Is the cat an important reservoir host for visceral leishmaniasis? A systematic review with meta-analysis

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Abstract

In recent years feline leishmanial infections (FLI) have been studied more than ever before in various parts of the world. However, evidence-based knowledge on FLI has remained unavailable. The main objectives of this study were to investigate the status of felines infected by Leishmania spp. worldwide. Data were extracted from 10 available databases over the period of 1982 to 2017. Overall, 78 articles fulfilled the inclusion criteria and were used for data extraction in this systematic review. The overall FLI prevalence by both serological and molecular methods was estimated at 10% (95% CI: 8%-14%). In Italy, both the seroprevalence (24 %) and PCR prevalence (21 %) were found to be higher than in other countries. The most common diagnostic test used was the indirect fluorescent antibody test (38.5%). Studies on mixed-breed felines were more common than those on other breeds, while the most common parasite species was L. infantum (63%). Our findings suggest that cats act as primary and/or secondary reservoir hosts in the transmission of the Leishmania spp. to humans and also to dogs, by sandflies, at least in endemic foci. Moreover, available data confirm the enzootic stability situation of FLI in several countries including some in Europe.

Article Info

Keywords:
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Background
The leishmaniases are neglected protozoal diseases caused by Leishmania spp. that occur in 98 countries [1], affecting 1.2 million in the form of cutaneous leishmaniasis (CL), and 400,000 in the form of visceral leishmaniasis (VL), leading to approximately 40,000 deaths per year [2]. The main route of VL transmission is through the bite of vectors infected with Leishmania donovani (L. donovani) complex, mainly Leishmania infantum/chagasi (L. infantum/chagasi). Both domestic and wild animals may serve as host reservoirs of Leishmania spp. [3]. Dogs are the main reservoir hosts of L. infantum/chagasi but sandflies, as the natural vectors of Leishmania spp., may also feed on the blood of cats [4]. Therefore, cats infected with the L. donovani complex may be urban reservoirs of VL and transmit the protozoan to other sandflies [5, 6]; therefore, cats are potential reservoirs of this zoonotic VL disease. Studies on feline leishmanial infection (FLI) are limited and several aspects of the disease in cats are still unclear [7]. Recently, reports of FLI have increased dramatically, achieving a prevalence of up to 60% in certain cat populations [8]. The most common clinical signs reported in FLI include lymphadenomegalgy, splenomegaly, weight loss, anorexia, as well as cutaneous, mucocutaneous and ocular lesions [8]. However, in endemic regions such as Mediterranean countries, the subclinical feline infection L. infantum/chagasi is common, whereas clinical illness is relatively uncommon [7–8].

Identification of Leishmania amastigotes in aspirated samples of bone marrow, spleen and lymph node is specific and considered the gold standard method for diagnosing FLI. Feline vector-borne pathogens have been increasingly recognized worldwide based on serological and/or molecular epidemiological investigations [9,10]. Most epidemiological studies demonstrated the presence of anti-Leishmania antibodies in feline sera by means of different techniques such as indirect fluorescent antibody test (IFAT), enzyme-linked immunosorbent assay (ELISA) or western blot (WB) [10–17]. Polymerase chain reaction (PCR) is recommended preferentially over other diagnostic tests, especially when blood samples and other clinical samples contain a low parasitic burden [13, 16, 18,19]. However little is known in reference to their diagnostic performance in cats with FLI.

Although an effective treatment for symptomatic cats has not yet been established, oral allopurinol administration followed by subcutaneous glucantime has been frequently used as chemotherapy regimens in cats affected by FLI [7, 8, 20].

However, there is still no available evidence-based knowledge about various epidemiological aspects of FLI. Therefore, the purpose of this study was to determine the global status of the infection in cats and introduce currently used diagnostic laboratory methods.

