

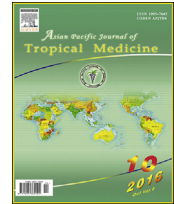
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## Epidemiology of the outbreak, vectors and reservoirs of cutaneous leishmaniasis in Mali: A systematic review and meta-analysis

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## ABSTRACT

**Objective:** To compile available data and to estimate the burden, characteristics and risks factors of cutaneous leishmaniasis (CL) in Mali.

**Methods:** Articles in English and French were searched in Hinari, Google scholar and PubMed. Unpublished studies were identified by searching in Google.com. Terms used were cutaneous leishmaniasis Mali; Leishmaniasis Mali, *Leishmania major* Mali; or Phlebotomus Mali or Sergentomyia Mali. We select descriptive studies on CL and sandflies in Mali. Data were extracted and checked by the author, then analyzed by region, by study population and type of biological tests, meta-analysis approach with STATA software was used.

**Results:** Nineteen published ( $n = 19$ ) and three unpublished were included. CL epidemiology was characterized by occurrence of clinical cases in different areas of Mali, outbreaks restricted to known areas of transmission and isolated cases diagnosed in travelers. In endemic areas, population at risk are young age persons, farmers, ranchers, housewives, teachers and military personnel. The annual incidence ranged from 290 to 580 cases of CL. *Leishmania major* is the main species encountered throughout the country (North Savanna, Sahel and Sub-Saharan areas), and *Phlebotomus duboscqi* has been identified as the vector and *Sergentomyia (Spelaemyia) darlingi* as possible vector. The overall estimated prevalence of positive LST (Leishmanin Skin Test) was 22.1%. The overall frequency of CL disease among suspected cases was 40.3%.

**Conclusions:** Although descriptive, hospital-based and cross-sectional studies are robust enough to determine the extent of CL in Mali; future well-designed eco-epidemiological studies at a nationwide scale are needed to fully characterize CL epidemiology and risk factors in Mali.

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## 1. Introduction

Leishmaniasis are zoonoses common in animals and human being. Globally, leishmaniasis are endemic in 98 countries, with an annual incidence of 0.7–1.2 million cases of cutaneous leishmaniasis (CL) and 0.2–0.4 million cases of visceral leishmaniasis (VL) causing 20000–40000 deaths each year [1]. In Sub-Saharan Africa, the estimated annual incidence of CL is between 770 and 1500 cases. The annual incidence of CL is between 40 and 80 cases in Senegal, and 30–50 in Nigeria [1].

In Mali, CL was described for the first time in 1948 by Lefrou [2]. Izri *et al.* reported for the first time the zymodeme

MON-26 of *Leishmania major* (*L. major*) in a Malian patient in 1989 [3]. Other authors described zymodemes MON-25, MON-17 and MON-117 of *L. major* in Mali [4,5]. Little data on CL have been reported by the national demographic and health survey [6]; given the difficulty of CL diagnosis, health facilities are not well equipped to diagnose CL, leading to under-notification. Research studies conducted on CL have used different diagnostic methods in different study populations within different regions of Mali. However, CL variation and its burden at the national level remain poor known.

## 2. Materials and methods

### 2.1. Search

A literature search was performed to identify records on CL in Mali. The search included original research done in Mali and publications in both French and English. Searches were performed on Hinari, Google scholar, and PubMed using the following terms: “Cutaneous leishmaniasis Mali”; “Leishmaniasis Mali”; “*Leishmania major* Mali”; or “*Phlebotomus* Mali”. Unpublished data on LC were searched for in [Google.com](http://Google.com) using the same search terms.

### 2.2. Study selection

Papers were screened manually. Descriptive studies (cross-sectional and cohort) conducted in communities or in health facilities were selected. Articles with full text available were preferred. When full text was not available, the published abstracts were included. Data from unpublished literature (Master's, MD dissertation defended at the Faculty of Medicine, Pharmacy and Dentistry of Bamako) have also been searched, checked and included in our review. Participants of any age were included. Citations were excluded when the full text or abstract were not available. Published papers on visceral leishmaniasis were not included.

