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The present situation of echinococcoses in Mongolia

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Abstract

This review presents the historical and current situation of echinococcoses in Mongolia. Since the collapse of the Soviet Union in 1991, Mongolia's health surveillance infrastructure has been very poor, especially as it pertains to chronic diseases, including neglected zoonotic diseases (NZDs). Although there is anecdotal evidence of people dying from hepatic disease due to infection with the larval stage of *Echinococcus* spp., there are very few published reports. All confirmed cases of echinococcoses in Mongolia are from hospitals located in the capital city of Ulaanbaatar. Cases of cystic echinococcosis (CE), caused by either *Echinococcus granulosus* sensu stricto or *Echinococcus canadensis* are believed to be relatively common throughout Mongolia. In contrast, cases of alveolar echinococcosis (AE), caused by *Echinococcus multilocularis*, are believed to be rare. Recent wild-animal surveys have revealed that wolves (*Canis lupus*) are the major definitive hosts of *E. canadensis*, whereas both wolves and red foxes (*Vulpes vulpes*) are the primary definitive hosts of *E. multilocularis*. Although wild-animal surveys have begun to elucidate the transmission of *Echinococcus* spp. in Mongolia, there have yet to be large-scale studies conducted in domestic dogs and livestock. Therefore, further epidemiological studies, in addition to education-based control campaigns, are needed to help combat this NZD.

Introduction

Echinococcoses, including cystic echinococcosis (CE) and alveolar echinococcosis (AE), are considered important yet neglected zoonotic diseases (NZDs) (Craig *et al.*, 1992, 2000, 2007; McManus *et al.*, 2003; Schantz *et al.*, 2003; Eckert & Deplazes, 2004; Ito *et al.*, 2006, 2013a; Budke *et al.*, 2006; Brunetti *et al.*, 2010, 2011; Torgerson *et al.*, 2010; Hotez & Alibek, 2011; Carmena & Cardona, 2013; Torgerson, 2013). The prevalence of these conditions is underestimated, due to a lack of reporting, in almost all developing endemic countries, including Mongolia. In addition to under-reporting, many endemic countries do not have access to appropriate diagnostic tools with which to confirm a diagnosis of CE or AE, with many cases likely misdiagnosed as liver cancer.

Due to their differing life cycles, distinct risk factors have been identified for the acquisition of CE and AE. Infection with CE has been associated with a pastoral life style, since the life cycle of *Echinococcus granulosus* sensu stricto is typically maintained between dogs and livestock. As the Mongolian people are largely nomadic, it is not unexpected that CE would be prevalent in this country (Ebright *et al.*, 2003). In contrast, AE is expected to be less common, since the *Echinococcus multilocularis* life cycle is typically maintained between wild canids (e.g. foxes and wolves) and small mammals. However, underdiagnosis may also contribute to the small number of reported AE cases (Ito *et al.*, 2014).

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While earlier works indicated the presence of echinococcoses in the former Soviet Union and Mongolia (Rausch, 1952, 1995, 2003; Abuladze, 1964; Bessonov, 1998, 2003), there are few actual case reports from this region (Bessonov, 1998, 2003; Wang *et al.*, 2005; Lukmanova *et al.*, 2007; Ito *et al.*, 2010, 2014; Jabbar *et al.*, 2011; Konyaev *et al.*, 2012a, b, 2013; Nakao *et al.*, 2013a, b). In fact, the number of AE and CE cases is likely underestimated for all of Central Asia (Torgerson & Budke, 2003; Hotez & Alibek, 2011; Torgerson, 2013; Zhang *et al.*, 2015). For example, while there are only approximately 200 AE cases confirmed in Kyrgyzstan, more recent reports indicate that this number should be closer to 700 (Sato, Japanese Embassy, Bishkek, pers. comm.). Similar situations are expected from other Central Asian countries, including Tajikistan, Turkmenistan, Uzbekistan and Mongolia (Torgerson *et al.*, 2010; Torgerson, 2013; Zhang *et al.*, 2015).

