Anoplocephala perfoliata in Horses in Sweden: Prevalence, Infection Levels and Intestinal Lesions

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¹Department of Parasitology, National Veterinary Institute and Swedish University of Agricultural Sciences, ²Swedish Animal Health Service, Kristianstad, and ³National Food Administration, Linköping, Sweden.

> Nilsson, O., B.-L. Ljungström. J. Höglund, H. Lundquist and A. Uggla: Anoplocephala perfoliata in horses in Sweden: Prevalence, infection level and intestinal lesions. Acta vet. scand. 1995, 36, 319-328. - Distal ileum, caecum and proximal colon of 470 horses were examined for helminths during 1 year at an abattoir in central Sweden. The infection levels of the horse tapeworm Anoplocephala perfoliata, their stage of development, site of attachment and gross pathological lesions caused by the worm were recorded. Faecal samples from 395 of the horses were examined specifically for tapeworm segments and eggs in order to correlate these findings with the numbers in the alimentary canal. In total 65% of the horses were infected with A. perfoliata and the mean intensity of infection was 79 worms per infected horse with a maximum of 912. The level of infection was significantly higher in (1) 3rd and 4th than in 1st and 2nd quarter of the year; (2) older horses than in yearlings; (3) females than in males and geldings; (4) thoroughbreed and cold-blooded horses than in Swedish standardbreeds and ponies. The level of infection was unaffected by the usage of anthelminthics against nematodes. Of the horses examined 51% had 1-100 worms whereas 14% were infected with more than 100 worms. Of the tapeworm positive horses 72% had mixed infections with both adult and juvenile worms, 20% solely juveniles, and 8% solely adults. The severity of intestinal lesions exacerbated by increasing numbers of A. perfoliata. About 11% of the intestines examined had severe lesions, but there was no history of acute abdominal distress in any of the horses included in this study. Although the number of detectable eggs was significantly higher for horses heavily infected with A. perfoliata, the egg recovery among infected horses was only 35%. An additional field survey comprising 218 horses on 88 premises in central and southern parts of Sweden showed that the prevalence of A. perfoliata egg positive horses was the same as found on faecal examination during the abattoir survey.

Cestoda; equine; Scandinavia; epidemiology.

Introduction

The cestode *Anoplocephala perfoliata* has an indirect life cycle including equine definitive hosts and soil-dwelling forage mites as intermediate hosts. The adult tapeworm has a scolex equipped with 4 suckers and lappets followed by a flattened strobila with numerous thin proglottids containing the reproduc-

tive organs. Full-grown parasites generally measure 2-3 cm in length and up to 1 cm in width. In the definitive host the parasite is most often found in the terminal ileum and in the caecum. For a review, see *French & Chapman* (1992).

Although A. perfoliata was previously regarded to be relatively harmless, recent reports have focussed on its potential pathogenicity in the horse. At the site of attachment of the scolex to the intestine pathological lesions characterized by mucosal thickening, oedema and ulcerations may be observed (*Pearson et al.* 1993). Massive infections with the parasite have also been associated with ileo-caecal colic in the horse (*Owen et al.* 1989, *Proudman* & Edwards 1993).

Prevalence studies have been carried out in several countries showing that *A. perfoliata* is distributed world-wide (*Owen et al.* 1988). Although the presence of the parasite is wellknown in Nordic countries (*Eydal* 1983, *Lütken* 1991), detailed information on infection levels and its clinical significance in horses in Sweden has not been available hitherto. Because of a poor sensitivity of faecal examination methods based on flotation techniques for *A. perfoliata* eggs (*Beroza et al.* 1985, *Proudman & Edwards* 1992), accurate surveys need to be based on direct worm counts in necropsy materials.

The aims of the present study were to investigate the levels of *A. perfoliata* infections and associated intestinal lesions in horses slaughtered in Sweden, and to correlate the presence of intestinal worms with demonstrable eggs in faecal samples. In addition, results from a field survey based on analysis of faecal samples are discussed.