### 2.3. Data collection process

The first author did the literature review, read the documents and checked citation eligibility. Data included in the meta-analysis were checked by the author and the statistician.

### 2.4. Data items

The data extracted from each study included: study location (community- or hospital-based study, geographic region), participants (tools of diagnosis, frequency of CL, age), and sandfly fauna (description of fauna, frequency).

### 2.5. Planned methods of analysis

Dates, type of study, diagnosis tools and localization were also included to place data in context. Data from each study (LST prevalence or frequency of confirmed CL, community or health facility-based studies) were considered separately. Data were analyzed by region and study population. A meta-analysis approach using STATA software was performed to compare frequencies and to compute the mean prevalence rate of LST-positive reactions in communities and the mean proportion of CL in suspected cases in health facilities.

## 3. Results

A total of 39 studies were identified, including 30 peer reviewed papers and nine from the unpublished literature. Five were removed mainly because of duplicating results, and 14 records were excluded after checking of eligibility criteria. Twenty studies have been included, among them six records (four full text articles, an abstract article and one MD dissertation) that have been included in meta-analysis (Figure 1).

The mean prevalence of positive LST in the general population was 22.1%; however, it varied by region and by study

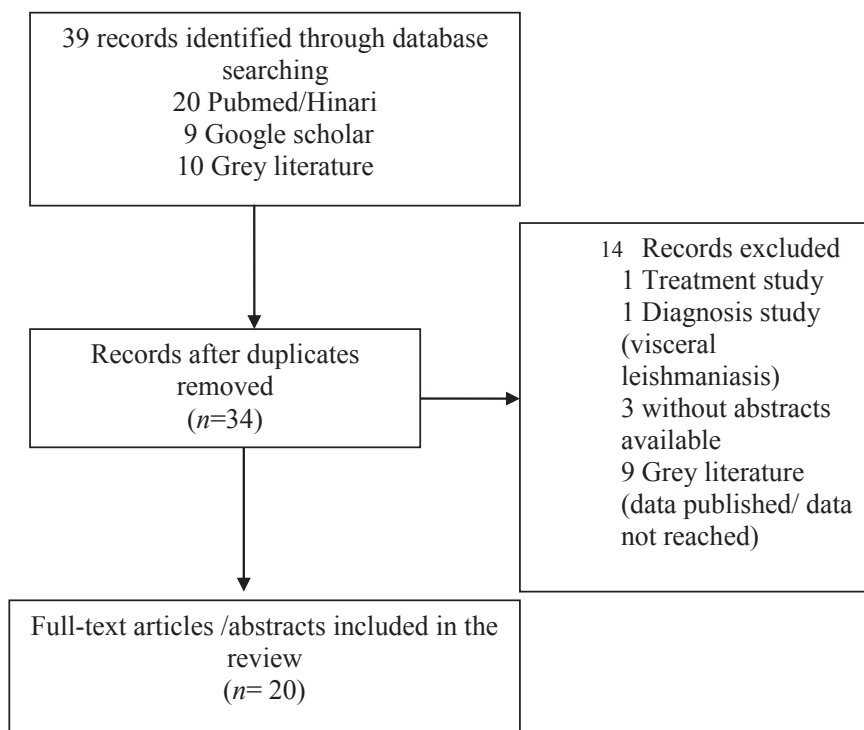


Figure 1. Study flow chart.

**Table 1**

Proportion of positive skin test by study population and proportion of LC among suspected cases in clinic.