Furthermore, recent molecular studies have revealed that *E. granulosus* sensu lato is not a single species, but rather a complex of five independent species: *E. granulosus* sensu stricto (G1–3), *Echinococcus equinus* (G4), *Echinococcus ortleppi* (G5), *Echinococcus canadensis* (G6–10) and *Echinococcus felidis* (Nakao *et al.*, 2007, 2013a, b; Alvarez Rojas *et al.*, 2014), with CE not only caused by *E. granulosus* sensu stricto (G1–3) but also by *E. canadensis* (mainly G6/7) (Lavikainen *et al.*, 2006; Hüttner *et al.*, 2009; Saarma *et al.*, 2009; Šnábel *et al.*, 2009; Nakao *et al.*, 2010, 2013a, b; Omer *et al.*, 2010; Jabbar *et al.*, 2011; Hailemariam *et al.*, 2012; Ito *et al.*, 2013b, 2014; Konyaev *et al.*, 2013; Mbaya *et al.*, 2014; Monteiro *et al.*, 2014; Rodriguez-Prado *et al.*, 2014; Schurer *et al.*, 2014) and *E. ortleppi* (Bowles *et al.*, 1992; de la Rue *et al.*, 2011; Grenouillet *et al.*, 2014).

CE due to *E. canadensis* is rather common in Europe (Dybicz *et al.*, 2013), Central Asia (Ziadinov *et al.*, 2008; Van Kesteren *et al.*, 2013; Zhang *et al.*, 2015), China (Bart *et al.*, 2006; Zhang *et al.*, 2014; Ma *et al.*, 2015; Yang *et al.*, 2015) and Mongolia (Jabbar *et al.*, 2011; Ito *et al.*, 2014). Although CE cases are known to be caused by multiple species, *E. granulosus* s.s. is believed to result in the majority of human cases. However, since the species have different life cycles and means of transmission to humans, molecular identification of the causative species for human CE cases is essential. Molecular identification will allow for improved understanding of the disease's pathogenesis and better-targeted control measures, since imaging and serology, or even histopathology, cannot provide a definitive diagnosis of the causative species.

Review

In Mongolia, there are very few hospital-based reports of human echinococcoses (Ebright *et al.*, 2003; Abmed *et al.*, 2005; Gurbadam *et al.*, 2010; Ito *et al.*, 2010, 2014; Jabbar *et al.*, 2011) and only a few community-based screening studies (Watson-Jones *et al.*, 1997; Lee *et al.*, 1999; Wang *et al.*, 2005; Huh *et al.*, 2006).

Human echinococcoses

Hospital reports of human echinococcoses

The vast majority of clinical cases of echinococcoses in Mongolia are managed by surgeons. In 1950, 7.8% of all surgical patients in Mongolia were diagnosed with CE, whereas this value had decreased to 1.9% by 1990 (Cross, 1995; Davaatseren *et al.*, 1995; Abmed *et al.*, 2005). However, CE was diagnosed in 18% of surgical cases treated at the First Hospital of Ulaanbaatar in 1993 (Cross, 1995). While this value is very high, it most likely reflects the fact that this is a referral hospital and is much more likely to see CE cases compared to smaller, more rural hospitals. To date, all surgical echinococcosis cases have been confirmed to be due to CE except for five cases of AE (Davaatseren *et al.*, 1995; Ebright *et al.*, 2003; Gurbadam *et al.*, 2010; Ito *et al.*, 2010, 2011). The first case of AE was reported in 1982. However, very little demographic information is available about this case. The other four cases were confirmed in 2002, 2006, 2007 and 2009, with the patients born in the provinces of Orkhon-Uul, Uvs, Khovd and Bayan-Ulgii, respectively (fig. 1) (Ito *et al.*, 2010).

