Cross Anthelmintic Resistance in Goats of Unorganized Sector in Haryana

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ABSTRACT

The present study was conducted to detect the status of anthelmintic resistance of fenbendazole and morantel against gastrointestinal nematodes in goats of village Badhra, district Charkhi dadri, Haryana. Forty five goats with eggs per gram of more than or equal to 150 were divided into three groups i.e. G1, G2 and G3 of 15 animals each. Group G1 and G2 were treated with fenbendazole @ 10 mg/kg b.wt. orally and morantel @ 20 mg/ kg b.wt. orally, respectively. Group G3 served as untreated control. Faecal egg count of goats was ascertained on day of treatment (0 day) and 12th day post treatment (PT) of all groups by the modified McMaster technique. Pooled faecal cultures were made to recover infective larvae on day 0 and 12 PT. Results revealed that fenbendazole (G1) and morantel (G2) reduced the faecal egg counts by 76.47% and 70.80% on 12th day PT with upper and lower confidence levels as 89.30% and 48.23% and 82.43% and 51.48%, respectively indicating moderate anthelmintic resistance against both drugs. The post-treatment coproculture showed larvae of Haemonchus contortus and Strongyloides sp. Thus, the present study revealed presence of cross anthelmintic resistance against fenbendazole and morantel in goats of unorganized sector in Charkhi Dadri district of Haryana.

Keywords: Anthelmintic resistance, Haemonchus contortus, goat

In India, small ruminants contribute in providing economic security to small, landless and marginal farmers. Parasitic diseases are important cause of production losses in small ruminants the world over. Of these, gastrointestinal (GI) nematodosis is a common parasitic infection of small ruminants in India (Yadav et al., 2009). It is caused by mixed infections of GI nematodes. Among the various GI nematodes, Haemonchus contortus is the predominant parasite found throughout the year and is considered as the most pathogenic GI nematode responsible for impaired productivity in small ruminants throughout the world (Khalafalla et al., 2011).

Anthelmintics are administered to animals even when they show non-specific clinical signs like diarrhoea or are found positive on faecal examination without estimation of intensity of infection. This has led to indiscriminate and frequent use of the drugs resulting in the emergence of drug resistance (Barton, 1980). The widespread use, incorrect dosing and increased frequency of treatment have often led to the development of resistance against anthelmintics in nematodes of sheep and goats (Meenakshisundaram et al., 2014). Efficacy of various anthelmintics must be monitored regularly so that proper selection of anthelmintic can be done otherwise there will be huge economic losses due to cost of anthelmintics, sustained parasitic load due to ineffective worm control strategies and increased selection of resistant worms. The growing importance of these anthelmintic resistant nematodes and the need for reliable information on their occurrence and spread has increased to rule out their occurrence in a particular area. However, there is no information on the prevalence of anthelmintic resistance in goats from villages of Charkhi Dadri district of Haryana. So, the present study was planned with the objective to know the status of anthelmintic resistance against GI nematodes in unorganized goat sector.

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MATERIALS AND METHODS
During September, 2018, a study was conducted at village Badhra, District Charkhi Dadri, Haryana to determine the efficacy of anthelmintics against GI nematodes of goats using faecal egg count reduction (FECR) test. Forty five goats naturally infected with GI nematodes and having eggs per gram (EPG) of faeces > 150 counts prior to treatment were used. The selected animals had not been administered any anthelmintic during the previous two months. These animals were weighed, identified, their EPG estimated and divided into three groups i.e. G1, G2 and G3 of 15 animals each. Group G1 and G2 were treated with fenbendazole (FENAZOL-150® tablets, Concept Pharmaceuticals Ltd., Animal Health Division, Mumbai) @ 10 mg/kg b.wt. orally and morantel (Banminth® Tab., Boehringer Ingelheim India Private Ltd. Mumbai) @ 20 mg/kg b.wt. orally, respectively. Group G3 served as untreated control.

