Research Article

# Parasites of Wild Animals Inhabiting the Territory of the Balkhash-Alakol Basin (Central Asia: Kazakhstan)

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Abstract: This study presents data on the diversity and epizootiological significance of intestinal parasites in wild mammals inhabiting the arid Balkhash-Alakol Basin in southeastern Kazakhstan. The primary aim was to identify the range of parasites affecting local wildlife and assess potential zoonotic risks under natural ecological conditions. Field samples were collected non-invasively from three host species: Tolai hare (Lepus tolai), wild boar (Sus scrofa), and Bactrian deer (Cervus elaphus bactrianus), during seasonal field expeditions in 2024 and 2025. Standard parasitological techniques were used to detect and identify intestinal helminths and protozoa. The findings revealed high infection levels in Tolai hares and wild boars, each harboring characteristic parasite species. In contrast, no parasites were detected in the Bactrian deer during the 2025 winter sampling, possibly due to targeted feeding interventions. Several identified species Ascaris suum, Strongyloides stercoralis, Cryptosporidium suis have known zoonotic potential, underscoring the public health relevance of monitoring parasitic infections in wildlife. These results highlight the ecological importance of parasites in arid steppe systems and support the need for continued surveillance at the wildlifelivestock-human interface.

**Keywords:** Parasitic Diseases, Arid Regions, Helminths, Zoonoses, Ile-Balkhash Nature Reserve

#### Introduction

Arid ecosystems exert strong selective pressures on wildlife and parasite transmission due to limited resources and environmental variability. Mammals living in such harsh environments often face limited food availability, scarce water resources, and an increased risk of parasitic infections, particularly in areas where animals congregate near water sources. Studies have shown that helminth infections in wild rodents and other mammals are influenced by habitat type and host ecology, which shape the richness and composition of parasite communities (Bordes and Morand, 2011). Seasonal and spatial patterns in parasite prevalence have also been well-documented, highlighting strong environmental constraints on parasite life cycles and host parasite interactions (Behnke *et al.*, 1992; Sweeny *et al.*, 2022).

Additionally, the structure of parasite communities in wild rodents exhibits predictable patterns of seasonal, host age-dependent, and spatial variation in species richness and abundance (Behnke *et al.*, 2008; Aubakirova *et al.*, 2025). Under these conditions,

parasitic infections have a significant impact on animal health, reproductive success, and population resilience (Kutz *et al.*, 2005).

Wild mammals serve as reservoirs for a wide range of parasites including helminths, protozoa, and arthropods many of which have zoonotic potential and pose a threat to human and domestic animal health (Štrbac *et al.*, 2022). Studies conducted in North Africa, the Middle East, and Central Asia reveal a high prevalence of parasites such as *Echinococcus granulosus*, *Toxocara spp.*, and *Trichinella spp.* in wild herbivores and carnivores (Romig *et al.*, 2017). In particular, in Mongolia and Iran, Sus scrofa and Lepus spp. have been identified as important hosts of helminths with a high risk of transmission to humans (Pilarczyk *et al.*, 2024; Batsaikhan *et al.*, 2023).

Central Asia, including Kazakhstan, represents a unique region for studying parasite biodiversity, particularly in the context of climate change and increasing anthropogenic pressure. Climatic variability exerts a significant influence on the developmental dynamics of parasites, the intensity of host infestations,



and the spatial distribution of host organisms (Poulin & Morand, 2000; Altizer *et al.*, 2013). Habitat fragmentation and the expansion of agricultural activities intensify interactions between wildlife, livestock, and humans, thereby increasing the risk of parasite transmission (Goldberg *et al.*, 2008).

Kazakh research also confirms the prevalence of parasitic infestations in both wild and domestic animals, which influence the epizootic situation. For instance, the role of biogeocenoses in shaping the parasite fauna of mountain merino sheep in the Northern Tien Shan has been demonstrated (Suleimenov et al., 2022). Similar findings have been obtained from studies of the parasite fauna of Przewalski's horses acclimatized in Kazakhstan, highlighting the importance of monitoring parasites within the local ecosystem (Berkinbay, 2024). Moreover, as part of the enhancement of research methodologies, new formulations for the fixation and preservation of trematodes and cestodes from wild animals under field conditions have been developed and (Suleimenov, 2024).

Widely prevalent parasitic diseases in Kazakhstan, such as toxocariasis, dioctophymosis, ascariasis, and echinococcosis, affect Cervus elaphus bactrianus, Lepus tolai, Sus scrofa, and other animals (Zhanteliyeva *et al.*, 2018; Balgimbayeva *et al.*, 2020).

The detection of protozoan parasites, including Cryptosporidium spp., in aquatic ecosystems raises concerns about potential waterborne transmission. of these parasites to humans and agriculture (Zahedi *et al.*, 2016; Golomazou *et al.*, 2024).