Methods
Searching strategy
This systematic review was performed according to the guidelines of the Preferred Reporting Items for Systematic Reviews and Meta-Analysis (PRISMA) [21]. To determine the prevalence of FLI, 10 English and Iranian databases including Google Scholar, Pub Med, Science Direct, Web of Science, Scopus, Elm net, Magiran, Barakatkns (formerly Iran medex), Iran doc, and Scientific Information Database (SID) were searched from 1982 to 2017 (36 years). The relevant keywords including “Leishmania spp.”, “Leishmania donovani”, “Leishmania infantum”, “feline leishmaniasis”, “feline leishmaniosis”, “cat”, “molecular”, “PCR”, “serology”, “ELISA”, “IFAT” were chosen using medical subject headings terms (MESH).

Inclusion and exclusion criteria
Data were extracted from studies with at least one of the following inclusion criteria: cross-sectional and case-control studies corresponding to determining prevalence of leishmanial infections that evaluated the presence of FLI based on serological and molecular tests among all types of cats. Also, summaries of articles presented as proceedings at conferences, studies that contained no qualified data, experimental studies, review articles, duplicates, and case reports were excluded. The PRISMA flowchart of the study plan is shown in Figure 1. Out of the retrieved articles, 78 papers were eligible for inclusion in this systematic review and meta-analysis. The recorded data included author name, year of publication, country, type of cat, sample size, Leishmania species, laboratory method, seroprevalence (%) and PCR prevalence and quality assessment. The above details were extracted separately by two researchers (SA and MF).

Meta-analysis
For each study, the prevalence and standard error (SE) were determined. We used forest plots to estimate pooled effect sizes and the effect of each study with 95% confidence intervals. The Cochran Q-test (p-value<0.1) and the I-squared index were employed to evaluate heterogeneity, with values between 25% and 50% as thresholds for low , between 50% and 75% for moderate, and above 75% for high heterogeneity. When heterogeneity was found, a random-effects model (Dersimonian–Laird model) was applied; if not, a fixed effects model (Mantel–Haenszel) was utilized to calculate overall effects.

Quality assessment
The quality of meta-analysis was evaluated with the STROBE checklist. A checklist including 22 items was considered for adequate reporting of observational studies. These items related to the article’s title, abstract, introduction, methods, results, and discussion sections. A score under 7 was defined as poor quality, 8 to 17 low, 18 to 28 moderate and more than more 28 high quality [22]. The mean score obtained via the STROBE checklist for 78 analyzed articles was 31, whereas 28 is considered high quality. Possible publication bias was explored using a funnel plot and Egger’s test, which evaluated whether the precision of studies was appropriate for the scale of their effect size. All data analyses were performed using the software Stata, v. 14 (Stata Corp LP, College Station, Texas, USA).
Results
Seventy-eight (78) cross-sectional studies published from 1982 to 2017 (36 years) were included in this meta-analysis. Most studies (30.8%) were performed in Brazil (Table 1). The total number of cases was 12,635 (ranging from 8 to 1,101). In Italy, both the prevalence by seropositivity (24%) and PCR positivity (21%) were found to be higher than in other countries. The overall seroprevalence of FLI in 4 European countries including Italy, Spain, Portugal, and Greece was estimated at 12.2% (see Table 2).

The most common diagnostic test was IFAT, used in 38.5% of studies. Among serological methods, WB and indirect hemagglutination test (IHT), being the least common diagnostic tests, were used in only 3.8% of studies (see Table 1).

The seroprevalence (15%) and PCR prevalence (23%) of FLI in mixed-type/breed cats (defined as cats descending from two or more breeds) were higher than in other cat types/breeds. Approximately 63% of *Leishmania* species were *L. infantum*, while the remainder frequently included *Leishmania* spp. (Table 2).

The pooled prevalence of FLI based on a random effect meta-analysis (was estimated at 10% (95% CI: 8%-14%). The estimate of prevalence based on seropositivity (11%), was significantly higher than PCR positivity (10%) (z=0.01, p=0.92) (Figure 2).

Not only funnel plot but also Egger’s test found no evidence a heterogeneity among effect size of studies for seroprevalence (b=1.36, p=0.180) and PCR prevalence (b=0.16, p=0.875) (see Figure 3).

