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Original paper

An epidemiological study of cutaneous leishmaniosis in human and dogs

Rasha Khalil Abduljalil ALSAAD[1,2], May Hameed KAWAN[1]

[1]Department of Parasitology, College of Veterinary Medicine, University of Baghdad, Baghdad, Iraq
[2]Department of Microbiology, Faculty of Medicine, University of Misan, Misan, Iraq

Corresponding Author: Rasha Khalil Abduljalil Alsaad; e-mail: rashak.mcm@uomisan.edu.iq

ABSTRACT. Cutaneous leishmaniosis (CL) is a vector-borne parasitic disease with widespread on the communities throughout the Mediterranean regions and the Middle East, including Iraq. The study aimed for detection and diagnosis of CL in humans and dogs. This is an epidemiological study included six districts in Misan province, during the period from December 2019 to November 2020. All samples were taken from patients and owned-dogs. A total of 436 patients suspected with CL enrolled. A total of 237 owned-dogs were examined. The mean age of patients was (18.15±16.92) years. The rate of CL infection was high in Al-Amarah district. About 409 (93.807%) patients were diagnosed as a new case of CL, with no significant difference (P>0.05). The duration from bites by sandflies to the time of being diagnosed with CL ranged from 1 to 18 weeks. Patients owned-dogs (direct contact) were 167 (38.302%), and those with indirect contact were 123 (28.211%). Dogs manifested with clinical features of CL were 74 (27.715%), while those were asymptomatic (193, 72.284%). The lesions in dogs were severe (47, 63.513%), moderate (22, 29.729%), and mild (5, 6.756%). In conclusion, to the best of our knowledge, this epidemiological study of CL was conducted for the first time in Iraq.

Keywords: cutaneous leishmaniosis, owned-dogs, parasitic diseases, vector-borne, zoonotic infection, *Leishmania*

Introduction

Leishmaniosis is one of the major vector-borne communicable diseases of the world [1]. It is a zoonotic infection caused by obligate intracellular protozoa of the genus *Leishmania*. Natural transmission of *Leishmania* parasites is carried out by sandflies of the genus *Phlebotomus* (Old World) or *Lutzomyia* (New World) [2]. The disease is widespread and may cause serious health problems in communities throughout the Mediterranean regions and the Middle East, including Iraq [3–5].

There are an estimated 12 million cases worldwide, and there are about 1.5 million new cases of CL each year, of which over 90% occur in Afghanistan, Algeria, Iran, Iraq, Yemen, Saudi Arabia, Syria, Brazil, and Peru [6–8].

Old World CL has three distinctive varieties in the Eastern Hemisphere: urban or dry, caused by *L. tropica*; a rural or wet type, caused by *L. major*, and a diffuse cutaneous type caused by *L. aethiopica*. Clinical, epidemiological, immunological, and biochemical differences among these varieties indicate that each is a distinct entity [9]. Epidemiological studies in the Middle East have shown that anthroponotic CL caused by *L. tropica* and zoonotic CL caused by *L. major* [10,11]. The WHO considers CL one of the six most important infectious diseases due to its high incidence rates and the potential to cause deformities in patients, and it is endemic in 88 countries [11–13].

It is called "Balkhsore" in northern Afghanistan, ''Delhi boil'' in India, and ''oriental sore'' or ''Baghdad boil'' in Iraq [14].

Several cases of cutaneous infection in *L. major* infected dogs have been reported in Saudi Arabia, Egypt, and Iraq [15,16]. Infected dogs act as a potential primary reservoir [17].

There are over 20 species of *Leishmania* that have been recorded as causative infectious agents to humans, the distribution of each is determined by the distribution of vector, reservoir host, or both [6,18].

The canine CL is a neglected disease, mainly because most of the cases occur far away from cities, that is, far from veterinary services. Dogs have been identified to be an important host for the parasite. Infected dogs normally develop both visceral and cutaneous lesions, whereas restriction to the skin occurs in some forms of human leishmaniosis (oriental sore) [19,20]. The disease in dogs is characterized by local, self-healing ulcerative lesions on the ears, scrotum, feet, nipples, and muzzle [19,21].

Here, the study tries to detect and diagnose CL in human, and dogs in the highly endemic area in Iraq. Besides, the study determines the effect of age, sex, and climate factors on the infection rates of CL and severity.

Materials and Methods

Study design, setting, and locations

This is an epidemiological study that included six districts in Misan province, Iraq: Ali Al-Gharbi, Al-Amarah, Al-Kehlaa, Al-Maymouna, Al-Majar Alkabeer, and Qal'at Saleh. The study period was 12 months, started from the beginning of December 2019 to the end of November 2020.

Sample collection

All samples were taken from patients and domestic dogs, who were clinically diagnosed with CL. A total of 436 patients suspected with CL were enrolled in this study, 234 males and 202 females. They attended to the Dermatological and Venereology Department in Al-Sadder Teaching Hospital, and public health laboratories in Misan Health Directorate.

History and data collection

A history was taken from each case according to a questionnaire sheet. Items included sex, age, address, months of detection, duration of the lesions, sites of lesions, other associated symptoms, other dermatological diseases, comorbidities, history of previous CL, home type (regarding building, roof, and floor), garden presentation, features near the house (river, branches, mini-branches, marshes, and settlements), family history of CL, number of family members infected, and previous infections with CL. Regarding dog owners, the questions included the number of dogs, sex, maturity, and any signs of CL seen by patients on their dogs.

Clinical dermatological examination

The dermatological examination was performed for all patients with suspected CL and included: examined sites, nature, number, consistency, the severity of lesions, and other associated signs.