Faecal egg count 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 were made at 27 ± 2°C for 7 days to recover infective larvae, from each group on day 0 and 12th day PT. The infective larvae were identified as per criteria (Keith, 1953). Faecal egg count reduction percentage and confidence intervals (95%) were determined following the method of the World Association for the Advancement of Veterinary Parasitology (WAAVP) using arithmetic mean egg counts (Coles et al., 1992). The drug was considered fully effective when it reduced the egg counts by more than 95% and lower confidence limits below 90%. The drug was considered moderately effective when it reduced the egg counts between 60% to 95% and considered severely resistant when the reduction in egg counts was between 60% along with lower confidence limits below 90%. All the recorded data was statistically analyzed by one way ANOVA test (SPSS software version 2.0).

RESULTS AND DISCUSSION
Faecal egg counts (Mean ± S.E.) on 0 and 12th day post-treatment (PT), percent reduction in faecal egg counts (FECR%), variance, upper and lower confidence limits (95%) of goats naturally infected with gastrointestinal nematodes and treated with different anthelmintics at Badhra village, Charkhi Dadri are given in table 1. The percent FECR and confidence intervals(95%) were determined following the method of WAAVP using arithmetic mean egg counts. The result of pre-treatment faecal egg counts indicated high prevalence of gastrointestinal nematodes with strongyles as the most predominant GI nematodes affecting goats. The finding is similar to that reported by other workers (Singh and Yadav, 1997; Das and Singh, 2005; Sarika, 2012; Ruchi, 2015; Chaudhari and Singh, 2003; Kumar et al., 2008; Vohra et al., 2018). The WAAVP guidelines give precise details and recommendations for the use of this detection method (Coles et al., 1992 and Coles et al., 2006). The faecal egg count reduction test provides a good estimation of anthelmintic resistance with comparatively low costs and labour input (Taylor et al., 2002 and Cabaret and Berrag, 2004). Furthermore, this test allows identifying problems with the application of the anthelmintic resistance under field conditions. The accuracy of faecal egg count reduction test in anthelmintic resistance survey in goats and sheep have been reported by various workers in India (Singwan et al., 2006; Singh and Gupta, 2009; Rialch et al., 2013; Kumar et al., 2017) and abroad (Falzon et al., 2013; Sheferaw et al., 2013; Pena-Espinoza et al., 2014). Reduction in faecal egg counts less than 95% and confidence level less than 90% indicates presence of resistant worm population (Coles et al., 1992).

Results revealed that fenbendazole @ 10 mg/kg b. wt. (Group G1) reduced the faecal egg counts by 76.47% on 12th day PT with upper and lower confidence levels as 89.3% and 48.23%, respectively indicating moderate anthelmintic resistance. Fenbendazole belongs to benzimidazole class and its resistance to GI nematodes in goats had been reported by many workers from our country (Kumar et al., 2017; Uppal et al., 1992; Singh et al., 2012; Singh et al., 2017) and as well as abroad (Howell et al., 2008 and Holm et al., 2014). The repeated exposure of the compound to nematodes inside the animal body results in development of resistance. History revealed that fenbendazole was frequently used depending upon availability and convenience of owner.

Further, morantel @ 20 mg/kg b. wt. (Group G2) caused 70.80% reduction in faecal egg counts with upper and lower confidence levels as 82.43% and 51.48%, respectively. The results indicate moderate anthelmintic resistance. The resistance of morantel against GI nematodes has also been reported by other workers (Singh and Yadav, 1997).
Cross anthelmintic resistance in goats of Haryana and Uppal et al., 1992) and (Elliott, 1987). History of use of anthelmintic by the animal owners due to government supply in veterinary hospitals results in anthelmintic resistance. The infected untreated control (G3) had significantly (P<0.05) higher faecal egg counts (3173.33 ± 556.13) than group G1 and G2 on day 12 (PT).