Materials and methods

Abattoir survey

During October 1992 to September 1993 a total of 470 horses slaughtered at the abattoir of Linköping in central Sweden (Lat. $58^{\circ}30'$, Long. 16°) were subjected to parasitological examination. For investigation of *A. perfoliata* 50 cm of distal ileum, the caecum and 50 cm of proximal colon were collected immediately after slaughter. These parts of the intestine were opened with a pair of scissors and the contents and the mucosa were scrutinized for tapeworms. Worms were collected and counted and later classified as small or large. The lengths of representative numbers in each class were measured. Gross pathological lesions in the intestinal mucosa were recorded, and selected tissue specimens were fixed in 10% neutral buffered formalin for histopathological examination. The severity of the intestinal lesions were graded 0-3 as follows 0 =no lesions observed; 1 = slight thickening and hyperaemia of the caecal mucosa; 2 =moderate thickening and hyperaemia of the caecal mucosa with scattered necrotic ulcera; 3 = severe lesions with multiple confluent necrotic ulcera mainly located to the ileocaecal iunction.

From the rectum of 395 of the horses faecal samples were taken for examination of helminth eggs. Nematode egg counts, based on 3 g of faeces, were carried out using a modified McMaster technique, which had a minimum detection level of 50 helminth eggs per gram of faeces (epg) (*Anon.* 1986). Each faecal sample was also examined specifically for tapeworm eggs, based on 30 g of faeces (see below).

Information on age, breed, sex and recent anthelminthic treatment was obtained from farm records, but particulars were missing from some individuals (Table 1). All data were entered into a specifically designed data base (FileMaker Pro; Claris, Santa Clara, USA) in a Macintosh computer (Apple Computer Inc., Cupertino, USA).

Field survey

During the period June-September 1993, faecal samples collected from 218 horses of different ages (≥ 1 year) and sexes on 88 different farms situated in the southern and central parts of Sweden, up to lake Mälaren, were analysed for *A. perfoliata* eggs as below.

	Prevalence (%)	Number of tapeworms per examined horse		
		Relative density*	Maximum	Transformed figures**
Quarter of the year				
4/92 (140) ***	70.0 a	57.7	648	8.8 ab
1/93 (111)	59.5 b	42.9	564	5.6 b
2/93 (115)	56.5 b	44.9	812	5.5 b
3/93 (104)	75.0 a	59.9	912	11.3 a
All quarters average (470)	65.3	51.5		7.4
Age				
1 year (16)	37.5 a	32.7	213	2.7 b
2-10 years (142)	66.9 a	54.2	912	8.5 a
11-20 years (194)	68.0 a	42.6	648	7.2 a
21-30 years (78)	60.3 a	56.4	812	7.0 a
Sex				
Female (217)	68.2 a	62.0	912	8.7 a
Male (27)	51.9 a	51.5	428	4.6 b
Gelding (191)	63.9 a	35.5	812	5.5 b
Breed				
Swedish Standard bred (194)	63.4 a	44.7	564	7.0 a
Thoroughbred (116)	73.3 a	60.8	812	10.8 b
Ardennes+North Swedish (20)	65.0 a	122.1	912	12.6 b
Ponies (109)	61.5 a	29.8	412	5.0 a
Others (22)	59.1 a	40.3	387	4.8 a
Treatment				
Ivermectin (110)	66.4 a	63.6	428	11.2 a
Pyrantelembonat (31)	61.3 a	66.5	912	8.5 a
Untreated (38)	52.6 a	85.9	812	7.0 a
Various drugs including benzimidazoles (260)	64.6 a	36.1	563	5.9 a

Table 1. Prevalence and infection levels of *Anoplocephala perfoliata* in Swedish horses according to time of slaughter, age, sex, breed, and recent anthelminthic treatment.

* Arithmetric mean.

