Cutaneous Leishmaniasis in Gharyan – Libya – a Case-Control Study

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Abstract: Leishmaniasis occurs in 88 countries throughout the Tropics and Temperate zones and is estimated to infect 1.5 million persons with the cutaneous form and 500 thousand persons with the visceral form yearly. Cutaneous leishmaniasis (CL) has a significant social impact as it may lead to severe stigmatization of affected individuals when lesions or scars occur on the face and exposed extremities. **In Egypt,** Cutaneous leishmaniasis is endemic in the Sinai region, and when the etiologic agent was identified, L. major has been the culprit parasite. **In Libya**, CL is widespread in the north-western region. Information about disease distribution combining risk factors is confined. The **objectives** of this study was to study socio-demographic risk factors associated with cutaneous leishmaniasis in Gharyan, Libya and to describe the characteristics of the lesions among cases enrolled in the study. The **results** of this study showed that cutaneous leishmaniasis is significantly associated with some socio-demographic factors as residence in rural areas, illiteracy, increased family size, decreased number of floor and rooms per house, and presence of garden and domestic animals. It was **recommended** to control rodents and sandfly vector and to decrease human vector contact by simple screening of windows.

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1. Introduction

Leishmaniasis is a parasitic disease caused by several species of the protozoan genus Leishmania. Species in this genus cause a spectrum of clinical diseases ranging from self-limited cutaneous ulcers, to devastating infection of the nasopharyngeal mucous membranes, to fatal visceral infections of the liver and spleen (Herwaldt, 1999). Infected female sand flies are believed to act as vectors, transmitting this parasite when a fly takes its blood meal (Sadlova *et al.,* 2003) specifically those from the genus Phlebotomus. Known reservoir hosts include infected humans, wild animals (i.e. rodents) and domestic animals (Williams *et al.,* 1991).

Cutaneous leishmaniasis (CL) is a vector-borne protozoan disease that is characterized by cutaneous lesions which develop at the site of the insect bite. Lesions can vary in severity, clinical appearance, and time to cure; in a proportion of patients lesions can become chronic, leading to disfiguring mucosal leishmaniasis. Cutaneous leishmaniasis can have a significant social impact as it may lead to severe stigmatization of affected individuals when lesions or scars occur on the face and exposed extremities (Bousslimi *et al.,* 2010).

Leishmaniasis occurs in 88 countries throughout the Tropics and Temperate zones and is estimated to infect 1.5 million persons with the cutaneous form and 500 thousand persons with the visceral form yearly. In 2004, the CDC reported that there are more than 12 million chronic cases worldwide. However, there is an increased incidence of disease that can be partially attributed to geographical expansion, population migration, international travel, and/or deployment of military forces (Desjeux, 2001 and Antinori, 2005).

In general, susceptibility to infection and disease is determined by a number of parasite, host and sand fly effects factors (Reithinger, *et al.,* 2007). Thus, infections can cluster within households, which is indicative of the short flight range of sand flies (Killick-Kendrick, 1999), or genetic susceptibility (Sakthian and Esware, *et al.,* 2009). Infection is also known to be dependent on host nutritional status and acquired immunosupression (e.g. HIV) (Weigel, *et al.,* 1995 and Alvar, 2008).

In Egypt, Cutaneous leishmaniasis is endemic in the Sinai region, and when the etiologic agent was identified, L. major has been the culprit parasite (Mansour *et al.,* 1991; Kamal *et al.,* 2003 and Dawoud, 2004). Leishmania major is a zoonotic parasite. In the Sinai, parasites are transferred to humans from infected rodent reservoir hosts (Meriones crassus and Gerbillus pyramidum) by the bite of the sand fly vector Phlebotomus papatasi. (Morsy *et al.,* 1987; Mansour, *et al.,* 1991 and Fryauff, 1993). *P. papatasi* is the principal sand fly vector throughout the Mediterranean basin, Middle East, Central Asia, and East Africa (Schlein, 1984 and Parvizi, *et al.,* 2003).