Molecular studies

There was no molecular identification of the causative species of echinococcoses in Mongolia until 2010. Molecular studies conducted on histopathological specimens from three AE patients (Ito *et al.*, 2010) revealed that the cases were caused by two distinct *E. multilocularis* genotypes, the Asian and Mongolian genotypes (Nakao *et al.*, 2009). In addition, molecular differentiation of CE cases caused by *E. granulosus* s.s. and *E. canadensis* G6/7 and G10 have been published (Jabbar *et al.*, 2011; Ito *et al.*, 2014) (fig. 2). These reports confirmed the presence of CE cases caused by both *E. granulosus* s.s. and *E. canadensis* from numerous provinces in western Mongolia. Specifically, CE cases due to *E. canadensis* (G10) have been confirmed from the provinces of Tuv (Ito *et al.*, 2013b) and Uvurkhangai (Jabbar *et al.*, 2011). In addition, CE cases caused by *E. canadensis* (G6/7) have been confirmed from 13 provinces, including Uvurkhangai. While human CE cases caused by *E. canadensis* (G10) have yet to be identified in Zavkhan, this species has been found in local wolves (*Canis lupus*) (Ito *et al.*, 2013b) (fig. 1). Therefore, human CE cases caused by *E. canadensis* (G10) are also likely to occur in this province.

Serological studies

Serology using recombinant antigen B (rAgB8/1) (Mamuti *et al.*, 2004) has been applied to CE cases in Mongolia. An enzyme-linked immunosorbent assay (ELISA) using rAgB8/1 positively identified 9 of 10 (90%) and 13 of 20 (65%) CE cases caused by *E. granulosus* s.s. and *E. canadensis* (G6/7), respectively (Ito *et al.*, 2014). Most of the evaluated CE cases were from Ulaanbaatar, where more than 50% of the population of Mongolia lives. All 18 CE cases in children were due to *E. canadensis*. Additional information is needed to evaluate differences in serological response, age and pathology of CE cases caused by *E. granulosus* s.s. and *E. canadensis*. Introduction of a rapid diagnostic kit (Ito & Budke, 2014) or routine ELISA and immunoblot using recombinant antigens would help improve diagnostics and case identification in Mongolia (Mamuti *et al.*, 2004, 2006, 2007; Tamarozzi *et al.*, 2013).

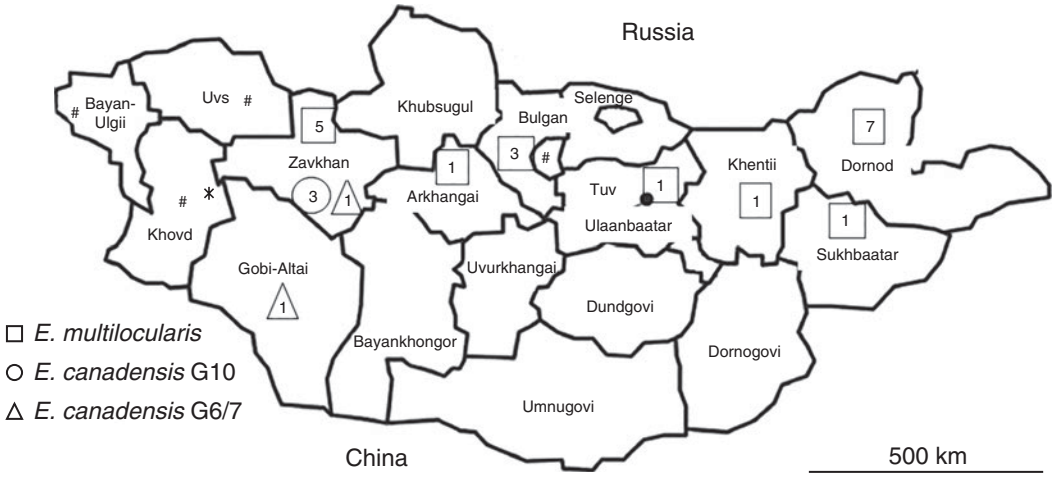

Fig. 1. Distribution of *Echinococcus* spp. from red fox (*Vulpus vulpus*) and wolves (*Canis lupus*) (Ito *et al.*, 2013b). *One lacustrine vole (*Microtus limnophilus*) confirmed to be infected with *E. multilocularis* (Gardner *et al.*, 2013). # Provinces where four AE patients were identified (Ito *et al.*, 2010).