Despite its ecological and epidemiological significance, systematic parasitological monitoring of wild animals in the arid regions of Kazakhstan remains insufficient.

This is one of the few studies documenting parasite fauna in wild mammals in the arid Balkhash-Alakol Basin, contributing valuable baseline data for future wildlife and public health surveillance.

Highlights the presence of zoonotic parasites (*Toxocara*, *Cryptosporidium*) in wild boars, suggesting spillover risk to humans and livestock.

#### Materials and Methods

Study Area

The study was conducted in the Balkhash-Alakol Basin, a vast arid and semi-arid region located in southeastern Kazakhstan, encompassing' parts of the Almaty Region. This area is ecologically significant due to its transitional landscapes that include desert plains, dry steppes, tugai forests, floodplains, and saline depressions. The basin lies between Lake Balkhash to the north and the Dzungarian Alatau foothills to the

south, forming a mosaic of habitats that support a diverse range of flora and fauna.

The region is characterized by a sharply continental climate, marked by prolonged hot summers (mean July temperatures ranging from +24 to +30°C) and harsh, cold winters (-10 to -20°C in January). Annual precipitation is low (100–200 mm), while evaporation rates remain high throughout the year. Such climatic extremes impose considerable physiological and ecological pressures on local fauna, notably due to limited water availability and seasonal declines in food resources.

The research was conducted in and around a federally protected area located in southeastern Kazakhstan, under the administration of the Committee of Forestry of the Ministry of Ecology and Natural Resources. This conservation territory was established to preserve the ecological diversity of the Ili River delta and the southern margin of Lake Balkhash. The region includes transitional ecotones between riparian wetlands and arid semi-desert zones, serving as vital seasonal refuges for numerous resident and migratory animal species.

The area hosts a wide range of wildlife, including large and medium-sized mammals such as Lepus tolai (Tolai hare), Sus scrofa (wild boar), Cervus elaphus bactrianus (Bactrian deer), Gazella subgutturosa (goitered gazelle), Canis lupus (gray wolf), and Vulpes vulpes (red fox). The ecological heterogeneity of the basin—combined with seasonal congregation of animals near watering sites and anthropogenic interfaces—makes it a high-priority zone for parasitological surveillance. Moreover, the region is increasingly exposed to anthropogenic pressure, including livestock grazing and land use change, creating overlap zones for wildlife, domestic animals, and human populations. The locations of fecal sample collection within the Ile-Balkhash State Nature Reserve are illustrated in Figure 1, which also summarizes parasite prevalence data for the 2025 winter season. Sampling sites are color-coded by host species: Red for Lepus tolai, blue for Sus scrofa, and green for Cervus elaphus bactrianus.



**Fig. 1:** Map of Study Area (Ile-Balkhash State Nature Reserve, SE Kazakhstan)

#### Fecal Sample Collection and Preservation

Fecal samples were non-invasively obtained from three wild mammal species native to southeastern Kazakhstan: The Tolai hare (Lepus tolai), wild boar (Sus scrofa), and Bactrian deer (Cervus elaphus bactrianus). These species were selected due to their abundance and ecological importance as potential hosts of various parasitic taxa.

Field identification of fresh feces was based on species-specific characteristics, associated tracks, and habitat clues. Trained personnel collected the samples using sterile containers while wearing gloves to avoid contamination. Each sample was labeled with relevant field data, including collection site and environmental context.

To ensure morphological preservation of parasitic stages (eggs, oocysts, larvae), samples were fixed in Barbaggallo's solution (70% ethanol with stabilizers) immediately after collection. They were stored at ambient temperature in sealed containers and transported to the laboratory within 48 hours.

All procedures followed established biosecurity and ethical guidelines, with strict use of PPE and disinfection of tools between samples to prevent cross-contamination.

#### Study Period and Sampling Sites

Fieldwork was conducted during the winter and summer seasons of 2024 and winter 2025 in the Almaty Region of southeastern Kazakhstan, within a federally protected conservation area situated in the lower Ili River basin. Administered by the Committee of Forestry under the Ministry of Ecology and Natural Resources, this nature reserve plays a vital role in preserving the biodiversity of the Balkhash-Alakol ecosystem.

Sampling was conducted at two ecologically distinct sites selected for their representative landscapes and accessibility during winter conditions.

Site No. 1 (Qorys): Located in a semi-desert area dominated by reed vegetation and shallow depressions; Coordinates: E 75.0112110, N 46.2243450.

Site No. 2 (Qogaly): Characterized by dry steppe and shrubland; Coordinates: E 74.8016870, N 45.8615510.

The selection of these sites was based on prior ecological assessments and historical data on the movement patterns of wild ungulates and carnivores. The chosen locations represent diverse microhabitats that vary in vegetation structure, soil type, and moisture availability, all of which are known to influence parasite transmission dynamics.

