

## A wide diversity of zoonotic intestinal parasites in domestic and stray dogs in rural areas of Kermanshah province, Iran

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**Abstract.** Dogs can act as reservoirs, carriers, and transmitters of several zoonotic intestinal parasites that can cause serious health problems for humans. The aim of this study was to determine the prevalence of intestinal parasites in dogs in Kermanshah Province, west of Iran. Faecal samples were collected from domestic and stray dogs from 30 rural areas of Kermanshah province from August 2014 to April 2015 and were analyzed by formalin-ether sedimentation, sucrose flotation technique and the modified Ziehl-Neelsen method. Out of 301 dogs examined, 230 (76.4%) were infected with at least one parasite. The incidence on the different types of intestinal parasitic species recovered from domestic and stray dogs are as follows: *Toxocara leonina* (20.8% and 27.6%), *T. canis* (7.5% and 9.4%), *Taenia* spp. (9.2% and 9.4%), hookworm spp. (18.3% and 33.7%), *Capillaria* spp. (0.8 and 1.7%), *Dicrocoelium dendriticum* (0.8% and 3.3%), *Fasciola* spp. (0.8% and 2.2%), *Acanthocephal* spp. (3.3% and 5.5%), *Trichuris vulpis* (0.8% and 1.7%), *Dipylidium caninum* (4.2% and 3.3%), *Physaloptera* spp. (6.7% and 6.6%), *Cryptosporidium* spp. (21.7% and 25.4%), *Eimeria* spp. (35.0% and 34.3%), *Giardia* spp. (6.7% and 12.7%), *Cystoisospora* spp. (7.5% and 5.5%), *Blastocystis* spp. (18.3% and 20.4%) and *Sarcocystis* spp. (6.7% and 7.2%), respectively. Significant difference in infection rates was observed between domestic and stray dogs ( $P>0.05$ ). Hookworm and *Eimeria* spp. were the most common intestinal helminth and protozoa detected with 83 (27.6%) out of 301 and 104 (34.6%), out of 301, respectively. There was no significant difference in prevalence and distribution of intestinal parasites between male and female dogs ( $P>0.05$ ). The wide range of zoonotic parasites indicated that people residing in rural areas in Kermanshah province are at risk of exposure to these pathogens. In this respect, appropriate implementation of control programs by public health authorities and veterinarians should be taken into account.

### INTRODUCTION

In Iran, action by government in providing information regarding the risks of zoonotic diseases transmitted by domestic dogs, and control of stray animals are practically non-existent, resulting in an increasing risk of exposure to zoonotic diseases transmitted by these animals (Traub *et al.*, 2005). Intestinal

parasites affecting dogs, i.e. roundworms, hookworms, whipworms, and protozoa have health-risk for animals and, especially to human beings. Both dogs and humans are infected by ingesting infective stages, (i.e. larvated eggs or larvae for helminths and cyst or oocysts for protozoa) which are present in the environment (Soriano *et al.*, 2010a). Young children, pregnant women,

immunosuppressed and elderly people are particularly at risk of severe disease (Irwin, 2002). Stray dogs constitute important source of human infection mainly due to environmental contamination with faeces containing infective parasitic forms (eggs, larvae or oocysts) (Okoye *et al.*, 2011).

Stray dogs are largest group of dogs in both urban and rural areas of Iran that they can usually become infected by roaming around human homes and feeding of residues and contaminated with these in some areas. Dogs are considered to be the main source of transmission of parasitic disease to human and livestock due to their ability to spread the parasite eggs and oocysts over a wide range of areas, especially where herbivores graze (Ranjbar-Bahadori *et al.*, 2008; Harandi *et al.*, 2011). Information regarding prevalence on various parasites of dogs are valuable to human health care providers, and is crucial for planning control programs to minimize the risk of transmitting diseases from dogs to humans and other animals (Rokni, 2009; Joffe *et al.*, 2011). This study aims to determine the prevalence of intestinal parasites in domestic and stray dogs in villages of Kermanshah province, west of Iran.

