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Parasites of farmed marals in Kazakhstan

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Abstract

The farming of maral (a type of red deer- *Cervus elaphus*) is an expanding livestock industry in Kazakhstan. However, to date there is little knowledge of the parasitic fauna that infect farmed deer in Kazakhstan. This study investigated the gastrointestinal parasites of 508 farmed marals from the south and east of Kazakhstan between spring and autumn 2015. When fecal samples were being collected, the presence of any ectoparasites were also noted and specimens taken for identification. This study revealed 6 species of nematodes: *Bunostomum phlebotomum, Capillaria bovis, Haemonchus contortus, Nematodirus spathiger, Oesophagostomum venulosum and Trichuris skrjabini.* Two cestode species were identified: *Moniezia benedeni* and *Moniezia expansa.* Three species of coccidia: *Eimeria cervi, E. gallivalerioi and E. robustus* were found. Ectoparasites recovered were the deer ked *Lipoptena cervi,* larvae of *Hypoderma diana* and *Booponus borealis* and the tick *Dermacentor marginatus.*

Key words:

Maral, *Cervus elaphus sirbiricus*, parasites, helminths, coccidia, Kazakhstan, *Bunostomum, Capillaria, Haemonchus, Nematodirus, Oesophagostomum, Trichuris, Eimeria, Lipoptena, Dermacentor, Hypoderma, Booponus*

Introduction

Kazakhstan is a large central Asian country. Livestock industry are an important part of agricultural production. The are approximately 23.3 million farmed ruminants in Kazakhstan and about 70% of the land area is permanent meadows and pastures (FAO 2013) . This includes extensive rangelands in semi arid areas.

The maral or Siberian wapiti - (*Cervus elaphus sibiricus)* is one of the easternmost subspecies of Red deer that is native to areas in Kazakhstan, China, Mongolia and Russia. They are large, strong animals: adult deer can be up to 150 cm high and weigh up to 260-330 kg for males and 150-250 kg for females (Korzhikenova et al., 2014). Phylogenetic analysis suggests that red deer should be divided into two species based on the western group from Europe, southern and western Asia and an eastern group from northern Asia and north America. Division into subspecies however, may not be warranted (Ludt et al., 2004). On this basis Marals would be a variant of the well known red deer.

Maral deer antlers are of high quality commanding a premium price (Kurmanabev and Dyuysenbaev 2004; Kim et al., 2015). Faming of red deer is a new industry in Kazakhstan and provides antlers, meat and by products. Parasitic diseases are potentially a major limiting factor for deer production in Kazakhstan. However as yet there is little data on the parasite fauna infecting marals under the husbandry conditions prevailing in Kazakhstan. This study details the parasitic fauna found in 508 farmed marals in eastern and southern Kazakhstan in 2015.

Materials and methods

Seven farms were selected in eastern and southern Kazakhstan. Six farms were located in the East Kazakhstan Oblast – 5 farms in the Katonkaragay District and one farm in the Ulan district. One farm was located in the Karasai District of Almaty Oblast. In total there are 18 farms in East Kazakhstan. Five of 12 farms from the Kantonkargay district were investigated and the only farm in Ulan district. The other 5 farms located in other districts of East Kazakhstan were not investigated. The farms in East Kazakhstan were primarily selected for ease of access as it is a remote region. In the Almaty region there is only one Maral farm.

Faecal samples were taken from 508 animals between May and November 2015. Age and gender of all animals was recored. There was no significant evidence of clinical issues caused by parasites in the sampled animals. For male animals, faeces were collected directly from the rectum at the time that antlers were harvested. For other animals, samples were taken from the ground immediately following an observed defecation. With such samples, only the upper portion of the freshly passed faces, which had not had direct contact with the ground, was taken. All adult animals were sampled on 6 of the 7 farms. On one farm it proved impossible to sample some of the animals.

All samples were immediately placed in plastic containers before dispatch to the laboratory.

For each sample 5 g of faeces was mixed and homogenized with 15ml of water. Following filtration through a metal sieve with aperture size of 0.25 mm, the mix was centrifuged for 3 minutes at 200g. The supernatant was discarded and the sediment was mixed an equal part of glycerine and saturated sodium chloride solution (Darling's solution). The precipitate was resuspended by stirring and the sample was centrifuged again for 3 minutes. Samples were then taken with an 8mm diameter loop from the surface of the tube. Three drops were placed on a slide with a cover slip and examined microscopically.