Community surveys

There are a few older reports using the Casoni skin test to screen for echinococcoses in Mongolia (Jezek *et al.*, 1971, 1973). More recently, there have been several studies that have used an ELISA to evaluate Mongolian populations. One study identified 5.2% (17/334) of semi-nomadic pastoralists in Bayan-Ulgii as strongly seropositive against *E. granulosus* native Antigen B using an ELISA (Watson-Jones *et al.*, 1997). Another study found that 8.5% (12/141) of the inhabitants of rural areas near Ulaanbaatar (Lee *et al.*, 1999), 2.1% (4/187) of the inhabitants of Dornod in eastern Mongolia, and 11.7% (58/496) of the inhabitants of Selenge in north-central Mongolia were positive against crude hydatid cyst fluid by ELISA (Huh *et al.*, 2006). However, the reliability of serology is based on the quality of the diagnostic antigens and the type of control population used to evaluate the test and its findings (Mamuti *et al.*, 2002; Ito, 2013, 2015).

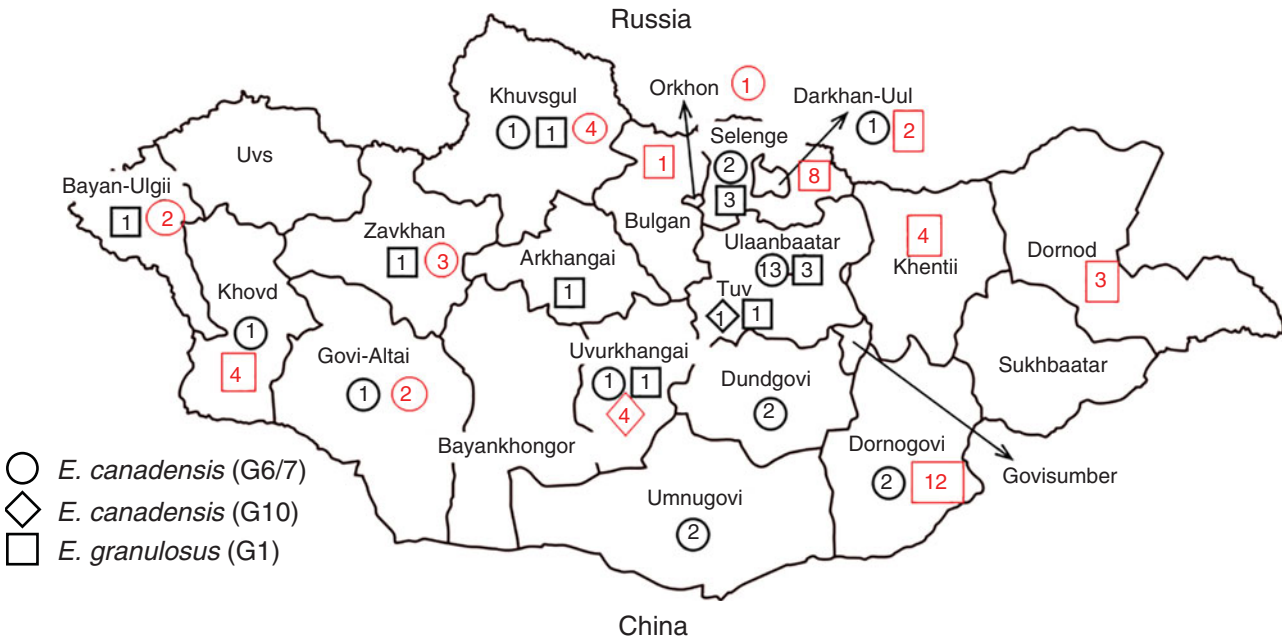

Fig. 2. Distribution of human CE cases in Mongolia differentiated into *E. granulosus* s.s., *E. canadensis* (G6/7) and *E. canadensis* (G10) (Jabbar *et al.*, 2011; Ito *et al.*, 2014). Black symbols represent samples from Ito *et al.* (2014) whereas red symbols represent samples from Jabbar *et al.* (2011).

