Clinco-Epidemiologic Study of Cutaneous Leishmaniasis in Bikaner, Rajasthan, India

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Abstract. Cutaneous leishmaniasis is endemic to the Thar Desert of Rajasthan, Bikaner, India. The present study describes clinico-epidemiologic data of all cases of cutaneous leishmaniasis CL in this region during 2001–2011. A total of 1,379 patients with 2,730 lesions were reported during the study period. Ages of patients ranged from 3 months to 86 years, and there was a predominance of infections in males. Most patients were from urban areas and lower middle socioeconomic groups. Lesions were dry, ulcerated nodules or plaques of different sizes commonly over face and upper limb. Skin smears were positive for parasites in 958 (69.5%) patients, and the remaining 45.8% (193 of 421) patients were positive by skin biopsy. Histopathologic analysis of the skin showed mixed granulomas consisting of macrophages, lymphocytes, epitheloid, and plasma cells. Species identification was conducted for 45 randomly selected patients by polymerase chain reaction, the infective species was *Leishmania tropica*. Most patients were treated with intra-lesional injections of sodium stibogluconate.

INTRODUCTION

Leishmaniasis is a vector-borne parasitic disease that is endemic to 88 countries. Global prevalence is approximately 12 million cases, with 1–2 million new cases each year throughout Africa, Asia, Europe, and North and South America. *Leishmania* causes different type of clinical manifestations ranging from cutaneous leishmaniasis and mucosal leishmaniasis to potentially fatal visceral leishmaniasis.1,2 According to the World Health Organization, the annual incidence of cutaneous and visceral leishmaniasis is 1–1.5 million and 500,000 cases, respectively.3,4

Cutaneous leishmaniasis in the Old World is caused by *Leishmania tropica*, *L. major*, and *L. aethiopica*. New World leishmaniasis is caused by *L. Mexicana*, *L. amazonensis*, and *L. braziliensis*.5 Clinical manifestations range from spontaneously healing nodulo-ulcerative lesions to mutilating cutaneous or mucosal ulcers.6 Rarely, diffuse cutaneous leishmaniasis occurs, particularly in immunocompromised patients.5

In India, cutaneous leishmaniasis is known to be endemic to the Thar Desert of Bikaner in Rajasthan State, which is located in northwestern India at an elevation of approximately 227 meters above mean sea level (Figure 1). Bikaner is situated in the middle of the Thar Desert. The area of the city is approximately 37.95 km²; the interior of the city is surrounded by a stone wall and is overcrowded. Most of the time climate of Bikaner is hot and dry with little rainfall (265 mm/year).6

The first evidence for cutaneous leishmaniasis in Bikaner was based on clinico-epidemiologic analysis of cases from a large-scale outbreak that occurred during 1971–1973.7–10 Agarwal and Chadda11 in 1981 and Vyas and others12 in 1994 reported the epidemiology of cutaneous leishmaniasis in Bikaner. In 2007, Kumar and others13 identified *L. tropica* as the causative species of cutaneous leishmaniasis in Bikaner in rural and urban areas. This disease, once restricted to the Thar Desert in India, is now showing a trend to spread to previously non-endemic areas, such as Himachal Pradesh. In 2005, Sharma and others14 reported a new endemic zone of cutaneous leishmaniasis in the Subalpine Valley, which is located along the Satluj River in the Kinnaur District of Himachal Pradesh.

Cutaneous leishmaniasis is an emerging public health problem in certain regions of India, particularly in the Bikaner Region of the State of Rajasthan. The current study was conducted because no data are available on the clinico-epidemiological profile of this disease in this region after 1994. Epidemiologic information for cutaneous leishmaniasis in this region is of paramount importance for implementing appropriate control strategies to prevent the spread of this disease to other regions.

MATERIALS AND METHODS

The present study included persons with clinically diagnosed consecutive cases of cutaneous leishmaniasis who came to the Dermatology Department of Sandar Patel Medical College in Bikaner, Rajasthan, India, during January 2001–December 2011. Demographic data were recorded on printed proforma sheets and included age, sex, occupation, religion, habitat, area of residence, family history, history of keeping domestic animals, and socioeconomic status. Clinical details were also recorded and included number, size, site, and type of lesions; duration of disease onset; confirmative investigatory modality; treatment given; and follow-up. Lesions were classified as early (duration less < 6 months) and late lesions (duration ≥ 6 months).

Lesions were also classified as papules, nodules, ulcerative nodules, plaques, and ulcerated plaques. Clinically diagnosed cases were confirmed by demonstration of parasites in Giemsa-stained skin slit smears from lesions. Skin biopsies were performed on patients who were smear negative. Parasite species was identified for some randomly selected patients and was determined by internal transcribed spacer 1 polymerase chain reaction–restriction fragment length polymorphism–(PCR) and kinetoplast DNA PCR at a molecular biology laboratory of the Indian Council of Medical Research in New Delhi. Relevant tests for individual patients were conducted before starting treatment and included routine hemograms, urine analysis, liver function tests, and renal function tests.

