Prevalence, risk factors and zoonotic potential of intestinal parasites in dogs from four locations in Morocco

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ABSTRACT

Dogs can harbor various intestinal parasites that have serious clinical, economic, and zoonotic impact. In Morocco, the epidemiological status of those parasites is largely unknown.

This study aimed to obtain data on the prevalence of intestinal parasites in various Moroccan dog populations, to identify associated risk factors, to evaluate people’s knowledge regarding zoonotic parasites and to estimate the risk of human infection.

A total of 291 fecal samples were analyzed using a 33% Zinc Sulphate (ZnSO4) centrifugal flotation and a sheather’s sugar simple flotation techniques. In addition, 100 dog owners were asked to fill out a questionnaire about their knowledge regarding canine zoonotic intestinal parasites.

Overall, 58% of sampled dogs were positive for at least one parasite species. *Ancylostoma/Uncinaria* spp. (31.9%), *Toxascaris leonina* (27.4%), *Toxocara canis* (27.1%), *Cystoisospora* spp. (13.4%) and *Giardia* spp. (7.2%) were the most frequently isolated parasites. The overall prevalence was associated with the dogs’ age, activity and feces consistency. More specifically, the prevalence of *Ancylostoma/Uncinaria* spp., *Toxascaris leonina* and *Cystoisospora* spp. was associated with dogs’ activity and feces consistency and, additionally with age for *Cystoisospora* spp., *Toxocara canis* prevalence was associated with age and feces consistency, while *Giardia* spp. prevalence was associated with dogs’ activity.

A weak awareness among dog owners regarding the zoonotic potential of canine intestinal parasites was noticed (33%) especially when their knowledge was compared to another endemic zoonotic disease, i.e. rabies (85%). Furthermore, the rate of respecting the vaccination protocol (82%) was significantly higher than the rate of respecting the deworming protocol (47%).

We conclude that intestinal parasites, including the zoonotic ones, were highly prevalent in the dogs sampled in this study whereas people’s knowledge about those parasites was very limited. These findings suggest that the risk of human infection is very likely, which highlights the need for effective control programs and health education.

1. Introduction

Dogs may harbor a wide range of intestinal parasites (Traversa, 2012). These parasites, whether helminths or protozoa, usually cause growth retardation, decreased performance, increased sensitivity to acquire other infectious diseases (Yacob et al., 2007; Raza et al., 2018), and some of them can lead to severe clinical symptoms (Traversa, 2012; Riggio et al., 2013). Heaviest infections and highest morbidity rates are seen in neonates and puppies, where intestinal parasites can be fatal (Traversa, 2012; Perera et al., 2013; Baneth et al., 2016), especially...
when associated as a co-morbidity with other infectious diseases such as viral gastro-enteritis due to parvovirus (Mazzaferrro, 2020). In addition, some canine intestinal parasites, such as *Taenia* spp. and *Sarcocystis* spp., can be transmitted to farm animals leading to important economic losses (Kohansal et al., 2017; Pennelegion et al., 2020).

These parasites may also represent a significant risk due to their zoonotic potential, since many of them, such as *Toxocara canis*, *Ancylostoma* spp., *Echinococcus* spp., *Giardia* spp., *Dipylidium caninum*, and *Cryptosporidium* spp., can be transmitted to humans either directly via contact with infected dogs or indirectly via contaminated water, foods or soil (Perera et al., 2013; Gharekhani, 2014; Traversa et al., 2014; Kohansal et al., 2017). Overall, children, pregnant women, and immunocompromised people are usually at higher risk for these zoonoses and their outcomes (Robertson and Thompson, 2002; Kohansal et al., 2017).

