

## A SURVEY OF ANTHELMINTIC RESISTANCE ON BELGIAN HORSE FARMS

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### ABSTRACT

A survey to determine the prevalence of anthelmintic resistance of cyathostomes was carried out on 13 horse farms distributed over five different Belgian provinces. Based on faecal egg count reduction tests, resistance to mebendazole (a benzimidazole) was demonstrated on 12 (92%) of these 13 farms. The efficacy ranged from 0% to 100%. The efficacy of pyrantel could be evaluated on only three farms, where the faecal egg count reduction varied from 83% to 96%, which suggested a reduced efficacy of this drug on one farm. The very high prevalence of mebendazole resistance in this study shows that drugs of the (pro-)benzimidazole family should not be used to control cyathostome infections. This survey also demonstrates that the efficacy of anthelmintics used for cyathostome control programmes on horse farms should be routinely evaluated.

**Keywords:** equine - small strongyles - anthelmintic resistance - benzimidazoles - pyrantel

### SAMENVATTING

Een onderzoek naar de prevalentie van anthelminticumresistentie van kleine strongylden bij het paard werd uitgevoerd op 13 bedrijven verspreid over 5 Belgische provincies. Steunend op faecale eireductietesten werd resistentie tegenover mebendazole (een benzimidazole) aangetoond op 12 (92%) van deze 13 bedrijven; de werkzaamheid van deze behandeling varieerde van 0% tot 100%. De werkzaamheid van pyrantel kon slechts op 3 bedrijven worden uitgetest, faecale eireductie lag tussen 83% en 96%, en resistentie werd vermoed op 1 bedrijf. De zeer hoge prevalentie van mebendazoleresistentie in deze studie toonde aan dat anthelmintica behorend tot de groep van de (pro-)benzimidazoles moeten uitgesloten worden voor de controle van cyathostominose. Verder wordt gewezen op het belang van het regelmatig beoordelen van de werkzaamheid van de anthelmintica gebruikt in wormcontroleprogramma's.

### INTRODUCTION

Larval cyathostominosis, the equine parasitic disease syndrome that results from the pathological changes induced by the rapid emergence of large numbers of inhibited cyathostome larvae from the mucosa of the large intestine, is being increasingly diagnosed (Herd, 1990). Because the diagnosis of this syndrome is difficult (Smets *et al.*, 1999) and the success rate of treatment unpredictable, the aim should be to keep pasture infectivity at a low level. Suppressive anthelmintic treatments of horses to reduce faecal egg output on pasture and non-chemical methods of reducing pasture infectivity such as the regular removal of faeces from pasture, are currently the most effective ways to prevent larval cyathostominosis (Abbott, 1998). The success of chemoprophylactic treatment depends largely on the efficacy of the drugs used and on determining the opti-

imum interval between treatments. This efficacy can however be affected by the development of anthelmintic resistance. The resistance of small strongyles against anthelmintics is being increasingly reported from all parts of the world, including Western Europe. The benzimidazole group of compounds is most under threat, with its prevalence reaching the dramatic proportions of over 80% of farms affected in the Netherlands and Denmark (Boersema *et al.*, 1991; Craven *et al.*, 1998). Resistance to pyrantel is also suspected (Craven *et al.*, 1998).

In Belgium, benzimidazole resistance was detected in 2 out of 7 horse farms examined in the province of Limburg, and in 2 out of 4 studs in East Flanders (Dorny *et al.*, 1988; Geerts *et al.*, 1988). The aim of the present study was to obtain more recent data on the occurrence of anthelmintic resistance against cyathostomes on Belgian horse farms.

## MATERIALS AND METHODS

The study was done between August and November 1998. A total of 15 horse farms in the provinces of Antwerp, Luxembourg, East Flanders, Flemish Brabant and West Flanders, including 302 horses of both sexes and all age classes, were used in this study. The addresses of the farms were acquired from private veterinarians. The farm selection criteria were that there should be a sufficient number of horses to enable statistically reliable efficacy testing and that the owners should be willing to collaborate. Anthelmintic resistance was not suspected on any of the farms prior to this study. The owners were questioned about the use of anthelmintics on the farms. These included stud farms, riding farms, trotter farms and jumping farms (Table 1). The number of horses on each farm varied from 13 to more than 100. Horses that had received an anthelmintic treatment in the two months preceding the study were excluded.