Study population/authors publication year	Study year	Age in year	Study sites	% LST/Leishmaniasis (95% CI)	% Weight
<b>General population</b>					
Imperato PJ <i>et al.</i> 1969	1967	6–20 years	Bamako	18.7 (16.7–20.6)	25.15
Oliveira F <i>et al.</i> 2009	2006–2008	All	Kemena	45.4 (41.6–49.2)	24.78
Oliveira F <i>et al.</i> 2009	2006–2008	All	Sougoula	20.0 (17.3–22.6)	25.04
Imperato PJ <i>et al.</i> 1974	<1974	All	Mopti	4.8 (2.2–7.5)	25.04
Subtotal (I-squared=99.0%. p=0.000)				22.1 (8.9–35.3)	100.00
<b>Suspected cases in clinic</b>					
Keita S <i>et al.</i> 2003	1997–2001	All	CNAM	78.4 (73.9–82.9)	33.66
Kampo OM 2009	1998–2001	All	Hospital	14.1 (5.5–22.6)	33.38
Kone AK <i>et al.</i> 2011	2010	2–55 years	Bandiagara	28.0 (15.6–40.4)	32.96
Subtotal (I-squared=99.0%. p=0.000)				40.3 (–6.1 to 86.7)	100.00

Weights are from random effects analysis.

population (Table 1). The frequency of CL in patients where there was clinical suspicion of CL was 78.4% (251/320) in Bamako (Mali) between 1997 and 2001 [7]. The prevalence of positive reactions to the LST in 1969 in school-aged children in Bamako city was 12.7% (90/705) [8], and in the city of Mopti in the general population was 4.8% in 1974 (12/249) [9]. In the general population in Kemena, Segou region, the prevalence rate of LST-positive reactions was 45.4% (301/663) in 2008 [10] and 30.4% (34/112) frequency of LST-positive reactions observed between 1957 and 1966 in residents of Bamako from the Segou region ( $P < 0.01$ ) [8].

A recent survey conducted in two villages of Segou region shows an annual incidence rate of 18.5% (53/287) in Kemena and 5.7% (32/366) in Sougoula in 2008 [10] ( $P < 0.01$ ).

Sandflies collected in rural and suburban areas in Mali have shown a variability of sandfly fauna. To date four species of *Phlebotomus* sp.: *Phlebotomus duboscqi*, *Phlebotomus rodhaini*, *Phlebotomus sergenti* and *Phlebotomus kazeruni* have been identified in Mali. *P. sergenti* has been cached in Mopti and Bamako areas. *P. kazeruni* has been identified in Mopti area [11–14].

Twenty two species of *Sergentomyia* sp. have been identified: *Sergentomyia schwetzi*, *Sergentomyia antennata*, *Sergentomyia dubia*, *Sergentomyia clyde*, *Sergentomyia africana*, *Sergentomyia squamipleuris*, *Sergentomyia affinis affinis*, *Sergentomyia affinis vorax*, *Sergentomyia balmicola*, *Sergentomyia bedfordi*, *Sergentomyia fallax*, *Sergentomyia buxtoni*, *Sergentomyia (Spe-laeomyia) darlingi*, *Sergentomyia christophersi*, *Sergentomyia wansoni*, *Sergentomyia magna*, *Sergentomyia davidsoni*, *Sergentomyia fretownensis*, *Sergentomyia herollandi*, *Sergentomyia congolensis*, *Sergentomyia ghesquierei* and *Sergentomyia (schoutedeni) schoutedeni* [*S. (S) schoutedeni*] *Sergentomyia wansoni*, *Sergentomyia magna*, *Sergentomyia davidsoni*, *Sergentomyia affinis affinis* and *Sergentomyia balmicola* were trapped in Mopti and Bamako areas. *Sergentomyia fretownensis*, *Sergentomyia herollandi*, *Sergentomyia congolensis*, *Sergentomyia ghesquierei* and *Sergentomyia (S) schoutedeni* have been identified in suburban area of Bamako [11–16].

## 4. Discussion

### 4.1. Burden

The mean prevalence of positive LST in the general population was 22.1%; however, it varied by region and by study population. The frequency of CL in patients in Bamako was

higher than that reported in Niamey (Niger): 66.7% (64/96) between 1985 and 1987 [17].