Table 1. Baseline characteristics of studies included in the meta-analysis of feline leishmanial infection

| Author         | Year of publication | Country | Type of cat | Sample size | Leishmania species | Lab test | Seropositive (%) | PCR positive (%) |
|----------------|---------------------|---------|-------------|-------------|-------------------|----------|------------------|------------------|
| Michael. SA    | 1982                | Egypt   | stray       | 80          | *Leishmania* spp. | IHAT     | 3.8              | .                |
| Morsy. TA      | 1988                | Egypt   | stray       | 28          | *Leishmania* spp. | IHAT     | 3.6              | .                |
| Bez. M         | 1992                | France  | stray       | 174         | *Leishmania* spp. | IFAT     | 0.6              | .                |
| Morsy. TA      | 1994                | Egypt   | mixed       | 60          | *Leishmania* spp. | IHAT     | 10               | .                |
| Sherlock. IA   | 1996                | Brazil  | mixed       | 53          | *Leishmania* spp. | IFAT     | 0                | .                |
| Pennisi. MG    | 1998                | Italy   | mixed       | 93          | *Leishmania* spp. | IFAT     | 59.1             | .                |
| Ozon. C        | 1998                | France  | stray       | 97          | *L. infantum*     | WB       | 12.4             | .                |
Table 1. Cont.