Dogs

A total of 237 domestic dogs of patients were examined during visiting their homes, of both sex, and maturity from areas of patients' addresses.

Pus and discharge aspiration

Each skin lesion was cleaned with iodine or ethanol, and then about 0.5 to 1 ml of Lock's solution subcutaneously injected using a disposable insulin syringe at the peripheral edges of the lesion. Then aspirated solution and stored in Eppendorf tubes, to be directly used for examination by direct smear and culture [22].

Skin scraping

A blunt blade scrape in the direction of hair growth was used to collect skin samples. Initially, we scraped the superficial layer and then subsequently scraped deeper (enough to cause capillary ooze). Then the small scraped pieces were transferred to a cup for examination [23].

Diagnosis

Direct smear methods

Exudative and purulent materials aspirated from lesions were smeared onto a clean slide and allowed to air-dry. They were then fixed with methyl alcohol for 1–2 minutes [24–26].

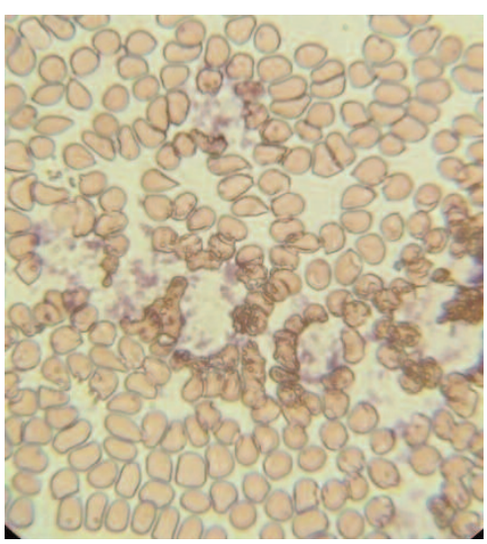

Figure 1. Smears from lesions stained with Giemsa stain show amastigotes in macrophages under oil immersion (100×)

Staining with Giemsa

The slides were washed with tap water. Then stained with Giemsa stain for 45 minutes after immersed in a solution of one part Giemsa stock solution (commercial liquid stain) to 10 parts D.W. (pH 7.2). then discarded the excess stain and rinsed the slides in buffered water. Then drained the slides thoroughly in a vertical position and allowed them to air-dry. Then mounted the stained smears in a neutral mounting medium (Canada balsam or DPX). After that, the slide examined under an oil immersion objective lens (100× magnification), according to Alsaad et al. [27], as shown in figure 1.

Staining with Leishman

We covered the air-dried blood smear with 10 drops of stain and leaved for one min. Then diluted the stain solution with double volume of drops of buffered water drops (pH=6.5). Then allowed it to stand for 20 minutes and then drained off the excess stain by washing with tap water. Finally, rinsed the stained slide with buffered water and stand it vertically for air-drying. Then examined under oil immersion objective lens (100× magnification), according to Al-Emarah et al. [28], as showed in figure 2.

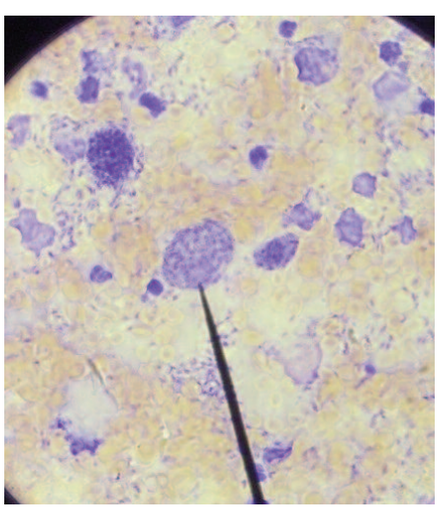

Figure 2. Smears from lesions stained with Leishman stain show amastigotes in macrophages under oil immersion (100×)

Ethical consideration

Written informed consent was obtained from all participants, or the parents of those aged less than 18 years. The Medical Ethical Committee at the College of Veterinary Medicine, University of Baghdad approved this study (Ref. No. 2650, 17/12/2019).

Statistical analysis

All data collected were entered for statistical analysis into the Statistical Package for Social Sciences version 24 (SPSS v24) (SPSS Inc., Chicago, Illinois, USA). Descriptive statistical analysis of clinical characteristics was performed. The mean, standard deviation, and ranges were used to described numerical data. Categorical data are presented as frequencies and percentages. A two-sided *P*-value <0.05 was considered statistically significant. Logistic regression was used where more appropriate and to incorporate multiple variables as well as their interactions in a single analysis.

Results

CL and patients demographic

The mean age of patients was (18.15±16.92) years and the median age was 12 years. There were thirteen patients aged less than one year. Ages ranged from 6 months to 75 years. High rates of CL infection were found in patients aged 6 months – 10 years group (198, 45.412%). Therefore, the rate of infection was higher in younger age groups and began to decline gradually in older age groups with some significant differences ($P<0.05$).

The rate of infection in males was higher than in females (53.669% vs 46.33%), with no significant difference between sexes ($P>0.05$).

The rate of infection was highest in Al-Amarah district (198 patients, 45.412%), followed by Al-Kehlaa (66, 15.137%), Ali Al-Gharbi (63, 14.449%), Al-Maymouna (51, 11.697%), Al-Majar Alkabeer (33, 7.568%), with the lowest rate recorded in Qal'at Saleh (25, 5.734%) (Fig. 3), with some significant differences among patient's area of residence ($P<0.01$). The high rate reflected the crowding pattern of patients with CL living in major cities.