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The following treatments were administered on a case-by-case basis. Most patients were given intralesional injections of sodium stibogluconate (SSG, Pentostam; Albert David Ltd., Kolkata, India) at a dose of 0.5 mL/cm² (100 mg/mL solution) of lesion once or twice a week for 5–7 injections. Some patients were treated with a single application of topical radiofrequency heat therapy (RFHT), which was introduced in 2009. The controlled heat was applied locally at 50°C for 30–60 seconds under local anesthesia, depending on the thickness of the lesion, with a localized current field radiofrequency generator (ThermoMed 1.8 device; Thermosurgery Technologies, Inc., Phoenix, AZ). Children and patients with multiple ulcers were treated with oral rifampicin at a dosage of 20 mg/kg of body weight/day for 4–6 weeks. Some patients who did not respond to intralesional SSG injections were treated with cryotherapy, oral dapsone, oral ketoconazole, topical paramomycin, or other antibiotics.

Complete cure was defined as total re-epithelization of the lesion with or without scarring and a negative skin smear. Data were tabulated and analyzed by using descriptive statistics in a Microsoft (Redmond, WA) Excel spreadsheet (2010). Records were analyzed for demographic profile, clinical data, method of diagnosis, and outcome of treatment given.

RESULTS

A total of 1,379 clinically diagnosed patients with cutaneous leishmaniasis with 2,730 skin lesions were reported during the study period. The incidence of cutaneous leishmaniasis increased every year up to 2009, after which there was a decrease in 2010 and 2011. The highest number of patients was seen in 2009. Cases of cutaneous leishmaniasis during winter (November–April) were higher compared with those during summer (May–October). The lowest number of cases for a month was reported in October (58 cases), and the maximum number of cases was reported in January (181 cases).

Epidemiologic data showed that cutaneous leishmaniasis was prevalent in persons of all ages; the youngest patient was 3 months of age and the oldest was 86 years (mean age = 23 years) (Table 1). The highest proportion of patients (n = 379, 27%) were 11–20 years of age. The ratio of male to female patients was comparable. In persons < 20 years of age, male patients were affected more frequently. In persons 21–50 years of age, infection prevalence was higher in female patients. Most patients in our study were from old-walled localities, including Kotegate (552 patients, 40%) and Rani Bazar (441 patients, 32%). These regions are heavily populated areas and have poor sanitary conditions.

Students and housewives of the socioeconomically lower middle class population were most commonly affected. A positive family history was seen for 173 patients (13%). A total of 139 patients (10%) with a history of keeping domestic animals (including dogs, cows, and buffaloes) on their residential premises were positive for cutaneous leishmaniasis. Most patients who kept animals were from rural areas. The infection ratio between urban and rural populations was 4:1.
The time between the appearance of a cutaneous leishmaniasis lesion and the first diagnosis ranged from 2 days to 38 months. The average time when the patient first visited our clinic for treatment was 16.5 weeks after lesion onset, and 1,168 patients came to our clinic between 4 and 24 weeks after lesion onset. Early and late lesions were observed in 1,191 (86%) and 188 (14%) patients, respectively.

A total of 2,730 lesions were present in 1,379 patients. Most patients had one lesion (805 patients, 58%), whereas two lesions were present in 275 patients (20%), three in 118 patients (9%), four in 71 patients (5%), five in 35 patients (3%), and only five patients had more than 10 lesions. Lesions varied in size from a few millimeters to 12 cm in diameter. A total of 1,938 (71%) of 2,730 lesions ranged in size between 0.5 cm and 3.0 cm, and only 48 lesions were > 5.0 cm. Most lesions were located on exposed parts of the body such as the face (33%), upper extremities (41%), and lower extremities (20%). The trunk was involved in only 2% of patients, and none of the patients had mucosal lesions. Of 2,730 lesions, nodular (26%) and ulcerated plaques (35%) were the most common (Figure 2). Papules (3%), ulcerated nodules (13%) and plaques (23%) comprised the remainder of the lesions.

Clinical diagnosis was confirmed in 958 (69%) patients by Giemsa-stained skin slit smears, which showed amastigotes of *Leishmania*. Skin biopsies were performed in 421 patients who were smear negative. Biopsy specimens were positive for *Leishmania* in 193 (45.8%) of 421 patients. Histopathologic analysis of lesions showed granulomatous inflammation comprised of parasitized macrophages, lymphocytes, epitheloid, and plasma cells. Internal transcribed spacer region 1 restriction fragment length polymorphism–PCR and kinetoplast DNA PCR were performed for 45 randomly selected patients and all patients were positive for *L. tropica*.