## MATERIALS AND METHODS

### *Study area*

The study was carried out from August 2014 to April 2015 in Kermanshah province, situated in West of Iran which spreads over an area of 25,000 km<sup>2</sup> which included 1.5 percent of the total area of the country. According to the 2011 census, its population was 1.945 million. This province has a cold climate and mountainous with a partially cold winter and rainy spring. The annual rainfall is 300-500 mm with the average temperature 15.6°C in spring, 25.3°C in summer, 11.3°C in autumn and 3.3°C in winter.

### *Sample collection and parasitological procedures*

The prevalence of intestinal parasites was determined in stray and domesticated dogs based on fecal examination of 301 samples

from 30 rural areas that were randomly selected using a cluster sampling method.

Fresh fecal samples of dogs were collected from 120 home dogs randomly and 181 fecal samples collected from stray dogs after defecation. Due to the lack of information with regards to age of dogs, age was not investigated in this study. Five grams of feces was collected from each dog and placed into sterile containers that were labeled and stored at -20°C for further examination. One gram of each fecal sample was examined by formalin-ether sedimentation technique to find eggs of helminths (Allen & Ridley, 1970). Two slides, each smeared with 20 µl of sediment were used for wet mount with and without Lugol's iodine stain. The slides were examined under the light microscope using 100X and 400X magnification. With formalin-ether sedimentation technique protozoa can be investigated in feces but for better identification protozoan, cysts and oocysts, fecal samples were examined using a sucrose flotation technique with specific density of 1.2 g ml<sup>-1</sup> as described by Beirumvand *et al.* (2013). A 20 µl of formalin-ether sediment spread on a clean microscope slide and then air dried at room temperature and slides were stained by the modified Ziehl-Neelsen method (Garcia *et al.*, 1983). Using a light microscope, the entire smear was examined at 100 × magnification for *Cryptosporidium* oocysts. Base of the size and shape of parasites that are seen under the light microscope, eggs and oocysts are differentiated.

### *Statistical analysis*

Statistical analysis was performed using SPSS version 16 software (SPSS Inc., Chicago, IL). Chi-square ( $\chi^2$ ) was used to evaluate the prevalence of infection and difference between domestic and stray dogs and *P*-values of less than 0.05 were considered as significant statistically.

## RESULTS

Faecal examination showed that 230 (76.4%) out of 301 dogs that included 84 of 120

domestic dogs (70.0%) and 146 of 181 stray dogs (80.7%) were positive with one or more of species of parasites. Significant difference in infection rates with intestinal parasites were observed between domestic and stray dogs ( $\chi^2=5.08$ , d.f.=1 and  $P=0.024$ ). Hookworm and *Eimeria* spp. were the most common intestinal helminth and protozoa with 83 (27.6%) and 104 (34.6%), respectively. *Capillaria* spp. and *Trichuris vulpis* with 4 (1.3%) and *Cystoisospora* spp. with 19 (6.3%) had the lowest prevalence, respectively (Table 1). The prevalence of hookworm was significantly different between domestic and stray dogs ( $\chi^2=8.53$ , d.f.=1 and  $P=0.003$ ) and other parasites revealed no significant difference. The distribution pattern of single and multiple infections of the various intestinal parasites are presented in Table 2. One hundred seventy four dogs (76.4%) had multiple infections. Of the 301 dogs, 56 (18.6%) were infected with one, 70 (23.3%) with two, 55 (18.3%) with three, 29 (9.6%) with four, 15 (5.0%) with five and 5 (1.7%) with more five species of intestinal parasite (Table 2).

Statistical analysis revealed that the distribution pattern of infection rate in domestic and stray dogs was not significant.

The prevalence of the various intestinal parasites recovered in relation to gender of dog is presented in Table 3. There was no significant difference in prevalence of intestinal parasites in male and female dogs ( $P>0.05$ ).