** Geometric mean.

*** (No. of horses examined). Related figures with different letters differ significantly from each other.

Faecal examination for A. perfoliata eggs

Faecal samples from slaughtered horses as well as from horses in the field were assayed by a centrifugation/flotation method for the presence of tapeworm eggs as follows. Thirty g of faces was soaked for at least 1 h in 4 dl tap water, homogenized with an electric stirrer and strained through a 40 mesh sieve into a 5 dl beaker. The homogenate was left for sedimentation for at least 3 h. After discarding of the supernatant the sediment was transferred to 1 or 2 flat bottomed centrigfuge tubes, depending on the volume of the sediment, and centrifuged for 2 min at $800 \times g$. The supernatant was sifoned off and the sediment resuspended in saturated NaCl solution containing 0.2 g sucrose per litre. After centrifugation for 2 min at $800 \times g$ material from the surface was transferred to a McMaster counting chamber by a Pasteur pipette and the total number of tapeworm eggs in each chamber, i.e. in 30 g of faeces, was counted at ×40 and identified at ×100 magnification. A standard positive faecal sample with a recovery rate of 10-20 *A. perfoliata* eggs was regularly processed in order to check that the analytic procedure showed consistent results. The faecal samples from slaughtered horses were coded so that the animals' tapeworm status was unknown at the time of faecal examination.

Statistical analyses

Statistical analyses were performed using StatViewTM version 1.0 (Abacus Concepts, Berkeley, USA) for Macintosh (Apple Computers). The data were analysed by Student's t-test on transformed $[\ln(X + 1)]$ data because of non-normal distribution. Geometric means were calculated by relogging the mean of $\ln(x + 1)$ and subtracting by one. Heterogeneity between frequencies was analysed using the χ^2 test. Linear regression analyses were made to show correlation between the number of *A. perfoliata* present and 1) the number of tapeworm eggs observed, 2) the severity of lesions, and 3) the strongyle epg.

Definitions

Definitions used were according to *Margolis* et al. (1982). Prevalence is the number of horses infected divided by the number of individuals examined. Relative density refers to the total number of *A. perfoliata* recorded in all horses divided by the number of horses examined, whereas mean intensity is per infected host. All of these terms describe the infection level. Site refer to the part of the host in which the parasite was found.

Results

Infection levels of A. perfoliata

Anoplocephala perfoliata was found in 65.3% of the horses. The maximum number of tapeworms recorded was 912, the relative density being about 51.5 and mean intensity 78.8. A higher prevalence (p<0.010) and geometric mean number of worms (p<0.025) were found in guarters 3 and 4 of the year than in guarters 1 and 2 (Table 1). There was no significant difference in prevalence between the age of the horses investigated (1-30 years), but the relative density was on average lower in yearlings than in older animals (p<0.050). The geometric mean number of worms was significantly higher in females than in males and geldings (p <0.050). Thoroughbred and cold-blooded horses had on average more tapeworms than Swedish standardbreds and ponies (p<0.001). There was no difference in infection levels between untreated horses and horses that had been regularly treated with different types of anthelmintics during the last year or longer.

Distribution and site of worms

The frequency distribution of tapeworms within the host population in class intervals of 100 was skewed, $s^2/x = 26$ (Fig. 1). Up to 100 worms were recorded in 239 (51%) of the horses examined and of these 106 (45%) harboured 1-10 worms, whereas 68 (14%) were infected with more than 100 worms. Caecum was the preferred site, and clusters of worms were frequently attached to the mucosa. The ileocaecal valve was the preferred site of large clusters. If the intestine was opened later than 20 min after slaughter, most worms were detached and found free in the caecal contents.