*T. canis*, *Ancylostoma caninum*, *Echinococcus granulosus* and *Giardia intestinalis* (assemblages A and B) are among the most important canine zoonotic intestinal parasites. The clinical outcome of human infection with *T. canis* ranges from an asymptomatic form to a severe organ injury, depending on the host’s immune response, the invaded tissues and the number of migrating larvae. Visceral and ocular larva migrans are the major clinical syndromes of toxocariasis with the first one considered more severe, especially in children (Traversa, 2012; Raza et al., 2018). Ocular larva migrans can also cause serious ocular damages ranging from impaired vision up to a total loss of sight in severe cases (Pinelli et al., 2011; Traversa, 2012; Raza et al., 2018). According to a survey conducted between 2009 and 2010, published in the ‘Morbidity and Mortality Weekly Report’ journal (MMWR), at least 68 cases of blindness and eye damages due to *T. canis* were estimated to occur yearly in the USA (Lum et al., 2011). Other syndromes such as neurotoxocariasis and covert toxocariasis are also reported (Pinelli et al., 2011; Traversa, 2012; Adeeel, 2020). Cutaneous infection by *A. caninum* may lead to a cutaneous larva migrans while oral infection may result in an eosinophilic enteritis. Both forms represent a serious threat to human health (Robertson and Thompson, 2002; Raza et al., 2018). The severity of human giardiasis is variable with many cases remaining asymptomatic (Raza et al., 2018). Finally, *E. granulosus* causes a severe and potentially life-threatening disease in humans called cystic echinococcosis (Pennelegion et al., 2020) which is considered a major zoonosis in Morocco (Azlaf et al., 2007). In 2012, the Moroccan Ministry of health reported 1627 surgical cases of cystic echinococcosis (Pennelegion et al., 2020) which is considered a major zoonosis in Morocco (Azlaf et al., 2007).

In spite of their significant health impact, little interest is attributed to dog’s intestinal parasites. There is currently limited data published on this topic in Morocco and other countries within the Mediterranean basin, including France, Israel, Malta, Slovenia and Lebanon. Currently, studies exclusively on *Echinococcus* spp. (Pandey et al., 1988; Azlaf et al., 2007; Dakkak, 2010; Dakkak et al., 2017; Amirar et al., 2020) were previously published in Morocco and, to the author’s knowledge, only one survey conducted in 1987, reported the prevalence of intestinal parasites in a small number (n = 57) of dogs from Rabat region (Pandey et al., 1987). However, these previous studies involved exclusively stray dogs (Pandey et al., 1988; Azlaf et al., 2007; Dakkak, 2010). Yet, in order to develop efficient and adapted control strategies, it is crucial to understand the epidemiology of these parasites in various canine populations (Dubná et al., 2007; Gharekhani, 2014), such as owned dogs, considering their close relationship with their owners and the increasing tendency of canine ownership in Morocco these last years (Bouaddi et al., 2018); working dogs because of their close contact with their trainers; and shelter/breeding dogs because of their close relationship with their care-givers and with the visitors of these centers.

To the best of our knowledge, this is the first study in Morocco reporting the prevalence of intestinal parasites, with particular attention to the zoonotic ones, in owned, shelter, breeding and service dogs with the identification of risk factors and an evaluation of people’s knowledge regarding those parasites and their zoonotic potential.

2. Materials and methods

2.1. Study population and location

Between January, 2019 and March 2020, 291 fecal samples were collected from various Moroccan dog populations including owned (n = 96), shelter (n = 129), breeding (n = 10) and working dogs (n = 56). Owned dogs were all collected at the Small Animal Clinic of the Hassan II Agromony and Veterinary Institute (IAV Hassan II), Rabat, Morocco. Shelter dogs were collected at refuges from the cities of Rabat (n = 70), Marrakech (n = 24) and Casablanca (n = 35). Breeding dogs were exclusively sampled from the city of Rabat while working dogs were recruited from a kennel located in Benslimane city.

Data regarding the age, sex, vaccination and deworming status of dogs were collected by interviewing the owners (owned dogs), breeding and shelters supervisors (shelter and breeding dogs) and dog trainers (working dogs). Because of logistical issues, sex of 84 shelter dogs was not recorded. Additionally, age of three dogs was not determined.

2.2. Samples collection and fecal analysis

Fresh fecal samples were collected from each dog into a clean sterile plastic bottle and stored at 4 °C until processing. The fecal analysis was performed within two days after collection, after which samples were preserved in a 10% formalin solution.

All the samples were first screened macroscopically to detect the presence of nematodes and proglottids and to record feces consistency. The macroscopic aspect of the feces was classified from 1 to 4 ascending levels depending on the consistency (1 = liquid stools, 2 = very soft, 3 = soft and 4 = formed stools). Then, samples were examined microscopically using a 33% zinc sulphate (ZnSO4) centrifugal flotation and a sheather’s sugar simple flotation techniques, allowing the identification of eggs and oocysts, in most cases to genus level (Zajac and Conboy, 2013). Parasites were identified according to a key of identification (Zajac and Conboy, 2013; CAPC guideline, 2019, 2020a, 2020b).