## DISCUSSION

Domestic dogs are not restricted to the house compound and are allowed to roam freely with stray dogs and increases the risk of zoonotic infection in rural habitats. In this study, the prevalence of intestinal parasites in dogs (76.4%) was high compared to other parts of Iran, such as 39.0% in Moghan plain (Northwest) (Zare-Bidaki *et al.*, 2010), 29.2% in Khorasan Razavi province and 66.0% in Chenaran County (Northeast) (Razmi, 2009; Beiomvand *et al.*, 2013), 7.1% in Kerman (South) (Mirzaei and Fooladi, 2012), But was relatively low compared with 80.0% Garmsar County (Central) (Eslami *et al.*, 2010), 90.0% Sari (North) (Gholami *et al.*, 2011), 83.0% Ilam city (west) (Abdi *et al.*, 2013). Also, the infection rate observed in dogs was higher compared to other countries such as Greece (26.0%) (Papazahariadou *et al.*, 2007),

Table 1. Intestinal parasites isolated from domestic and stray dogs in villages of Kermanshah province, Iran

Parasite species	Domestic dogs (n=120)	Stray dogs (n=181)	Total (n=301)	95% CI* of total infection	<i>p</i> -value†
<i>Toxocara leonina</i>	25 (20.8)	50 (27.6)	75 (24.9)	20-29	Ns
<i>Toxocara canis</i>	9 (7.5)	17 (9.4)	26 (8.6)	5-11	Ns
<i>Taenia/Echinococcus</i> spp.	11 (9.2)	17 (9.4)	28 (9.3)	6-12	Ns
Hookworm	22 (18.3)	61 (33.7)	83 (27.6)	22-32	0.003
<i>Capillaria</i> spp.	1 (0.8)	3 (1.7)	4 (1.3)	0-2	Ns
<i>Dicrocoelium dendriticum</i>	1 (0.8)	6 (3.3)	7 (2.3)	0-4	Ns
<i>Fasciola</i> spp.	1 (0.8)	4 (2.2)	5 (1.7)	0-3	Ns
<i>Acanthocephal</i> spp.	4 (3.3)	10 (5.5)	14 (4.7)	2-7	Ns
<i>Trichuris vulpis</i>	1 (0.8)	3 (1.7)	4 (1.3)	0-2	Ns
<i>Dipylidium caninum</i>	5 (4.2)	6 (3.3)	11 (3.7)	1-5	Ns
<i>Physaloptera</i> sp.	8 (6.7)	12 (6.6)	20 (6.6)	3-9	Ns
<i>Cryptosporidium</i> spp.	26 (21.7)	46 (25.4)	72 (23.9)	19-28	Ns
<i>Eimeria</i> spp.	42 (35.0)	62 (34.3)	104 (34.6)	29-39	Ns
<i>Giardia</i> spp.	8 (6.7)	23 (12.7)	31 (10.3)	6-13	Ns
<i>Cystoisospora</i> spp.	9 (7.5)	10 (5.5)	19 (6.3)	3-9	Ns
<i>Blastocystis</i> spp.	22 (18.3)	37 (20.4)	59 (19.6)	15-24	Ns
<i>Sarcocystis</i> spp.	8 (6.7)	13 (7.2)	21 (7.0)	4-9	Ns

\* Condence intervals.

†:  $\chi^2$ -test; Ns: Not significant ( $P > 0.05$ ).

Table 2. Number of single and multiple infections in dogs in villages of Kermanshah province, Iran