| Author               | Year of publication | Country       | Type of cat | Sample size | Leishmania species | Lab test      | Seropositive (%) | PCR positive (%) |
|----------------------|---------------------|---------------|-------------|-------------|---------------------|---------------|------------------|------------------|
| Pennisi. MG [8]      | 2000                | Italy         | mixed       | 89          | Leishmania spp.     | IFAT, PCR     | 68.5             | 60.7             |
| Simões-Mattos. L [49]| 2001                | Brazil        | stray       | 84          | Leishmania spp.     | ELISA         | 10.7             | 0                |
| Poli. A [31]         | 2002                | Italy         | domestic    | 110         | Leishmania spp.     | IFAT          | 0.9              | 0                |
| Portús. M [50]       | 2002                | Spain         | domestic    | 117         | *L. infantum*       | ELISA         | 1.7              | 0.5              |
| Zárate-Ramos. JJ [51]| 2002                | Spain         | domestic    | 50          | *L. infantum*       | DAT           | 42               | 0                |
| Vila. S [52]         | 2005                | Italy         | mixed       | 203         | Leishmania spp.     | IFAT, PCR     | 16.3             | 100              |
| Solano-Gallego. L [37]| 2007               | Spain         | mixed       | 445         | *L. infantum*       | ELISA         | 6.3              | 3                |
| Martin-Sánchez. J [36]| 2007              | Spain         | domestic    | 183         | *L. infantum*       | IFAT, PCR     | 70.5             | 25.7             |
| Nasereddin. A [53]   | 2008                | Israel        | stray       | 104         | Leishmania spp.     | ELISA         | 6.7              | 0                |
| Huebner. [54]        | 2008                | Greece        | mixed       | 389         | Leishmania spp.     | IFAT          | 21.6             | 0.5              |
| Tabar. MD [55]       | 2008                | Spain         | domestic    | 100         | *L. infantum*       | PCR           | 0.9              | 0                |
| Ayllon. T [40]       | 2008                | Spain         | domestic    | 233         | *L. infantum*       | IFAT, PCR     | 4.29             | 0.4              |
| Maia. C [56]         | 2008                | Portugal      | stray       | 23          | *L. infantum*       | IFAT, PCR     | 17.4             | 30.4             |
| Da Silva. AV [42]    | 2008                | Brazil        | domestic    | 8           | *L. infantum*       | IFAT          | 25               | 5.8              |
| Sarkari. B [57]      | 2009                | Iran          | stray       | 40          | *L. infantum*       | IFAT, DAT     | 27.5             | 0.5              |
| Diakou. A [58]       | 2009                | Greece        | stray       | 284         | Leishmania spp.     | ELISA         | 3.9              | 0.5              |
| Figueiredo. FB [11]  | 2009                | Brazil        | domestic    | 43          | Leishmania spp.     | IFAT, ELISA   | 2.4              | 0.5              |
| Hatam. GR [59]       | 2010                | Iran          | domestic    | 40          | *L. infantum*       | PCR           | 0.9              | 0.5              |
| Veronesi. F [60]     | 2010                | Italy         | mixed       | 95          | Leishmania spp.     | IFAT, PCR     | 9.5              | 3.9              |
| Cardoso. L [35]      | 2010                | Portugal      | domestic    | 316         | *L. infantum*       | ELISA, DAT    | 2.8              | 0.5              |
| Duarte. A [61]       | 2010                | Portugal      | stray       | 180         | *L. infantum*       | IFAT          | 0.6              | 0.5              |
| Maia. C [62]         | 2010                | Portugal      | domestic    | 142         | *L. infantum*       | IFAT, PCR     | 1.3              | 20.4             |
| Costa. TA [63]       | 2010                | Brazil        | stray       | 200         | Leishmania spp.     | ELISA         | 11.5             | 0.5              |
| Dahroug. MA [64]     | 2010                | Brazil        | mixed       | 16          | *L. infantum*       | PCR           | 0.5              | 37.5             |
| Bresciani. KD [65]   | 2010                | Brazil        | domestic    | 283         | Leishmania spp.     | IFAT          | 0.9              | 0.5              |
| Sherry. R [36]       | 2011                | Spain         | mixed       | 105         | *L. infantum*       | ELISA, PCR    | 12.4             | 8.6              |
| Milián. J [66]       | 2011                | Spain         | mixed       | 86          | *L. infantum*       | WB, PCR       | 15.7             | 25.6             |
| Miró. G [67]         | 2011                | Spain         | .           | 20          | *L. infantum*       | IFAT          | 15               | 0.5              |
| Vides. JP [12]       | 2011                | Brazil        | .           | 55          | *L. infantum*       | IFAT, ELISA   | 27.3             | 0.5              |
| Da Silveira Neto. L [68]| 2011             | Brazil        | .           | 113         | *L. infantum*       | ELISA         | 34.5             | 0.5              |
| Coelho. WM [13]      | 2011                | Brazil        | .           | 70          | Leishmania spp.     | IFAT, ELISA   | 4.2              | 0.5              |
| Coelho. WM [13]      | 2011                | Brazil        | .           | 52          | *L. infantum*       | PCR           | 0.9              | 0.5              |
| Pennisi. MG [69]     | 2012                | Italy         | mixed       | 431         | Leishmania spp.     | IFAT, PCR     | 6.9              | 18.3             |
| Ayllon. T [70]       | 2012                | Spain         | mixed       | 680         | *L. infantum*       | IFAT, PCR     | 3.7              | 0.6              |
| Sobrinho. LS [14]    | 2012                | Brazil        | stray       | 302         | *L. infantum*       | IFAT, ELISA   | 15.23            | 0.5              |
| Longoni. SS [17]     | 2012                | Mexico        | stray       | 95          | *L. infantum*       | ELISA, WB     | 31.6             | 0.5              |
