LEISHMANIASIS CONTROL STRATEGIES

A CRITICAL EVALUATION OF
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This series includes meeting documents, internal reports, and preliminary technical documents that may later form the basis of a formal publication. A Manuscript Report is given a small distribution to a highly specialized audience.

La présente série est réservée aux documents issus de colloques, aux rapports internes et aux documents techniques susceptibles d’être publiés plus tard dans une série de publications plus soignées. D’un tirage restreint, le rapport manuscrit est destiné à un public très spécialisé.

Esta serie incluye ponencias de reuniones, informes internos y documentos técnicos que pueden posteriormente conformar la base de una publicación formal. El informe recibe distribución limitada entre una audiencia altamente especializada.
Leishmaniasis control strategies
Leishmaniasis control strategies:
A critical evaluation of IDRC-supported research

Proceedings of a workshop held in Mérida, Mexico, November 25–29, 1991, sponsored by the International Development Research Centre, in collaboration with the Universidad Autónoma de Yucatán (UADY) and the Universidad Peruana Cayetano Heredia (UPCH)

Edited by
Pandu Wijeyaratne, Tracey Goodman
and Carlos Espinal
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The Ecology of Visceral and Cutaneous Leishmaniases in Tunisia

M.S. Ben Rachid, R. Ben-Ismaili, M. Ben Saïd

Introduction

Visceral (kala-azar) and cutaneous leishmaniases have been known in Tunisia for many years. The first cases of cutaneous leishmaniasis were reported in the Governorate of Gafsa in 1884 by Deperet and Boinet. The world's first case of infantile mediterranean kala-azar was reported in the suburbs of Tunis in 1904 by Laveran and Cathoire. Through a chronicle of the leishmaniases (published in the records of the Institut Pasteur de Tunis) started by Charles Nicolle (1912) and regularly updated by others, we are able to follow the development of the epidemiological situation of these diseases in Tunisia from the turn of the century to the present. Beginning in the 1980s, this situation has gradually changed as a result of a severe epidemic of zoonotic cutaneous leishmaniasis in the mid-south of the country (Bouratbine-Belma 1988), where the disease had not previously been observed (Rachid et al. 1983), and the unexpected appearance of new foci of kala-azar in the same areas.

On the basis of clinical, epidemiological criteria and the occasional typing of strains, four forms of leishmaniases can be found in Tunisia:

(i) infantile kala-azar (KA) (Map 1);
(ii) sporadic cutaneous leishmaniasis (SCL) in the North;
(iii) zoonotic cutaneous leishmaniasis (ZCL) found in rural areas; and
(iv) anthroponotic cutaneous leishmaniasis (ACL) in the Southeast (Map 2).

The general objective of this project investigation was to study the role of various vectors and animal reservoirs in the transmission of these forms of human leishmaniases in Tunisia in order to implement an appropriate strategy for the rational control of the leishmaniases. The specific objectives were:

(i) - to determine the prevalence of canine leishmaniasis in the foci of kala-azar and establish its relationship with human visceral leishmaniasis.

(ii) - to confirm the theory that dogs can host strains related to cutaneous leishmania.

(iii) - to determine, in endemic strains, the role of various rodents as reservoirs of visceral and cutaneous leishmaniases.

(iv) - to study vector species, their density and association with the various clinical forms of leishmaniasis.

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2 The Lesihmaniases Project, IDRC 3-P-86-0308.
ALGERIA

KALA AZAR INDEX DISTRIBUTION
ACCORDING BIOCLIMATIC STAGES

- Humid and subhumid
- Semi-arid
- Arid
- Saharian

= 1/100,000 children

MAP 1
GEOGRAPHICAL DISTRIBUTION OF THE VARIOUS CUTANEOUS LEISHMANIASIS FORMS OBSERVED IN TUNISIA

SCL: Northern sporadical cutaneous Leishmaniasis
ZCL: Zoonotic cutaneous Leishmaniasis
ACL: Anthroponotic cutaneous Leishmaniasis
Methodology

The Relationship of KA with Canine Leishmaniasis

Detection of KA (kala-azar) was passive, based on samples (bone marrow smears or serodiagnosis) that were received by us and annual statistics collected by the various pediatric health care services in Tunisia (Tunis and suburbs, Sousse, Kairouan, Sfax) and occasionally some adult services.

The relationship between canine leishmaniasis and human leishmaniasis was studied in a recent focus of KA: the Governorate of Sidi Bouzid, located in the mid-south, where six cases of kala-azar were reported between 1986 and 1988 and an epidemic of ZCL has been raging since 1982.

The survey of the dog population was conducted in two stages, always during the diapause seasons of the sandfly (January and April). The first stage took place in 1987 and 1988 and focused on the first four cases (I and IV), grouped in the same area; this we have called the focus area. The second was carried out in 1990, to evaluate the results of the control methods implemented during the first study and to complete the data on the other two cases (V and VI).