The differences observed in disease estimates between the general population (community-based surveys) and suspected cases (hospitals and outbreaks in Dogon villages) could be explained by the study populations, area and the tools used for *Leishmania* infection detection. In the suspected cases group, the CL cases were diagnosed using PCR or microscopy, while in the general population, the LST (Leishmanin Skin Test) was used for detecting cases contact with *Leishmania*.

#### 4.1.1. CL in urban areas

In addition to the scarcity of studies on *L. major* transmission in urban areas of Mali, data on CL in urban areas are old, in Bamako, the city study population consisted of primary and secondary schools, while in Mopti city, it was the general population (no cases of CL have been detected where the LST has been used). The study in Bamako city with school-aged children from different regions gives the range of the frequency of positive LST in Mali [8].

At the Centre National de Lutte contre la Maladie (CNAM, the dermatology reference center in Bamako), suspected cases were ill patients carrying chronic skin wounds that were diagnosed as CL on *Leishmania* positive thin smears at microscopy. The higher proportion of confirmed CL within suspected patients in CNAM could be explained by the fact that this center is the country-wide reference facility for the diagnosis and the treatment skin diseases [7]. This result differs from that of the Center of Dermatology and Venereology in Segou, where the frequency of 14,06% LC was reported [18].

#### 4.1.2. CL in rural areas

Descriptive studies on *L. major* transmission in rural areas indicated that populations older than one year from two villages of Segou region have positive reactions to Leishmanin [10]. Outbreaks of CL may occur frequently in rural foci. In 2010, an outbreak of cutaneous wounds in Dogon villages was investigated. Suspected cases of CL were examined by a dermatologist, and samples were collected. PCR of scrapings from wounds edges was performed, and Western blot of sera was used to detect antibodies against *Leishmania*. Out of 50 patients examined, 28% (14/50) were diagnosed as CL cases. Microscopy examination was negative and frequency of antibody detection by IFA was lower than that of Western blot [19].

Data collected in the Segou region in 2008 [10] and in the Mopti region in 2010 [19] suggest an increasing contact with *L. major* when the results of a positive LST in Segou and a positive Western blot in Bandiagara are compared to previous results on reactivity to LST in 1967 [8,9]. However these data show that transmission of CL remains significant over time in the Segou region.

Besides the possibility that the prevalence rate of LST-positive reaction observed in these studies could be less than the rate of *Leishmania* infection in the population, as individuals harboring *L. major* infection are often LST negative, the rate may also be higher because more than a 5% LST false positive rate is expected [8]. In addition, a recent analysis of the geographic distribution of PCR-confirmed cases of CL in the data collected by the CNAM reveals that the disease is endemic in the north Sudan savanna, Sahelian areas, and the Sub-Saharan region spanning parts of the Kayes, Koulikoro, Segou, Mopti and Tombouctou regions [20]. These findings support previous results on geographic distribution of positive LST in Mali [20,21].

Among CL cases reported, 70.1% (413/589) are from Kayes region (Nioro, Kayes, Bafoulabe, and Yelimane) from 1957 to 1966 [9], showing that western rural areas of Mali could be more endemic compared to others areas of Mali. Recent findings support these previous data and show also that CL cases come from all the country [22].

Although the accurate incidence rate of CL in Mali is not known, Alvar and colleagues reported an annual incidence of 290–580 cases from 2004 to 2008. The same report suggested an incidence of 2–5 times the number of reported cases [1]. An incidence rate of 6.7 per 1000 was reported in CNAM in 2003 [7].

Data reviewed suggest a lower transmission of CL in urban areas [8] compared to rural area of Segou [13]. The transmission may vary considerably between villages in rural area. A discrepant prevalence and incidence rates [10] and discrepant frequency [19] of CL within villages in rural area have been observed.

#### 4.1.3. Limitations

We did a meta-analysis to measure the burden of CL; few studies have been conducted on CL in Mali. We analyzed studies conducted in different areas in Mali; these studies may have different designs and may use different diagnosis tools conducted at different times. The differences in the estimates and risk of CL in Mali could be explained by the heterogeneity of the studies and design effect. In spite of these differences, this review shows that CL is a rural disease and that transmission occurs mainly in rural areas of the North Savanna and Sahel regions of Mali.