Only nine patients (9/436) had at least one dermatological disease other than CL. This finding had no effect on the rate of CL infection ($P>0.05$).

Thirty-six out of 436 patients who were known to have diabetes mellitus (DM), suffered from a severe form of CL, although there were no significant differences ($P>0.05$).

Just 27 (6.192%) patients were known to have been previously infected with CL, with 409 (93.807%) patients were diagnosed as a new case of CL, with no significant difference ($P>0.05$) (Tab. 1).

Table 1. Rate of infection of CL by demographic characters of patients (n=436)

Variables		No.	%	P-value
Age groups (years)*				
	6 months–10	198	45.412	<0.05
	11–20	98	22.477	
	21–30	49	11.238	
	31–40	34	7.798	
	41–50	30	6.88	
	51–60	17	3.899	
	61–70	8	1.834	
	>70	2	0.458	
Gender	Male	234	53.669	NS
	Female	202	46.33	
Districts	Ali Al-Gharbi	63	14.449	<0.01
	Al-Amarah	198	45.412	
	Al-Kehlaa	66	15.137	
	Al-Maymouna	51	11.697	
	Al-Majar Alkabeer	33	7.568	
	Qal'at Saleh	25	5.734	
Other dermatological diseases**	Positive	9	2.064	NS
	Negative	427	97.935	
Comorbid conditions***	Positive	36	8.255	NS
	Negative	400	91.743	
Previous CL infection	Yes	27	6.192	NS
	No	409	93.807	

Explanations: *13 patients aged below one years; Mean±SD (Median)=18.15±16.92 (12); Range (min-max): 6 months–75 years; **Scabies, and acne vulgaris; ***Diabetes mellitus; NS=non-significant

CL and climate

According to the months of the year of the study, higher infection rates were observed in December 2019, January 2020, and February 2020 (25.917%, 21.1%, and 16.743%, respectively). After that the rates dropped, which may be due to the effect of the climate on the parasite's vector, there was a highly significant difference (P<0.001).

The results showed the rate of infection was higher in months with moderate mean temperature (ºC) in the months December, January, and February, whereas the rate declined during the hotter months of April, May, June, August, and September (infection rates: 2.293%, 1.605%, 0.229%, 0.458%, and 6.192%, respectively), but in July, the rate was zero. These findings showed significant differences in the rate of CL (P<0.05).

In relation to precipitation rate (mm), the rates were moderately high in the rainy months of November, December, January, and February, while the rates were lower in the remaining months, except September and October when the rates were 6.192%, and 8.486%, respectively, despite zero percipitation, with highly significant differences (P<0.01).

The high mean humidity (%) resulted in a high rate of CL infection in Misan province. November, December, January, and February had high humidity associated with high rates of infection, with significant differences. September and October had

Table 2. Rate of infection of CL according to months and climate factors

Month	Mean temperature (C°)	Precipitation amount (mm)	Mean humidity (%)	Wind speed (m/s)	Wind direction	No.	%
December 2019	20	37.1	78	2	NW	113	25.917
January 2020	18.6	33.9	71	1.9	EE	92	21.1
February 2020	20.7	44.5	66	2.6	EE	73	16.743
March 2020	27.1	24	57	2.8	EE	34	7.798
April 2020	32.1	6.6	44	2.9	EE	10	2.293
May 2020	39.9	0	29	3.5	N	7	1.605
June 2020	44.9	0	23	3.9	NW	1	0.229
July 2020	48.3	0	22	2.9	NW	0	0
August 2020	45.7	0	23	3	NW	2	0.458
September 2020	45.2	0	25	2.2	NW	27	6.192
October 2020	26.6	0	34	1.5	NW	37	8.486
November 2020	37.5	65.3	60	2.3	EE	40	9.174
Total/mean±SD	33.88±11.072	21.73±17.617	44.33±20.986	2.625±0.657		436	99.995
P value	<0.05	<0.01	<0.01	NS	NS		<0.001

Explanations: NW: north-west; EE: east-east; N: north; NS: non significant

moderate humidity but had moderately high rates of infection, with highly significant differences (P<0.01).

Different wind speeds and directions showed no association with differences in CL infection rates, (P>0.05) (Tab. 2).

CL and patients' accommodation

In relation to patients' accommodation, the rates of CL infection are shown in table 3. Most of the infected patients lived in fired-bricks houses (299, 68.577%), followed by people lived in buildings made of blocks (86, 19.724%), with fewest living in mud-bricks houses (51, 11.697%), with significant difference (P<0.05).

Patients living in houses with a *cashi* roof had the highest rate of CL infection (55.275%), while patients who lived in houses with roofs made of clay had the lowest rate of infection (5.504%), with no significant difference (P>0.05). Patients living in houses with concrete floors had a slightly higher rate of CL infection compared with those who lived in houses with *chinko* floors (52.293% versus 47.706%), with no significant difference (P>0.05). Patients who had gardens with their houses were infected by CL more than those who did not have gardens (67.201% versus 32.798%); this difference was significant (P<0.01).