2.2.1. Centrifugal flotation with 33% Zinc Sulphate (ZnSO4) (specific gravity 1.18)

Five grams of feces were mixed with 33% zinc sulphate solution (Zajac and Conboy, 2013). Then, 14 ml of the suspension were poured through a sieve into a 15 ml centrifuge tube and centrifuged at 1500 RPM for 5 min. Zinc sulfate solution was then filled to the rim of the centrifuge tube, forming a positive meniscus, and a coverslip was placed on top. After 10 min, the coverslip was removed and placed onto a microscope slide. The entire slide was examined under a light microscope at 100× and 400× magnifications.

2.2.2. Sheather’s sugar simple flotation (specific gravity 1.25)

Five grams of feces were mixed with sheather’s sugar solution (Zajac and Conboy, 2013). The suspension was poured through a sieve into a 15 ml centrifuge tube until forming a positive meniscus, and a coverslip was placed on top. After 10 min, the coverslip was removed and placed onto a microscope slide. The entire slide was examined under a light microscope at 100× and 400× magnifications.

2.3. Questionnaires

After obtaining an informed consent, one hundred dog owners were interviewed about their knowledge regarding canine zoonotic intestinal parasites and other zoonoses, such as rabies, and prevention measures including deworming and vaccination. They were also asked whether they respected their dog’s vaccination protocol and whether they regularly dewormed their pet dogs. Finally, they were questioned about their perception regarding canine zoonotic intestinal parasitosis.
2.4. Statistical analysis

The prevalence of the presence of a parasite (any of the reported parasites) and specific parasites as a function of different risk factors was analyzed by a logistic regression model. Risk factor groups were compared pairwise by the likelihood ratio test in order to test whether the odds ratio differs significantly from zero. All analyses were performed using SAS Version 9.4 (Copyright (c) 2020 by SAS Institute Inc., Cary, NC, USA) at a global significance level of 5% and using the Bonferroni technique to adjust for multiple comparisons. The answers recorded in the questionnaire are binary and thus are also analyzed by logistic regression.

3. Results

The study population comprised 60.8% males (n = 126) and 39.1% females (n = 81). Dogs were distributed in four age categories including puppies (<6 months) (24.3%, n = 70), young adults (6–12 months) (10.7%, n = 31), adults (1–6 years) (56.9%, n = 164) and seniors (>6 years) (7.9%, n = 23).

The overall prevalence of intestinal parasites in the 291 studied dogs was 58% (95% CI 52.1–63.8). *Ancylostoma/Uncinaria* spp. (31.9%, 95% CI 26.6–37.6) (Fig. 1), *Toxascaris leonina* (27.4%, 95% CI 22.4–33) (Fig. 2), *T. canis* (27.1%, 95% CI 22.1–32.6) (Fig. 3), Cystoisospora spp. (13.4%, 95% CI 9.7–17.8) (Fig. 4) and *Giardia* spp. (7.2%, 95% CI 4.5–10.8) (Fig. 5) were the most isolated parasites (Table 1). Among positive dogs, 47.3% (80/169 dogs) were infected with a single parasite genus, while 29.5% (50/169 dogs) and 23.0% (39/169 dogs) harbored two and 3 to 5 different genera, respectively.

The prevalence of any of the parasites was the lowest in owned dogs and significantly lower than the prevalence in shelter and working dogs. For *Ancylostoma/Uncinaria* spp., *T. leonina* and Cystoisospora spp. the highest prevalence levels were found in shelter dogs, with significant differences from the other categories, except from the breeding category for Cystoisospora spp. (Table 2). However, for *Giardia* spp., breeding dogs had by far the largest and significantly different prevalence from each of the three other categories (Table 2).

The prevalence of any of the parasites did not differ significantly between the age groups. For *T. canis* and Cystoisospora spp., the highest prevalence levels were found in the puppy group, which differed significantly from the other age groups except from the senior group for Cystoisospora spp. (Table 3).

Significantly more parasites were detected in liquid stools compared to other fecal consistency levels. We observed a significant decrease of parasite prevalence in more consistent feces for the overall prevalence, *Ancylostoma/Uncinaria* spp., *T. leonina*, *T. canis*, and Cystoisospora spp. (Table 4).