Leishmania tropica also occurs widely in the Middle East. The transmission cycle for *L. tropica* is usually anthroponotic with zoonotic transmission reportedly occurring in some areas (Jacobson, 2003 and Jacobson *et al.,* 2003). Phlebotomus sergenti is an

often implicated sand fly vector in both transmission scenarios and can be found along the Mediterranean coast from Libya, through Egypt, Jordan, and Iraq (Seccombe, *et al.,* 1993). Egypt is not a known focus for *L. tropica*, although there are some old reports from the Nile Delta region (Cahill, 1965). The only recent Egyptian case of CL definitively attributed to *L. tropica* was an infection acquired outside of Egypt by a worker returning from Saudi Arabia (Mohareb, *et al.,* 1996).

In September 2006, physicians from El Barth community hospital (near Rafah, Northern Sinai, Egypt) collected case histories and tissue biopsies from 10 patients with suspected CL. Using culture methods, the Ain Shams University Research and Training Center (RTC) verified the presence of Leishmania in six of these samples. The RTC also surveyed the local sand fly fauna to identify vector species, and successfully cultured or visually observed Leishmania from lesions of 14 wild-caught rodents. The RTC subsequently contacted the U.S. Naval Medical Research Unit No. 3 (NAMRU-3), Cairo, to determine the species identity in Leishmania-positive cultures. The results of this study verified the pathogens, vector, and potential reservoir involved in the leishmaniasis disease cycle in the study area as follows:

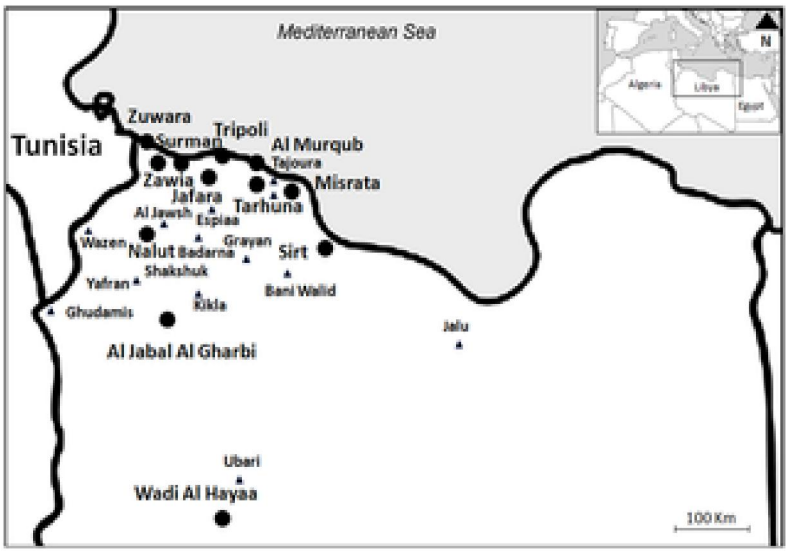
1) human cases of CL were caused by either L. major or L. tropica, 2) known sand fly vectors of both parasites were identified, and 3) wild-caught rodents harbored Leishmania parasites, infected with L. tropica (Magdi *et al.,* 2009).

In Libya, CL is widespread in the north-western region. The first case of CL was reported in 1930, followed by recording of 40 cases in 1971 in Nalut near the Tunisian border (Ashford, *et al.,* 1976; Ashford, *et al.,* 1977 and El-Buni, *et al.,* 2000). In the following years several CL cases have been subsequently occurred in the west and south-west of Tripoli, Al-Badarna (El-Buni and Ben-Darif ,1996) and Yafran areas (El-Buni, *et al.,* 1997 and El-Buni, *et al.,* 2000).

The diagnosis of CL in Libya is based on clinical signs of the disease and microscopic observation of parasites in stained skin biopsies (El-Buni *et al.,* 1997 and El-Buni *et al.,* 2000). Information about disease distribution combining risk factors is deficient.

All CL cases in Libya were originated from the north-western districts of the country exclusively (Amro *et al.,* 2012).