Length structure of A. perfoliata

Measurements of 20 worms in the small and large classes gave the following average lengths: small worms 6.8 ± 3.6 mm, large

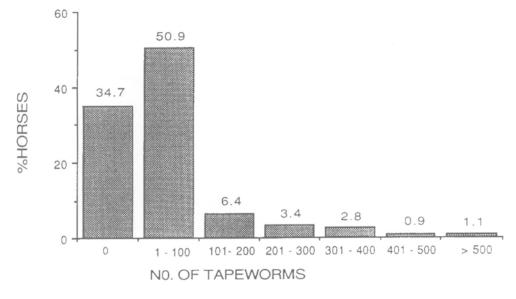


Figure 1. Frequency distribution of the numbers of *Anoplocephala perfoliata* from 470 Swedish horses, 1-30 years of age.

worms 32.6 ± 6.4 mm (Fig. 2). Most large worms had gravid segments whereas all the small worms were sexually immature. A high proportion of the small worms was juveniles of the lancet type, but triangular shaped worms were also recorded regularly. The morphology of the latter type indicated that they were adults with discharged distal segments.

The majority of the horses, 72%, harboured both adult and juvenile worms, the prevalence of infections with large and small worms only being higher in quarters 3 and 4 than in quarters 1 and 2 (p<0.05). Horses solely infected with juveniles were found on average in 20% of the material, whereas solely adult worms were seen in only about 8% of the horses examined. Consequently the generations seemed to be overlapping as no clear seasonal pattern of occurrence was observed.

Eggs in faecal samples from infected horses $A = \frac{1}{2} \int \frac{1}{2$

About 65% of the examined horses were infected with A. perfoliata, but only 23% of them were also egg-positive on faecal examination. Thus the sensitivity of the test was low. The A. perfoliata egg recovery rate in 90 infected horses was 35%, the quarterly range being 26–44% (p≤0.3). No egg-positive sample was obtained from horses negative to A. perfoliata at necroscopy. There was no correlation between the number of detectable intestinal tapeworms and the number of tapeworm eggs in individual horses ($R^2 = 0.023$). The result of comparing horses with low and high worm burdens is shown in Table 2. The egg recovery rate and the number of detectable eggs were significantly higher for the high than for the low worm burden group (p<0.001). There was no correlation between

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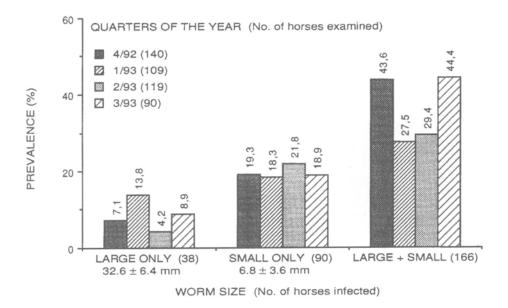


Figure 2. Prevalence of large and small Anoplocephala perfoliata in Swedish horses 1-30 years of age, according to quarter of the year at slaughter.

the strongyle epg value and occurance of adult tapeworms in individual horses ($R^2 = 0.004$).

Intestinal lesions

About 11% of the 470 horses examined had severe mucosal changes in the caecum and the terminal ileum suggestive of *A. perfoliata* lesions (Table 3). The lesions comprised mucosal thickening, hyperaemia, oedema and ulcerations. Histological examination of caecal lesions from selected animals revealed extensive inflammatory cell infiltration of eosinophils and lymphocytes surrounding ulcerative mucosal changes and diptheric membranes on the mucosal surface, consistent with the descriptions of *Pearson et al.* (1993). Nine horses with more than 100 worms had no visible or only very slight lesions. Four worm-free horses had moderate to severe lesions. There was a significant correlation between the numbers of worms and the severity of lesions (R-squared = 0.293). The average graded lesion score was 1.39 for horses having 1–100 worms (n = 239) and 2.30 for horses with more than 100 worms (n = 68). No severe intestinal lesions were seen in 12 slaughtered foals, out of which 4 were infected with *A. perfoliata*. A single necrotic lesions occurred at the ileocaecal orifice of a foal having 198 tapeworms located to the distal part of ileum.

