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## Epidemiology of fish borne trematodiasis in Kazakhstan

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#### ABSTRACT

Fish borne trematodes are an important cause of morbidity in Kazakhstan. The number of human cases of opisthorchiidosis (infection with parasites of the family opisthorchiidae) reached a peak of 2521 recorded cases (17 cases per 100,000) in 2002 with a gradual decline to 1225 cases (7.4 cases per 100,000) in 2011. Most human cases are found in the north and north east part of Kazakhstan in areas drained by the Irtysh River and it tributaries. A further focus is found in the north west in the Ural river basin in the European part of Kazakhstan. The most common occupations of patients with opisthorchiidosis included the manual laborers, those employed in the home or unemployed. Necropsy investigations of village dogs in an endemic region revealed 37 of 51 (72%) village dogs infected with either Opithorchis felineus or Methorchis bilis. Likewise an investigation of 242 cats consisting of strays, village, suburban and city cats revealed 79 (33%) animals infected with O. felineus. Higher prevalences were seen in strays and village cats compared to suburban cats. No urban cats, which lived in apartments, were found to be infected. Other important zoonoses included Echinococcus granulosus, detected in 2 of the 51 necropsied village dogs and E. multilocularis was found in 2 out of 124 necropsied stray cats. Investigations of locally caught fish revealed 10 of 107 (9%) roach (Rutilus rutilus), 49 of 68(72%) ide (Leuciscus idus) and 2 of 79 (2.5%) bream (Abramis brama) infested with trematode metacercariae. No metacercariae were found in 609 crucian carp (Carassius carassius), 35 tench (Tinca tinca), 79 carp (Cyprinus carpio), 46 perch (Perca fluviatilis) or 20 zander (Sander lucioperca).

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#### 1. Introduction

Food borne trematodes are a number of parasitic species transmitted by the consumption of undercooked, usually aquatic products. (Keiser and Utzinger, 2009) and infections with such trematodes cause a high global burden of disease. Over 56 million people are infected with food borne trematodes, 7.9 million with severe sequelae. Over 7000 deaths per year can be attributable to infection with foodborne trematodes (Fürst et al., 2012). The Wold Health Organization also recognises food borne trematode infections as a neglected tropical disease (World Health Organisation, 2010).

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Kazakhstan is the largest landlocked country in the world with the western most district being geographically part of Europe and eastern districts bordering China. The country has an area of 2.7 million square kilometers and a population of approximately 17 million, of which 46% is rural, and population density of 5.9 per km². Agriculture represents 5% of economic output. The climate is continental, with warm summers and colder winters. Precipitation varies between arid and semi-arid conditions.

Opisthorchis felineus is known to be endemic in the new independent states of the former Soviet Union (Ilyinskikh, 2002; Kochetkov, 2010; Syskova et al., 2001). The first intermediate hosts of O. felineus are species of freshwater snails from the genus Bithynia (Brusentsov et al., 2013; Mordvinov et al., 2012). Fish of the Cyprinidae family serve as the second intermediate hosts (Pozio et al., 2013). Metacercariae, the encysted and resting stage of a fluke larva, accumulate in muscles of susceptible fish. When final hosts ingest raw fish, the young trematode escapes from the metacercarial cyst and migrates through the common bile duct into the bile duct and gall bladder,

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where it develops into an adult. Mature flukes live in the liver of definitive hosts (fish-eating mammals, including humans). Human infections occur by consuming raw or undercooked fish infested with metacercariae.

O. felineus affects 1.6 million people (Yossepowitch et al., 2004). In humans, acute O felineus infection may result in hepatitis-like symptoms including fever abdominal pain, nausea and emesis. In chronic infections, inflammation, fibrosis and obstruction of the biliary tract, liver abscesses, pancreatitis and supperative cholangitis may occur (Keiser and Utzinger, 2009). The carcinogenicity of chronic O. felineus infections is yet to be proven. However, available epidemiological and clinical data in humans and animals suggest that O. felineus can be the cause of neoplasia. There is a clear increase in the incidence cholangiocarcinoma in endemic areas of Siberia. In Russia, the highest incidence of bile duct cancer in humans was documented in the same area (i.e., Tyumen Region) with the highest incidence of O. felineus infection in humans (Mordvinov et al., 2012) and a previous estimates of the health impact of this parasite have assumed that infection carries the same relative risk of carcinogenesis as O. viverrini infection (Parkin, 2006).