The results of the questionnaires showed that owners respected their dogs’ vaccination protocol (82%) significantly more (p < 0.001) than they did so for the deworming protocol (47%). Also, significantly (p < 0.001) more owners (85%) knew about rabies and its zoonotic potential than about canine zoonotic intestinal parasites (33%). When questioned about their perception of zoonotic intestinal parasites, 43% of owners responded that these zoonoses can cause serious diseases in humans.
4. Discussion

Our study aimed at reporting prevalence of intestinal parasites in a large dog population from various Moroccan locations. Such studies were needed because of lack of large and recent data on this topic. This study revealed a high overall prevalence of intestinal parasites (58%) in the sampled dogs. A study from 1987 conducted in Morocco by Pandey et al. (1987), showed an even higher prevalence (100%). This can be explained by the fact that the aforementioned study was conducted exclusively on stray dogs, and parasites were screened by necropsy, probably allowing a better sensitivity comparing to the microscopic fecal examination alone (Lahmar et al., 2017). Prevalence rates reported in other regions from the Mediterranean Basin were widely different and ranged from 22% in France to 71.3% in Spain (Franc et al., 1997; Awadallah and Salem, 2015; Gurler et al., 2015; Kostopoulos et al., 2017; Matalall et al., 2018; Scaramozzino et al., 2018; Sanchez-Thevenet et al., 2019). This variation can be attributed to several factors including the study area, the diagnostic techniques used for the detection of parasites, the studied populations and their living conditions.

Table 1
Prevalence of intestinal parasites detected in the feces of 291 dogs.

<table>
<thead>
<tr>
<th>Parasite</th>
<th>Positive</th>
<th>Prevalence % (CI)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ancylostoma/Uncinaria spp.</td>
<td>93</td>
<td>31.9 (26.6–37.6)</td>
</tr>
<tr>
<td>Toxocara canis</td>
<td>80</td>
<td>27.4 (22.4–33)</td>
</tr>
<tr>
<td>Toxascaris leonina</td>
<td>79</td>
<td>27.1 (22.1–32.6)</td>
</tr>
<tr>
<td>Cystoisospora spp.</td>
<td>39</td>
<td>13.4 (9.7–17.8)</td>
</tr>
<tr>
<td>Giardia spp.</td>
<td>21</td>
<td>7.2 (4.5–10.8)</td>
</tr>
<tr>
<td>Dipylidium caninum</td>
<td>2</td>
<td>0.6</td>
</tr>
<tr>
<td>Trichuris vulpis</td>
<td>1</td>
<td>0.3</td>
</tr>
<tr>
<td>Tamarisc spp.</td>
<td>1</td>
<td>0.3</td>
</tr>
</tbody>
</table>

Table 2
Prevalence of intestinal parasites according to activity level. Prevalences in the same row not sharing a letter differ statistically significantly from each other.

<table>
<thead>
<tr>
<th>Parasite</th>
<th>Owned dogs (n = 96)</th>
<th>Shelter dogs (n = 129)</th>
<th>Breeding dogs (n = 10)</th>
<th>Working dogs (n = 56)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Overall (%)</td>
<td>32.2 ± 0.3</td>
<td>75.1 ± 0.5</td>
<td>70 ± 0</td>
<td>60.7 ± 0.4</td>
</tr>
<tr>
<td>Ancylostoma/Uncinaria spp. (%)</td>
<td>9.3 ± 0.3</td>
<td>59.6 ± 0.5</td>
<td>0 ± 0</td>
<td>12.5 ± 0.3</td>
</tr>
<tr>
<td>Toxocaris leonina (%)</td>
<td>2 ± 0.3</td>
<td>49.6 ± 0.5</td>
<td>0 ± 0</td>
<td>25 ± 0.3</td>
</tr>
<tr>
<td>Toxocara canis (%)</td>
<td>20.8 ± 0.3</td>
<td>32.5 ± 0.5</td>
<td>10 ± 0</td>
<td>28.5 ± 0.3</td>
</tr>
<tr>
<td>Cystoisospora spp. (%)</td>
<td>7.2 ± 0.6</td>
<td>23.2 ± 0.5</td>
<td>20 ± 0</td>
<td>0 ± 0</td>
</tr>
<tr>
<td>Giardia spp. (%)</td>
<td>5.2 ± 0.7</td>
<td>6.9 ± 0.7</td>
<td>50 ± 0</td>
<td>3.5 ± 0</td>
</tr>
<tr>
<td>Dipylidium caninum (%)</td>
<td>1 ± 0.7</td>
<td>0.7 ± 0.7</td>
<td>0 ± 0</td>
<td>0 ± 0</td>
</tr>
<tr>
<td>Trichuris vulpis (%)</td>
<td>0 ± 0.3</td>
<td>0 ± 0.3</td>
<td>0 ± 0</td>
<td>1.7 ± 0</td>
</tr>
<tr>
<td>Tamarisc spp. (%)</td>
<td>0 ± 0.3</td>
<td>0.7 ± 0.3</td>
<td>0 ± 0</td>
<td>0 ± 0</td>
</tr>
</tbody>
</table>