Field survey based on faecal examinations

Tapeworm eggs were demonstrated in 58 (27%) of 218 horses originating from 88 different farms in central and southern Sweden. The difference between the observed and the expected value, based on data from the examination of slaughtered horses, was not significant (p = 0.30). The prevalence of infected

	Low worm burden (< 100 worms)	High worm burden (≥ 100 worms)	
No. of horses included	193	65 *	
% egg positive horses	21.8 a	73.8 b	
No. of eggs per 30 g of faeces			
Arithmetic mean	2.4	17.7	
Maximum	86	183	
Geometric mean	0.5 a	5.9 b	

Table 2. The occurrence of tapeworm eggs in faecal samples from horses with low and high worm burdens.

Related figures with different letters differ significantly from each other.

* 4 extreme values (100, 150, 150 and 800 EPG) are excluded in the quantitative calculations.

Table 3. The presence of intestinal lesions in 470 1 to 30 – year – old Swedish horses according to numbers of *Anoplocephala perfoliata*.

Lesions Grade number	No 0	Slight 1	Moderate 2	Severe 3
No. of horses	116	205	98	51
% horses	24.7	43.6	20.9	10.9
Numbers of worms				
Arithmetic mean	7.1	21.8	67.1	239.9
Maximum	327	348	381	912
Geometric mean	0.4a	4.8b	34.5c	133.2d

Figures with different letters differ significantly from each other.

farms was not calculated because too few animals per farm were examined. However, at least 47 (53%) of the farms accommodated egg-positive horses. According to farm records there had been no clinical signs related to the alimentary tract in any of the herds for a long time.

Discussion

The present investigation showed that about 65% of 470 horses slaughtered in Sweden were infected with *A. perfoliata*. The maximum number of tapeworms observed in a single horse was 912, whereas the relative density was about 52, and the corresponding mean intensity was about 79. Although relatively few European prevalence studies on slaughtered

or necropsied horses have been published, the present infection levels correspond with what has been demonstrated in other parts of the world, i.e. 69% in England (*Owen et al.* 1988), 51% in Ireland (*Fogarty et al.* 1994), 60% in Kentucky (*Lyons et al.* 1987), 82% in New Zealand (*Bain & Kelly* 1977) and 52% in Japan (*Yoshihara et al.* 1993).

The infection levels of *A. perfoliata* in the present study were of equal magnitude irrespective of age and sex, as also observed in other investigations (*Lyons et al.* 1983, *Fogarty et al.* 1994). This indicates that neither acquired protective immunity nor age resistance to *A. perfoliata* seem to be of any importance in horses. Furthermore, there was no evidence of interspecies competition between

sexually mature strongyles and *A. perfoliata* as the numbers of nematode eggs in the faeces and tapeworms in the intestine were uncorrelated. The level of infection appeared also to be unaffected by the usage of anthelmintics against nematodes, as suggested by *Torbert et al.* (1986). Consequently it does not seem like the increased usage of ivermectin, which has no documented cestocidal effect, is a probable explanation to a possibly increased number of horses with colic and associated heavy tapeworm infection, which has been proposed (*Edwards* 1986, *Owen et al.* 1989, *Geering & Johnson* 1990).

Juvenile worms were found throughout the year. This implies that the infective stage may be picked up indoors or that the turnover of tapeworms is slow and the prepatent period of *A. perfoliata* is probably much longer than 6-8 weeks as generally accepted (*French & Chapman* 1992). In 1 study it was shown that it may take up to 12 months for the cysticercoid to develop into the adult worm (*Sanda & Tsukada* 1985).

Every faecal sample included in this study was examined grossly for the presence of tapeworm segments, with negative result. Four samples contained extremely high numbers of A. perfoliata eggs, up to 800 epg. In a few instances cestode eggs were surprisingly recovered from 3 but not 30 grams of the same faecal batches. Taken together it can be concluded that the discharge of gravid tapeworm segments is sporadic, that their integrity is lost before shed with the faeces, and that the eggs are not equally distributed in the faecal mass. Consequently, there is usually a low probability that eggs are found in infected horses, and thus current faecal examination techniques are clearly inaccurate for a definitive diagnosis of tapeworm infection in individual animals. Moreover, there was no individual correlation between the number of intestinal

tapeworms and the number of eggs, which supports previous findings (*Proudman & Edwards* 1992). However, the likelihood of finding eggs is significantly higher in horses with high (>100 worms) than low worm burdens. Therefore faecal examinations may be acceptable for diagnosis and epidemiological studies on a herd level.