The parasite has been reported to be endemic in Aktobe, Karadanda, Pavlodar, Akmola and Kostanay districts (World Health Organisation, 1995). Wild caught fish are commonly consumed raw or slightly salted. Consequently, there have previously been a number of reports in the Russian literature on opithorchosis in humans, dogs and cats. Pantiykhov and Vinogradova (1963) described up to 100% of cats and 70% of dogs infected in the Pavlodar and Semipalatinsk regions. In addition there were a large number of human cases of infection diagnosed in Pavlodar which were associated with the habit of eating salted dried fish. In some villages along the Irtysh river human prevalences of 42% have been recorded in the past (Terentyeva, 1985) and Zhumabekova and Kamyeshyeva (2008) reported that the annual incidence of human opithorchosis had increased from under 10 per 100,000 to over 140 per 100,000 between 1995 and 2006 in the Kashir district (population 21,000). Studies of fish from the Irgiz and Turgay river basins have previously found the prevalence of infection in ide of up to 50% (Khokhalkova, 1957). Older data has indicated foci of opithorchosis in the Karaganda and Tamirtau regions (Kreptogorskaya, 1932). More recently there is a report of 4.5% of Prussian carp (Carassius gibelio) recovered from the Baikon lakes, Kashir district in the Pavoldar Oblast in north Easter Kazakhstan were infected with metacercariae of Opithorchis felineus (Zhumabekova and Kamyeshyeva, 2008).

The objective of this study was to report the incidence of infection in humans in Kazakhstan. This study reports the numbers of officially reported cases of human opisthorchiidosis in Kazakhstan between 1997 and 2011 and analyses the trends in the disease. We also give details of the geographical origin and occupations of cases since 2007. Therefore a second objective to this study was to investigate the prevalence of zoonotic trematodes in potential definitive and intermediate hosts in one of these endemic districts, namely Akmola district. Consequently, we report surveillance results from domestic definitive and intermediate hosts of *O. felineus* from the northern part of Kazakhstan.

#### 2. Materials and methods

#### 2.1. Human data

Human faecal samples are supplied by patients who are suspected clinical cases of opisthorchiidosis (or indeed possibly other diseases) and the samples submitted to regional diagnostic laboratories by the patients' physician between 1997 and 2011. The Kato-Katz technique and/or the ether sedimentation technique (as

described by WHO, 1991) was used for routine diagnosis of human helminthoses. All data is sent to the Scientific Practical Center of Sanitary and Epidemiological Expertise and Monitoring, Almaty. From these records data human opisthorchiidosis are reported together with details of the origin of the case and occupation.

#### 2.2. Dogs

A total of 51 dogs from villages were necropsied from rural areas of Akmola district. These dogs were from a village environment and euthanised at the request of their owners and hence were a convenience sample. All helminth parasites were removed from the gastrointestinal tract and liver and identified morphologically, with the use of microscopy where necessary. The bile ducts were incised specifically to find any infection with hepatic trematodes. Total parasite counts of each species identified were recorded.

#### 2.3. *Cats*

We undertook necropsy of 242 cats from rural areas of Akmola district and from Astana. Astana, the capital of Kazakhstan is completely surrounded by Akmola district. These cats were urban cats living in apartments (n=56), suburban cats (n=85), village cats (n=128) or stray cats (n=72). All cats were humanely euthanised by the request of their owner, and were also inevitably a convenience sample. All helminth parasites were recovered from the gastrointestinal tract and liver and identified.

#### 2.4. Fish

In total we investigated 1043 specimens of freshwater fish representing 8 species. Fish were purchased from various markets and shops in Astana or Akmola district, or caught directly from the Korgaldjin lakes (also in the Akmola district). The species of freshwater fish examined were carp (Cyprinus carpio), crucian carp (Carassius carassius), roach (Rutilus rutilus), tench (Tinca tinca), ide (Leuciscus idus), rudd (Scardinius erythropthalmus), bream (Abramis brama), perch (Perca fluviatilis) and zander (Sander lucioperca). Visual inspection and microscopy was performed to the internal organs of the body cavity, the eye and the edible parts of the fish's body (muscles). The skin was separated from underlying subcutaneous tissue. Thin layers of muscle of no more than 2-3 mm thickness were scraped off with a sharp scalpel or cut and squeezed between glass slides and examined microscopically. To identify the larval stages of the parasites in the muscles of fish examined by the compressor. The parasites in fish were identified microscopically according to Bayer and Shulman (1984). The studies on dogs, cats and fish were undertaken between 2008 and 2010.

#### 2.5. Statistical analysis

Trends in the numbers of human cases were examined by  $\chi^2$  for trend. Abundances and prevalences and their confidence intervals were examined by generalized linear models using R (R Core Team, 2013).

#### 2.6. Ethical statement

All euthanised animals were at the specific request of their owners with a further request to dispose of the animal carcass with permission given for collection of any relevant samples for surveillance purposes. All human data was from routine collected surveillance data and anonymised so that no individual patient details can be identified.

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