	Parasite speciesNumber of parasite species in total dogs (%)						Total
	1	2	3	4	5	>5	
<i>Toxocara leonina</i>	7 (9.3)	24 (32)	16 (21.3)	14 (18.7)	9 (12)	5 (6.7)	75 (100)
<i>Toxocara canis</i>	3 (11.5)	3 (11.5)	12 (46.2)	3 (11.5)	3 (11.5)	2 (7.7)	26 (100)
<i>Taenia/Echinococcus</i> spp.	5 (17.9)	4 (14.3)	9 (32.1)	6 (21.4)	2 (7.1)	2 (7.1)	28 (100)
Hookworm	4 (5.5)	19 (26)	29 (39.7)	19 (26)	8 (11)	4 (5.5)	73 (100)
<i>Capillaria</i> spp.	0 (0)	0 (0)	1 (25)	2 (50)	1 (25)	0 (0)	4 (100)
<i>Dicrocoelium dendriticum</i>	0 (0)	1 (14.3)	1 (14.3)	3 (42.9)	1 (14.3)	1 (14.3)	7 (100)
<i>Fasciola</i> spp.	0 (0)	0 (0)	2 (40)	1 (20)	0 (0)	2 (40)	5 (100)
<i>Acanthocephal</i> spp.	0 (0)	3 (21.4)	2 (14.3)	3 (21.4)	5 (35.7)	1 (7.1)	14 (100)
<i>Trichuris vulpis</i>	0 (0)	0 (0)	1 (25)	1 (25)	1 (25)	1 (25)	4 (100)
<i>Dipylidium caninum</i>	1 (9.1)	2 (18.2)	2 (18.2)	3 (27.3)	3 (27.3)	0 (0)	11 (100)
<i>Physaloptera</i> sp.	1 (5)	4 (20)	5 (25)	5 (25)	3 (15)	2 (10)	20 (100)
<i>Cryptosporidium</i> spp.	6 (8.3)	19 (26.4)	23 (31.9)	12 (16.7)	9 (12.5)	3 (4.2)	72 (100)
<i>Eimeria</i> spp.	16 (15.4)	23 (22.1)	32 (30.8)	20 (19.2)	10 (9.6)	3 (2.9)	104 (100)
<i>Giardia</i> spp.	3 (9.7)	10 (32.3)	7 (22.6)	4 (12.9)	6 (19.4)	1 (3.2)	31 (100)
<i>Cystoisospora</i> spp.	1 (5.3)	4 (21.1)	8 (42.1)	2 (10.5)	3 (15.8)	1 (5.3)	19 (100)
<i>Blastocystis</i> spp.	9 (15.3)	20 (33.9)	10 (16.9)	11 (18.6)	7 (11.9)	2 (3.4)	59 (100)
<i>Sarcocystis</i> spp.	0 (0)	3 (14.3)	5 (23.8)	6 (28.6)	4 (19)	3 (14.3)	21 (100)
Total number of dogs (%)	56 (18.6)	70 (23.3)	55 (18.3)	29 (9.6)	15 (5)	5 (1.7)	301 (100)

Table 3. Prevalence of intestinal parasites infection in relation to gender of domestic and stray dogs in villages of Kermanshah province, Iran

Gender	No. of examined dogs	Domestic dogs (n=120)		Stray dogs (n=181)		p-value†
		Infected	Not infected	Infected	Not infected	
Male	208 (69.1)	71 (59.2)	29 (24.2)	89 (49.1)	19 (10.5)	Ns
Female	93 (30.9)	13 (10.8)	7 (5.8)	58 (32)	15 (8.3)	Ns
Total	301(100)	84 (70)	36 (30)	147 (81.2)	34 (18.8)	0.024

†:  $\chi^2$ -test; Ns, Not significant ( $P > 0.05$ ).

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Belgium (54.7%) (Claerebout *et al.*, 2009), Jordan (70.3%) (El-Shehabi *et al.*, 1999), Italy (31.0%) (Riggio *et al.*, 2013), Island (71.4%) (Gingrich *et al.*, 2010) and Argentina (37.8%) (Soriano *et al.*, 2010b), but lower than Ethiopia (84.78%) (Abera *et al.*, 2013) and Nigeria (93.8%) (Umar, 2009).

Some factors such as demographic characteristics, geographical location, public health education and socioeconomic status of animal ownership, usage of anti-helminths, sampling methods and fecal examination techniques are responsible for the wide range of intestinal parasites prevalence (Katagiri & Oliveira-Sequeira, 2008).

Common parasitological techniques that used for detection of parasites in fecal samples are not able to identify all parasites. Therefore, we used two diagnostic methods (formalin-ether concentration and sucrose flotation) that increased the chance of parasites observation, especially protozoan parasites.

In our study, the most prevalent gastrointestinal helminth parasites were hookworm (27.6%) and *Toxocara leonina* (24.9%). The studies in Nigeria, Venezuela and different regions in Iran (Ramirez-Barrios *et al.*, 2004; Dalimi *et al.*, 2006; Okoye *et al.*, 2011; Beiromvand *et al.*, 2013) reported high

levels of these helminths and emphasized the zoonosis of the disease.

In deprived communities, in which people sleep in environments and are unable to afford proper footwear, the level of contamination by larvae of different *Ancylostoma* species is very high (Traub *et al.*, 2005). Although parasitic worms can be seen in rural and urban areas as well as in industrial areas, most of them are related to poor areas. Because the funds allocated to health in these communities are very limited and low and the program for controlling parasitic diseases is not successful. The presence of a large number of infected animals, as well as the lack of awareness of people about parasitic diseases and their prevention, are another important factor in increasing these diseases in poorer areas.

