Short communication

Gastrointestinal parasites in greater rheas (*Rhea americana*) and lesser rheas (*Rhea pennata*) from Argentina

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ABSTRACT

Few data exist on the parasites of ratites, especially from regions within their natural range. It is only recently that extensive studies on the parasites of ostriches (*Struthio camelus*) have been published, mainly from European countries where commercial farming has expanded. Two species of ratites are native in South America: the lesser rhea also known as Darwin's rhea (*Rhea pennata*) and the greater rhea (*Rhea americana*). Both species are considered near threatened by the IUCN and are included in the CITES' Appendices I and II, respectively. Parasitological studies have conservation implications, as they allow us to assess the risk of transmission of pathogens from farmed ratites to wild populations. In this study 92 faecal samples from greater rheas and 55 faecal samples from lesser rheas from different localities in Argentina were analyzed to determine their gastrointestinal parasites. In greater rheas the protozoa (*Balantidium coli*-like and *Entamoeba* spp.) and helminths (*Fasciola hepatica* and *Deletrocephalus* spp.). The protozoa had not previously been cited as parasites of greater rheas in South America. Cysts and/or trophozoites of *B. coli*-like were found in 16.3% of the samples, while the prevalence of the remaining parasites was below 10%. Lesser rheas harbored the protozoa *B. coli*-like, *Entamoeba* spp. and *Chilomastix* spp. as well as *F. hepatica* and nematode eggs and larvae. *B. coli*-like cysts were found in 20.0% of the samples, while the prevalence of the other parasites remained below 5%. Some of them had not been cited as infecting lesser rheas yet.

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1. Introduction

The geographic range of the greater rhea (*Rhea americana*) extends from Eastern Brazil to the Río Negro river in central Argentina, whereas the lesser rhea (*Rhea pennata* = *Pterocnemia pennata* following SACC, 2012) inhabits the plains of the Patagonia region in the south of Argentina and Chile, as well as the Andean regions of these two countries and Bolivia and Peru (BirdLife International, 2012a,b).

Both species are considered near threatened in the red list of the IUCN (BirdLife International, 2012a,b) and are included in the CITES' Appendices I and II.

Parasitological studies have conservation implications for threatened and endangered species (Pedersen et al., 2007). With regard to rheas, the Argentinean Rhea Project (Martella and Navarro, 2006) considers the reintroduction of farmed animals in the wild. Thus, the identification of parasites that might affect rheas would allow to assess the risk of transmission of pathogens from farmed birds to wild populations. Few works on the endoparasites of greater rheas have been published in the last 20 years. Several protozoa have been described from rheas

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in Europe (*Cryptosporidium* sp., *Eimeria* sp., *Balantidium* sp., *Entamoeba* sp., *Endolimax* sp., *Trichomonas* sp., *Giardia* sp. and *Pleuromonas* sp.; Ponce-Gordo et al., 2002) and several helminths have been described mostly from South American birds: the nematodes *Deletrocephalus dimidiatus*, *Deletrocephalus cesarpintoi*, *Paradeletrocephalus minor* (Strongyloidea), *Trichostrongylus colubriformis* (Trichostrongyloidea), *Odontospirura* (=Vaznema) *zschokkei*, *Sicarius uncinipenis*, *Torquatooides crotophaga* (Habronematoida), *Dicheilonema rhea* (Diplostriaenoidea) and *Capillaria venteli* (Trichinelloidea), the cestodes *Houttuynia struthionis*, *Chapmania tauricolis* (Davaineidae) and *Monococcestus* sp. (Anoplocephalidae) and the trematodes *Philophthalmus aweerensis* (Philophthalmidae) and *Fasciola hepatica* (Fasciolidae) (Giossa et al., 2004; Zettermann et al., 2005; Comolli et al., 2006; Schuster, 2011). References to parasitism in lesser rheas are even more scant and only three nematodes (*D. dimidiatus*, *Trichostrongylus* sp. and *Capillaria* sp.) and one protozoon (*Eimeria* sp.) have been recorded (Ewing et al., 1995; Chang Reissig et al., 2001). The aim of the present study was to contribute to the knowledge on parasites of ratites by describing the gastrointestinal parasites of farmed and wild greater and lesser rheas in Argentina.

2. Materials and methods

Faecal samples of captive-bred and wild birds were collected during 2007–2009. Captive birds were raised in farms or zoos whose typology varied depending on the rearing scheme adopted (intensive or semi intensive). In general, the intensive model (smaller paddocks with a greater density of birds and a controlled diet) is implemented by most zoos and farms, and the semi intensive model (larger facilities where birds graze freely in the pasture and receive a comparative smaller quantity of controlled food) is implemented in few farms (Navarro and Martella, 2002). The wild greater rheas were in areas with pastures and grasslands; cattle, horses, sheep and hares (*Lepus* sp.) were also present in the area. Wild lesser rheas were located in areas of medium to low shrub steppes; co-occurring with hares, sheep and guanacos (*Lama guanicoe*). The location and characteristics of the sampling sites are given in Supplementary Files 1 and 2.

Supplementary data associated with this article can be found, in the online version, at <http://dx.doi.org/10.1016/j.vetpar.2012.12.021>.