#### 4.1.4. Risk factors

The risk of CL is likely to be important in the rural areas of Kayes, Koulikoro, Segou, and Mopti where higher LST-positive reaction has been reported [8]. CL cases recorded ( $n = 251$ ) between 1997 and 2001 show that CL is endemic in the eight regions of Mali; few cases ( $n = 4$ ) were from Tombouctou–Gao–Kidal in the north of Mali compared to Bamako–Kayes–Segou, with ( $n = 233$ ) cases at the center-west of Mali. Infected people were between 20 and 40 years old; men were more infected than women [19], CL cases occurred in travelers visiting endemic areas of Mali [23] where locals populations

such as farmers, ranchers, and military personnel are mostly infected by *L. major* [7,10,19,23,24]. Although the disease affects all age groups, the risk of *Leishmania* infection increases with age [9]; children aged less than three years have lower infection rates compared to adults [10].

In West Africa, as in Dogon villages in Mali, outbreaks were also reported in Ouagadougou city, Burkina Faso [25] and in Ghana within communities presenting significant population movements between endemic and non-endemic areas [26]. Mass migration of unimmunized people in endemic areas and domestication of zoonotic foci due to rapid urbanization and agro-industrial projects such as dams, wells, roads, trash deposits, irrigation systems, and deforestation may have contributed to increased risk of parasitic diseases such as CL [25–27].

#### 4.1.5. Leishmania-HIV co-infection

Co-infection with *Leishmania* and HIV was reported Mali [7,28]; the risk factors associated with HIV-*Leishmania* co-infection are not well documented in Mali. Descriptive studies at CNAM show that in 261 skin diseases in HIV-positive, there were two (0.7%) CL cases [28], and among 251 CL cases, six (2.3%) were HIV-positive [7]. These results could be explained by the overall lower prevalence of HIV infection in Mali (1.1%) [6]. In Cameroon, 4.8% of CL (7/146) cases were HIV-positive; women were more co-infected than men, likely due to the higher prevalence of HIV in women [29]. However, in Burkina Faso, such differences were not observed [30]. HIV-positive patients were not more exposed than HIV-negative patients to *Leishmania* infection, and co-infected patients showed a lack of healing of skin lesions [31]. Severe and atypical clinical spectrum of CL has been observed in HIV-positive patients and children [7,31], co-infection with *Leishmania* and HIV has caused death ( $n = 1$ ) in CNAM [7].

#### 4.1.6. Leishmania parasite

The enzyme electrophoresis analysis identified four strains of *L. major* in Mali. The most prevalent strain is MON-26, (15 out of 30), followed by MON-74, which is the most prevalent strain described in Burkina Faso [5,31]. Attempts to culture parasites from samples collected in Dogon country for identification of *L. major* strains was unsuccessful, but Izri *et al.*, were able to successfully grow *L. major* from a patient in Bamako and described the first case of CL due to *L. major* MON-26 [3]. *L. major* DNA was detected in wound scrapings and sand flies in Dogon country and Segou [13,14,19]. PCR-confirmed CL cases demonstrated that *L. major* was the main species in Mali, with a large geographical distribution [19]. However in regions of Mali where LST-positive reaction has been reported without documented PCR-confirmed CL, *Leishmania* sp. screening should be done.

#### 4.1.7. Vectors

The first collection of sandflies was done in Mali in 1943 [11]. Ranque *et al.*, in 1972 described sandfly fauna on the mountain of Point G around Bamako and have identified *P. sergenti* and several species of *Sergentomyia* sp. [12]. Other studies have shown an important variability of sandfly fauna within surveyed areas where the *P. duboscqi* vector of *L. major* was frequently trapped in rural and suburban areas. *S. schwetzi* was most abundant in the Segou region but very rare in the Mopti region; *Sergentomyia* (*Spelaemyia*) *darlingi* was more



frequently trapped in the Bandiagara area (Mopti) than the Segou region [13–15].