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It is, therefore, important to confirm cases with ultrasound and histopathology whenever possible.

Animal echinococcoses in Mongolia

There is one review article focusing on the animal hosts of *Echinococcus* spp. in Mongolia (Abmed *et al.*, 2005). In addition, there are two reports of dog surveys for *Echinococcus* spp. in the country (Zoljargal *et al.*, 2001; Wang *et al.*, 2005). Zoljargal *et al.* (2001) used a copro-antigen test to evaluate 67 dogs and two red foxes in the town of Altai, with 17 dogs and one fox found positive. Since the monoclonal antibody used for this copro-antigen test was not species or genus specific, it was not possible to evaluate the degree to which false positives and false negatives were reported without additional molecular and/or necropsy evidence. In the second study, Wang *et al.* (2005) reported that 35.7% (5/14) of necropsied dogs were infected with *E. granulosus* s.s. (G1) in Bulgan Province.

Very little direct evidence exists of *Echinococcus* species infection in livestock. There is only a single study that reported the findings from sheep ($n = 590$), goats ($n = 338$) and cattle ($n = 779$) screened serologically using recombinant Antigen B (8/1) (rAgB8/1) (Chinchuluun *et al.*, 2014). All serum samples were collected from the serum bank at the Institute of Veterinary Medicine in Ulaanbaatar, with samples available from 19 of the 22 Mongolian provinces. Seropositive cattle were identified from 13 provinces, with 18.0% (9/50) of cattle samples from Ulaanbaatar seropositive. Since molecular studies have not been conducted in Mongolian cattle, it is not known if any of these infections were due to other species, such as *E. ortleppi* (G5).

While camels are an important livestock species in some regions of Mongolia, serological studies have not been conducted for camels since, at present, there is no good secondary antibody to detect antibody responses. However, camels have been found to be infected with *E. granulosus* s.l. in Mongolia (Chinchuluun, unpublished) and other endemic regions of the world. Therefore, molecular identification of *Echinococcus* spp. infection in all locally important livestock species is needed to better understand the life cycles of the circulating *Echinococcus* species.

Recent surveys of wild animals have revealed that both wolves (*C. lupus*) and red foxes (*Vulpes vulpes*) are definitive hosts of *E. multilocularis* in Mongolia, whereas thus far only wolves have been identified as definitive hosts of *E. canadensis* (G6/7 and G10) (Ito *et al.*, 2013b). The Mongolian *E. multilocularis* genotype has been found in wild canids throughout Mongolia (fig. 1). In contrast, *E. canadensis* has only been identified in a few provinces in western Mongolia. In 2012, Gardner *et al.* (2013) found one lacustrine vole (*Microtus limnophilus*) infected with *E. multilocularis* from Khovd Province (fig. 1). To the authors' knowledge, this is the only Mongolian small mammal confirmed to be infected with *E. multilocularis* (GenBank #AB271235) (Gardner *et al.*, 2013). Brandt' vole (*Microtus brandtii*) is a known intermediate host of the Mongolian genotype in Inner Mongolia, China (Tang *et al.*, 2001, 2004, 2006, 2007). However, to date, this small mammal species has not been identified as an intermediate host in Mongolia.