There was a significant relationship between the numbers of intestinal tapeworms and the severity of associated lesions. In the caecum and at the ileocaecal junction, about 11% of the horses in the abattoir study had severe necrotic ulcerations and granulomatous inflammations as typically seen in A. perfoliata infection (Pearson et al. 1993). Nevertheless, there was no history of acute abdominal distress in any of the animals studied. Therefore the finding of intestinal tapeworms during necropsy or of tapeworm eggs in faecal samples is not necessarily incriminating evidence of a causal relationship with intestinal disorders observed, which has been suggested (Lyons et al. 1983, Beroza et al. 1986, Owen et al. 1989). Although the relationship between tapeworm infection and intestinal dysfunction in the horse is at present unclear, it seems unacceptable from an animal welfare point of view that heavily infected animals are left untreated. Consequently, specific therapeutic treatment and a specific control programme against A. perfoliata would be advocated. Therefore, it is an urgent need to develop more specific and sensitive diagnostic methods in order to make future clinical and epidemiological work on A. perfoliata successful. For this purpose, serological diagnosis may be an alternative (Höglund et al. In press).

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Sammanfattning

Förekomst av Anoplocephala perfoliata och associerade tarmskador hos svenska hästar.

Distala ileum, caecum och proximala colon hos 470 hästar slaktade i Linköping under tiden oktober 1992 - september 1993, undersöktes på förekomst av Anoplocephala perfoliata. Maskarnas antal, storlek och lokalisation liksom utseende och utbredning av bandmaskskador i tarmen registrerades. Träckprov från 395 av hästarna undersöktes speciellt avseende förekomsten av bandmaskägg för att utvärdera det diagnostiska värdet av koprologiska undersökningar. Av de undersökta hästarna var 65% infekterade med A. perfoliata. I genomsnitt påvisades 79 maskar hos infekterade hästar och som mest 912. Av de infekterade hästarna hade 22% mer än 100 maskar. Den genomsnittliga infektionsnivån var högre under tredje och fjärde än under första och andra kvartalet. Föl och åringar var angripna i mindre omfattning än äldre hästar. Ston var infekterade i något högre grad än hingstar och vallacker. Fullblods- och kallblodshästar var infekterade i högre grad än halvblodshästar och ponnyhästar. Infektionsnivån var inte påverkad av tidigare behandlingar med anthelmintika mot nematoder. Både adulta och juvenila maskar påträffades oberoende av årstid hos 72% av de infekterade hästarna, medan 20% hade endast juvenila och 8% endast adulta. Bandmaskägg påvisades endast hos 35% av de infekterade hästarna, trots att varje prov utgjordes av 30 g träck, dvs 10 gånger mer än vid koprologisk rutinundersökning. Det fanns inget samband mellan antalet påvisade ägg och antalet maskar i tarmen. Koprologisk undersökning kan således inte ligga till grund för säkerställande av diagnosen hos enskilt djur men väl för besättningsutredningar och epidemiologiska studier. Cirka 11% av hästarna hade relativt omfattande bandmaskskador i tarmen. Skadornas omfattning ökade med ökat antal maskar. Trots att 51 hästar med i genomsnitt 240 maskar ingick i studien förelåg ingen klinisk rapport om tarmstörningar. Parallellt med slaktstudien utfördes en inventering med hjälp av träckprover avseende förekomst av bandmask i besättningar i Syd- och Mellansverige. Undersökningen kompletterades med material från Skellefteå slakteri. Det visades att *A. perfoliata* är vanligt förekommande i hela Sverige.

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