The anthelmintics used in the study were the commercially available oral paste formulations of Telmin™, Janssen-Cilag, Belgium (Mebendazole 20%) at a dosage of 6.6–8 mg kg<sup>-1</sup>LW and Horseminth™, Pfizer, Belgium (Pyrantel Embonate 43.9%) at a dosage of 19 mg kg<sup>-1</sup>LW). These anthelmintics were administered according to the manufacturers' recommendations. Individual horse weights were obtained from the horse owners.

During the first visit (Day 0) the horses available for the study were randomly allocated to two or three groups of at least 6 animals. Faecal samples were collected from the rectum or, if not possible, fresh faeces was collected from the floor in the stable. When three groups were formed, the animals in two of these groups were treated with either mebendazole or pyrantel and the third group was not treated (control group). When only two groups could be formed, one group was treated with mebendazole and the other group served as a control. Two weeks after the first visit, the farm was visited again (Day 14) and faecal samples were taken from the same horses.

Individual faecal samples were examined for strongyle eggs by a modified McMaster technique with a sensitivity of one egg representing 50 eggs per gram of faeces (EPG) (Thienpont *et al.*, 1979). Faecal cultures were made from pooled group samples using 10 g faeces from each animal with a positive egg count, and incubated for 10 days at 28°C, followed by collection of the third stage larvae by the Baerman technique and identification of at least 200 larvae (MAFF, 1986).

Faecal egg count reduction (FECR) was calculated according to the guidelines of the World Association for the Advancement of Veterinary Parasitology for the detection of anthelmintic resistance in sheep, i.e. less than 95% reduction in egg count and a lower 95% confidence limit (LCL) less than 90% (Coles *et al.*, 1992). If only one of these conditions was met, resistance was suspected, not confirmed. This method uses arithmetic means to calculate FECR.

## RESULTS

Of the 302 horses sampled on the first visit, only 185 (61%) were passing strongyle eggs in the faeces. Therefore, the number of animals in each treatment group had to be reduced and 2 of the 15 farms were excluded because the low number of animals with positive egg counts compromised the calculation of the FECR. The efficacy of mebendazole could be evaluated in 13 farms and that of pyrantel in only 3 farms.

The results of the FECR are given in Table 1. There was a wide variation in egg counts between farms, mean egg counts ranging from 45 to 1475 EPG. On 12 of the 13 farms evidence of mebendazole resistance was found: FECR was <95% and the lower 95% confidence limit less than 90%. On 5 farms, the efficacy was ≤ 50%. Only on one farm, a riding farm in the province of West Flanders, was mebendazole fully effective. On 2 of the 3 farms where pyrantel was tested, the efficacy was >95%, but the lower 95% confidence limit was less than 90%, and thus resistance was suspected. On the third farm (farm 4), the efficacy was only 83%.

In the faecal cultures from Day 0, larvae of cyathostominae, *Gyalocephalus* spp., *Oesophagodontus* spp., *Poteriostomum* spp. and *Strongylus vulgaris* were identified. After treatment (day 14), only larvae of small strongyles were recovered, suggesting full activity of the drugs against *S. vulgaris*.

On 8 farms only ivermectin was used for the control of strongyle infections. The other farms used drugs of different anthelmintic families, including benzimidazoles. On none of the farms was a proper anthelmintic control programme used, the treatments being administered at irregular frequency and interval.

## DISCUSSION

The finding of mebendazole resistance on 12 of the 13 farms investigated (92.3%) is in accordance with the high prevalence of benzimidazole resistance in cyathostomes in other European studies (Bauer *et al.*, 1986; Boersema *et al.*, 1991; Craven *et al.*, 1998; Fisher *et al.*, 1992; Nillson *et al.*, 1989). However, the low number of animals in each treatment group may have compromised the interpretation of the FECR %, an optimal number of animals per group being between 10 and 15 (Coles *et al.*, 1992). Therefore, the results of this survey should be interpreted with caution.