Echinococcoses in neighbouring countries

A number of studies evaluating the prevalence of AE and CE have been conducted in the neighbouring country of China (Craig *et al.*, 1992, 2000, 2006, 2008; Andersen *et al.*, 1993; Schantz *et al.*, 2003; Tiaoying *et al.*, 2005; T. Li *et al.*, 2008, 2011; Zhang *et al.*, 2014; D. Li *et al.*, 2015). However, only a few of these studies have identified the causative species of CE (Y.R. Yang *et al.*, 2005; Bart *et al.*, 2006; Li *et al.*, 2008; Nakao *et al.*, 2010; Zhang *et al.*, 2014; Ma *et al.* 2015; D. Yang *et al.*, 2015). As both CE and AE are highly endemic in the Chinese provinces of Xinjiang, Gansu and Inner Mongolia, which share a border with Mongolia, it is suspected that there are additional unreported cases occurring in the border areas of Mongolia.

Limited comparative studies are available on the frequency of *Echinococcus* spp. infection in ethnically Mongolian communities located in Mongolia and across the border in China. Wang *et al.* (2005) evaluated the ultrasound-based prevalence of human CE in the communities of Hobukesar, in Xinjiang Province, China and Bulgan, in western Mongolia, and found a significantly higher prevalence in the community located in China (2.7% (49/1844)) compared to the community in Mongolia (0.2% (4/1609)). This same study also evaluated necropsy-based infection prevalence in dogs located in these two communities, but did not find a significant difference in the frequency of infection. The authors attributed the lower prevalence of CE in residents of Bulgan to Soviet Union administered dog deworming programmes that were common in Mongolia until the mid-1980s. There are relatively few reports, in the English language literature, on CE and AE in bordering regions of Russia (Rausch, 1952, 1995, 2003; Abuladze, 1964; Bessonov, 1998, 2003; Konyaev *et al.*, 2012a, b, 2013). However, recent molecular studies have confirmed the presence of all four *E. multilocularis* genotypes in Russia, as well as the presence of *E. granulosus* s.s. and *E. canadensis* (G6/7 and G10) (Nakao *et al.*, 2013b).

General discussion

There is now evidence that both *E. granulosus* s.s. and *E. canadensis* G6/7 and G10 are distributed in Mongolia (Jabbar *et al.*, 2011; Ito *et al.*, 2013b, 2014). However, additional species such as *E. ortleppi* may yet be detected. Due to the presence of these zoonotic parasites, there is an urgent need to establish a centralized repository for data on Mongolian echinococcoses cases. As part of this initiative, all histopathology specimens should be further identified using molecular tools. Questions remain on whether or not cases of CE caused by *E. canadensis* are more benign compared to cases caused by *E. granulosus* s.s. Evidence from Alaska and Canada also suggests that lung cysts caused by *E. canadensis* are typically smaller than those caused by *E. granulosus* s.s. (Wilson *et al.*, 1968; Pinch & Wilson, 1973; Finlay & Speert, 1992; Lamy *et al.*, 1993; Rausch, 2003). Thus far, there has been no differentiation between CE cases caused by *E. granulosus* s.s. and *E. canadensis* when it comes to standardization and assessment of pathology, imaging, treatment and evaluation of serology (Ito, 2015). Therefore, Mongolia

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may be an ideal location to investigate these differences between the circulating *Echinococcus* species.

Further systematic studies are essential to better elaborate the epidemiology of, and to guide control measures for, echinococcoses in Mongolia. Although CE caused by both *E. canadensis* and *E. granulosus* s.s. is found in residents of Ulaanbaatar, the age distribution of cases appears to differ between the two species (Ito *et al.*, 2014). Thus far, there have been no studies to try to identify risk factors associated with *Echinococcus* spp. infection in and around Ulaanbaatar. Evaluation of stray dogs and wild canids for intestinal infection, and molecular differentiation of the infecting species, would also aid in identifying how human infection may be occurring locally.