| Spada. E [71]        | 2013                | Italy         | stray       | 233         | Leishmania spp.     | IFAT, PCR     | 25.3             | 0.5              |
| Vilhena. H [9]       | 2013                | Portugal      | domestic    | 320         | *L. infantum*       | PCR           | 0.3              | 0.5              |
| Cardia. DF [72]      | 2013                | Brazil        | stray       | 386         | Leishmania spp.     | IFAT          | 0.5              | 0.5              |
| Silva. RD [73]       | 2013                | Brazil        | .           | 153         | *L. infantum*       | ELISA         | 3.9              | 0.5              |
| Chatzis. MK [10]     | 2014                | Greece        | domestic    | 100         | *L. infantum*       | IFAT, ELISA, PCR | 11            | 41              |
| Silaghi. C [74]      | 2014                | Albania       | stray       | 146         | Leishmania spp.     | IFAT, PCR     | 0.7              | 0.5              |
| Miro. G [75]         | 2014                | Spain         | stray       | 346         | *L. infantum*       | IFAT, PCR     | 3.2              | 0.5              |
| Maia. C [76]         | 2014                | Portugal      | mixed       | 649         | *L. infantum*       | PCR           | 0.5              | 0.5              |
| Maia. C [76]         | 2014                | Portugal      | mixed       | 271         | *L. infantum*       | DAT           | 3.7              | 0.5              |
| Nimsuphan. B [77]    | 2014                | Bangkok       | pet         | 237         | *L. donovani*       | DAT           | 0.8              | 0.5              |
| Moreno.I [78]        | 2014                | Spain         | stray       | 43          | *L. infantum*       | IFAT          | 9.3              | 0.5              |
| Dorbadam. SM [79]    | 2014                | Iran          | stray       | 50          | *L. infantum*       | DAT           | 2.3              | 0.5              |
| Author          | Year of publication | Country | Type of cat | Sample size | Leishmania species | Lab test                  | Seropositive (%) | PCR positive (%) |
|-----------------|---------------------|---------|-------------|-------------|-------------------|----------------------------|------------------|------------------|
| Sousa. KC [80]  | 2014                | Brazil  | domestic    | 151         | L. infantum       | IFAT                       | 6.6              | .                |
| Fatollahzadeh. M [81] | 2014          | Iran    |             | .           | L. infantum       | DAT, PCR                  | 23.0             | 0                |
| Braga. AR [82]  | 2014                | Brazil  | domestic    | 50          | Leishmania spp.   | IFAT                       | 4                | .                |
| Costa. AP [83]  | 2014                | Brazil  | domestic    | 52          | L. infantum       | IFAT                       | 3.8              | .                |
| Braga. AR [84]  | 2014                | Brazil  |             | 100         | Leishmania spp.   | IFAT, PCR                  | 15               | 0                |
| Nemati. T [85]  | 2015                | Iran    |             | 65          | L. infantum       | DAT                        | 27.7             | .                |
| Maia. C [86]    | 2015                | Portugal| mixed       | 271         | L. infantum       | IFAT, PCR                  | 30               | 2.2              |
| Dincer. E [87]  | 2015                | Turkey  |             | 22          | L. infantum       | PCR                        | 4.5              | .                |
| Oliveira. TM [88]| 2015               | Brazil  |             | 52          | Leishmania spp.   | PCR                        | 13.5             | .                |
| Spada. E [89]   | 2016                | Italy   | stray       | 90          | L. infantum       | IFAT, PCR                  | 30               | 2.2              |
| Figueiredo. FB [90]| 2016            | Brazil  | domestic    | 34          | L. braziliensis   | ELISA                      | 20.6             | .                |
| Persichetti. MF [91]| 2016            | Spain   |             | 42          | L. infantum       | IFAT, PCR                  | 2.4              | 7.1              |
| Can. H [16]     | 2016                | Turkey  | stray       | 1101        | L. infantum, L. tropica | IFAT, ELISA, PCR          | 10.5             | 0.54             |
| Persichetti. MF [15]| 2017            | Italy   |             | 161         | L. infantum       | ELISA, IFAT, WB            | 29.2             | .                |
| Otranto. D [92] | 2017                | Italy   |             | 330         | L. infantum       | IFAT, PCR                  | 25.8             | 3.9              |
| Mylonakis. ME [93]| 2017            | Greece  |             | 100         | L. infantum       | PCR                        | .                | 41               |
| Mohabili. M [94]| 2017                | Iran    | stray       | 103         | L. infantum       | DAT                        | 24.3             | .                |
| Metzendorf. IP [95]| 2017           | Brazil  | domestic    | 100         | L. infantum       | PCR                        | .                | 6                |
| De Mendoça. IL [96]| 2017           | Brazil  | domestic    | 83          | L. infantum       | ELISA                      | 4                | .                |
| Lopes. AP [97]  | 2017                | Portugal| domestic    | 102         | Leishmania spp.   | DAT                        | 0                | .                |
| Poffo. D [98]   | 2017                | Brazil  | domestic    | 88          | Leishmania spp.   | PCR                        | .                | 0                |
| Akhtardanesh. B [99]| 2017          | Iran    | stray       | 60          | L. infantum, L. tropica | ELISA, PCR                | 6.7              | 16.7             |
| Benassi. JC [100]| 2017               | Brazil  | mixed       | 108         | L. infantum       | PCR                        | .                | 1.85             |