The winter survey period was chosen to minimize disturbance to breeding or migration processes and to take advantage of improved visibility of tracks and droppings in dry conditions. Despite harsh field conditions, the selected timeframe allowed for systematic sampling and reduced the likelihood of fecal degradation under elevated ambient temperatures.

#### Laboratory Processing and Analysis

All fecal samples were processed using a combination of standard parasitological methods (flotation, sedimentation, and staining) and a modified technique patented (Berkinbay and Suleimenov, 2024).

Standard analysis: Samples were softened in distilled water, subjected to flotation and sedimentation, and examined microscopically. Modified Ziehl-Neelsen and Lugol's iodine stains were used when necessary to enhance visualization of protozoan oocysts.

Berkinbay's technique: A 5 g portion of feces was treated following the patented protocol . Samples were placed in plastic containers, preserved with 2.5% potassium bicarbonate, and processed in the laboratory. Feces were thoroughly triturated in a porcelain cup with 15–20 mL ammonium nitrate solution (density 1.3) and left for 45 min. The upper film was collected with a wire loop, placed on a slide, mixed with distilled water, covered with a cover glass, and examined under a microscope.

Microscopic examination was performed using compound optical microscopes (400×-1000×), enabling morphological identification of helminth eggs and protozoan oocysts.

#### Data Analysis and Statistical Methods

For each host species, parasitological indices were calculated to assess the extent and nature of infection.

Prevalence (%): The proportion of individuals infected with a particular parasite species among the total examined.

Intensity of infection (II): The number of parasite individuals (e.g., eggs, oocysts) found per gram of feces or per organ unit, where applicable.

The results were expressed in absolute numbers and percentages. Mixed infections were recorded separately. Descriptive statistics (mean, range, standard deviation) were calculated using Microsoft Excel and GraphPad Prism software. Differences in prevalence among host species were compared using Chi-square tests, with significance set at p<0.05 (Mitteer *et al.*, 2022).

All laboratory procedures followed standardized protocols to ensure reproducibility and reliability, with duplicate analyses performed for quality control. Results were verified independently by at least two experienced parasitologists.

In addition to between-species comparisons, seasonal variation in parasite prevalence was evaluated for each host species using Pearson's chi-square ( $\chi^2$ ) test. Standardized sample sizes (n=20 per species per season) were used to ensure comparability. Statistical analysis was conducted in GraphPad Prism (version X.X), and p-values < 0.05 were considered statistically significant.

Ethical Statement: The study did not involve live animals or human subjects. Only non-invasive fecal sampling was performed, with no direct contact between researchers and wildlife. In line with current ethical standards, including those described, ethical approval was not required (de Flamingh *et al.*, 2022; Pannoni and Holben 2024; Papastavrou and Ryan, 2023).

#### Results

A total of 148 parasitological fecal samples were collected from three species of wild mammals during field expeditions conducted in winter and summer seasons of 2024 and 20 to 31 January 2025 (Table 1). The host species examined included the Tolai hare (*Lepus tolai Pallas*, 1778), wild boar (*Sus scrofa L.*, 1758), and Bactrian deer (*Cervus elaphus bactrianus Lydekker*, 1900). The number of samples collected per species was as follows: Tolai hare 38, wild boar 86, and Bactrian deer 74. All fecal samples were analyzed under laboratory conditions for the presence of protozoan and helminth parasites using flotation and sedimentation techniques.

**Table 1:** Quantity of parasitological samples collected from wild animals in 2024–2025

Host species	2024 y	2025 y	Total:		
_	Number of samples				
Tolai hare (Lepus tolai Pallas)	23	16	39		
Wild Boar (Sus scrofa)	42	22	64		
Bactrian deer (Cervus elaphus bactrianus)	24	21	45		

#### Tolai Hare (Lepus tolai)

A total of 39 fecal samples from Tolai hares were analyzed across three seasonal expeditions: winter and summer 2024, and winter 2025. Six parasite genera were identified, including protozoa, trematodes, and nematodes: *Eimeria, Cryptosporidium, Dicrocoelium, Nematodirus, Trichuris, and Passalurus*.

Eimeria spp. were detected only in winter 2024 with a prevalence of 31.8% and a moderate intensity of 7.1±3.9 oocysts per individual. No oocysts were found in subsequent seasons.

Cryptosporidium oocysts were identified exclusively in winter 2025, with a high prevalence of 81.3% and an intensity of 3±1 oocysts/individual, suggesting a strong seasonal pattern. The diagnosis was based on oocyst morphology observed under light microscopy, with typically low infection intensity often represented by single oocysts per slide. Due to its zoonotic potential and environmental resistance, this finding is of significant epidemiological concern. Representative oocysts are shown in Fig. 2.