. **Figure (1): Geographical distribution of CL in Libya**



The map of Libya showing the areas endemic for CL. •Districts. ▲Endemic areas. Source: Amro, *et al.* (2012)

Objectives

1- To study the socio-demographic risk factors associated with cutaneous leishmaniasis in Gharyan, Libya.
2- To describe the characteristics of the lesions among cases enrolled in this study.

2.Subjects and Methods

A case control study was conducted to evaluate risk factors for cutaneous leishmaniasis in Gharyan, Libya.

Data were collected through personal interview by using structured questionnaire after being pre-tested in a pilot study. Data were collected by well

trained interviewers under close supervision of the researchers.

Demographic and household data were included in the questionnaire. Demographic data included information on nationality, age, sex, education, occupation and family size. Household data included information on household design (i.e. number of rooms and floors per house), and ownership of garden and domestic animals (i.e. goats, sheep and cattle).

Patients with CL attending at outpatient clinic of Gharyan teaching hospital, Gharyan, Aljabal Algharbi, Libya, during a four-month period from January, 1st, till April, 30th, 2010 were included in this study. Gharyan is a sub-province of Aljabal Algharbi which is located south west of Libyan Capital, Tripoli. Total population of Aljabal Algharbi is 302,000 individuals. Gharyan is the administrative city of Al-jabal Al-Gharbi province. At Gharyan clinic, those CL cases were clinically diagnosed and confirmed by laboratory investigations.

This study included 135 cases of cutaneous leishmaniasis in Gharyan, Libya. Controls were individuals who had been selected at random from attendants of outpatient clinic of Gharyan teaching hospital. Two controls were matched according to residence, age group and sex to every case. Clinical characteristics of the lesions as site, size of the lesions and treatment received by the patients were included in the questionnaire.

Verbal consent was obtained from both cases and controls before participation in this study. All participants were informed that the purpose of the study was to evaluate and assess the risk factors associated with the occurrence of cutaneous leishmaniasis.

The statistical package for social sciences (SPSS) software version 11 was used for entry and analysis of data. Chi square and T tests were used to evaluate the difference between cases and controls regarding presence of risk factors. A significant P-value was considered if less than 0.05.

3. Results

The mean age of both cases and controls was about 22 years. There was no significant difference between cases and controls regarding age, sex and nationality (P = 0.673, 0.160 and 1.0 respectively).

51.1 % of cases were residing in rural areas compared to 37.8% of controls (P =0.01). 23% of cases were illiterate compared to 8.1% of controls (P <0.001). The mean family size of cases was 6.8 compared to 5.9 for controls (P = 0.001). The mean number of floors per house was 1.4 for cases compared to 1.8 for controls (P < 0.001). The mean number of rooms per house was 2.4 for cases compared to 3.8 for controls (P < 0.001). The presence of screen for windows was 37.8% for cases compared to 58.5% for controls (P < 0.001). Garden was present in the residence area of 67.4% of cases compared to 43% of controls (P <0.001). Domestic animals was present in 60.7% of residence area of cases compared to 40% of controls (P <0.001). Rodents and sand-fly were present in significantly higher percentage of cases than controls (49.6% versus 25.2% and 23.7% versus 8.9% respectively) (P <0.001 for both).

Regarding type of lesion, it was papule (or ulcerated papules) and ulcers in 48.9% and 32.6% of cases respectively. The size of lesions was less than 2 by 4 cm in about three quarters of cases. The most common site of lesions was the upper limbs (32.6%), followed by the face (25.9%) and lower limps (19.3%). The number of lesions was one in 40.7% and two in 29.6% of cases. Complications like inflammation, was present in 26.7% of cases. Laboratory diagnosis was present in 14.8% of cases. 87.4% of cases received treatment.

59.3% of cases had other family member affected by leishmaniasis. The most common family member affected were, brothers and sisters (24.4%) followed by son and daughters (11.9%) and parents (9.6%) (Figure 2).

4. Discussion

This study included 135 cases and 270 controls with the aim to study the sociodemographic characteristics associated with cutaneous leishmaniasis in Gharyan, Libya.

