



An Evaluation of Transmission Dynamics of *Cryptosporidium* Using Molecular Methods

Dear Editor-in-Chief,

The members of genus *Cryptosporidium* are intracellular parasites that infect mammals, poultry, reptiles and amphibians. From the total of 30 valid species mentioned currently, 14 have been determined to infect the human being. Two species, *Cryptosporidium hominis* (*C. hominis*) (Anthroponotic species) and *Cryptosporidium parvum* (*C. parvum*) bovine genotype (Zoonotic species), are considered of major public health importance¹.

Commonly, the ubiquitous oocysts of *Cryptosporidium* are transmitted *via* direct contact with infected hosts or indirectly *via* contaminated food and water². The low infectious dose and its resistance against common water disinfectants make it a challenge for the drinking water plants³. Common laboratory techniques which are being used for diagnosis of *Cryptosporidium* cannot discriminate it at species and genotype level. However, the genetic tools allow species determination of the parasite as well as tracing its transmission routes².

In this study, a total of 55 drinking water samples were collected from 11 different areas of Tabriz, the largest city in North West of Iran. Each sample contained 30 L of water. To collect the suspended particles, samples were filtered through a membrane filter with 1.2 μm pore size (Sartorius, Germany). The pellets trapped on the filter were collected⁴. All water pellets were subjected to DNA extraction by a method described previously⁵. Then, the amplification of small ribosomal subunit RNA (SSU-rRNA; 18S rRNA) gene was done using a two step nested PCR method⁶. A sample of *C. parvum* DNA that was extracted by the extraction method was included in each round of PCR as a positive control. The multi copy nature of 18S rRNA gene and nested format of the PCR make it a very sensitive method for detection of *Cryptosporidium* oocysts in water samples⁷. However, the expected amplicon (826-864-bp) was not detected in any of the water samples.

Various reports from many areas of the world provide strong evidence that contaminated water is an important risk factor for cryptosporidiosis⁸. Several factors, including water source, location of sampling, number and volumes of samples, type of ecosystem, climate and detecting procedures are effective for detection of *Cryptosporidium* oocysts in the water samples⁹. The drinking water supplied from surface water sources is more susceptible to contamination. Thus, underground water is a more protected source¹⁰. In our study area, the main part of the drinking water supplies comes from rivers.

Table 1. *Cryptosporidium* in different sources and identified species

Sample types	Sample size	Frequency (%)	<i>Cryptosporidium</i> species
Children (2)	113	2 (1.6)	<i>C. parvum</i>
Cattle (6)	104	11 (10.5)	<i>C. parvum</i> <i>C. andersoni</i>
Drinking water	55	0.0 (0.0)	-

The role of rainfall as a determining risk factor for the waterborne transmission of *Cryptosporidium* can be significant. The relationship between increased rainfall and an increase in the concentration of *Cryptosporidium* oocysts in nearby river waters has been reported¹⁰. The North West part of Iran is an area with lower than average rainfall, which could be the most important reason for the lack of parasites in the water.

There are not many reports about the prevalence of *Cryptosporidium* in water samples in Iran. Meanwhile, the prevalence of *Cryptosporidium* on the surface and recreational water was 36 and 20%, respectively^{4,9}. The two researches conducted in Ardabil and Chaharmahal va Bakhtiyari provinces as well as our study are limited studies with small sample size. Thus, more comprehensive studies with large samples from different sources and in different seasons are required to assess the real risk of waterborne cryptosporidiosis in Iran.

Previous studies in Tabriz showed that 1.76% of diarrheic children and 3.8% of cattle have been infected with *C. parvum*^{2,6} (Table 1). The presence of *C. parvum* in children, as a sensitive group and in cattle, as a major source for zoonotic disease may be associated with zoonotic transmission of the parasite in the study area. Lack of parasite in drinking water may indicate that this cannot be an important route for transmission; instead, it can be a reason for the low prevalence of the infection in children. Lack of *C. hominis* (Anthroponotic species) in children and the prevalence of *C. parvum*, potentially zoonotic species, in cattle and its presence in diarrheic rural children would raise the possibility that zoonotic transmission originally occurs through direct contact with farm animals in this region. Therefore, cattle and other domestic animals should be considered as important sources of infection in the North-western part of Iran.

Conflict of Interest

All authors declare that there is no conflict of interests.

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