The coproculture of pooled faecal cultures of infective third stage larvae in different groups and untreated control on day 0 and 12 (PT) are depicted in Table 2. A total of 100 infective larvae in each group (G1, G2 and G3) were counted. The result showed different genera of GI nematodes of goats with the predominance of *H. contortus* (86-87%) followed by *Strongyloides* sp. (6-7%), *Trichostrongylus* sp. (5-6%) and only 1% *Oesophagostomum* spp. larvae in all the treated and untreated control groups on day 0. This finding is in agreement with earlier works (Das and Singh, 2005; Kumar et al., 2008; Garg et al., 2004; Chaudhri et al., 2007; Sharma et al., 2009; Kumar and Singh, 2016). Previously, Yadav, 1997 had also reported *H. contortus* to be most prevalent and pathogenic species among various GI nematodes which is responsible for high mortality and morbidity in India. After 12 days of treatment, there was predominance of *H. contortus* larvae in fenbendazole and morantel treated animals. The presence of *H. contortus* and *Strongyloides* sp. larvae was also reported by workers (Rialch et al., 2013 and Singh et al., 2012).

There are many factors due to which resistance appears including extensive use of anthelmintics (Coles, 1999; Sangster and Gill, 1999; Prichard, 1994). It has been observed that frequent usage of the same group of anthelmintic may result in the development of anthelmintic resistance (Singh and Yadav, 1997; Das and Singh, 2005; Sarika, 2012; Singh et al., 2012; Martin et al., 1982).

Table 1: Response to various anthelmintics in goats naturally infected with gastrointestinal nematodes at Badhra village, Charkhi Dadri

| Group | Anthelmintic | Dose (mg/kg) | No. of goats treated | Route of administration | Faecal egg counts on days (Mean ± S.E.) | Faecal egg counts reduction on 12th day post treatment | Confidence limits at 95% |
|-------|-------------|--------------|----------------------|------------------------|----------------------------------------|-----------------------------------------------------|-------------------------|
|       |             |              |                      |                        | 0                                      | 12 (%)                                              | Variance               |
|       |             |              |                      |                        |                                         |                                                     | Upper      |
|       |             |              |                      |                        |                                         |                                                     | Lower      |
| G1    | Fenbendazole| 10           | 15                   | Oral                   | 3526.67±678.55 746.67±255.95            | 74.47 ± 0.15                                      | 89.30       |
| G2    | Morantel    | 20           | 15                   | Oral                   | 1913.33±142.72 926.67±162.56            | 70.80 ± 0.06                                      | 82.43       |
| G3    | Control     | —            | 15                   | —                      | 2560.00±334.49 3173.33±556.13           | 0 ± —                                              | —           |

Means with same superscripts in column are not significantly different (p<0.05).

The selection pressure exerted by regular use of anthelmintic is responsible for the development of anthelmintic resistance. Another important factor which may have contributed to the development of anthelmintic resistance in goats is under dosing. Under dosing occurs when a host is administered dose that is less than the therapeutic dose recommended by the manufacturer (Smith et al., 1999). It has recommended that goats require higher dosage than sheep to achieve similar efficacy (Coles, 1997). Goats metabolize anthelmintics
much more rapidly than other livestock (Hennessy, 1994; Conder and Campell, 1995) and have lower bioavailability of drugs after oral administration than sheep. So, higher doses of anthelmintics are required for goats compared to sheep (Coles et al., 1989; Sangster et al., 1991; Hennessy et al., 1993) which is generally not followed. Dosages 1.5 (for levamisole) to 2 (benzimidazoles and avermectins) times higher than those given to sheep are now recommended for treating goats (Pomroy, 1996; Silvestre et al., 2002). Many workers had reported dosing of goat with sheep dose to be responsible for anthelmintic resistance in goats (Jabbar et al., 2006; Kumsa and Abebe, 2009; Saeed et al., 2010; Chandrawathani et al., 2013).

CONCLUSION

It may be concluded that the choice of anthelmintic in a flock should be based on the previous history of use of drug, frequency of use of drug, dose of drug and status of anthelmintic resistance. A higher doses of anthelmintics is required in goats as compared to sheep. This is the first report of cross anthelmintic resistance against fenbendazole and morantel in a goat flock from Badhra village in Charkhi Dadri district of Haryana.

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