Distribution of the disease in Libya:

CL is considered as an endemic disease in Libya. All CL cases in Libya were originated from the north-western districts of the country exclusively. These districts have typical Mediterranean costal climate in the upper northern districts like Tripoli, and semiarid and arid climate in Al Jabal Al Gharbi (including Ghrayan) and Wadi Al Hayaa to the south. Like many other countries around the Mediterranean Sea, climatic and environmental conditions and development of agricultural activities in these districts may be favourable for transmission of Leishmania (Ashford *et al.,* 1976, El-Buni, Ben-Darif, 1996 El-Buni *et al.,* 2000, Ben-Ahmed *et al.,* 2009 and Bousslimi, 2010).

The geographical nature of Gharyan province makes it a suitable place for breeding and spread of many kinds of reservoir hosts for leishmania, especially dogs and rodents as well as the transmission vector, sandfly.

Risk factors of the disease:

In this study, it was found that risk factors of disease are: sex (e.g. sex bias usually points to behavioral patterns that increase vector exposure); age, residence in rural areas, illiteracy, farmers as an occupation and household design and construction material (e.g. number of floors, number of rooms).

Our study found that, the male: female ratio indicated that the infection rate among males was slightly higher than females (52.6: 48.4%). The possible explanation is that men have a habit of sleeping outside their homes during hot nights and more engaged in outdoor activities so they may be more prone to get bitten by infected sand flies compared to women who tend to have fewer activities outside their homes. These results are in consistence with a previous study conducted in Yafren region, Al-jabal Al-Gharbi (Libya) (El-Buni *et al.,* 2000, Abdellatif *et al.,* 2012 and Amro, *et al.,* 2012).

Table (1): Distribution of participants according to some socio-demographic characteristics in Gharyan, libya, 2010.

Characteristics	Cases	Controls	Total	*P* – value
Sex:				
- Male	71 (52.6)	136 (50.4)	207 (51.1)	0.673
- Female	64 (47.4)	134 (49.6)	198 (48.9)	
Age:				
- 0 –	40 (29.5)	77 (28.5)	117 (28.9)	0.437
- 10 –	25 (18.5)	59 (21.9)	84 (20.7)	
- 20 –	23 (17.0)	52 (19.3)	75 (18.5)	
- 30 –	23 (17.0)	43 (15.9)	66 (16.3)	
- 40 and more	24 (17.8)	39 (14.4)	63 (15.6)	
Mean age:	22.9 ± 17.5	22.8 ± 16.1	21.8±17.3	0.160
Nationality:				
- Libyan	130 (96.3)	260 (96.3)	390(96.3)	1.0
- Non- Libyan	5 (3.7)	10 (3.7)	15 (3.7)	
Residence:				
- Urban	66 (48.9)	168 (62.2)	234 (57.8)	0.010
- Rural	69 (51.1)	102 (37.8)	171 (42.2)	
Occupation:				
- Farmers	42 (31.3)	58 (21.5)	100 (24.7)	0.020
- Students	30 (22.2)	62 (23.0)	92 (22.7)	
- Employee	10 (7.4)	30 (11.1)	40 (9.9)	
- House wife	8 (5.9)	26 (9.6)	34 (8.4)	
- Worker	17 (12.6)	22 (8.1)	39 (9.6)	
- Others	3 (2.2)	26 (9.6)	29 (7.2)	
Education:				
- Illiterate	31 (23.0)	22 (8.1)	53 (13.1)	<0.001
- Literate	104 (77.0)	248 (91.9)	352 (86.9)	
Family size:				
- 2 – 4	20 (14.8)	56 (20.7)	76 (18.8)	0.001
- 5 – 7	63 (46.7)	158 (58.6)	221 (54.5)	
- 8 and more	52 (61.5)	56 (20.7)	108 (26.7)	
Mean family size (±SD)	6.8 ± 2.3	5.9 ± 1.7	6.2 ± 2.0	0.001
No. of floor/house:				
- One	92 (66.1)	120 (44.4)	212 (52.3)	<0.001
- Two	36 (26.7)	92 (34.1)	128 (31.6)	
- Three or more	7 (5.2)	58 (21.5)	65 (16.0)	
Mean No. of floor (±SD)	1.4 ± 0.58	1.8 ± 0.78	1.64 ± 0.74	<0.001
No. of rooms/house:				
- One	33 (24.4)	12 (4.4)	45 (11.1)	<0.001
- Two	39 (28.9)	60 (22.2)	99 (24.4)	
- Three	43 (31.9)	80 (29.6)	123 (30.4)	
- Four or more	20 (3.7)	118 (43.8)	138 (34.1)	
Mean No. of rooms	2.4 ±1.1	3.8±1.9	3.4±1.8	<0.001
Presence of screen:				
- Yes	51 (37.8)	158 (58.5)	209 (51.6)	<0.001
- No	84 (62.2)	112 (41.5)	196 (48.4)	
Presence of garden:				
- Yes	91 (67.4)	116 (43.0)	207 (51.1)	<0.001
- No	44 (32.6)	154 (57.0)	198 (48.9)	
Presence of domestic animals:				
- Yes	82 (60.7)	108 (40.0)	190 (46.9)	<0.001
- No	53 (39.3)	162 (60.0)	215 (53.1)	
Presence of rodents:				
- Yes	67 (49.6)	68 (25.2)	125 (33.3)	<0.001
- No	46 (34.6)	134 (49.6)	180 (44.4)	
- Do not know	22 (16.3)	68 (25.6)	90 (22.2)	
Presence of sand-fly:				
- Yes	32 (23.7)	24 (8.9)	56 (13.8)	<0.001
- No	51 (37.8)	124 (45.9)	175 (43.2)	
- Do not know	52 (38.5)	122 (45.2)	174 (43.0)	
Total	135 (100.0)	270 (100.0)	405 (100.0)	