Many treatments, including intralesional SSG, oral rifampicin, thermotherapy, as well as cryotherapy were used on

<table>
<thead>
<tr>
<th>Table 1</th>
<th>Epidemiologic characteristics of patients with cutaneous leishmaniasis, Bikaner, Rajasthan, India</th>
</tr>
</thead>
<tbody>
<tr>
<td>Characteristic</td>
<td>No. (%) patients</td>
</tr>
<tr>
<td>Age, years</td>
<td></td>
</tr>
<tr>
<td>0–20</td>
<td>732 (53)</td>
</tr>
<tr>
<td>21–40</td>
<td>430 (31)</td>
</tr>
<tr>
<td>41–60</td>
<td>160 (12)</td>
</tr>
<tr>
<td>&gt; 60</td>
<td>57 (4)</td>
</tr>
<tr>
<td>Sex</td>
<td></td>
</tr>
<tr>
<td>M</td>
<td>764 (55)</td>
</tr>
<tr>
<td>F</td>
<td>615 (45)</td>
</tr>
<tr>
<td>Regional distribution</td>
<td></td>
</tr>
<tr>
<td>Kotegate</td>
<td>552 (40)</td>
</tr>
<tr>
<td>Rani Bazar</td>
<td>441 (32)</td>
</tr>
<tr>
<td>Peripheral colonies</td>
<td>124 (9)</td>
</tr>
<tr>
<td>Rural area</td>
<td>262 (19)</td>
</tr>
<tr>
<td>Occupation</td>
<td></td>
</tr>
<tr>
<td>Farmer</td>
<td>41 (3)</td>
</tr>
<tr>
<td>Labor</td>
<td>187 (14)</td>
</tr>
<tr>
<td>Serviceman</td>
<td>185 (13)</td>
</tr>
<tr>
<td>Housewife</td>
<td>323 (23)</td>
</tr>
<tr>
<td>Student</td>
<td>643 (47)</td>
</tr>
<tr>
<td>Economic status</td>
<td></td>
</tr>
<tr>
<td>Higher</td>
<td>4 (0.3)</td>
</tr>
<tr>
<td>Middle</td>
<td>1,142 (83.8)</td>
</tr>
<tr>
<td>Poor</td>
<td>233 (16.9)</td>
</tr>
<tr>
<td>Habitat</td>
<td></td>
</tr>
<tr>
<td>Rural</td>
<td>275 (20)</td>
</tr>
<tr>
<td>Urban</td>
<td>1,104 (80)</td>
</tr>
</tbody>
</table>

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![Figure 2](image-url) Morphology of skin lesions in patients with cutaneous leishmaniasis from Bikaner, India. A, Nodule; B, ulcerated nodule; C, ulcerated plaque; D, crusted plaque.
case-by-case basis, and complete cure rates in response to these treatments are shown in Table 2. Of 1,005 patients treated with intrallesional SSG, 609 patients were given two SSG injections weekly and 396 patients were given injections once a week. Only 361 patients in the twice a week group and 297 patients in the once a week group completed the study; thus, 347 patients were lost to follow-up. Of 200 patients treated with RFHT, 15 patients were lost to follow-up. A total of 56% patients in RFHT group and 54% patients in SSG group came for follow-up at 18 months post-treatment and were found to be disease free. Forty three of 139 patients in the rifampicin group were lost to follow-up. Treatment with N2O cryosurgery showed cure rates of 85% at 12 weeks. Ten of 10 patients who were treated with paramomycin topically twice a day for 10 days were cured completely after 12 weeks. Oral 1.24:1. This finding is similar to those in studies conducted in Bikaner.6,12 In contrast, Yemisen and others16 and Gurel and others17 reported a higher incidence of females. Students and housewives were most commonly affected in this study, which is similar to that in the study by van der Meide and others.15 A positive family history was seen in 173 (12.5%) patients in the present study, while 17% of patients had a positive family history of cutaneous leishmaniasis in Bikaner.6,12 These data suggest that the clinic-epidemiologic profile of cutaneous leishmaniasis in Rajasthan and Himachal Pradesh could be different.

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reported 75% and 25%, respectively. The average number of lesions was 1.98 in our study; Srivastava and others6 and Sharma and others14 reported averages of 1.4 and 1.58, respectively. Similar to earlier studies5,7 in the same location, our study found that exposed parts of the body are most affected, likely because sand flies can easily bite exposed parts during sleep. The most frequent type of lesion in the present study was the ulcerated plaque, and Sharma and others14 reported ulcerated nodules as the most frequent lesion.

Our study showed that 69.5% of skin smears were positive for *Leishmania* parasites, which is similar to findings in studies by Yemisen and others16 and Al Samarai and Allobaidi.19 Tissue smears from skin biopsies were positive in 45.8% of skin smear–negative patients in the present study, and Sharma and others14 reported only 37% positivity in tissue smears. On the basis of our PCR findings, *L. tropica* was identified as the causative organism of cutaneous leishmaniasis in Bikaner whereas *L. donovani* was found to be the causative species in most patients from Himachal Pradesh, India.14

Although the incidence of CL cutaneous leishmaniasis has decreased in the past two years, it is still a public health problem in this region of India. A closer look at sanitation programs to control vectors and improvement of living conditions for the population is needed for better control of the infection and preventing its spread to other regions.

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