Regarding to the features near the houses, we noted that the rate of CL infection was high in regions near settlement, and marshes as 48.853%, and 25.917%, respectively. The lowest rate of

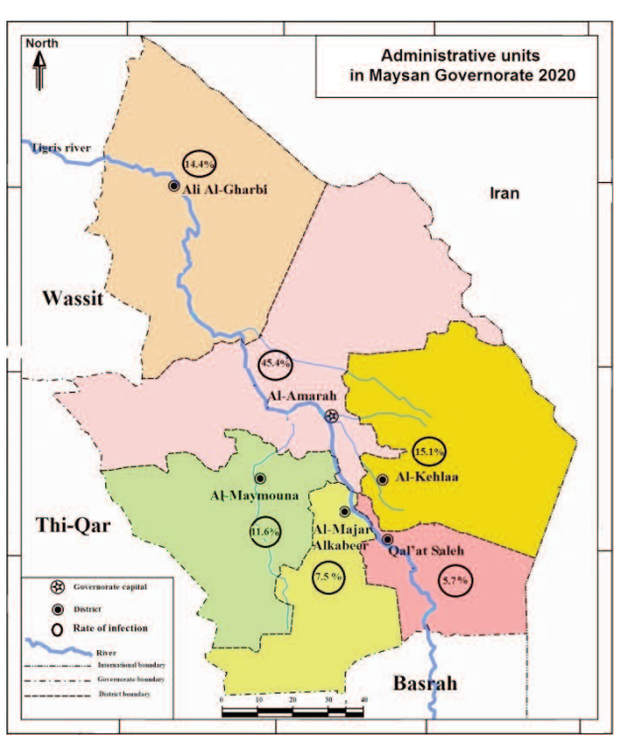

Figure 3. Maysan government map showing the rates of infection of CL according by district

Table 3. Rate of CL infection and patients' accommodation (n=436)

Variables		No.	%	*P*-value
House	Mud bricks	51	11.697	<0.05
	Blocks building	86	19.724	
	Fired bricks	299	68.577	
Roof	Clay	24	5.504	NS
	Sabah	171	39.22	
	Cashi (ceramic)	241	55.275	
Floor	Chinko	208	47.706	NS
	Concrete	228	52.293	
Garden	Yes	293	67.201	<0.01
	No	143	32.798	
Nearby water sources	Settlement	113	25.917	<0.05
	Marshies	213	48.853	
	Mini-branches of rivers	55	12.614	
	Branches of river	37	8.486	
	Rivers	18	4.128	

infection (4.128%) was reported in patients that lived near the River Tigris (one of the main rivers in Iraq), with a significant difference (P<0.05).

CL and patients' families

CL may be run in family regarding to the questions we asked. There were 143 (32.798%) patients who had family members infected with CL, whereas 293 (67.201%) did not have infected family members; this was a significant difference (P<0.05). There were 109 of 143 (76.223%) patients who had one member of their family infected with CL, 21 (14.685%) patients who had two infected family members, 12 (8.391%) patients who had three infected family members, and just one patient who had four infected family members, with a significant difference (P<0.05). Regarding previous infection in the family, 64 (14.678%) patients had previous infection in the family and 372 (85.321%) had no previous history of infection in the family, with no significant difference (P>0.05) (Tab. 4).

Clinical results of CL

Clinically, the upper limbs were the sites most commonly affected by CL, in 110 (25.229%) patients, followed by the head (including the face, ears, and nose) in 90 (20.642%), and the lower limbs in 72 (16.513%). In addition, CL lesions were detected in 153 (35.091%) patients in other sites of

Table 4. Rate of CL infection in patients' families (n=436)

Variables		No.	%	*P*-value
Is there any members of your family infected with CL?	Yes	143	32.798	<0.05
	No	293	67.201	
How much members infected with CL?(n=143)	1	109	76.223	<0.05
	2	21	14.685	
	3	12	8.391	
	4	1	0.699	
Is there any previous leishmaniosis cases in your family?	Yes	64	14.678	NS
	No	372	85.321	

Figure 4. A 3-years-old child with a single large cresting nodule on the left face (cheek)

Figure 5. An 11-months-old baby with two cresting papules lesions on the face (chin)

the body. These findings were statistically significant (P<0.01), as shown in Figs 4–7.

Regarding the nature of CL lesions, papules were the most common lesions diagnosed, in 190 (43.577%) patients, followed by plaques in 119 (27.293%) patients. The least common types were nodules and pustules in 28 (19.266%) and 15 (3.44%) of cases, respectively. Ulcerative lesions were present in 84 (19.266%) patients. These findings showed a significant difference (P<0.05).

A total of 385 out of 436 patients (88.302%) had fewer than ten lesions, while 51 (11.697%) patients had more than ten lesions at the time of

Figure 6. A 13-years-old boy with six multiple CL lesions (cresting pustules on arms and feet)

Figure 7. A 9-months-old baby with a single large cresting ulcerative lesion on the left arm

examination; this was a significant difference (P<0.05).

In this study, the duration from a bite by sandfly to the time of being diagnosed with CL ranged from 1 to 18 weeks, with a mean duration of 5.69±3.22 weeks. Indeed, these different durations highly impacted the severity CL, and this was a highly significant association (P<0.000). The nature of most of the CL lesions was wet in 379 (86.929%) patients, while the remainder (57, 13.073%) were dry in nature, with a significant difference (P<0.05). CL lesions manifested with pus discharge in 119 (27.293%) patients; lesions associated with itching were seen in 105 (24.082%) patients, with bleeding from lesion sites in 70 (16.055%) of patients. In addition, mixed symptoms and signs were presented in 135 (30.963%) patients. The fewest lesions were associated with redness, in just 7 (1.605%) patients. These findings were not significant (P>0.05). In terms of the severity of CL lesions, the most common presentation was as moderate lesions, in 283 (64.908%) patients, followed by severe forms (132, 30.275%), and mild lesions (21, 4.816%),

Table 5. Rate of CL infection according to lesions characters (n=436)