Table (2): Characteristics of the lesions of cutaneous leishmaniasis among cases included in the study in Gharyan, Libya, 2010.

Characteristics	Frequency
Type of lesions (n = 135):	
- Papule (or ulcerated papule)	66 (48.9)
- Ulcer	44 (32.6)
- Scar	25 (8.5)
Size of lesions (n = 135):	
- 1 X 1 cm	49 (36.3)
- 1 X 2 cm	19 (14.1)
- 1 X 3 cm	14 (10.4)
- 2 X 3 cm	14 (10.4)
- 2 X 4 cm	5 (3.7)
- 3 X 5 cm	7 (5.2)
- More than this size	27 (20.0)
Site of lesions (n = 135):	
- Face	35 (25.9)
- Upper limb	44 (32.6)
- Lower limb	26 (19.3)
- Trunk	12 (8.8)
- More than one site	18 (13.3)
Number of lesions (n = 135):	
- One	55 (40.7)
- Two	40 (29.6)
- Three – Four	32 (23.7)
- Five and more	8 (6.0)
Complications (e.g. inflammation, increase in size and disfigurement) (n = 135):	
- Present	36 (26.7)
- Absent	99 (73.3)
Diagnosis (n = 135):	
- Clinical	115 (85.2)
- Laboratory	20 (14.8)
Treatment (n = 135):	
- Yes	120 (87.4)
- No	15 (12.6)
Type of treatment (n =120):	
- Cryo-therapy	81 (67.5)
- Antimony compounds	14 (11.7)
- Both cryo-therapy and systemic antibiotics	25 (20.8)
Follow up (n = 120):	
- Yes	107 (79.3)
- No	13 (13.6)