Benzimidazoles were the main anthelmintics used for treatment of strongyle infection in horses in the seventies and the eighties, and their frequent use has selected for resistant populations. In the nineties benzimidazoles were substituted on most farms by ivermectin and pyrantel. This change in preference of anthelmintics resulted from the awareness of the problem of benzimidazole resistance and from the increased efficacy of these modern drugs, which allows a longer interval between treatments. This study shows that reversion to susceptible worm populations is not

**Table 1. Evaluation of anthelmintic efficacy on Belgian horse farms: faecal egg reductions (FECR) and 95% lower confidence limits (LCL) following treatment with mebendazole and pyrantel**

Farm Type	No. of animals in		Mean FEC Control †	Mebendazole			Pyrantel		
	Control group*	Treated groups*		FECR %	LCL%	Res	FECR %	LCL %	Res
Stud	6	7	1475	59	0	R			
Stud	7	7	950	45	0	R			
Merchant	7	7	275	21	0	R			
Riding	6	6	325	92	72	R	83	0	R
Trotter	21	22	464	85	0	R			
Riding	6	6	192	0	0	R	96	53	SR
Jumping	7	7	45	67	0	R			
Riding	6	7	313	79	0	R			
Riding	14	13	297	93	66	R			
Riding	8	7	285	100	99	S			
Riding	12	12	309	0	0	R	96	81	SR
Riding	8	8	433	73	0	R			
Riding	7	8	850	50	0	R			

R: Resistance; SR: Resistance suspected; S: Susceptible

\* only animals with eggs in their faeces at the moment of treatment were used in the calculation of the efficacy

† Arithmetic mean faecal egg count of control group on Day 14

likely to occur in the short span of time that has passed since the cessation of benzimidazole use. The very high prevalence of mebendazole resistance and the common feature of side resistance within the same anthelmintic family suggest that the use of drugs of the benzimidazole family for controlling small strongyle infections in horses in Belgium should be discontinued. On 7 of the 15 farms visited in this study, benzimidazoles were among the anthelmintics used.

The efficacy of pyrantel on the three farms investigated was lower than expected. Resistance was suspected on two farms and confirmed (FECR 83%) on one farm. Reports of pyrantel resistance in horse cyathostomes are scarce. Chapman *et al.* (1996) described a case of pyrantel resistance in the US and Craven *et al.* (1998) found evidence of pyrantel resistance in 20% of Danish farms investigated. These authors suggested from their results that routine testing for pyrantel resistance should be carried out, as the prevalence is possibly much higher than previously thought.

Resistance to ivermectin or moxidectin has, to our knowledge, never been reported in horse strongyles, and therefore has not been investigated in this study. However, the high frequency of anthelmintic treatments and the shift from benzimidazole to macrocyclic lactone compounds for

the treatment of horses may select for resistance, which could lead to drug failure in the future (Sangster, 1999). In small ruminant strongyles, resistance to ivermectin is already widespread (Waller, 1997).

The suppressive anthelmintic treatment of horses on pasture is currently the main method of controlling cyathostomes. In order to reduce selection for resistance and preserve the activity of the available drugs, the frequency of the treatments should be reduced. An appropriate anthelmintic control programme that takes into account the "egg reappearance period" of the drug used (Demeulenaere *et al.*, 1997) and involves all horses grazing on the same paddock results in a dramatic reduction of pasture infection and consequently in a more effective suppression of worm infection. Other non-chemical means of control should also be encouraged. The twice-weekly manual removal of faeces from pastures is an excellent means of controlling strongyle infections in horses (Herd, 1986). While it is considered rather time consuming, it has the other positive effect that typical defecation areas do not appear, which may increase the grazing area on the paddocks (Boersema *et al.*, 1991). Finally, this survey has also demonstrated that the efficacy of anthelmintics used for cyathostome control programmes on horse farms should be routinely evaluated.

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