Anoplocephala sp. (Cestoda, Cyclophyllidea) infection in horses in Central Italy

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Summary
Species of Anoplocephalidae are cestodes affecting equines worldwide and causing many intestinal disorders. Between October 2010 and September 2013, 284 faecal samples of horses originating from Lazio Region (Central Italy) were tested for the presence of *Anoplocephala* sp. eggs by a classic copromicroscopic technique. Data regarding pasture-dependence/non dependence, age, and sex of the horses were also collected. Statistical analysis, aimed at detecting possible associations between these variables and *Anoplocephala* sp. prevalence, was performed. *Anoplocephala* sp. eggs were found in 13.0% of tested horses. Higher values of prevalence were recorded in females (15.2%), younger animals (< 6 months old) (15.4%), and animals dependent on pasture for their diet (18.58%), the latter being the only statistically significant variable. Prevalence recorded in the present study is comparable to those reported in previous surveys based on coprological methods.

Anoplocephala sp. (Cestoda, Cyclophyllidea) in cavalli dell'Italia centrale

Parole chiave
Anoplocephala sp.,
Cavallo,
Cestode,
Italia,
Parassita.

Riassunto
I cestodi del genere *Anoplocephala* sono parassiti ubiquitari degli equini, nei quali possono provocare disturbi intestinali di varia gravità. Nel periodo compreso tra ottobre 2010 e settembre 2013, 284 campioni fecali di cavalli provenienti dal Lazio sono stati testati per la presenza di uova di *Anoplocephala* sp. utilizzando la tecnica copro-parassitologica classica. È stata effettuata un'analisi statistica per valutare l'eventuale associazione tra prevalenza di *Anoplocephala* sp. e accesso al pascolo, età e sesso degli animali. La prevalenza totale è risultata del 13,0%, significativamente più alta (18,6%) negli animali che avevano accesso al pascolo per la loro alimentazione. Femmine (15,2%) e animali giovani (15,4%) sono risultati i più infestati, sebbene la differenza riscontrata non sia statisticamente significativa.

Anoplocephala perfoliata (Cestoda, Cyclophyllidea) is the commonest cestode affecting equines worldwide (Gasser *et al.* 2005, Matthews *et al.* 2004). It is considered responsible for many intestinal disorders (Bohórquez *et al.* 2012, Meana *et al.* 1998, Scala and Cancedda 1996), sometimes leading to fatalities. In horses, chronic infections with high worm burdens have been associated with enteritis, colics, intussusceptions, ileal impactions, and intestinal perforations and obstructions (Gasser *et al.* 2005, Traversa *et al.* 2008, Trots-Williams *et al.* 2008). Epidemiological data about tapeworms distribution in equine populations report prevalence ranging from 20% to 80% (Abbott and Barret 2008). According to some authors, *A. perfoliata* prevalence increased in the last decades due to the growing use of anthelmintics ineffective for tapeworms (Gasser *et al.* 2005), facilitating higher tapeworm burdens for the reduced competition with other gastrointestinal parasites (*i.e.,* strongylids, *Parascaris equorum*, etc.) affected by treatments (Meana *et al.* 1998).

Validity and comparability of prevalence data regarding equine tapeworms are negatively conditioned, on the one hand, by the low sensitivity of copromicroscopic diagnostic techniques and, on the other, by the many different kinds of diagnostic approaches (such as necropsy, copromicroscopic, serological, molecular) used in the different studies (Traversa *et al.* 2008). Copromicroscopic detection

of horse cestodes is still an open problem (Rehbein *et al.* 2011). Different techniques for the detection of tapeworm eggs in horse faeces have been proposed over the years, none of them reaching a satisfying sensitivity, especially in case of low worm burdens (less than 100 worms/host) (Rehbein *et al.* 2011). To date, the more sensitive copromicroscopic technique for the diagnosis of cestodosis in equines is the centrifugation/flotation technique developed by Proudman and Edwards (Proudman and Edwards 1992). According to the authors, this technique reaches a 61% sensitivity, increasing to 92% in horses harbouring more than 20 *A. perfoliata* specimens (Gasser *et al.* 2005, Proudman and Edwards 1992).

At present, in Italy epidemiological data about the distribution of these parasites are quite scanty, mainly because of the inability of the common flotation techniques to detect tapeworm eggs in faeces (Meana *et al.* 2005). In addition, despite the, by now, well-known clinical relevance of these cestodes (in particular *A. perfoliata*) (Meana *et al.* 2005), private practitioners are rarely aware of the importance of detecting these parasites. Two studies conducted in Sardinia (Italy) reported prevalence data ranging from 8 to 16% (Scala and Cancedda M. 1996, Scala *et al.* 2001), while in Umbria Region (Central Italy) a 25.6% prevalence was recorded (Veronesi *et al.* 2008). More recently (Campigli *et al.* 2009), a study carried out in Tuscany on 118 horse faecal samples reported an overall prevalence of 20.5%. The situation being so, it was considered relevant to carry out a survey about *Anoplocephala* sp. in horses in Lazio Region (Central Italy), in order to have a wider picture of the presence and distribution of these parasites throughout Italy.