Variables		No.	%	*P*-value
Sites	Head	90	20.642	<0.01
	Neck	6	1.376	
	Upper limbs	110	25.229	
	Chest	3	0.688	
	Lower limbs	72	16.513	
	Back	2	0.458	
	Different sites	153	35.091	
Nature	Nodule	15	3.44	<0.05
	Plaque	119	27.293	
	Papule	190	43.577	
	Pustule	28	6.422	
	Ulcer	84	19.266	
No. of lesions*	<10	385	88.302	<0.05
	>10	51	11.697	
Duration (weeks)**	1–4	214	49.082	<0.000
	5–8	146	33.486	
	9–12	58	13.302	
	>12	18	4.128	
Consistence of lesion	Moist or wet	379	86.926	<0.05
	Dry	57	13.073	
Other symptoms and signs	Redness	7	1.605	NS
	Itching	105	24.082	
	Bleeding	70	16.055	
	Pus discharge	119	27.293	
	Mixed	135	30.963	
Severity	Mild	21	4.816	<0.01
	Moderate	283	64.908	
	Severe	132	30.275	

Explanations: *Mean±SD (median)=3.02±2.247 (2); Range: 1–15; **Mean±SD (median)=5.69±3.22 (5) weeks; Range: 1–18

with a significant difference (P<0.01) (Tab. 5).

Relationship between patients and their contact with dogs

There were 167 (38.302%) patients who owned-dogs (direct contact), while those with indirect and no contact numbered 123 (28.211%) and 146 (33.486%), respectively. These results were significantly different (P<0.05) (Tab. 6).

There were 167 patients who were dog owners, and they owned 237 dogs in total. There were 102 (61.077%) patients who had one dog each (total 102 dogs), 23 (13.772%) patients who owned two dogs each (total 46 dogs), 10 (8.982%) patients who owned three dogs each (total 30 dogs), four (5.389%) patients who owned four dogs each (total 16 dogs). In addition, two (1.197%) patients owned five dogs each (total 10 dogs), and three patients owned 8, 10, and 15 dogs each, respectively. These findings showed significant differences (P<0.01) (Tab. 7).

Table 6. Relationship between patients and their contact with dogs (n=436)

Variables		No.	%	*P*-value
Patients' owners	Direct contact	167	38.302	<0.05
	Indirect contact	123	28.211	
	No contact	146	33.486	

Table 7. Patients and the number of dogs they owned

No. of dogs' owned	No. of patients	%	No. of owned-dogs (Totally)	%	*P*-value
1	102	61.077	102	43.037	<0.01
2	23	13.772	46	19.409	
3	10	5.988	30	12.658	
4	4	2.395	16	6.751	
5	2	1.197	10	4.219	
8	1	0.598	8	3.375	
10	1	0.598	10	4.219	
15	1	0.598	15	6.329	
Total	167		237		

CL and dogs

Of the owned-dogs, samples were collected from 169 (63.295%) males and 98 (36.704%) females. There were 189 (70.786%) adult dogs and 78 (29.213%) puppies. Following clinical examination, owned dogs manifesting clinical features of CL numbered 74 (27.715%), while there were 193 (72.284%) that appeared asymptomatic. All these findings of no significance (*P*>0.05) (Tab. 8).

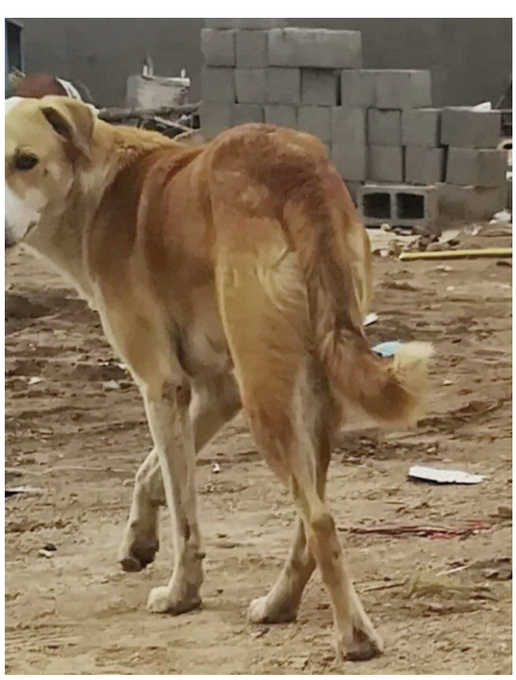

Figure 8. Adult dog with multiple nodular lesions in the legs

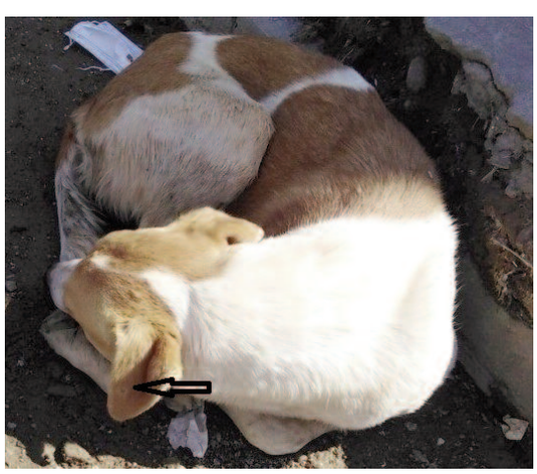

Figure 9. Adult dog with a single papule in the ear

Clinical manifestations of CL in owned-dogs were most commonly seen in the head and neck, in 30 (40.54%) dogs, the tail (21, 28.378%), legs (18, 24.324%), and trunk (5, 6.756%), with a significant difference (*P*<0.05), as shown in figures 8 and 9.