Species identification of *Eimeria* spp oocysts was according to Kheysin (1967) and Svanbaev (1979). Helminth eggs were identified according to Kapustin (1953), Boev et al., 1962 and Demidov (1987). Strongyl eggs were further cultured for larvae and the larvae isolated by Bearman method and subsequently identified (Demidov, 1987) . To obtain stage 3 larvae, faeces were placed in glass jars and kept moist at 25-27^{o} C for 10-20 days. The feces were placed in a gauze pouch and dipped in a glass with warm water at a temperature of 35 -38 ° C and left for 10-12 hours. The gauze pouch was removed, supernatant gently decanted and the sediment examined for larvae. After culturing the larvae were microscopically examined under a microscope for species identification. Any ectoparasites present on deer at the time of sampling were identified morphologically according to Kapustin (1955).

Possible variations of parasite prevalence with age or sex of the marals was analysed using prevalence ratio regression in R (R Core Team, 2016.). That is binomial regression with a logarithmic link. Both age and sex of the marals were used as covariates in the analysis.

Results

Eggs from a total of 5 nematode species, 3 protozoa spp, 2 cestode species were isolated from farmed deer in East Kazakhstan region in summer and autumn 2015. In addition one species of tick and two dipteran species were recovered from animals (table 1 and 2). In the Almaty region an additional nematode species was recovered and a dipteran species (deer ked) (tables 3-4). The notable differences was that *Haemonchus contortus* was only recovered in the spring in the Almaty region, but only in the autumn in east Kazakhstan. *Bunostomum phlebotomum* was recovered from a number of animals in the Almaty region in the summer but not from east Kazakhstan region.

The only parasite that appeared to have a variation in prevalence with age was Eimeria cervi isolated from deer in East Kazakhstan in the autumn. There was a significant decrease in the prevalence as age increased ($p<0.001$). The only parasite that appeared to have a significant variation with sex was *Oesophagostonum venulosum* from deer in the Almaty region in the summer. Here, female animals appeared to have a higher prevalence of infection ($p<0.001$), No other

parasites appeared to have any statistical association of prevalence with age or sex of their hosts.

Table 1. Parasites recovered from 117 marals (63 females, 54 males) in early summer (June) 2015 from East Kazakhstan region of Kazakhstan. These consisted of animals of age groups 3 years to 7 years with a mean age of 4.3 years.

Parasite Species	Numbers positive	Number females positive	Prevalence (%)	Exact Binomial CIs (%)
Nematodirus spathiger	2	1	1.7	0.2-6
Oesophagostonum venulosum#	37	21	31.6	23.2-40.9
Trichuris skrjabini	1	1	0.8	0.02-5.7
Eimeria cervi	1	1	0.8	0.02-5.7
Eimeria gallivalerioi	3	2	2.6	0.5-7.3
Eimeria robusta	1	1	0.8	0.02-5.7
Dermacentor marginatus*	1	0	0.8	0.02-5.7
Hypoderma diana	1	0	0.8	0.02-5.7
Larvae of *Booponus borealis* – antler flies	4	0	3.4	0.94-8.5

#It was only possible to analyze any variation in parasite prevalence by age with *O. venulosum* and none was detected.
*6 specimens were recovered from a single animal
Parasites which were not isolated (ie point prevalence of 0%) have 95% CIs of 0% to 3.1%

Table 2. Parasites recovered from 226 marals (107 females and 119 males, aged 1 to 7 years, mean 4.9 years) in autumn (September/October) 2015 from East Kazakhstan region of Kazakhstan.

Parasite Species	Numbers positive	Number females positive	Prevalence (%)	Exact Binomial CIs (%)
Capillaria bovis	26	15	11.5	7.6-16.4
Haemonchus contortus	31	19	13.7	9.5-18.9
Nematodirus spathiger	2	1	0.9	0.11-3.2
Oesophagostomum venulosum	65	36	28.8	22.9-35.1
Trichuris skrjabini	3	1	1.3	0.3-3.8
Moniezia benedeni	5	2	2.2	0.7-5.1
Moniezia expansa	2	2	0.9	0.11-3.2
Eimeria cervi#	11	7	4.9	2.5-8.5
Eimeria gallivalerioi	34	18	15.0	10.7-20.4
Eimeria robusta	13	6	5.8	3.1-9.6
**Dermacentor marginatus*	1	0	0.4	0.01-2.4