Table 2. Subgroup meta-analysis for seroprevalence and PCR prevalence of *Leishmania* infection in cats

| Characteristics | Factors | n | Prevalence (%) (95%CI) | I-square (%) | p  | n | Prevalence (%) (95%CI) | I-square (%) | P<0.001 |
|-----------------|---------|---|------------------------|--------------|----|---|------------------------|--------------|---------|
| Country         |         |   |                        |              |    |   |                        |              |         |
| Iran            |         | 6 | 17.0 (8.0-28.0)        | 83.8         | 0.06 | 3 | 6.0 (0.0-21.0)        | -            |         |
| Egypt           |         | 3 | 6.0 (2.0-10.0)        | 98.3         | -   |   |                        |              |         |
| Greece          |         | 3 | 11.0 (2.0-26.0)       | 95.2         | -   |   |                        |              |         |
| Italy           |         | 10| 24.0 (13.0-37.0)      | 97.1         | 7   | 21.0 (10.0-61.0)      | 99.5         | P<0.001 |
| Spain           |         | 12| 12.0 (4.0-23.0)       | 97.7         | 8   | 6.0 (1.0-14.0)       | 96.7         |         |
| Portugal        |         | 7 | 2.0 (1.0-4.0)        | 70.1         | 4   | 11.0 (2.0-26.0)      | 96.7         |         |
| Brazil          |         | 17| 8.0 (3.0-13.0)       | 93.7         | 7   | 5.0 (1.0-11.0)       | 85.3         |         |
| ELISA           |         | 12| 9.0 (5.0-13.0)       | 87.8         | P<0.001 | - | - | - | - |
| IHAT            |         | 3 | 6.0 (2.0-10.0)       | 97.5         | -   |   |                        |              |         |
| IFAT            |         | 30| 11.0 (6.0-17.0)      | 97.7         | -   |   |                        |              |         |
| Diagnostic test |         |   |                        |              |    |   |                        |              |         |
| IFAT, ELISA     |         | 6 | 11.0 (7.0-16.0)      | 97.5         | -   |   |                        |              |         |
| WB              |         | 3 | 14.0 (9.0-20.0)      | 96.6         | -   |   |                        |              |         |
| DAT             |         | 9 | 10.0 (3.0-19.0)      | 94.9         | -   |   |                        |              |         |
| PCR             |         | 12| 7.0 (3.0-14.0)       | 93.5         | -   |   |                        |              |         |
| Stray           |         | 22| 10.0 (6.0-14.0)      | 95.2         | 0.46 | 7 | 2.0 (0.0-5.0)       | 90.5         |         |
| Breed of cat    |         |   |                        |              |    |   |                        |              |         |
| Domestic        |         | 17| 7.0 (2.0-16.0)       | 97.6         | 9   | 8.0 (1.0-19.0)      | 96.8         | P<0.001 |
| Mixed           |         | 13| 15.0 (8.0-24.0)      | 95.6         | 16  | 23.0 (4.0-50.0)     | 99.4         |         |
| L. infantum     |         | 36| 12.0 (8.0-17.0)      | 95.4         | 0.09 | 23 | 8.0 (4.0-14.0)      | 95.9         |         |
| Leishmania species |       |   |                        |              |    |   |                        |              |         |
| L. infantum, L. tropica |       | 25| 8.0 (4.0-14.0)      | 96.7         | 9   | 15.0 (5.0-48.0)     | 99.4         | P<0.001 |

NR=not reported, NE=not enough studies
Figure 2. Forest plot for the prevalence of Leishmania infection in cats by PCR and serology tests.

Figure 3. Funnel plot for seroprevalence (a) and PCR prevalence (b) of Leishmania infection in cats.
**Discussion**

Zoonotic VL (ZVL) is a zoonosis that occurs in the Old and New World. Research studies on FLI are limited but have become more numerous internationally in recent years, especially in Brazil [23]. In this study the highest numbers of reported studies (30.8%) were found in Brazil, where leishmaniasis is a major public health problem. Brazil is one of the countries with the highest prevalence and widest geographical distribution of the disease [2, 24].