*Taenia/Echinococcus* spp. with relatively high prevalence rate (9.3%) in dogs might be a potential source for infection of resident in the studied rural areas. Infection prevalence, with different strains of *Echinococcus granulosus* has been reported among 5.0% and 45.0% of dogs (Hosseini-Safa *et al.*, 2016). Also, seroprevalence of hydatid cyst have been reported within 1.2–21.4% of the population in different parts of Iran (Fasihi Harandi *et al.*, 2012). A lack of knowledge and education about life cycle of the *Echinococcus* and lack of meat inspection and offal disposal such as infected liver and lung at home slaughter and local abattoirs, significantly contribute to domestic cycles of transmission (Azami *et al.*, 2013). The most important way of contaminating carnivorous animals, especially dogs, is consumption of waste and internal parts of body of slaughtered animals, especially the liver and lungs infected with the parasite (Ezatpour *et al.*, 2014). Like mentioned above agents, the eggs of *Dicrocoelium dendriticum* and *Fasciola* spp. may have been excreted after ingestion of raw liver. Also, dogs as definitive host can be infected through eating grass with an infected ant and metacercariae on aquatic vegetation (Nesvadba, 2006). According to contamination of dogs in studied

areas with *Dicrocoelium dendriticum* (2.3%) and *Fasciola* spp. (1.7%), this problem should be considered as an environmental risk for resident's people in rural areas.

In our study, *Eimeria* spp. (35.0%, 34.3) and *Cryptosporidium* spp. (21.7%, 25.4%) showed highest prevalence among intestinal protozoa in domestic and stray dogs and it was higher than data obtained in Chenaran County (Northeast of Iran) (Beiromvand *et al.*, 2013). In developing countries, cryptosporidiosis is a frequent cause of diarrhea in humans, mostly in children younger than 5 years (Mohaghegh *et al.*, 2017; Jafari *et al.*, 2016; Ghomashlooyan *et al.*, 2015b; Mohaghegh *et al.*, 2015) and dogs can be responsible for the transmission of cryptosporidiosis among human and dogs. In our study, according to high rate of *Cryptosporidium* spp., it can be a major environmental problem in studied rural areas. Dogs harboring of *Giardia* spp. are considered responsible of human giardiasis. The global prevalence of *Giardia* spp. is 5.4 to 55.2% (Hannes *et al.*, 2007) and in patients and healthy dogs was reported to be between 5.0% and 15.0%, respectively (Tangtrongsup and Scorza, 2010). In the present study, 10.3% of dogs were infected with *Giardia* spp. that was relatively high compared to what has been reported in Iran (Jafari Shoorijeh *et al.*, 2008; Sardarian *et al.*, 2015) and other countries (Capelli *et al.*, 2006; Becker *et al.*, 2012), but lower than prevalence reported in Brazil (Katagiri and Oliveira-Sequeira, 2008) and Japan (Itoh *et al.*, 2011).

In the current study, infection rate of dogs with *Blastocystis* spp. (19.6%) was lower compared to Australia (70.8%) (Duda *et al.*, 1998) and Sari in north of Iran (28%) (Daryani *et al.*, 2008), but higher than prevalence reported in USA (9.7%) (Ruau & Stang, 2014). Despite the risk factors in the studied areas, many dogs were not infected, and most infected dogs had two infections, and only a few dogs had five or more infections. Multi-infectious patterns, play a very important role in the design and implementation of control programs.

## CONCLUSION

The high prevalence of some zoonotic helminthes and protozoan such as *T. leonina* and *canis*, Hookworm, *Taenia/Echinococcus* spp., *Cryptosporidium* spp., *Blastocystis* spp. and *Giardia* spp. could indicate that humans in rural areas in Kermanshah are seriously at risk and the human infection may spread vastly and cause severe damage. In this regard, health education for dog owner, anti-parasitic regimes and preventive programs such as elimination of the stray dogs from rural areas and prevention of feeding slaughter offal from home slaughter by domestic dogs is essential to reduce human infection by public health authorities and veterinarians in Kermanshah province.

### Conflict of interest

The authors have no financial or personal relationship with other people or organizations that could inappropriately influence or bias this paper.

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