Table 3
Prevalence of intestinal parasites by age group. Prevalences in the same row not sharing a letter differ statistically significantly from each other.

<table>
<thead>
<tr>
<th>Parasite</th>
<th>&lt;6 months (n = 70)</th>
<th>6–12 months (n = 31)</th>
<th>1–6 years (n = 164)</th>
<th>&gt;6 years (n = 23)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Overall (%)</td>
<td>57.1 ± 0.6</td>
<td>67.7 ± 0.6</td>
<td>58.5 ± 0.6</td>
<td>43.4 ± 0.6</td>
</tr>
<tr>
<td>Ancylostoma/Uncinaria spp. (%)</td>
<td>22.8 ± 0.6</td>
<td>19.3 ± 0.6</td>
<td>39.6 ± 0.6</td>
<td>26 ± 0.6</td>
</tr>
<tr>
<td>Toxocaris leonina (%)</td>
<td>21.4 ± 0.6</td>
<td>35.4 ± 0.6</td>
<td>31.7 ± 0.6</td>
<td>8.6 ± 0.6</td>
</tr>
<tr>
<td>Toxocara canis (%)</td>
<td>45.7 ± 0.6</td>
<td>16.1 ± 0.6</td>
<td>22.5 ± 0.6</td>
<td>13 ± 0.6</td>
</tr>
<tr>
<td>Cystoisospora spp. (%)</td>
<td>28.5 ± 0.6</td>
<td>3.2 ± 0.6</td>
<td>9.7 ± 0.6</td>
<td>8.6 ± 0.6</td>
</tr>
<tr>
<td>Giardia spp. (%)</td>
<td>5.7 ± 0.6</td>
<td>6.4 ± 0.6</td>
<td>7.9 ± 0.6</td>
<td>8.6 ± 0.6</td>
</tr>
<tr>
<td>Dipylidium caninum (%)</td>
<td>1.4 ± 0.6</td>
<td>0 ± 0.6</td>
<td>0.6 ± 0</td>
<td>0 ± 0</td>
</tr>
<tr>
<td>Trichuris vulpis (%)</td>
<td>0 ± 0.3</td>
<td>0 ± 0.3</td>
<td>0.6 ± 0.3</td>
<td>0 ± 0</td>
</tr>
<tr>
<td>Tamarisc spp. (%)</td>
<td>0 ± 0.3</td>
<td>0 ± 0.3</td>
<td>0.6 ± 0.3</td>
<td>0 ± 0</td>
</tr>
</tbody>
</table>

Table 4
Prevalence of intestinal parasites depending on the feces consistency. Prevalences in the same row not sharing a letter differ statistically significantly from each other.