*2 specimens were recovered from a single animal

#Prevalence significantly decreased with increasing age

Parasites which were not isolated (ie point prevalence of 0%) have 95% CIs of 0% to 1.6%

Table 3. Parasites recovered from 34 marals (17 female, 17 male, aged 2-5 years, mean age 4.1 years) in Spring (May) 2015 from Almaty region of Kazakhstan

Parasite Species	Numbers positive	Number females positive	Prevalence(%)	Exact Binomial CIs (%)
Haemonchus contortus	18	10	52.9	35.1-70.2
Nematodirus spathiger	1	0	2.9	0.07-15.3
Trichuris skrjabini	1	1	2.9	0.07-15.3
Eimeria robusta	1	0	2.9	0.07-15.3

Parasites which were not isolated (ie point prevalence of 0%) have 95% CIs of 0% to 10.3%

Table 4. Parasites recovered from 131 marals (36 female, 95 male, age 1-7 years , mean age 4.8 years) in Summer (July) 2015 from Almaty region of Kazakhstan

Parasite Species	Numbers positive	Number females positive	Prevalence (%)	Exact Binomial CIs (%)
Bunostomum phlebotomum	10	0	7.6	3.7-13.6
Capillaria bovis	5	1	3.8	1.3-8.6
Nematodirus spathiger	1	1	0.8	0.02-4.2
Oesophagostomum venulosum	39	21#	29.8	22.1-38.3
Trichuris skrjabini	6	3	4.6	1.7-9.7
Eimeria cervi	1	1	0.8	0.02-4.2
Eimeria gallivalerioi	6	3	4.6	1.7-9.7
Eimeria robusta	3	3	2.3	0.5-6.6
**Lipoptena cervi*	1	1	0.8	0.02-4.2

*32 specimens were recovered from a single animal
#Significantly higher prevalence in female animals compared to males
Parasites which were not isolated (ie point prevalence of 0%) have 95% CIs of 0% to 2.8%

Discussion

This simple surveillance study of 7 deer farms in two regions of Kazakhstan presents some data on the parasite fauna that infects marals or red deer in central Asia. These include a number of potentially pathogenic species. *Haemonchus contortus* is documented as a parasite of red deer with reports from Italy (Manfredi et al., 2007) Poland (Pilarczyk et al., 2005), Portugal (Maia, 2001) and elsewhere. *H. contortus* is one of the most pathogenic nematode parasites of sheep and is widespread in Kazakhstan (Morgan et al., 2006). This parasite has been hypothesized as one of the most likely parasites to infect red deer if they have access to pasture where sheep have grazed (Tapia-Escarate et al., 2015) . Although our study was limited, there may be some patterns in the data. *H. contortus* eggs were only isolated in the spring (May) in deer from the Almaty region and not in the summer. In East Kazakhstan this parasite was only found at the autumn sampling. It is difficult to compare the two as there was no sampling in May in East Kazakhstan and none in the autumn in the Almaty region. However mean temperatures in Almaty region in May are comparable to those in June of East Kazakhstan as the latter is considerably further north and thus the climate in the south may suit the transmission of *H. contortus*. Both regions have cold winters and thus overwintering of this parasite could be reliant on hypobiosis and this may be the reason that a high proportion of animals in the Almaty region were infected with this parasite in the spring as larvae emerged from the hypobiotic state and matured into adults. In Canada, which has similar winter conditions, free living *Haemonchus* larvae are not able to successfully overwinter(Falzon et al., 2014)

Oesophagostomum venulosum is found world wide in livestock and is believed to be of low pathogenicity. Infective larvae of *Oesophagostomum* spp. do not survive the winter in Lithuania (Sarkunas et al., 2007). Therefore this is more unlikely in Kazakhstan where the winters are more severe. Consequently nematodes are reintroduced onto the pastures by adult deer in the spring. *O. venulosus* has been isolated from red deer (*Cervus elaphus elaphus*) in Poland (Drozdz et al. 1997)

Capillaria bovis is known to be parasitic to 30 species of ruminants worldwide, mainly in the families bovidae and cervidae (Justine and Ferté, 1989) and is believed to be of low pathogenicity.

