as it provides nutritional and economic basis for local or even national welfare. In Tanzania livestock production is constrained by several factors, which hinder its optimal productivity. As a result, the contribution of livestock industry to both, nutritional and economy is below the optimal potential level. The major constraints include diseases, poor nutrition, poor breeding policies and poor management (Mpelumbe, 1984; Mtenga *et al.*, 1986; Kusiluka, 1995). In sheep industry in Tanzania, about 40-60% of production losses are due to diseases and of all the diseases, helminthoses is considered to be the most important (Fison, 1987; Mtenga *et al.*, 1986).

Control of gastrointestinal nematode parasites in sheep in Tanzania has based on the use of anthelmintics for some decades. Unfortunately, routine use of these anthelmintics results in development of resistant strains of nematodes (Kelly and Hall, 1979; Waller, 1994). At the moment there are several anthelmintics with different mode of action available in the market in Tanzania further increase in the use of anthelmintics over the past 10 years. This has compromised the effectiveness of anthelmintics in sheep and other animals.

Despite the threat to the development of anthelmintic resistance, there have been only few studies to monitor the effectiveness of these anthelmintics. Even these few studies conducted have concentrated in Morogoro region and focused on resistance to some anthelmintics like thiabendazole, fenbendazole, oxfendazole and thiophanate (Ngomuo et al., 1990; Msangi *et al.*, 1990; Kassuku and Tibaijuka, 1987; Monrad *et al.*, 1987). These reports are interesting because the emergence of resistant nematodes could pose problems to both economic and practical importance to sheep enterprises. To the best of our knowledge, there are no available reports of survey on the extent of this problem in other parts of Tanzania. The purpose of this study therefore, was to determine the efficacy of commonly used anthelmintics (albendazole, levamisole and oxyclozanide and ivermectin) against naturally occurring nematode burdens in Northern Tanzania, particular, in selected sheep farms in peri urban areas of Arusha municipality.

that are widely used in the field. Their advocation and administration is mainly in the hands of paravets and other people who do not know the potential hazard of drug resistance. In addition, poor management of worm infestation has resulted into severe infections in sheep and other animals. This has led to

MATERIALS AND METHODS

Study area

The study was conducted in peri urban areas in Arusha municipality between September and October 2005. Arusha is located in northern part of Tanzania at latitude 03° south and longitude 36° east. Its altitude is 1387 m above sea level. The average annual rainfall is 888 mm spread over three months of short rains; October, November and December and three months of long rains; March, April and May. The rest of the months form the dry season. Arusha has a minimum temperature of 14.0°C in July and a maximum temperature of 25.5°C in February. This study was carried out in 800 sheep from eight randomly selected farms. Selection of the farms was based on two requirements; firstly, the farm must have a minimum of 100 sheep of three months of age and above for examination of feacal

samples; and secondly, the animals must not have been treated with any anthelmintic for at least 12 weeks prior to the study.

Samples collection and sample processing

In each farm a total of 100 sheep above three months of age were identified and ear tagged. Faecal samples were collected directly from the rectum and stored in cool box with ice parks for laboratory examination on the same day. Egg counts were done using a modified McMaster technique (Anon, 1986). Briefly, 3 g of faeces were homogenized in 42 ml of flotation fluid (saturated sodium chloride). The faecal suspension was passed through a mesh sieve to remove coarse materials. After thorough mixing, two chambers of the Universal McMaster slide were filled and all eggs under the two ruled grids (total volume 0.3 ml) counted at x40 magnification. The number of eggs obtained was multiplied by 50 to give the eggs per gram (epg) in the faecal sample.

Treatment regimen

In each farm, animals were randomly allocated into four groups of 25 animals each. Three groups were treated with three different anthelmintics according to the manufacturer's recommended doses, while the fourth groups were left as untreated controls. Three groups of animals were treated each with a single dose of albendazole (Univet, Ireland), levamizole and oxyclozanide (Interchem, Kenya) and ivermectin (Ivermectin^{R}, UK) at a dose of 5 mg/kg orally; 7.5 mg/kg orally and 0.2 mg/kg subcutaneously respectively. Ten days after treatment, the farms were visited again, all animals were sampled and the eggs were counted.