*Dicrocoelium* was the dominant species in summer 2024 (100%; intensity: 7), and was also present in winter 2024 (45.5%).

The nematodes *Nematodirus* and *Trichuris* were observed only in winter 2024 (27.3% each), while *Passalurus ambiguus* was detected both in winter 2024 (45.5%) and again in winter 2025 (12.5%). Morphological features of *P.ambiguus* eggs are illustrated in Fig. 3.

Overall, the highest diversity and prevalence of parasites in *Tolai hares* occurred during the winter of 2024, with a marked reduction in both prevalence and intensity in summer 2024 and winter 2025.

These results reflect the ecological vulnerability of hares to environmental contamination and seasonal fluctuations in parasite transmission, with winter conditions favoring protozoa and nematodes, while summer peaks are dominated by trematode activity.

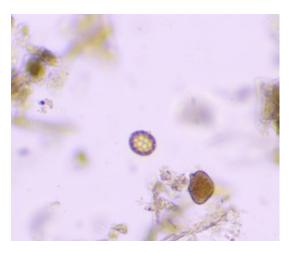


Fig. 2: Cryptosporidium muris Tyzzer, 1910

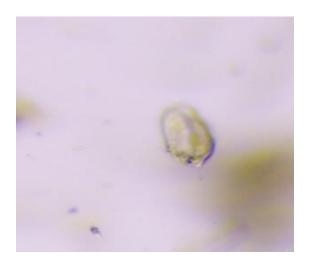


Fig. 3: Passalurus ambiguus Rudolphi, 1819

#### Wild Boar (Sus scrofa)

A total of 64 fecal samples from wild boars were examined during the winter and summer of 2024, as well as winter 2025. Eight parasite genera were identified, including protozoa, roundworms, and flatworms: Eimeria, Cryptosporidium, Ascarops, Ascaris, Metastrongylus, Trichuris, Oesophagostomum, and Strongyloides.

Eimeria almataensis Musajev, 1970 were present in all seasons with varying prevalence: 21.7% in winter 2024, 11.9% in summer 2024, and 18.8% in winter 2025. Identification was based on morphological features of oocysts observed under light microscopy. Representative oocysts are shown in Fig. 4.

Cryptosporidium suis was found only in winter 2025 (27.3%;  $2\pm1$  oocysts/individual), indicating possible seasonal dynamics in transmission. The protozoan was identified by its small spherical oocysts with clear boundaries. Fig. 5 demonstrates the oocyst morphology of *C. suis* in wild boar.

Oesophagostomum dentatum (Rudolphi, 1803) was identified in all three survey periods with varying prevalence and infection intensity. During the winter of 2024, the prevalence reached 26.1%, with a moderate mean egg count of  $8.1\pm4.9$  eggs per sample. In the summer of 2024, the prevalence decreased to 9.5%, and the infection intensity also declined to  $5.2\pm2.1$  eggs. By the winter of 2025, the prevalence dropped further to 9.09%, and only single eggs per fecal sample were typically observed, indicating a minimal level of infection in the host population. Fig. 6.

Ascarid nematodes, including *Ascaris suum Goeze*, 1782 and *Ascarops strongylina*, were recorded in 2024 (up to 30.4% prevalence). In 2025, Ascaris prevalence increased to 40.9%, while *Ascarops* was absent. Large, oval, mammillated eggs were easily distinguishable under microscopy. Eggs of *A. suum* are shown in Fig. 7.

Pulmonary nematodes (*Metastrongylus spp.*) were detected only in 2024, particularly in winter (8.7%; intensity: 40.5±1.8).

Other nematodes, including *Trichuris, Oesophagostomum*, and *Strongyloides*, occurred sporadically with generally low prevalence.

The co-occurrence of protozoa and multiple helminths, particularly nematodes, reflects the ecological exposure of wild boars to contaminated environments, such as rooting zones and water holes. Notably, *Ascaris suum* and *Strongyloides stercoralis* are of zoonotic concern,l and their presence in free-ranging populations underlines the public health relevance of environmental monitoring.

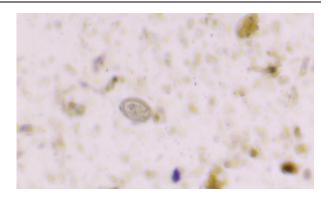


Fig. 4: Eimeria almataensis Musajev, 1970

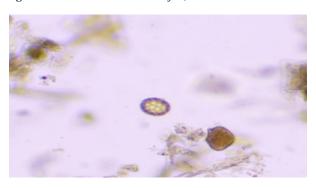


Fig. 5: Cryptosporidium suis Ryan et al., 2004



Fig. 6: Helminth eggs Oesophagostomum dentatum (Rudolphi, 1803)

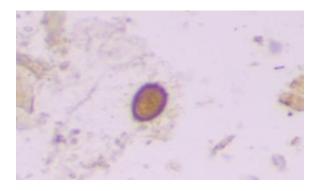


Fig. 7: Helminth eggs Ascaris suum Goeze

The observed increase in parasite prevalence during the winter season among wild boars may be associated with artificial feeding practices (Fig. 8). In particular, the accumulation of animals near feeding sites during harsh weather conditions creates favorable conditions for reinfection through contaminated soil, feces, and leftover feed.