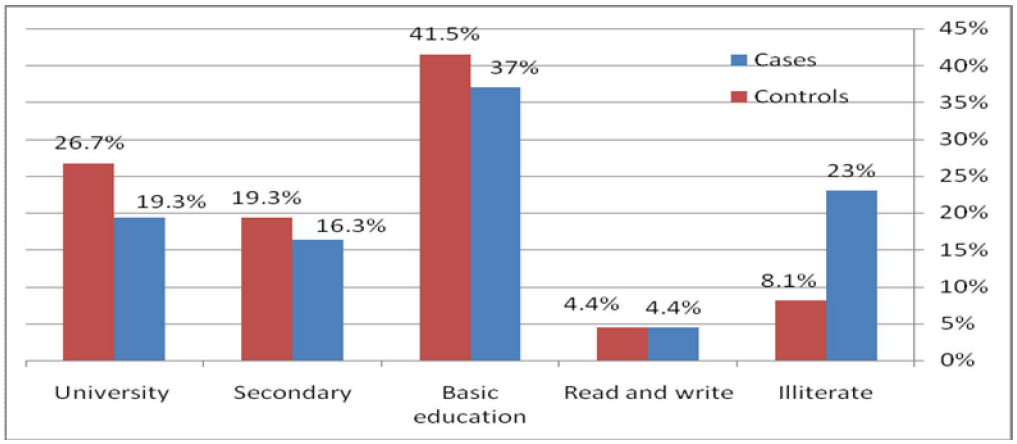


Figure (1): Distribution of participants by education in Gharyan, Libya, 2010.

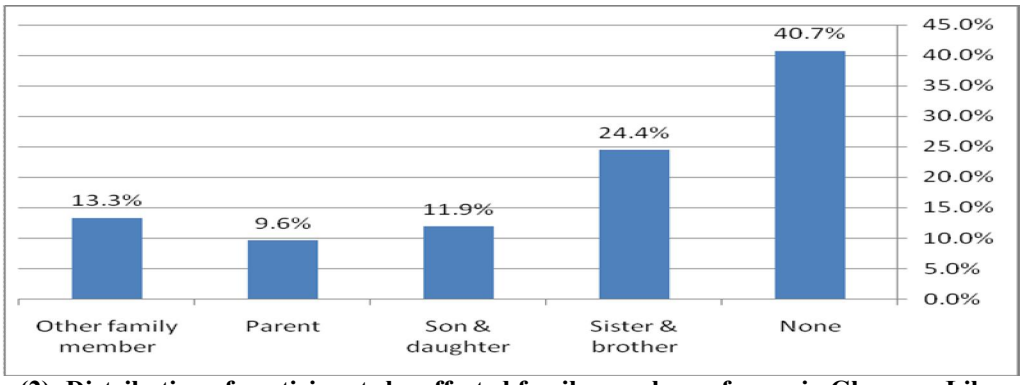


Figure (2): Distribution of participants by affected family members of cases in Ghrayan, Libya, 2010.

In this study, the mean age was 22.9 years, the most affected age group was less than 10 years (29.5%), followed by those aged 10 to less than 20 years (18.5%). In established endemic areas, CL prevalence typically increases with age up to 15 years, after which prevalence levels off, presumably because of the acquisition of immunity (Reithinger, *et al.,* 2007). The results of studies conducted in Al-jabal Algharbi (Libya) (Abellatif *et al.,* 2012), Southern Sri Lanka (Rajapaksa, 2007), Pakistan (Anwar *et al.,* 2007) and Turkey (Sucakli and Saka, 2007) found that the most affected age groups were (11-35), (10 – 19), (10 – 14) and (9 - 14) years, respectively.

Our study found that 59.3% of cases had other family member affected by leishmaniasis. The most common family member affected were, brothers and sisters (24.4%) followed by son and daughters (11.9%) and parents (9.6%). CL risk is strongly associated with the presence of disease in other household members, confirming previous findings that the disease is highly focal at the household level (Reithinger, *et al.,* 2003). A likely explanation for this is that sand fly distribution and abundance is patchy (Hewitt, *et al.,* 1998), but stable over time, and vector exposure is strongly dependent on household design. The sand fly flight range is generally short (Killick-Kendrick, 1999) and it is likely that Leishmania transmission does not occur beyond a defined cluster of households. Also, individuals of the same family are exposed to the same risk factors as cases for a long duration of time.

Zoonotic transmission has been shown for L. major in all areas of its distribution including the Middle East and North Africa where the parasites are transmitted by *P. papatasi* from its reservoir hosts (rodents) to humans (Belazzoug, *et al.,* 1983, Saliba *et al.,* 1994 and Ghawar, et al., 2011). CL due to L. tropica is considered as an anthroponosis in many countries, especially in densely populated cities (Ashford, 1999 and Ashford, 2000) and transmitted via its natural vector *P. sergenti* between humans, nonetheless, zoonotic transmission of *L. tropica* was recently proven in the Middle East (Talmi *et al.,* 2010) and suggested to occur in less populated rural areas in the Middle East and North Africa.