In the current study, the overall prevalence of FLI was estimated to be 10%. The relatively high prevalence of infection in cats demonstrates a similarity with dogs exposed to the leishmanial infections in some endemic areas [25–28]. A larger prevalence of *L. infantum* infection in dogs compared to cats is related to the immune-system differences in these two species and a more efficient Th1 immune response in cats compared to dogs [25,28]. The different FLI reports and the increased cat populations in diverse areas highlight the ability of these animals to maintain and spread the infection in natural and urban environments [28]. Overall, little information is available on the adaptive immune response of cats naturally exposed to *L. infantum* infection and mechanisms responsible for susceptibility or resistance of feline hosts. However, some evidence suggests that large numbers of clinical cases of FLI are reported in cats that are probably immunocompromised [8], although it seems that asymptomatic cases have an immunocompetent condition and act as cryptic reservoir hosts.

Based on our findings, FLI in the mixed-type/breed cats were higher than in other feline types/breeds. The role of domestic cats has been controversial in leishmaniasis epidemiology because they live in close contact with humans. Domestic cats can act as primary, secondary or accidental hosts [7, 28].

Seroprevalence rates from 0.9% to 28.5% and PCR detection rates between 0.43% and 30% have been reported in some regions and countries such as Spain, Portugal, France, and Italy in Southern Europe, as well as in North Africa, Iraq, Iran, Turkey and Central and South America, where canine leishmaniasis is endemic [29–36].

In our study, both seroprevalence (24%) and PCR prevalence (21%) of FLI were found higher in Italy than in other countries. Moreover, our data show the high seroprevaence rate (12.2%) of FLI in Southern European countries including Italy, Spain, Portugal, and Greece. However, this could be justified by the increased finding of active cases in cats, development of simple and rapid diagnostic tests and elevated rate of disease prevalence in these countries.

Diagnosis is usually based on the results of cytology, histopathology, immunohistochemistry (IHC), culture, serology and PCR. Apart from the advantages and limitations inherent to each of these methods, their diagnostic value depends on many factors, including the biological sample being used, the reagents and the particular technique employed.

In our study the most common diagnostic laboratory method was IFAT (38.5%). IFAT and ELISA are the most common serological techniques used for diagnosis and for clinical and research studies on canine and feline leishmanial infections [10–16]. In areas endemic for *Trypanosoma* spp. or other *Leishmania* spp., cross reactions with *L. infantum* must be taken into account for interpretation of serological tests [16]. However, some attention is needed before confirming leishmaniasis with IFAT in cats, whereas cats that present clinical symptoms for leishmaniasis but are found negative by IFAT should be subjected to other serological tests or complementary diagnostic tools such as WB and PCR [7]. WB analysis, a qualitative serological method, distinguishes the molecular weight of the *L. infantum* antigens stimulating antibody production, but is less frequently used for the diagnosis of leishmaniasis [37]. One potential application of the WB method is the discrimination between subclinical and clinical infections [38].

Three different species of *Leishmania* have been found in cats in Brazil: *L. amazonensis* [39], *L. braziliensis* [40] and *L. infantum* [34, 41]. Five *Leishmania* species have been reported in cats worldwide, although most cases involved *L. infantum* [8]. This is in agreement with our findings in the current study in which approximately 63% of species were *L. infantum*.

In conclusion, our data provide substantial evidence that cats can be considered sentinel reservoir hosts at least in endemic foci of zoonotic visceral leishmaniasis. Moreover, the current data demonstrate enzootic stability of FLI in several countries of the world particularly in some European countries. Furthermore, our results show the most common lab method for diagnosing FVL is the IFA test. In general, control of cat populations is recommended to reduce the transmission of *Leishmania* spp. among human populations in the endemic areas especially among nomadic tribes [42].

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**Abbreviations**

Not applicable.

**Availability of data and material**

All data extracted and or analyzed during this study are included in this published article.

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Competing interests
The authors declare that they have no competing interests.

Authors' contributions
SA wrote the preliminary draft of the manuscript and extracted all data. MF designed all steps of the study and contributed to writing and revising of the final manuscript. SHT contributed to meta-analysis of the extracted data. All authors read and approved the final manuscript.

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