



Original Research

Status of Anthelmintic Resistance of Fenbendazole against Gastrointestinal Nematodes in Cattle of Haryana

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Abstract

The present study was conducted to detect the status of anthelmintic resistance for fenbendazole against gastrointestinal nematodes in cattle under field conditions. For this, 10 cattle from village Bhimewala, district Fatehabad and 12 cattle from village Talwandi Rana, district Hisar were selected having eggs per gram (EPG) of faeces ≥ 150 . The cattle from each district were divided into two groups of equal number i.e. T1, T2 of Fatehabad and T3, T4 of Hisar. Animals of T1 and T3 were administered fenbendazole @ 5 mg/kg b. wt., orally while T2 and T4 served as untreated control. Faecal samples were collected on 12th day after treatment from animals of all groups and individual faecal egg counts were determined by modified McMaster technique. Faecal egg count reduction percentage in both groups T1 and T3 was 100%. Identification of infective larvae from pre-treatment faecal cultures revealed the predominance of *Haemonchus* spp. followed by *Trichostrongylus* spp. and *Strongyloides* spp. while no larvae were recovered from post treatment in treated groups.

Key words: Anthelmintic Resistance, Cattle, Fenbendazole, *Haemonchus* spp.

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Introduction

India had the largest cattle inventory in the world in 2017 with 30.39% of the total world population (Beef 2 live, 2018). The cattle are predominantly reared for milk, milk products (butter, ghee etc), leather, as well as they provide a large proportion of energy required (draft purpose) in the agriculture farm sector. Parasitic diseases are widely distributed in the world but their impact varies according to the region (Njongui *et al.*, 2016). Gastrointestinal (GI) nematodes are one of the major constraints for profitable rearing of these animals especially in tropical and sub-tropical countries of the world including India. Use of anthelmintics is the only practical field applicable method to control these GI nematode infections. The widespread



excessive, frequent and indiscriminate use, suppressive dosing and misuse of anthelmintic drugs usually lead to the development of anthelmintic resistance (Wolstenholme *et al.*, 2004; Gilleard, 2006). Anthelmintic resistance against *Haemonchus* spp. in cattle has been reported from many countries in the past (Areskog *et al.*, 2014; Cotter *et al.*, 2015; Ramos *et al.*, 2016) including India (Yadav and Verma, 1997; Singh and Singh, 2018). Thus, regular monitoring of status of anthelmintic resistance is required as an integral part of worm control programme. Therefore, the present study was planned to know the status of fenbendazole resistance against GI nematodes of cattle under field conditions in selected villages of Hisar and Fatehabad districts of Haryana.

Materials and Methods

Pre-Treatment Faecal Egg Counts

Before the commencement of actual anthelmintic resistance trial, pre-treatment faecal egg count (FEC) were performed on total 142 cattle which include 35 from Lala Lajpat Rai University of Veterinary and Animal Sciences (LUVAS) Cattle Breeding Farm, Hisar; forty-seven from Agroha Gaushala, Agroha, twenty six from village Bhimewala, Fatehabad and 34 from village Talwandi Rana, Hisar, respectively, to ensure that sufficient eggs per gram (EPG) of faeces were present in the animals to warrant their inclusion in the trial. Only three cattle from LUVAS farm were found positive for strongyle eggs whereas no sample from Agroha Gaushala was positive for GI nematodes. Therefore, the animals kept in villages at Hisar and Fatehabad showing $EPG \geq 150$ were included in the present study, based on the guidelines published by the World Association for the Advancement of Veterinary Parasitology (WAAVP) (Coles *et al.*, 1992). The fresh faecal samples were collected from all animals and faecal egg count was performed by the modified McMaster technique.

Anthelmintic Resistance Test

Twenty two cattle (10 from village Bhimewala, district Fatehabad and 12 from village Talwandi Rana, district Hisar) which were naturally infected with GI nematodes and had $EPG \geq 150$ prior to treatment were used for faecal egg count reduction test (FECRT). The selected animals had not been administered any anthelmintic during the previous two months. Ten positive cattle from Fatehabad were divided into two groups of equal number (T1, T2) and twelve positive animals from Hisar were also divided into two groups of equal numbers (T3, T4). T1 and T3 were administered with Fenbendazole @ 5 mg/kg b. wt., orally and T2 and T4 served as untreated control. Faecal egg count (FEC) of each animal was ascertained on 0 day and 12th day post treatment (PT), by the modified McMaster technique to an accuracy of one egg counted representing 50 EPG. Pooled faecal cultures at $27 \pm 2^\circ\text{C}$ for 7 days were made to recover infective larvae,

L3, from each group on day 0 and 12 PT by standard protocol. The infective larvae were identified as per criteria of Keith (1953).