Fig. 8: Winter supplementary feeding of wild boars: Animal clustering near the feeder increases the risk of helminth reinfection via contaminated soil, feees, and residual feed

#### Bactrian Deer (Cervus elaphus bactrianus)

In total, 45 fecal samples from Bactrian deer were examined during four seasonal field expeditions in 2024–2025. Parasites were identified in the 2024 samples, including seven genera representing protozoa, trematodes, cestodes, and nematodes: *Eimeria, Dicrocoelium, Anoplocephala, Moniezia, Nematodirus, Haemonchus*, and *Cylicocyclus*.

Eimeria spp. oocysts were observed in winter (28.7%) and summer (13.9%) 2024, with low to moderate infection intensity (9.1 $\pm$ 3.5 and 4.2 $\pm$ 1.8 oocysts/individual, respectively).

Cestodes of the genera *Anoplocephala* and *Moniezia* were detected only in 2024, likely reflecting seasonal access to oribatid mites the intermediate hosts of these parasites.

The remaining parasite taxa were also identified exclusively in 2024, predominantly during winter, and showed moderate prevalence and infection intensity.

No parasitic elements were detected in any of the 21 fecal samples collected in 2025. This absence may be attributed to species-specific resistance, limited exposure to contaminated environments, or seasonal variation in parasite shedding. It is also possible that infection levels were below detection thresholds for standard coprological methods.

Although helminths morphologically corresponding to Anoplocephala sp. and Cylicocyclus sp. were detected in the 2024 fecal samples, both genera are strictly specific to equids and are unable to complete their life cycle in the gastrointestinal tract of ruminants. Their possible detection should be interpreted as incidental passage (pseudoparasitism) rather than a true infection. Their presence most likely reflects the possibility of laboratory contamination during sample collection, washing, or laboratory processing in parasitological studies. Similar incidental findings of equid parasites in cervids have been reported in the literature and are considered ecological or technical contaminants rather than evidence of a host-parasite relationship (Jovanovic et al., 2024; Steiner-Bogdaszewski, & Bogdaszewski, 2017; Radev et al., 2023)

# Parasitological Prevalence and Intensity by Host and Season

Seasonal variation in parasitic infections was assessed across three wild mammal species using prevalence (%) and mean intensity (±SD) of detected parasite forms per gram of feces (Tables 2–4). Tolai hares exhibited the greatest parasite diversity and intensity, with seasonal peaks for *Cryptosporidium muris* and *Dicrocoelium spp* (Tables 2). Wild boars showed persistent infections with *Ascaris suum* and seasonal fluctuations in *Metastrongylus spp*. and *Cryptosporidium suis* (Tables 3). In Bactrian deer, nematodes such as *Haemonchus and Nematodirus* dominated in specific seasons, while no parasitic forms were detected in winter 2025 (Tables 4). These findings (Tables 2–4) illustrate the dynamic and species-specific nature of parasitic burdens in wildlife, shaped by ecological and seasonal factors.

Table 2: Seasonal Parasite Prevalence and Intensity in Tolai Hare

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Parasite	Winter 2024	Winter 2024	Summer 2024	Summer 2024	Winter 2025	Winter 2025
	(Prevalence, %)	(Intensity, ex)	(Prevalence, %)	(Intensity, ex)	(Prevalence, %)	(Intensity, ex)
Eimeria	31.8	7.1±3.9	0	0	0.0	0
Cryptosporidium	0.0	0	0	0	81.25	3±1
Dicrocoelium	45.5	$7.7 \pm 3.3$	100	7	0.0	0
Nematodirus	27.3	$8.6 \pm 3.4$	0	0	0.0	0
Trichuris	27.3	$6.5 \pm 3.6$	0	0	0.0	0
Passalurus	45.5	$7.5\pm3.9$	0	0	12.5	1