Human AE cases from Mongolia have been confirmed to be caused by both the Mongolian and Asian genotypes (Ito *et al.*, 2010). Bretagne *et al.* (1996) was the first to report the existence of three different *E. multilocularis* genotypes (North American, Asian and European). The Mongolian genotype was first reported from Inner Mongolia, China as the Inner Mongolian genotype (Nakao *et al.*, 2009). Prior to molecular characterization, the parasite had been described as the new species *Echinococcus sibiricensis* or *Echinococcus russicensis* (Tang *et al.*, 2001, 2004, 2006, 2007). However, molecular studies revealed that it was, in fact, an intra-species variant of *E. multilocularis* (Nakao *et al.*, 2007, 2009).

Thus far, all *E. multilocularis* adult worms from red foxes in Mongolia have been the Mongolian genotype (Ito *et al.*, 2013b), with 7.9% (15/191) of red foxes, but no corsac foxes (0/111), found to be infected with *E. multilocularis*. However, since there has been a confirmed case of human AE in Mongolia caused by the Asian genotype (Ito *et al.*, 2010), Mongolian wild canids are expected to be infected with this genotype as well. The Mongolian and Asian genotypes have been identified from corsac foxes in Inner Mongolia, China (Tang *et al.*, 2004).

The main definitive host of the Mongolian *E. multilocularis* genotype has been hypothesized to be the red fox. However, additional molecular testing of adult worms from both red and corsac foxes is needed to confirm or reject this hypothesis. This is especially true since worm abundance may be smaller in corsac foxes as compared to red foxes, and currently available tools (Matoba *et al.*, 2006) may not be sensitive enough to detect very small worm burdens (Ito *et al.*, 2013b). Additional wild canid studies need to be conducted to determine which animals are currently acting as definitive hosts for *E. multilocularis* in Mongolia.

Human echinococcoses may be substantially underdiagnosed in Mongolia, due to the chronic nature of the disease and the inability of patients to seek care in the large referral hospitals in Ulaanbaatar. Therefore, community-based studies using serology and diagnostic imaging are needed to better elaborate the frequency of infection in the population. Stray dogs were routinely killed when Mongolia was strongly influenced by the former Soviet Union (Wang *et al.*, 2005). While this practice might seem cruel to some, it did help control a number of dog-associated zoonotic diseases, including echinococcoses. In an effort to decrease disease spread among dogs and to people, the city council of Ulaanbaatar re-introduced the culling of stray dogs in 2013. However, no parasitological studies have been associated with the culling programme.

Hegglin *et al.* (2015) and Liccioli *et al.* (2015) have discussed that the urban red fox population is increasing in many European cities, as is the risk of AE to those who live in these cities. As Mongolia industrializes, the influx of foxes and other wildlife into the cities will also become more of a problem. It is, therefore, essential that the population is educated about the disease risks that foxes and other wildlife present. Along those same lines, the importance of wolves and free-roaming dogs as definitive hosts should be recognized.

Perspectives

Mongolia currently lacks the resources to assess and address NZDs, including echinococcoses, adequately. Thus far, three expert meetings on echinococcoses have been conducted in Ulaanbaatar, in June 2009, 2012 and 2014 (Gurbadam *et al.*, 2010). In addition, the World Health Organization (WHO) held a one-day meeting on NZDs in September 2012. To date, most human AE and CE data are from the major referral hospitals (State Central First Hospital and National Centre of Maternal and Child Health, etc.) and the National Centre of Pathology in Ulaanbaatar. The National Centre of Pathology has been strongly encouraged to conduct molecular identification of all *Echinococcus* spp. specimens that it receives. In the spirit of One Health, all personnel involved in the evaluation and control of echinococcoses in Mongolia (National Centre of Communicable Diseases, Mongolian National University of Medical Sciences, National Centre of Zoonotic Diseases, National Institute of Veterinary Medicine, etc.) should work together for the common good. A WHO-supported project aimed at the control of echinococcoses in Mongolia, which would provide education to the general population and train local researchers towards the goal of controlling these diseases, would be very beneficial.

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Conflict of interest

None.

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