Faecal culture

From each farm both pre-treatment and post-treatment faecal samples were pooled and processed for faecal culture. Briefly, the faecal samples were ground in a mortar, mixed with sterile vermiculite to form a consistency of horse faeces. The mixture transferred into plastic cups covered with muslin cloth, and then incubated at 25°C for 7 days. The third stage larvae (L_3) were recovered by partially immersing the plastic cups down into water in conical flasks. This allowed the L_3 to swim and sediment to the bottom of the conical flasks. Consequently, the L_3 were recovered using Pasteur pipette and then transferred into clean petri dishes assessed examined under stereomicroscope before further examination under x40 magnification in light microscope. The larvae were identified and differentiated according to the Manual of Veterinary Parasitological Laboratory Techniques, MAFF (Anon, 1986).

Data analysis

The criteria used to evaluate anthelmintic resistance were based

on the W.A.A.V.P. recommendations for detecting anthelmintic resistance (Coles *et al.*, 1992). Resistance is present when the faecal egg count reduction post treatment is less than 95% and the lower limit of the 95% confidence interval for the percentage reduction is equal to or less than 90%. Resistance is suspected when only one of these two criteria is satisfied (Coles *et al.*, 1992).

RESULTS

Efficacy of anthelmintics

The results of the efficacy of anthelmintics are shown in Table 1. As shown in the Table, albendazole had faecal egg count reduction ranging from 67% - 87% and the 95% lower confidence limit ranging from 43% - 78%, levamizole and oxyclozanide had faecal egg count reduction ranging from 69% - 87% and the 95% lower confidence limit ranging from 37% - 78% and ivermectin had faecal egg count ranging from 74% - 90% and the 95% lower confidence limit of 46% - 78%. Albendazole has shown to exhibit highest resistance followed by levamizole and oxyclozanide. Ivermectin was the least. However in some farms the trend was different, which showed the same level of resistance to albendazole and levamizole and oxyclozanide.

Identification of L_3

Identification of L_3 before anthelmintics treatment revealed the presence of *Trichostrongylus, Haemonchus, Bunostomum* and *Ostertagia* species, whereas *Trichostrongylus and Haemonchus* were predominant (Table 2). *Trichostrongylus* and *Haemonchus* were encountered in all eight farms; *Ostertagia* in four farms; and *Bunostomum* in only one farm. Post treatment cultures consisted solely of *Trichostrongylus* and *Haemonchus* larvae (Table 3). Table 3 further shows that albendazole cleared *Haemonchus* in two farms, while the drug failed to clear *Trichostrongylus* in all eight farms. Levamisole and oxyclozanide was able to clear *Haemonchus* in three farms and *Trichostrongylus* in one farms whereas ivermectin cleared *Haemonchus* in six farms and *Trichostrongylus* in two farms.

Table 1. Efficacy of anthelmintics

	Njoolay			Tengeru			Solomon			Manyara		
	AL	LO	IV	AL	LO	IV	AL	LO	IV	AL	LO	IV
Reduction (%)	73	69	74	77	85	85	67	78	83	87	87	90
LowerConfidence limit (95%)	43	37	46	62	69	78	45	63	70	78	78	76

	LRC			Gomba			Ndoibo			Lamaiyan		
	AL	LO	IV	AL	LO	IV	AL	LO	IV	AL	LO	IV
Reduction (%)	73	83	86	78	87	84	68	82	78	71	75	81
LowerConfidence limit (95%)	57	69	70	57	71	70	48	66	60	50	59	68

Table 2. Identification of L_3 before treatment

	Njoolay	Tengeru	Solomon	Manyara
Trichostrongylus	39.7	55.3	45.0	49.0
Haemonchus	53.4	38.2	55.0	51.0
Bunostomum	6.9	0	0	0
Ostertagia	0	6.5	0	0