Papules were the most common types of lesions, seen in 32 (43.243%) cases. Ulcerative lesions were present in 29 (39.189%) of owned dogs. In addition, nodular lesions were found in 13 (17.567%) dogs, with a significant difference (*P*<0.05). There were 28 (37.837%) owned dogs with fewer than five

Table 8. Dogs infected with CL (n=237)

Variables		No.	%	*P*-value
Sex	Male	139	58.649	NS
	Female	98	41.35	
Maturity	Adult	159	67.088	NS
	Puppy	78	32.911	
CL diagnosed	Clinical	74	31.223	NS
	No signs	163	68.776	

NS: non-significant

Table 9. CL clinical features in domestic dogs (n=74)

Variables		No.	%	*P*-value
Lesion sites	Head and neck	30	40.54	<0.05
	Body	5	6.756	
	Legs	18	24.324	
	Tail	21	28.378	
Lesion nature	Nodule	13	17.567	<0.05
	Papule	32	43.243	
	Ulcer	29	39.189	
No. of lesions*	<5	28	37.837	<0.05
	>5	46	62.162	
Consistence of lesion	Wet	59	79.729	<0.05
	Dry	15	20.27	
Severity	Mild	5	6.756	<0.05
	Moderate	22	29.729	
	Severe	47	63.513	

Explanations: *Mean±SD (median)=2.56±1.98 (3); Range:1–12

lesions, while there were 46 (62.162%) dogs with more than five lesions, with a significant difference (P<0.05). Wet lesions were the most commonly diagnosed, in 59 (79.729%) of cases, whereas dry lesions were present in 15 (20.27%) dogs, with a significant difference (P<0.05). Severe CL lesions were seem in 47 (63.513%), moderate lesions in 22 (29.729%), and mild lesions in 5 (6.756%) owned dogs, with a significant difference (P<0.05) (Tab. 9).

Discussion

Cutaneous leishmaniosis (CL) is a vector-borne parasitic disease that is widespread and causes serious health problems in communities throughout the Mediterranean regions and the Middle East, including Iraq [1,3–5]. It is endemic in Iraq and is widespread throughout nearly all provinces of the country.

In 2018, of the 200 countries and territories that reported to WHO, 97 (49%) were considered to be endemic for CL, with four having no previously reported cases of leishmaniosis. In the WHO Eastern Mediterranean Region (EMR), 82% (18/22) of countries and territories were endemic for CL, in the Americas Region (AMR) 58% (21/36), in the European Region (EUR) 47% (25/53), in the African Region (AFR) 40% (19/47), and in the South-East Asia Region (SEAR) 36% (4/11) [1,29].

WHO weekly reports from 2014 to 2018 revealed that in 2018, Iraq ranked in fifth position

worldwide for CL cases reported (11426), after Syrian Arab Republic (80215), Pakistan (19361), Brazil (16432), and Iran (15486) [1].

In Misan government, CL was the most common endemic disease, with a high prevalence. During this study, 436 patients were detected and diagnosed clinically and in the laboratory to be infected with CL.

Younger age groups are more susceptible to CL infection. We found 13 cases aged less than one year and a baby aged six months with severe and multiple lesions of CL. The mean age of patients was 18.15±16.92 years and the median age was 12 years. CL was most frequent in the age group 6 months to 10 years, with 198 cases (45.412%). These finding agree with the findings of another study in the Iraqi city of Kirkuk, with 43% of cases in the age groups 1 day–5 years and 6–10 years [30].

While our percentage of CL infection was higher than that reported by Shahatha and Saleh [31], of 35.5% in Anbar province. In another study, 20% of CL occurred in the 1–10-year age group in Salah-Adeen and Baghdad provinces [32]. In Al-Diwanyiah city, Abdlkadim [33] found that 31% of 10–20-year-olds were infected with CL [33].

Recently, a study by Ghezzai et al. [34] in Al-Najaf city reported 6.2% of CL patients were aged <10 years.

Our study showed that the ages between 6 months and 10 years are the most susceptible age for CL infection, while individuals aged more than 60 years were less affected. This depended on the nature of children's skin in these age groups, as it is very thin compared to other groups.

This result is similar to that of Abdulwahab [35] in Diyala, she found the age group most affected by CL was 0–14 years in both sexes (31.7%) and the lowest frequency was 11.6% in the age group >50 years.

In addition, this could be due to children at this age playing outdoors for a long time and as a result they would have more exposure to being bitten by sandflies, making them more susceptible or preferred by the carrier host (sandfly) to penetrate the skin during feeding.

Aldifaei [36] reported that the highest rate of infection occurred in the age group 1–10 years, at a rate of 47.27%, with the lowest rate in the age group 70–80 years, at a rate of 1.8%, in Al-Diwanyiah city.

In neighboring countries such as Iran, Mehdi et al. [37], reported a similar pattern. In Turkey, Eksi et al. [38], said CL can infect individuals at any age.

Both sexes were exposed to CL infection with no significant differences. This could be explained by the influence of society, customs, and traditions on sex. Males are more susceptible to infection because of movement and work in fields and farms outside, whereas in general women remain at home, and particularly in Islamic countries the women wear long clothes. As a result, the exposed parts of the body in women are less than those of men. All of these are reasons to make men more susceptible to being bitten by sandflies. These findings were supported by other studies in Iraq [35,36].

In Iran, a study by Mohammed et al. [39] found more males with CL (61.8%) than females (38.2%). Similar results were reported in Turkey by Mustafa et al. [40]. However, our results differ from another study done by Saki et al. [41], in Iran, which found that CL was more common in females (54.68%) than males (45.31%).