Between October 2010 and September 2013, 284 faecal samples of horses originating from Lazio Region (Central Italy) were tested for the presence of *Anoplocephala* sp. eggs by means of the centrifugation/flotation technique described by Proudman and Edwards (Proudman and Edwards 1992). Data regarding pasture-dependence/non dependence for a significant part of the diet (considered the major risk factor for *Anoplocephala* sp. infection), age (categorised into 3 classes: 1, < 6 months; 2, between 6 months and 5 years, and 3, > 5 years), and sex (male, female or gelding) were available for 247, 140, and 181 horses, respectively.

Chi square and the Fisher exact tests were used to analyse the statistical significance of possible associations. Epi Info (Epi Info™ 3.3.2, 2005) software was also used to determine the Odds ratios (OR) of the associations between the recorded variables and prevalence of *Anoplocephala* sp. in horses. Level of significance was set at $p = 0.05$.

Anoplocephala sp. eggs were found in 13.0% of tested horses. Higher values of prevalence were recorded in females (15.2%), younger animals (< 6 months) (15.4%), and animals dependent on pasture for their diet (18.6%) (Table I). Prevalence differences were statistically significant only in the association with the variable pasture dependence/non-dependence ($p < 0.005$, OR = 14.38).

Anoplocephala sp. prevalence reported in literature ranges from 0 to 100%, depending on the study area and the diagnostic approach (Abbott and Barret 2008). Prevalence recorded in the present study is similar or higher than those reported in previous works based on copromicroscopic detection of eggs in faeces (Trots-Williams *et al.* 2008). Moreover, prevalence values are quite variable also depending upon the geographic area of study, with highest prevalence reported in Northern Europe (Meana *et al.* 2005) and values similar to or lower than those recorded in this study reported in Mediterranean areas, such as Sardinia (Italy) and Greece (14% and 0.4%, respectively) (Scala and Cancedda 1996, Sotiraki *et al.* 1997).

As expected and as reported in previous studies (Gasser *et al.* 2005, Kornas *et al.* 2010, Trots-Williams *et al.* 2008), *Anoplocephala* sp. was more prevalent in horses depending on pasture for a significant part of their diet, this being the only risk factor significantly and positively related to the prevalence of infection. Even though not statistically significant ($p = 0.65$), the prevalence recorded in foals of less than 6 months (15.4%) was most interesting, resulting higher than those recorded in older horses (9.1% and 14.3% for class 2 and class 3 horses, respectively; Table I). In fact, previous studies reported horses over 1 and 4 years of age

Table I. *Prevalence (%) of* Anoplocephala *sp. detected in 284 horse faecal samples between October 2010 and September 2013, in Central Italy.* Data are presented in relation to sex, age class (1: < 6 months; 2: between 6 months and 5 years; 3: > 5 years), and dependence/non-dependence on pasture. Different letters denote a $p<0.05$. In brackets the number of samples for each category and the confidence interval for each prevalence value.

	Sex			**Age class**			**Pasture dependence**	
	Females (118)	Males (48)	Geldings (15)	1 (26)	2 (44)	3 (70)	Yes (183)	No (64)
Prevalence (%) (95% CI)	15.2 (8.8-21.7)	8.3 (0.5-16.1)	6.7 (0.0-19.3)	15.4 (1.5-29.2)	9.1 (0.6-17.6)	14.3 (6.1-22.5)	18.6[a] (12.9-24.2)	1. 6[b] (0.0-4.6)

to be at higher risk of infection (Campigli *et al.* 2009, Lyons *et al.* 2000). All the 4 positive foals of less than 6 months of age were kept on pasture with mares since their birth. As the pre-patent period of *A. perfoliata* is reported to be 6-10 weeks (Gasser *et al.* 2005), we concluded that these foals acquired infection when 14-18 week old at most.

Prevalence recorded in this study (13.0% in horses) is not negligible, especially because the real prevalence of infection must be expected to have been underestimated, given the low sensitivity of copromicroscopic methods in detecting equine tapeworms eggs in faeces (Trots-Williams *et al.* 2008). Thus, the results of this study stress the importance of including this parasite in control programs aimed at improving equine health and welfare (Campigli *et al.* 2009) and at making aware private practitioners of this poorly known and scarcely considered parasite, which, in some cases, can be considered responsible for serious health problems in horses.

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