	LRC	Gomba	Ndoibo	Lamaiyan
Trichostrongylus	32.4	61	52.8	48.3
Haemonchus	64.9	39	42.9	48.3
Bunostomum	0	0	0	0
Ostertagia	2.7	0	4.3	3.4

DISCUSSION

In this study, the results of the faecal egg count reduction test indicated that albendazole, levamisole and oxyclozanide and ivermectin resistance were present in the eight farms screened. The resistance to albendazole was highest. Several other scholars have demonstrated this resistance. Keyyu *et al.*, 2002 revealed the presence of albendazole resistant *Haemonchus* in Morogoro, supporting the results obtained in 1991 by Bjorn *et al.*, (1991), Kassuku and Tibaijuka, (1987) and Ngomuo *et al.*, (1987).

Albendazole belongs to a group of benzimidazoles together with fenbendazole and thiophanate that are as well, commonly used. Since benzimidazoles have the same mode of action, resistance to albendazole could result to the development of side resistance. Benzimidazole resistant nematodes of sheep have also been reported in Australia, other parts of Africa, Europe, North and South America, whenever animals are regularly

treated with anthelmintics (Waller, 1986; Maingi, 1991; Waruiru *et al.*, 1991; Eady *et al.*, 1998).

Table 3. Identification of L_3 after treatment

	Njoolay			Tengeru			Solomon			Manyara		
	AL	LO	IV	AL	LO	IV	AL	LO	IV	AL	LO	IV
Trichostrongylus	71	67	100	75	50	100	75	100	100	100	100	100
Haemonchus	29	33	0	25	50	0	25	0	0	0	0	0
Bunostomum	0	0	0	0	0	0	0	0	0	0	0	0
Ostertagia	0	0	0	0	0	0	0	0	0	0	0	0

	LRC			Gomba			Ndoibo			Lamaiyan		
	AL	LO	IV	AL	LO	IV	AL	LO	IV	AL	LO	IV
Trichostrongylus	100	100	100	60	50	0	18	50	100	33	0	0
Haemonchus	0	0	0	40	50	100	82	50	0	67	100	100
Bunostomum	0	0	0	0	0	0	0	0	0	0	0	0
Ostertagia	0	0	0	0	0	0	0	0	0	0	0	0

Key note
AL- albendazole
LO- levamosole and oxyclozanide
IV- ivermectin

We noted further in this study a high level of resistance to levamisole and oxyclozanide. This resistance was slightly lower than that of albendazole. Ngomuo *et al.*, (1990), Waruiru *et al.*, (1991) and Maingi (1991), noted an increasing trend in the incidence of resistant strains of strongylid parasites to levamisole and tetramizole in East Africa. Results on the resistance to levamizole observed in this study differ with those obtained in a study conducted in Morogoro in 1990 which revealed that levamisole was effective against the gastrointestinal nematodes of sheep and goats at Sokoine University of Agriculture (Nyangi *et al.*, 1990; Ngomuo *et al.*, 1994). At that time levamizole had not been used extensively as compared to the benzimidazoles in the control of gastrointestinal worms in small ruminants.

The present study showed that, ivermectin at its recommended dose was not effective against gastrointestinal nematodes in sheep. Our results on one hand differ from those of Ngomuo *et al.* (1987) who found that, ivermectin

was 100% effective in suppressing faecal egg output in sheep and goats at SUA in Morogoro. Similarly Barragry (1987) revealed that, ivermectin was highly effective against nematodes resistant to the benzimidazole. On the other hand, these results are in harmony with other scholars who reported resistance to ivermectin in South Africa, Brazil and in the United State (van Wyk and Malan, 1988; Giordano et al., 1988; Echevarria and Trindade, 1989; Bowman, 2003).

We conclude in this study that, anthelmintic resistance problem is more widespread and serious than previously anticipated. The magnitude of this problem in this country and other African countries should be assessed and remedial solutions found in order to save the farmer from economic losses attributed by this problem.

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