Results and Discussion

The FECRT is the major and most widely used *In vivo* test for resistance detection against anthelmintics and is suitable for all anthelmintic classes (Coles *et al.*, 2006). The WAAVP has established guidelines that give precise details and recommendations for the use of this detection method (Coles *et al.*, 1992; Coles *et al.*, 2006). The FECRT provides a good estimation of anthelmintic resistance with comparatively low costs and labour input (Taylor *et al.*, 2002).

Furthermore, this test allows identification of problems with the application of anthelmintic under field conditions. The simplicity and accuracy of FECRT in anthelmintic resistance survey in cattle have been reported by various workers from different states of India (Gill, 1996; Yadav and Verma, 1997; Lalchandama, 2010). Fenbendazole is one of the widely used broad spectrum benzimidazole compound effective against GI parasites. The report of low level of anthelmintic resistance to fenbendazole in cattle under small holder dairy units by *Haemonchus* spp. in village Ludas district Hisar (Haryana) made the situation worse (Singh and Singh, 2018). But in the present study the percent reduction in FEC by fenbendazole @ 5mg/ kg bwt. was 100% at Hisar and Fetehabad and 95% upper and lower confidence limits were 100% and 100%, respectively (Table 1).

Table 1: Response to fenbendazole in cattle naturally infected with gastro-intestinal nematodes

Group	No. of Cattle	Faecal Egg Count on Days(Mean \pm SE)		Percentage Reduction and Variance on 12 th day PT		Confidence Limits at 95%	
		0	12	%Reduction	Variance	Upper	Lower
T1	5	220 \pm 17.89	0.00 \pm 0.00	100	0.0037	100	100
T2	5	170 \pm 12.25	180 \pm 10.95	0	-----	-----	-----
T3	6	358.33 \pm 93.11	0.00 \pm 0.00	100	0.0037	100	100
T4	6	341.67 \pm 104.36	358.33 \pm 136.63	0	-----	-----	-----

Group T1= Treated with fenbendazole @ 5 mg/kg b. wt. orally; Group T2= Untreated control; Group T3= Treated with fenbendazole @ 5 mg/kg b. wt. orally; Group T4= Untreated control

The result based on larval culture showed that no GI nematode species survived after treatment with fenbendazole, whereas, in pre-treatment and untreated groups the predominance of *Haemonchus* spp. was observed. The other genera of nematode identified (*Trichostrongylus* spp. and *Strongyloides* spp.) were present only in smaller proportions. This finding is consistent with findings of earlier workers from different parts of the world (Tsoetsi *et al.*, 2013; Aruna *et al.*, 2016; Marskole *et al.*, 2016; Vanisri *et al.*, 2016). Previously, Yadav (1997) had also reported *Haemonchus* spp. to be most prevalent and pathogenic species among various GI nematodes which is responsible for high mortality and morbidity in India.

Thus, the present study revealed that fenbendazole is fully effective against GI nematodes of the selected village of respective districts, indicating no development of resistance in *Haemonchus* spp. of cattle under village conditions. Although benzimidazole resistance to GI nematodes in cattle had been reported by many workers worldwide (Suarez and Cristel, 2007; Cotter *et al.*, 2015), but in Bhimewala villagers, Fatehabad and Talwandi Rana village, Hisar, no resistance against the compound was observed during the present study. This can be correlated with history of deworming in these villages which revealed that fenbendazole was infrequently used only in calves below one year. Thus, anthelmintic resistance of fenbendazole against *Haemonchus* spp. in cattle at village Bhimewala, Fatehabad and village Talwandi Rana, Hisar has not developed yet.

Conclusion

Based on the results of this and previous studies (Singh and Singh, 2018), it may be concluded that choice of anthelmintic should be based on the previous history of use of drug and status of anthelmintic resistance, which may be estimated at least once in two years.

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