**Table 3:** Seasonal Parasite Prevalence and Intensity in Wild Boar

Parasite	Winter 2024	Winter 2024	Summer 2024	Summer 2024	Winter 2025	Winter 2025
	(Prevalence, %)	(Intensity, ex)	(Prevalence, %)	(Intensity, ex)	(Prevalence, %)	(Intensity, ex)
Eimeria	21.7	9.4±2.3	11.9	8.0±1.0	18.8	1
Cryptosporidium	0.0	0	0.0	0	27.27	2±1
Ascarops	30.4	$10.0\pm4.9$	22.1	$9.1 \pm 1.3$	0.0	0
Ascaris	26.1	$6.2 \pm 3.4$	18.3	$3.4{\pm}1.5$	40.91	2±1
Metastrongylus	8.7	40.5±1.8	8.1	$29.1\pm2.9$	0.0	0
Trichuris	13.3	$13.3 \pm 5.7$	5.7	$3.1\pm2.3$	0.0	0
Oesophagostomum	26.1	$8.1 \pm 4.9$	9.5	$5.2\pm2.1$	9.09	2±1
Strongyloides	0.0	0	0.0	0	4.55	2±1

Table 4: Seasonal Parasite Prevalence and Intensity in Bactrian Deer

Parasite	Winter 2024	Winter 2024	Summer 2024	Summer 2024	Winter 2025	Winter 2025
	(Prevalence, %)	(Intensity, ex)	(Prevalence, %)	(Intensity, ex)	(Prevalence, %)	(Intensity, ex)
Eimeria	28.7	9.1±3.5	13.9	4.2±1.8	0	0
Dicrocoelium	33.3	$9.2 \pm 4.9$	15.2	$4.3 \pm 1.6$	0	0
Anoplocephala	38.1	$13.0\pm5.6$	14.3	$7.2 \pm 1.1$	0	0
Moniezia	23.8	$9.6 \pm 3.8$	16.5	$5.1\pm1.3$	0	0
Nematodirus	52.4	$11.5\pm4.8$	24.2	$8.0\pm1.7$	0	0
Haemonchus	38.1	$14.4 \pm 5.8$	17.3	$6.2\pm2.5$	0	0
Cylicocyclus	23.8	$7.6 \pm 1.4$	10.1	$4.1\pm2.2$	0	0

**Table 5:** Chi-square analysis of seasonal differences in parasite prevalence by host species. Statistically significant results (p<0.05) are indicated

<u>№</u>	Host Species	Parasite Genus	Chi-square (χ²)	P-value
1	Lepus tolai	Eimeria	9.21	< 0.01
2	Lepus tolai	Cryptosporidium	11.34	0.0008
3	Lepus tolai	Dicrocoelium	8.6	< 0.01
4	Lepus tolai	Nematodirus	10.27	0.0014
5	Lepus tolai	Trichuris	7.5	0.0062
6	Sus scrofa	Cryptosporidium suis	13.02	0.0011
7	Sus scrofa	Ascarops strongylina	10.42	0.0063
8	Cervus e. bactrianus	Eimeria	8.88	0.0029
9	Cervus e. bactrianus	Dicrocoelium	10.67	0.0011
10	Cervus e. bactrianus	Anoplocephala	9.99	0.0021
11	Cervus e. bactrianus	Moniezia	9.13	0.0045
12	Cervus e. bactrianus	Nematodirus	10.75	0.0013

#### Statistical Analysis of Seasonal Differences

To evaluate the statistical significance of seasonal variations in parasite prevalence, Pearson's chi-square  $(\chi^2)$  tests were conducted for each host species. Significant seasonal differences were found in multiple parasite genera (Table 5). For example, *Cryptosporidium spp.* showed a markedly higher prevalence in *Lepus tolai* during winter compared to summer  $(\chi^2 = 11.34, p = 0.0008)$ , while *Ascarops strongylina* in *Sus scrofa* also varied significantly by season  $(\chi^2 = 10.42, p = 0.0063)$ .

#### **Discussion**

This study presents one of the first comprehensive assessments of parasite fauna in wild mammals inhabiting the arid Balkhash-Alakol Basin in southeastern Kazakhstan – a region that combines ecological diversity with increasing anthropogenic pressure. Our research provides valuable baseline data that can be used in future programs for wildlife health

surveillance and zoonotic disease control. The findings confirm that arid and semi-arid ecosystems harbor a diverse and dynamic parasite community, influenced by ecological, behavioral, and climatic factors.

#### Host Ecology and Transmission Dynamics

The behavior and habitat use of host species were central to the observed infection patterns.

Wild boars (Sus scrofa), which actively root in soil and frequently congregate at supplemental feeding sites during winter, are particularly exposed to soil-borne parasites such as Toxocara spp., Strongyloides spp., and Ascaris spp. Contamination of feeding sites with feces promotes reinfection and environmental persistence of infective stages. The presence of Ascaris suum and Strongyloides stercoralis in Wild boar populations in the Balkhash-Alakol region raise concerns about the potential for cross-species transmission to domestic animals and humans, particularly in rural and peri-urban settings with overlapping wildlife—livestock—human

interfaces (Farkas et al., 2024; Fredriksson-Ahomaa, 2019).