<table>
<thead>
<tr>
<th>Parasite</th>
<th>Liquid (n = 46)</th>
<th>Very soft (n = 98)</th>
<th>Soft (n = 63)</th>
<th>Formed (n = 84)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Overall (%)</td>
<td>86.9 ± 0.7</td>
<td>69.3 ± 0.6</td>
<td>50.7 ± 0.5</td>
<td>34.5 ± 0.5</td>
</tr>
<tr>
<td>Ancylostoma/Uncinaria spp. (%)</td>
<td>45.6 ± 0.7</td>
<td>41.8 ± 0.6</td>
<td>26.9 ± 0.6</td>
<td>16.6 ± 0.6</td>
</tr>
<tr>
<td>Toxocaris leonina (%)</td>
<td>41.3 ± 0.7</td>
<td>39.7 ± 0.6</td>
<td>22.2 ± 0.6</td>
<td>9.5 ± 0.6</td>
</tr>
<tr>
<td>Toxocara canis (%)</td>
<td>52.1 ± 0.7</td>
<td>27.5 ± 0.6</td>
<td>22.2 ± 0.6</td>
<td>16.6 ± 0.6</td>
</tr>
<tr>
<td>Cystoisospora spp. (%)</td>
<td>32.6 ± 0.7</td>
<td>10.2 ± 0.6</td>
<td>12.6 ± 0.6</td>
<td>7.1 ± 0.6</td>
</tr>
<tr>
<td>Giardia spp. (%)</td>
<td>6.5 ± 0.7</td>
<td>11.2 ± 0.7</td>
<td>9.5 ± 1.1</td>
<td>1.1 ± 0.1</td>
</tr>
<tr>
<td>Dipylidium caninum (%)</td>
<td>0 ± 0.3</td>
<td>1 ± 0.3</td>
<td>0 ± 0.3</td>
<td>1.1 ± 0.1</td>
</tr>
<tr>
<td>Trichuris vulpis (%)</td>
<td>0 ± 0.3</td>
<td>0 ± 0.3</td>
<td>1.5 ± 0.5</td>
<td>0 ± 0.5</td>
</tr>
<tr>
<td>Tamarisc spp. (%)</td>
<td>0 ± 0.3</td>
<td>0 ± 0.3</td>
<td>1.5 ± 0.5</td>
<td>0 ± 0.5</td>
</tr>
</tbody>
</table>
conditions, their ownership status, the anthelmintics used, hygienic conditions and, their proximity to other animals (Mundim et al., 2007; Katagiri and Oliveira-Sequeira, 2008; Bahrami et al., 2011).

_Ancylostoma/Uncinaria_ spp., _T. leonina, T. canis, Cystoisospora_ spp. and _Giardia_ spp. were the most frequently detected parasites in our study, which is in agreement with other reports from several Mediterranean countries (Pandey et al., 1987; Martínez-Moreno et al., 2007; Riggio et al., 2013; Awadallah and Salem, 2015; Kostopoulou et al., 2017; Scaramozzino et al., 2018). The prevalence rates of _Trichuris vulpis_ (Fig. 6) and cestodes, such as _D. caninum_ and _Taenia_ spp. (Fig. 7), were very low in the current study, which is in line with the findings of some other surveys (Awadallah and Salem, 2015; Kostopoulou et al., 2017) but contradicts others (Pandey et al., 1987; Martínez-Moreno et al., 2007). However, it is worth mentioning that the diagnostic technique used in our study (centrifugal flotation) is more specific for nematodes than for cestodes (CAPC guideline, 2016; Adolph et al., 2017), which can at least partially explain the very low prevalence rate of cestodes in our study. Also, most of sampled dogs were under controlled diet, with no access to raw meat or carcasses, which reduces the risk of contamination by Taeniid tapeworms (Pennelegion et al., 2020).

A large proportion of shelter dogs (76.9%) harbored at least one species of intestinal parasite, and showed a significantly higher prevalence of _Ancylostoma/Uncinaria_ spp., _T. leonina_ and _Cystoisospora_ spp. This is in agreement with previous studies revealing a high level of parasitism in this dogs’ category (Palmer et al., 2008; Gharekhani, 2014; Simonato et al., 2015; Kostopoulou et al., 2017; Raza et al., 2018). These findings can easily be explained by the lifestyle of these animals, characterized by a lack of health control measures and anthelmintics use, poor hygienic conditions, direct contact with other dogs and their stools, and stressful conditions such as overcrowding, loud noises, poor diet and frequent dietary changes, causing an immunosuppressive effect (Palmer et al., 2008; Pesavento and Murphy, 2014; Kostopoulou et al., 2017; Raza et al., 2018). Although breeding and service dogs are usually well cared for and receive more veterinary care compared to shelter dogs, they were highly infected in this study. The results obtained from previous surveys are comparable to ours, indicating that service and working dogs frequently harbor intestinal parasites (Senlik et al., 2006; Ahmed et al., 2014), since these dogs go to exercise daily and the feces are not removed from training area, which increases the risk of soil contamination and infection of other dogs (Senlik et al., 2006). Additionally, working dogs enrolled in this study received anthelmintic treatment twice a year which may be insufficient according to the European Scientific Counsel Companion Animal Parasites (ESCCAP) guidelines (ESCCAP, 2021). Breeding dogs showed significantly higher prevalence of _Giardia_ spp. compared to the three other categories. This could be due to the inefficacy of the anthelmintic administered against protozoa or perhaps the lack of compliance of the owners. Moreover, the high stocking density may lead to horizontal spread of protozoan parasites (Gharekhani, 2014). Even if owned dogs showed a noteworthy prevalence of intestinal parasites (32.2%), this rate was significantly lower than that of shelter and working dogs, confirming previous results (Awoke et al., 2011; Perera et al., 2013; Gharekhani, 2014; Kostopoulou et al., 2017). Domestic dogs are usually well cared for by their owners, live in better hygienic conditions in low density, have limited contact with other animals and have a better veterinary follow-up, which explains their lowest frequency of infection (Awoke et al., 2011; Hinney et al., 2017).