Fresh faeces (up to 12 h old) were randomly collected from the soil avoiding contamination with soil debris. All birds within each farm/wild group appeared healthy at the time the samples were collected, except in one farm raising lesser rheas (Supplementary File 1, site code 24; faeces were greenish and two birds died the week before the sampling). Samples were fixed in 70% ethanol and sent to Spain for further processing. Faecal concentrates were made following the formaldehyde–ether concentration technique (Levine and Estevez, 1983) and the sediments were examined on temporary slides stained with Lugol's iodine; when necessary, permanent slides stained with trichromic stain were made.

3. Results and discussion

Protozoal and helminth parasites were present in both species of rheas; their morphological characteristics and prevalences are given in Supplementary Files 3–5.

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3.1. Greater rhea

The most frequent parasite found was *Balantidium*, which had previously been cited in greater rheas bred in Spain (Ponce-Gordo et al., 2002). The taxonomic status of *Balantidium* from ratites is not clear and it is possible that the species *B. struthionis* described in ostriches may be a synonym of *Balantidium coli* (Ponce-Gordo et al., 2011). Presently available data do not support considering the species from rheas as different from that of ostriches. We therefore record it as *B. coli*-like (following Ponce-Gordo et al., 2011) until further comparative analyses can be performed.

The morphology of the one nucleated cysts of *Entamoeba* spp. is compatible with that of *E. struthionis*, described in ostriches and rheas raised in Europe (Ponce-Gordo et al., 2002) and also reported in pigs and humans (Martínez-Díaz and Ponce-Gordo, 2010). However they are also morphologically compatible with the cysts of other one nucleated mature cyst-forming species of *Entamoeba*. For this reason, we prefer to identify the organism as *Entamoeba bovis*-like, using the name of the one nucleated mature cyst-forming *Entamoeba* group as proposed by Levine (1961), until further, more detailed studies could determine the species.

The trematode eggs found resemble those of *F. hepatica*, which has previously been reported from common rheas in Brazil (Soares et al., 2007). The morphology and size of the strongylid-like eggs found are compatible with *Deletrocephalus* spp. (150–160 µm × 70 µm; Vaz, 1936; Taylor et al., 2000); they are slightly smaller than the eggs of *P. minor* (190–200 µm × 60–70 µm; Acomolli et al., 2006) and larger than those of *O. zschokkei*, *Sicarius* sp., *T. crotophaga* or *Trichostrongylus* sp., which were in all cases less than 100 µm in length (Vaz, 1936; Freitas and Lent, 1947; Pence and Casto, 1976; Yong, 2012). The most frequent intestinal nematode in greater rheas is *D. dimidiatus* (Zettermann et al., 2005), which is considered of veterinary relevance due to its blood-feeding habits. In heavy infections it is responsible for the onset of an anaemia syndrome in rheas (Craig and Diamond, 1996).

3.2. Lesser rhea

The most prevalent parasite was *Balantidium* sp. The cysts and trophozoites are indistinguishable from those in greater rheas and have been identified as *B. coli*-like until further comparative analyses have been done.

Two different *Entamoeba* spp. cysts were found in this study. One of them corresponds to a one nucleated cyst-forming species which has been tentatively identified as *E. bovis*-like as in the greater rheas. The second species found

corresponds to an eight nucleated cyst-forming *Entamoeba*. *Entamoeba gallinarum* has been described from poultry. Its cysts are slightly smaller than those found in lesser rheas (12–15 μm vs. 14–22 μm) (McDowell, 1953). However, the present cysts are similar in size to the eight nucleated cysts of an *Entamoeba* sp. found in greater rheas in Spain (20 μm ; Ponce-Gordo et al., 2002). They also fall within the range described for *E. coli* and *E. muris* (10–33 and 9–20 μm , respectively; Levine, 1985). We have identified these cysts as *E. coli*-like, using the name of the morphological group as proposed by Levine (1961) until further analyses have been done.

The morphology of the *Chilomastix* cysts observed in two samples is in agreement with that of *C. gallinarum*, the only species of this genus described in birds. However, the cyst morphology is very similar among the species of this flagellate and we currently prefer not to assign our specimens to a definitive species. All species of *Chilomastix* are considered non-pathogenic (Kulda and Nohýnková, 1978; Levine, 1985).

The morphology of the trematode eggs found in a single sample clearly resembles that of *F. hepatica* eggs. Flukes had not previously been described in lesser rheas and this is the first report of this parasite in this host species. The positive sample was obtained from the farm where two adult birds had died the week before the samplings. The owner reported whitish lesions in the liver. Their association with a *Fasciola* infection could, however, not be established because the carcasses were destroyed prior to our arrival.

Capillariinae eggs were present in one sample from the Córdoba zoo. This subfamily includes several genera in birds that can, however, not be identified by egg morphology (Yabsley, 2008). *Capillaria parvumspinosa* has been described from rheas in Europe (Railliet and Henry, 1911) and Chang Reissig et al. (2001) reported *Capillaria* sp. eggs from lesser rheas.

Two different types of nematode eggs and of larvae were found in farmed birds. In one sample, the egg size and morphology found resemble that of *Trichostrongylus* spp. previously reported from lesser rheas by Chang Reissig et al. (2001). The other sample contained eggs resembling those of spirurids. Several spirurids have been recorded from greater rheas (see above) but none from lesser rheas. The nematode larvae could not be identified and it is possible that they are non-parasitic, belonging to free-living species.

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