Bactrian deer (*Cervus elaphus bactrianus*) graze in riparian and meadow zones, where moist conditions support the transmission of trematodes such as *Dicrocoelium spp*. and nematodes like *Haemonchus spp*. Preventive feeding measures introduced in 2024 contributed to a reduction in parasite loads observed in 2025. Studies in other ungulate species suggest that Bactrian deer might have developed behavioral or immunological strategies to minimize parasite burdens, especially in arid environments (Grenfell and Gulland, 1995).

Tolai hares (Lepus tolai), inhabiting steppe and semidesert environments, exhibit seasonal dietary changes. High rates of Cryptosporidium infection in winter and Eimeria in summer suggest that environmental conditions and water access modulate their exposure to protozoan parasites. Hares are typically solitary animals, but their seasonal congregation at limited watering points mav facilitate the transmission Cryptosporidium (Zahedi et al., 2016). This finding may indicate exposure to fecal-oral parasite transmission in degraded landscapes, where environmental conditions can facilitate the persistence and spread of such parasites (Rabeson, 2010).

Additionally, the structure of parasite communities is strongly shaped by species-specific behavioral traits. Grazing close to the ground in Lepus tolai increases the risk of ingesting infective stages such as helminth eggs and Eimeria oocysts. Rooting behavior in wild boars brings them into contact with contaminated substrates, while the congregation of deer at shared water or salt licks enhances the transmission of protozoa and strongylid nematodes. These behavioral patterns, combined with seasonal shifts in resource availability, play a critical role in shaping host–parasite dynamics in arid and semi-arid ecosystems like the Balkhash-Alakol Basin.

Ecology, feeding behavior, and access to water sources have a significant impact on the seasonal and interspecific variation in parasite infection. These factors have been taken into account and thoroughly analyzed in the revised version of the manuscript, thereby enhancing the interpretative strength of the study.

#### Zoonotic Risk and Public Health Implications

The study revealed the presence of several zoonotic parasites, particularly in wild boars.

Cryptosporidium spp. –a waterborne protozoan that causes diarrhea in humans.

*Toxocara spp.* –a nematode with potential to cause visceral and ocular larva migrans.

Ascaris suum and Strongyloides stercoralis -well-documented zoonotic helminths.

These parasites pose a risk to rural communities and farmers, especially where there is overlap between wild animal ranges and human settlements or livestock pastures. Inadequate fencing, shared water sources, and the use of untreated animal manure as fertilizer can facilitate the transmission of zoonotic agents agents (Agustina *et al.*, 2023; Katakam *et al.*, 2016).

To mitigate these risks, the following measures are recommended:

- Veterinary inspection and sanitation of supplemental feeding sites for wildlife
- Environmental monitoring of water bodies used by both wildlife and livestock
- 3. Awareness campaigns in rural communities to promote hygiene and parasite prevention
- 4. Avoiding direct handling or consumption of raw meat/organs from wild animals without inspection

These steps are crucial in limiting spillover events and ensuring the health of both animals and humans in ecologically sensitive areas.

Methodological Relevance and Integrated Approach

The findings of this study underscore the value of integrated field-laboratory approaches in wildlife parasitology. Standardized fecal sampling, combined with morphological identification, remains a reliable and cost-effective method for detecting parasitic infections. This approach is particularly valuable in field settings where resources may be limited, and it continues to provide essential insights into host-parasite dynamics and ecological health (Hass *et al.*, 2024; Rojas *et al.*, 2024; Kamel and Abdel-Latef, 2021).

Over the past few decades, nucleic acid-based methods have been developed for the diagnosis of intestinal parasitic infections. Molecular diagnostics improves sensitivity and specificity in detecting parasitic infections, especially for species that are morphologically similar and difficult to differentiate using traditional microscopy (Verweij et al., 2014). We plan to incorporate these molecular tools in future studies.

## Ecological Impact and Conservation Context

Ecologically, parasite burdens can significantly influence wildlife population dynamics, individual fitness, and interspecific interactions. In arid environments such as the Balkhash region, where water and forage are scarce, even moderate levels of parasitism intensify physiological stress and reduce reproductive output. Previous studies have shown that parasitic infections alter host can survival, reproduction, and susceptibility other to

environmental pressures, thereby affecting population stability and ecological resilience (Louvard *et al.*, 2025; Lee, 2025; Dickinson *et al.*, 2024).

The observed prevalence rate across species (61.0%) –combining 2024 and 2025 data –supports the notion that parasitic infections are widespread and ecologically embedded in this arid environment. While most infections were subclinical, the potential impact on host fitness and interspecies interactions remains significant and warrants further long-term monitoring.

#### Limitations and Future Research Directions

We acknowledge several limitations.