In this study, 52.6% of infections were mixed, and among them, 79.7% occurred in shelter dogs. This can be attributed to increased exposure of shelter dogs to various parasites, due to high environmental contamination with infectious parasitic developmental stages, and exacerbated by the lack of veterinary care and inappropriate anthelmintic use (Pesavento and Murphy, 2014; Raza et al., 2018).

This study showed that the prevalence of _T. canis_ and _Cystoisospora_ spp. were age-dependent, with the highest infection rates in puppies (<6 months). This is in agreement with data reported in numerous previous studies (Pandey et al., 1987; Fontanarrosa et al., 2006; Martínez-Moreno et al., 2007; Gates and Nolan, 2010; Barutzki and Schaper, 2013; Riggio et al., 2013; Gharekhani, 2014; Hinney et al., 2017; Stafford et al., 2020) and could be due to the immature immune system of young animals. Indeed, parasite-specific immunity is usually acquired with age, probably as a consequence of single or repeated exposures (Fontanarrosa et al., 2006; Palmer et al., 2008; Gharekhani, 2014). Furthermore, the transplacental and transmammary routes of transmission of _T. canis_ promote its occurrence in puppies (Palmer et al., 2008; Gharekhani, 2014).

Parasite’s prevalence, especially _Ancylostoma/Uncinaria_ spp., _T. leonina, T. canis_ and _Cystoisospora_ spp. significantly increased in less consistent feces. _T. leonina_ and some hookworms (e.g. _Uncinaria stenocepha_) are known to mainly cause asymptomatic infections (ESCCAP, 2021) but it should be considered that in this study, these species were mainly detected in shelter dogs and most of them were polyinfected, therefore they were more susceptible to have less consistent feces probably due to coinfections with other parasites such as _T. canis_.

Fig. 6. _Trichuris vulpis_ egg. Fecal flotation, magnification x100.

Fig. 7. _Taenia_ spp. egg. Fecal flotation, magnification x100.
Cystoisospora spp. and Giardia spp. or possibly other agents such as viruses and bacteria. T. canis and Cystoisospora spp. are found in puppies (<6 months), which are known to be more sensitive to these two parasites and hence more susceptible to have diarrhea when infected (Bartuzki and Schaper, 2013; Kostopoulou et al., 2017; ESCCAP, 2021).

The zoonotic helmint, T. canis, was significantly prevalent in our study and was detected in the four canine populations, including owned dogs (20.8%). Noteworthily, 29% of detected hookworms were suspected to be from the zoonotic species A. caninum, according to the eggs length and width measurements (Lucio-Forster et al., 2012; Zajac and Conboy, 2013). In Morocco, published data on human infection with T. canis and A. caninum are sporadic and limited to case reports (Laghmari et al., 2003; Guamri et al., 2009; Daoudi et al., 2014; Bembella et al., 2016; Elyadari et al., 2018; Adeel, 2020; Galosso et al., 2020; Kaibi et al., 2020). However, when taking in consideration the significant prevalence of T. canis among dogs of this study and the suspected cases of A. caninum, in addition to the Moroccan investigations that have revealed vegetable and wastewater contamination with these parasites (Hajami et al., 2012, 2013; Hajami, 2013; Adeel, 2020), the risk of human infection seems to be very likely and thus, human cases may be under-diagnosed or under reported in Morocco. Giardia spp. was also commonly found in dogs, but molecular-based tests and genotyping of the parasite are needed to determine the species and assemblages implicated in, and to estimate their zoonotic potential (El Fatni et al., 2014; Pallant et al., 2015).