In Shujan's [42] epidemiological study, he found in Tuz, females were more infected with CL than males, and the same was reported by Klein and Roberts [43].

In this study, we considered Maysan city to be a rural area, as a result of its climate, the nature of its environment, the distribution of streams and marshes, and the Tigris river, which divides the city into two parts. It is the city with the highest prevalence of CL in Iraq. In recent years, the spraying of insecticides has diminish or not been carried out by the local government, which has led to the increasing spread of sandflies. In addition, there are no strategies for the control of stray animals, including rodents and dogs, which are the reservoir hosts of the parasite, as are domestic dogs. Ali [44] mentioned that the highest rate of CL in rural areas was 87.9% in all the years from 2012 to 2016 in his study, while the lowest rate in urban areas was 12.1% in Diyala province.

Aldifaei [37] in her thesis, showed that a low percentage of people with CL might have other dermatological diseases, such as scabies and acne vulgaris; she suggested this might be due to the low immunity of the skin, making the patient more vulnerable to another skin infection. There are some common skin diseases that should be distinguished and differentiated from CL, including scabies, skin cancers, and boils. In this study, also people with CL suffering from scabies and acne.

When asked about their medical history, 8.255% of patients reported having diabetes mellitus. In one study conducted in Saudi Arabia, the authors

recorded 13.4% of CL cases in Saudis who also had diabetes mellitus [45].

The highest infection rate of CL was recorded during months of low rainfall, when the temperature was moderate but the humidity was very high. These conditions are appropriate for the growth and activity of the vector host (i.e. sandflies). We did not record any infections in the hot months, such as July, when there were high temperatures, a lack of rain, low humidity, and strong, dry winds. These conditions affect the presence, growth, and reproduction of the sandfly and thus the rate of infection.

These results were similar to those of other studies conducted in Iraq, where it was found that the highest frequency of CL recorded was in February (36.66%) then January (23.33%), with the lowest rate in May (1.66%) [35]. Moderate temperature, humidity, and wind speed provide an appropriate environment for the emergence of sandflies, with the population density of these insects then decreasing during the hot months of the year, when the high temperatures do not allow these insects to appear [46].

A study of the seasonal distribution of CL, from 2012 to 2016, showed the highest rate of leishmaniosis recorded in the winter, which was 68.3%, followed by the spring (19.3%), then a decline in the summer (7.3%), with the lowest rate recorded in the autumn (5.1%) [44].

These findings are similar to those reported by Al-Obiadi et al. [47], from a study in Tikrit, but not similar to those reported in Iran [48] and Afghanistan [49]. This variation in monthly peaks could be due to the activity of sandflies or the aggregate population of sandflies in the wet season, or it may be related to the development of female insects and their requirement for blood during their life cycle for the maturation and development of eggs, especially in the spring [50,51].

Furthermore, we investigated patients' accommodation, regarding the type of house, floor, roof, garden, and water sources. A thesis by Ali [44] showed that the house and the nature of its components, such as the floor, ceiling, and walls, have an effect on the spread of the sandfly, and thus have a potential effect on the rate of CL infection. The presence or absence of a garden beside the house has a great impact on the presence of sandflies, as well as the proximity of the house to ponds, swamps, branches of rivers, and marshes, because they provide a humid environment suitab. for insect growth.

Poor housing and domestic sanitary conditions, such as lack of waste management or open sewerage, may increase sandfly resting sites as well as their access to humans. Sandflies are attracted to crowded houses as these provide a good source of blood-meals [44].

Asmaa et al. [52] found the highest percentage of CL cases were related to the geographical site, which was near water flowing in a stream all year and abundant freshwater holes, that provide sandflies with a suitab. environment to complete their life cycle and increase reproduction activities.

Thus, the typical pattern of housing materials and settlements varies from one area to another. Generally, settlements, mud houses, and farmhouses can be found in clustered villages and scattered traditional groups of houses and sometimes in scattered individual houses, whereas modern houses and traditional houses are found in cities and towns [45].

In terms of bites, the upper limbs were the most common site of bites, due to the exposed parts of the body that are not covered with clothes being the most exposed to sandfly attacks; these results are similar to the results of other studies. Some authors mentioned that the main part of the body infected with CL was the upper limbs and hands (38.5%), followed by the face (25%), then the lower limbs and feet (21.5%), while the lowest frequency of infection (15%) was in other parts of the body [35].

The most common sites of cutaneous lesion were on the upper and lower limbs in all years of one study (50.1%), followed by the face (49.6%), and the lowest rate in the trunk (0.2%) [44], but Al-Obaidi et al. [47], found the face and feet were the sites most commonly affected by lesions. Other studies in different parts of the world, in Turkey [53], Colombia [54], and Iran [48] reported similar data. However, there was disagreement with our results mentioned by [55], who found that lesions mainly occurred on the lower limbs and less frequently on the face.

CDC recommend that, to minimize the amount of skin exposed (uncovered) to sandfly biting, to the extent that is tolerable in the climate, wear long-sleeved shirts, long pants, socks, and tuck your shirt into your pants [3].

In this study, papules represented the most common type of skin lesion recorded, followed by plaques. This may be affected by the medical educational level of the patients, their level of

awareness, the use of herbs, chemicals, and others without consulting a doctor, which in summation represented all factors leading to increases in the degree of skin damage. The appearance of different skin manifestations mainly depends on the length of time before patients seek medical care or visit a dermatologist to receive the correct treatment [22].