Trichuris skrjabini was first described in the former USSR and has been found in many different host species including deer over a wide geographical location (Knight and Tuff, 1984, Drozdz et al 1997) . Whipworms are rarely of any clinical significance in ruminants (Wideman, 2004).

Nematodirus spathiger has long been known to cause growth retardation and diarrhoea in young lambs (Turner, 1953) and may have similar effects in deer. It is also a cold adapted parasite and the free living stages are have been shown to have the ability to overwinter in the cold winter conditions that occur in Canada (Falzon et al., 2014) . Thus it is likely that free living stages also

overwinter in Kazakhstan

The hookworm, *Bunostomum phlebotomum*, is more commonly found in cattle and in areas of warm and moist climate throughout the world. Chief clinical signs are anemia, emaciation, and marked weakness in calves (Williams et al., 1983). There are few reports of this parasite in deer although it has been described in fallow deer in Bosnia (Sinanović and Zuko, 2012) .

Moniezia expansa has been observed in approximately 10% of deer calves in New Zealand (Mason and Moore, 1983). In Italy *M. benedeni* were found in red deer from the Stelvio National Park (Manfredi et al., 2003).

There are numerous reports of *Eimeria* spp isolated from red deer. However, they have been rarely differentiated into species. In Poland, 91 wild red deer were examined and 33 (36%), 12 (13%) and 7 (8%) were infected with *E. sordida*, *E. elephi* and *E. cervi* respectively (Balicka-Ramisz et al., 2000). *E. gallivalerioi* was first described in 1930 with the type host as *Cervus elaphus* from a zoo in Leningrad (now St Petersburg) (Levine and Ivens, 1970). Otherwise there is little information on its distribution or prevalence in red deer. *E. robusta* has been reported from *Cervus elaphus* in Slovenia (Brglez and Bidovec, 1978). The significance of coccidia in cervids is not known. When coccidia have been identified in adult deer and elk there have been no clinical effects, except perhaps in mule deer, where clinical disease has been experimentally induced in deer fawns giving chronic diarrhea and weight loss (Abbas and Post, 1980).

The deer ked (*Lipoptena cervi*) is a hematophagous ectoparasite of cervids in Europe, Asia, Africa and North-America (Bequaert, 1942). Infestations may induce inflammatory skin reactions and alopecia and the keds may act as vectors for the transmission of pathogens such as *Bartonella* spp (Paakkonen et al., 2014).

The larvae of antler flies *Booponus borealis* is known to cause myiasis in domesticated and wild deer from the Altai region of Russia (Raabe, 2008). This region of Russia borders directly on East Kazakhstan Oblast so it is unsurprising that deer from this part of Kazakhstan may become infested. The adult fly lays eggs in dense or scattered groups on the undamaged skin of the young antlers and hatch within 24 hr. The larvae penetrated the antlers close to their hatching sites, progressively destroying the connective tissue resulting in serious myiasis. Further eggs may be subsequently laid adjacent to the areas of damage, which are further extended by larval feeding (Sadovnikocva, 1969).

Hypoderma diana is a widely distributed warble fly of deer and differs from other warble flies species because it has several hosts. In addition to the European roe deer (*Capreolus capreolus*), which is its main host, *Hypoderma diana* also infests other cervids, e.g. the red deer (*Cervus elaphus*), sika deer (*Cervus nippon*), fallow deer (*Dama dama*), Eurasian elk (*Alces alces*), and rarely the chamois (*Rupicapra rupicapra*) and mouflon (*Ovis musimon*). Second-instar larvae

have also been found in the reindeer (*Rangifer tarandus*) (Pavlasek and Minar (2014). This warble fly has been described in Marals from the neighboring Altai region of Russia (Solopov and Zharkov, 1988).

Dermacentor marginatus, the ornate sheep tick, is distributed from Portugal through southern Europe up to Iran, Kazakhstan, and the mountain areas of central Asia(Parola et al., 2009). The parasite has a wide host range, including domesticated and wild mammals (Rubel et al., 2016) and thus the finding of this tick on marals in eastern Kazakhstan is unremarkable, although it is from the eastern most part of its known range.

Conclusions

This study documents the presence of a number of parasite species in marals in Kazakhstan. Some species are potentially pathogenic and hence warrant further investigation in the epidemiology and potential control of these parasites.

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