In Dogon country, four *Phlebotomus* species (*P. duboscqi*, *P. rodhaini*, *P. kazeruni*, and *P. sergenti*) and 16 *Sergentomyia* species were found, including *S. (S) darlingi* which contained *L. major* DNA and could play a role in endemic transmission of CL in Mali [14]. These data call for investigating the role of *S. (S) darlingi* in *L. major* transmission in Mali. It has been shown that *S. schwetzi* is refractory to *L. major*, *Leishmania donovani* and *Leishmania infantum* [32], but in Segou area, no *Sergentomyia* species were incriminated as a possible vector of *L. major*, compared to Bandiagara [14], Ghana [33] and Portugal [34] where respectively *S. (S) darlingi*, *Sergentomyia ingrami*, *Sergentomyia hamoni* and *Sergentomyia minuta* were naturally infected by *L. major*.

Few data were found on *L. major* transmission in urban and suburban areas of Mali. Bamako sandfly surveys showed the presence of two genera: three *Phlebotomus* species (*P. duboscqi*, *P. rodhaini* and *P. sergenti*) and several *Sergentomyia* species [12,15].

A residual local transmission of *L. major* in Bamako and surrounding areas might be possible. CL cases reported in residents of Bamako [7], the presence of *P. duboscqi*, *P. sergenti* and *P. rodhaini* [12,15] in suburban area and the presence of *P. duboscqi* in surrounding villages [16] support the possible transmission of *L. major* in suburban areas of Bamako. *P. rodhaini* captured around Bamako has been described recently as a possible vector of *Leishmania* spp. [27,34]. So far, urban and suburban transmission of *Leishmania* spp. has not been established in Mali. *P. duboscqi* has been confirmed as the vector of *L. major* in rural areas of Mali [13,14].

#### 4.1.8. Parasite reservoirs

The reservoirs of *L. major* in West Africa are rodents (Table 2). In Senegal *Mastomys erythroleucus*, *Tatera gambiana* and *Arvicanthis niloticus* have been found to be infected by *L. major* [35,36]. To our knowledge, there is no data found on identification of *Leishmania* parasites in mammals in Mali, and an attempt to identify *Leishmania* in rodents was unsuccessful [24]. However, in Kenya *Mastomys natalensis*, *Tatera robusta* and *Arvicanthis niloticus* are reservoirs of *L. major* [37]. Future investigations on *Leishmania*'s reservoirs in Mali should cover rodents and other mammals such as hedgehogs, as *L. major* has been isolated from the two species of hedgehog *Atelerix algirus* and *Paraechinus aethiopicus* in Algeria [38].

In Mali, CL is widely distributed, and the prevalence rates of positive reactions to LST are variable; age, residence and occupation are risk factors. Because of the wide geographic

distribution, climate and ecological changes, and the risk of outbreak, health workers and authorities should be aware of possible transmission of *L. major* and occurrence risk of CL outbreaks in Mali. Sparse data on geographic distribution of CL, population movement, HIV/AIDS co-infection and climate change call for implementation of epidemiological surveillance and more research, with the aim to better understand the epidemiology of CL in Africa. Identification of reservoirs and transmission paths in urban and suburban areas of Mali are required. This would help in the design of new and better adapted strategies for CL prevention and treatment.

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**Table 2**

Identified *Leishmania major* reservoirs in West Africa.

Species of rodent	Countries
<i>Mastomys erythroleucus</i> and <i>Tatera gambiana</i>	Senegal
<i>Mastomys natalensis</i> and <i>Tatera gambiana</i>	Nigeria
<i>Tatera gambiana</i> , <i>Mastomys natalensis</i> , <i>Taterillus</i> sp. and <i>Cricetomys gambianus</i>	Republic of Guinea Burkina Faso (Zida, A., personal communication)

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