The evaluation of owners’ knowledge through the questioners allowed us to highlight a weak level of knowledge regarding zoonotic intestinal parasites, their severity and the prevention measures. In contrast, the majority of the interviewed dog owners had great knowledge about rabies. Also, most of them respected their dogs’ vaccination protocol. Our results showed poor owner awareness and high prevalence of intestinal parasites, this together with the close proximity between humans and their pet dogs, emphasizes the major human health threat.

Veterinarians have a crucial role in education of pet owners, veterinary staff and people interacting with dogs about canine zoonotic intestinal parasites and prevention measures (Traversa, 2012; Kostopoulou et al., 2017). More widely, veterinarians are considered a more congruous source of knowledge on zoonoses than physicians (Traversa, 2012).

People in regular contact with dogs should be clearly informed about transmission routes of zoonotic intestinal parasites, to both dogs and humans, and about the prevention practices and behaviors to adopt to reduce the risk of contamination (Palmer et al., 2008; Traversa, 2012; Traversa et al., 2014).

According to the European Scientific Counsel Companion Animal Parasites (ESCCAP, 2021) and the North American Companion Animal Parasite Council (CAPC) guidelines, deworming 1–3 times a year does not provide sufficient protection and does not necessarily eliminate patent infections. Thus, it is recommended to deworm pets at intervals not exceeding 3 months, or even monthly, depending on their individual risk and the risk for their owners (Traversa, 2012; Pennelegion et al., 2020; ESCCAP, 2021).

Regular fecal examinations, at least four times in the first year of life and at least two times per year in adulthood, should be considered when, for whatever reason, a year-round-control is not possible to detect any parasitic stage and monitor the efficacy of the preventive treatment used (Traversa, 2012; Traversa et al., 2014; CAPC guideline, 2020a, 2020b; ESCCAP, 2021; von Samson-Himmelstjerna et al., 2021).

Epidemiological studies on human infections with zoonotic ascarids, hookworms and Giardia species are also strongly needed in order to investigate the real impact of these zoonotic parasites on human health and to guide preventive measures following the One Health approach (Adeel, 2020). The collaboration of physicians and veterinarians following the One Health approach is essential to address zoonotic diseases in both humans and animals and hence contribute to a more effective control/prevention.

5. Conclusion

To our knowledge, this is the first report of intestinal parasites in owned, shelters, breeding and working dogs from Morocco. Our results revealed that intestinal parasites were highly prevalent in all dog categories. We also highlighted a high prevalence of the zoonotic parasites T. canis, that can be a serious threat to public health. People’s knowledge about zoonotic intestinal parasites of dogs was found very limited. These results stress the need of improved control programs against canine intestinal parasites to protect both dogs and humans and the need of increasing knowledge of dog owners and caregivers on this subject. These observations also highlight the central role of veterinarians in preventing both canine and human intestinal parasitic infections not only by prescribing anthelmintic preventive treatments but also by educating dog owners and caregivers. We also recommend investigating the situation in humans by performing epidemiological studies on human infection with canine zoonotic parasites to apply the One Health approach, compare data, and study the possibility of zoonotic transmission.

Ethical statement

The work described in the manuscript ‘Prevalence, risk factors and zoonotic potential of intestinal parasites in dogs from Morocco’ involved the use of non-experimental animals only (owned, shelter, breeding and service dogs) and non-invasive approaches were performed. Oral informed consent was obtained from all dog owners, shelter, breeding and kennel managers, before inclusion of dogs in the study and before the start of any procedure described in the manuscript. Dogs were subjected to a routine clinical examination following the established internationally recognized high standards (‘best practice’) of individual veterinary clinical patient care. Freshly passed fecal samples were collected by dog owners (owned dogs), breeding and shelters supervisors (shelter and breeding dogs) and dog trainers (working dogs), then stored at 4 °C. Fecal analysis were performed within two days after collection. No invasive or painful procedures were performed on the dogs included in this study.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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