A previous study has shown that P. papatasi is the most abundant sand fly species in Libya, followed by P. sergenti (El-Buni *et al.,* 2000).

It is believed that sand flies get infected with Leishmania when biting people with active CL. Thus, a household with a high proportion of people with scars is –from an epidemiological point of view- less 'infectious' to its inhabitants than a household that includes a high proportion of CL cases (Killick-Kendrick, *et al.,* 1995).

In this study presence of CL was significantly associated with residence in rural areas, presence of domestic animals and rodents. This agreed with finding reported by studies conducted in Bolivia, Argentina and Brazil. In these studies it was reported that household presence of domestic animals (e.g. cattle), household proximity to agricultural areas or other areas where sand flies are known to aggregate, and migration of household members is associated with CL (Alcais *et al.,* 1997, Davis *et al.,* 1997, Yadon *et al.,* 2003 and Pedrosa and Ximenes, 2009). Another reason could be the nature of rural areas where the people are engaged in farming activities, working outdoors and staying there for long till night time

Our finding showed that CL was significantly associated with illiteracy and farmers as an occupation which is usually more common in rural population. Similar findings were reported in India and Libya (Ranjan *et al.,* 2005 and Abdellatif *et al.,* 2012)

Our findings indicate that household design (e.g. number of floors and rooms or number of persons per room) can significantly influence cutaneous leishmaniasis risk in Libya. Cutaneous leishmaniasis was significantly associated with decreased number floors and rooms per house. Observed associations are probably due to sand fly and human behaviour, and

ultimately increased or decreased sand fly exposure. A similar finding was reported in Kabul, Afghanistan (Killick-Kendrick, *et al.,* 1995).

Finally, our data also suggests that means to reduce vector exposure can be highly successful in reducing the risk of CL, with simple screening of windows being effective. As the presence of screen for windows was 37.8% for cases compared to 43% of controls ($P<0.001$). In other studies, it was found that, use of textile fabrics, whether insecticide-treated or not, used as bednets, have consistently shown efficacy in reducing indoor CL transmission in endemic areas (Kolaczinski, *et al.,* 2004, Yaghoobi-Ershadi, *et al.,* 2006, and Moosa-Kazemi *et al.,* 2007). Whether bednets are protective against Leishmania infection will be dependent on a number of factors besides use, including whether the nets are impregnated with insecticide, net shape and size, or wear and tear (Das *et al.,* 2007).

This study revealed that lesions were more common in exposed parts (upper, lower limbs and face). A similar finding was reported by Magdi *et al.,* 2009 and Abdellatif *et al.,* 2012). The type and size of lesions in this study agreed with study conducted in Pakistan (Khan and Muneeb, 2005). Also, the number of lesions may be single or multiple which is in consistence with findings reported by Abdellatif *et al.,* 2012.

Conclusion and Recommendations

The results of this study concluded that cutaneous leishmaniasis is an important health problem in Gharyan. The presence of CL was significantly associated with:

- Residence in rural areas
- Lack of education (illiterate individuals)
- Increased family size
- Decreased number of floors and rooms per house
- Lack of screening of windows
- Presence of garden, domestic animals, rodents and sandfly in the residency area

The quantification and the containment of these risk factors are major challenges and should be considered by health policy makers and health professionals in order to evaluate the CL burden and to highlight priority actions for the disease control. Therefore, it was **recommended** to:

- Massive rodent control
- Vector control: control of sand fly vectors by all possible means
- Early detection and proper treatment of cases. Lesions should be covered to avoid infection of vectors.
- Decrease human – sand fly contact by simple measures as screening of windows, avoiding sleeping outdoors in endemic areas and increased urbanization as far as possible.
- Further studies are needed to identify species of Leishmania parasite responsible for CL in Gharyan.

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