The lesions appear as sores, which can change in size and appearance over time. They may start out as papules (bumps) or nodules (lumps), and may end up as ulcers (like a volcano, with a raised edge and central crater); skin ulcers may be covered by a scab or crust. The sores are usually painless but can be painful [22,56].

In this study, the number of lesions on the bodies of patients was divided into less than ten, where the patients received local treatment (88.302%), while 11.697% of patients had more than ten lesions. This was in agreement with the data of Abdulwahab, (2013) [35], who reported 79.9% of cases had 1 to 9 skin lesions, while 20% had ≥10 lesions. Here, the frequency of multiple lesions in CL patients was in agreement with another Iraqi study [51].

Younis [22] documented one ulcerated lesion in 49 (65.33%) of patients, while multiple lesions (2–9) were observed in 24 (32%) patients. This could be due to the long periods of exposure to sandflies and the high population density of sandflies in Iraq [51].

The period from the appearance of signs to the time of visiting the dermatologist was divided into five periods. The most frequent period was from 1 to 4 weeks, followed by 5 to 8 weeks. The earlier the diagnosis, the better the management, and the less cost and duration of the treatment. Kadir and El-Gorban [57] documented in their paper, published in the Iraqi Journal of Veterinary Medicine in 2006, that the duration of lesions to diagnosis ranged from 2 to 16 weeks.

Wet lesions were highest percentage of lesions examined in this study, with fewer dry lesions detected. These results were in agreement with a previous Iraqi study [51], and with other countries studies, such as Pakistan [58], Iran [48], and Afghanistan [49], but in disagreement with another study in India [59]. The high frequency of wet lesions may be due to the presence of large numbers of reservoir animals in some areas in Iraq, especially rodents and dogs [51].

Regarding the severity of CL, the moderate pattern was the most frequent, over severe and mild disease. This may depend on patients' immunity, the period before patients consult a dermatologist, duration of CL, number and nature of lesions, and efficacy of treatment.

According to the relationship between patients and the reservoir host (dogs), a direct relationship was most common, whereas indirect contact was noted in 28.211% of cases and refers to dogs found near homes as neighborhood or stray dogs. Those with CL who had no contact with dogs was 33.486%. The rate of infection among patients who had domestic dogs or were close to dogs was high, as a result to their epidemics with the presence of the sandfly. Some of them owned one dog, some owned two, three, and even fifteen dogs. Ali [44] documented that the rate of CL was higher in those who had dogs (24.7%). There were 43.2% of respondents who saw dogs around their homes. In Saudi Arabia, Aldossari [45] suggested that the region had a combination of suitab. climate conditions, dense natural vegetation cover, CL host species, and scattered farming and grazing lands with the population living scattered amongst them in very mixed housing styles. In such environmental conditions, people are more likely to interact with CL vectors and reservoirs making Misan province unsurprisingly one of the highest endemic areas for CL.

According to the sex and age of dogs, adult males were most common, because the owners prefer males to guard and herd animals.

In relation to CL clinical signs appearing on domestic dogs, about 31.223% showed signs, including large papules to ulcerated lesions, in different sites of the body, such as the head and neck (40.54%), body (6.756%), legs (24.324%), and tail (28.378%), whereas the rest (68.776%) showed no symptoms or signs. A study in Iran by Bamorovat et al. [60] reported that 229 (48.61%) male and 242 (51.38%) female stray dogs were clinically examined and showed alopecia, footpad hyperkeratosis, and pustular dermatitis of the snout.

In Saudi Arabia, Alanazi et al. [23] documented 40 dogs had thick cutaneous lesions in areas such as the mouth, nose, ear, muzzle, abdomen, legs, and between claws, but ulceration in the left hind foot was also noted in a few dogs.

Due to the lack of concern of breeders for the health status of their dogs and their treatment, papules and ulcerative lesions converted to suppurative and purulent lesions in the area of infection.

This infection is a result of dogs being exposed

more than once to the sandfly, because the dogs are a suitab. environment for the growth and reproduction of the insect vector because they are always moist. Five or more CL lesions that were wet were the most prevalent seen in this study. According to the severity of CL in dogs, the highest percentage of lesions were severe, then moderate, and mild. This is due to the exacerbation and development of infection as a result of lack of follow-up and lack of treatment in the locality, frequent infection, and the abundance of sandflies [60].

The increase in leishmaniosis prevalence is mainly attributab. to several risk factors that are man-made. Generally, these factors include environmental conditions, demographics, large migrations, deforestation, urbanization, and immunosuppression [61].

The environment and population movements possibly lead to variations in the number, range, and density of the vectors and reservoirs; thus, they may increase human exposure to infected sandflies. CL affects the poorer population, and outbreaks occur during harvesting seasons. Other factors including living in houses with cracked mud, damp earthen floors, sleeping on the floor or outside, and vegetation near house, which can assist sandfly survival and enhance vector abundance by providing diurnal resting places, breeding sites, and humidity [62].

To the best of our knowledge, this epidemiological study of CL among human and dogs was conducted for the first time in Iraq, and in the Middle East area, and maybe even over the world. We concluded that CL is a highly endemic disease in Iraq, especially in Maysan city. The younger age has a high susceptibility to having CL than older age. The rate was of infection not different from male to female. The high rate of infection high prevalent in big crowding and major cities. The winter months of the year showed higher infection rates of CL. The patients' living places, styles, locations, and house components have an important role in spreading and prevalence of CL. CL may be run in family according to the questions that asked patients during the time of the study. The more the exposed parts of the body and the long duration of disease before the intervention, the more number, and the more severe lesions. There is a close relationship between CL in humans and dogs, whether dogs are domestic or stray.

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