Sampling was restricted to winter and summer seasons, as spring and autumn fieldwork was impeded by flooding and mud, limiting year-round surveillance.

Parasite identification relied primarily on morphological criteria; molecular tools (e.g., PCR) will be incorporated in future research.

The study focused on three host species; expanding to other mammals and birds would provide a broader ecological understanding.

Further research is needed to understand the dynamics of parasite transmission in these habitats and their implications for wildlife and human health. In conclusion, this study contributes valuable data on the parasitic infections of wild mammals in the Balkhash-Alakol region, providing a baseline for future ecological and epidemiological research. The detection of zoonotic parasites in wild boars emphasizes the need for continued surveillance and risk assessment, particularly in regions where wildlife habitats overlap with human agricultural activities. Furthermore, the ecological implications of parasitism underscore the importance of considering parasite dynamics in conservation and ecosystem health assessments.

#### Conclusion

This study provides novel and comprehensive insights into the parasitic fauna of wild mammals inhabiting the arid ecosystems of the Balkhash-Alakol Basin in southeastern Kazakhstan. The findings revealed a relatively high overall prevalence of parasitic infections (61.0%) and notable diversity of helminths and protozoa, particularly among wild boars (*Sus scrofa*) and Tolai hares (*Lepus tolai*), thus confirming the ecological importance of parasitism in wildlife populations under harsh environmental conditions.

A total of seven parasite taxa were identified across the study, with wild boars harboring five, including zoonotically significant species such as *Ascaris suum, Cryptosporidium suis*, and *Strongyloides stercoralis*. In contrast, no parasites were detected in Bactrian deer (*Cervus elaphus bactrianus*) during 2025, possibly due

to species-specific resistance, altered foraging behavior, preventive feeding measures, or seasonal variability in parasite shedding.

The study demonstrated the effectiveness of an integrated methodological approach, combining non-invasive fecal sampling, standard flotation and sedimentation techniques, and a patented modification to improve detection efficiency. This approach enabled reliable assessment of parasite prevalence and diversity under challenging field conditions.

From a public health perspective, the detection of zoonotic parasites in wild boars emphasizes the risk of cross-species transmission in areas where wildlife, livestock, and human activities overlap. These findings support the need for proactive surveillance and targeted interventions aimed at reducing environmental contamination and preventing spillover events.

The results of this research can serve as a scientific foundation for long-term ecological monitoring, wildlife health management, and the development of preventive strategies against parasitic infections in Kazakhstan's arid-steppe zones. Future studies should expand the seasonal and taxonomic scope of host species, incorporate molecular diagnostics for improved parasite identification, and explore environmental reservoirs and behavioral drivers of parasite transmission.

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#### **Author's Contributions**

Zhanteliyeva Laura: Conceived and supervised the research project on parasitic infections in wild animals. She was responsible for coordinating all aspects of the study, including research planning, sample collection, laboratory analysis, data interpretation, and manuscript drafting. She made critical revisions to the manuscript

following peer review and approved the final version for publication.

**Jussupbekova Nurgul**: Contributed to the critical review and scientific editing of the manuscript, including refinement of structure and argumentation, improvement of scientific style, and the accurate use of parasitological terminology and relevant literature. She helped ensure the clarity, logical consistency, and scientific integrity of the text, and approved the final version of the manuscript.

**Suleimenov Maratbek**: Contributed to the development of the research concept, hypotheses, and data analysis. He played a key role in interpreting the results, revising the manuscript for intellectual content, and providing critical insights during manuscript revision.

Omarhan Berkinbay Participated in fieldwork by collecting biological samples from wild animals and assisted in the preliminary identification and interpretation of parasitological findings. He made a substantial contribution to the *Materials and Methods* and *Results* sections, including performing statistical analysis using GraphPad Prism and providing detailed descriptions and interpretations of the data. He was actively involved in formulating the research topic, drafting the *Introduction*, and conducting an extensive literature review on parasitic diseases in wild animals in Central Asia and globally. Additionally, he contributed to the *Conclusion* section by partially writing and fully editing the final version. He also participated in the manuscript revision process.

Omarov Baizhan: Supervised and coordinated field activities, ensuring systematic collection of biological material from various wild animal species. He ensured adherence to veterinary and parasitological research protocols and was involved in manuscript revisions for intellectual content. Thoroughly addressed the reviewers' comments by expanding the methodology, clarifying key results, and refining the discussion. These changes significantly improved the rigor and clarity of the manuscript. The accepted article serves as evidence of these substantial improvements.

#### **Ethics**

The study did not involve any experiments on live animals or humans. Only non-invasive fecal sampling was conducted in compliance with national regulations and institutional guidelines. No ethical issues are anticipated following the publication of this manuscript.

#### **Conflict of Interest**

The authors declare that there are not conflicts of interest.

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