The first survey involved the Imadat de Saddaguia and the Imadat de Touila (Fig. 1), an area of irrigated crops covering 57 km², separated from each other by El Fekka Wadi. The total child population is 834, i.e. 14.63 children per km². Each survey took place within a 3 km radius of each patient's home (in theory, a distance equal to that of the flight of the sandfly). Three control areas without any cases of kala-azar were also investigated: Adlet on the border, Abadlia 20 km away with the same biotope as the focus and Jemel 25 km away where irrigated farming is relatively undeveloped.

After the dogs with owners had been surveyed (place, name, age, numbers), a systematic serological study was made using the IFI test (significant titer at 1/100 level, the antigen used by L. infantum, Tunisian strain). Samples were taken only from dogs over four months.

In 1987, we collected the data using a method that proved somewhat ineffective. In 1988, we adopted a door-to-door method with the help of (in all cases) the staff of the Basic Health Care Division (BHCD). A total of 677 dogs were tested. Positive dogs were, with their owners' consent, either bought or replaced by a healthy dog; they were then put down and autopsies were carried out to obtain smears, cultures in modified NNN medium (Brain Heart Agar Blood (BHAB)) and typing of isolated strains.

Data on agriculture and water resources programs was obtained from the appropriate divisions of the Ministry of Agriculture.
Kala Azar Focus and Control Areas in the Sidi Bouzid Governorate

Control areas

KA focus (I to IV)

FIGURE 1
Zoonotic Cutaneous Leishmaniasis (ZCL): A Study of Rodents as Reservoirs of *L. major*

One of the main tasks of the Basic Health Care Division has been the passive detection of human cases. The role of rodents as reservoirs of *L. major* was very carefully studied in the focus of Douera located in southwest Sidi Bouzid (Governorate of Gafsa), a mixed focus of *Psammomys* and *Meriones* (Ben-Ismail et al. 1987).

*Psammomys* are captured manually during the day following the destruction of their burrows; other rodents are caught during the night using a Sherman metal trap placed near the openings of the burrows.

Specimens causing identification problems are sent in formalin to the Natural History Museum (Prof. F. Petter) for accurate determination.

We looked for Leishmania in superficial cutaneous lesions, mainly on the ears but also at the visceral level: liver and spleen. Imprints were prepared and stained with MGG. A culture was made in modified NNN medium (BHAB) as well as an inoculation on the plantar cushion of a BALB/C mouse. When lesions developed on the mouse, samples were placed in a culture and the isolated strains typed.

In order to evaluate the extent to which the reservoir rodents were affected, we also tested the humoral response in *P. obesus*, *M. shawi* and *M. lybicus* by using the IFI test with *L. major* as the antigen and antoglobulin homologous markers and/or a commercial anti-rat preparation.

**Sporadic Cutaneous Leishmaniasis (SCL) of the North and Anthroponotic Leishmaniasis of the Southeast (ACL)**

The problems caused by KA and ZCL relegated these two forms of leishmaniases to secondary importance in the study. A survey of foci and some limited entomological studies were the main activities. We encountered, however, a specific problem in maintaining strains obtained from the primary culture of SCL lesions. We tried several methods: culture in BHAB and EMTM media, inoculating an immunodeficient golden hamster with cortisone, inoculating the BALB/c mouse infected with TG 180 sarcomatous cells (Monjour's method) and inoculating the muzzle of a three-month old dog.

We were able to obtain only one strain from a sixty-year old woman from El Haouaria (Cape Bon) who had two lesions on her lower limbs. A biopsy was performed, then crushed under sterile conditions and inoculated in Evans’ "sloppy" medium. The promastigotes collected after a month were cultured in ordinary media and typed. The inoculated dog did not develop lesions during the year it was kept under observation.

The application of specific DNA probes represents undeniable progress in determining this strain.
A comparative study of the intensity of the humoral response was carried out on 173 serums from three cutaneous forms of leishmaniasis found in Tunisia. It was conducted with a dosage of IgG and IgM assays; at the same time we looked for specific antibodies using the IFI and ELISA tests with L. major, L. tropica and L. infantum as antibodies for each serum sample (Ben Saïd, Khaled, et al. 1987).

**Entomological Studies**

Two types of studies were conducted. The first type was designed to establish the density of the various species of sandfly. Two such studies were conducted: one in July-August 1987 in Tataouine, an area of ACL and the other in September 1988 in Sidi Thabet, located 20 km north of Tunis, a focus of KA and SCL. The method used was that of sticky paper traps: papers measuring 20 cm by 20 cm were soaked in castor oil and left at the foci before sunset and collected the following morning. The sandflies caught were put in 70% alcohol. They were then cleared in Amann lactophenol and mounted in chloral gum for identification.

The second type of study was designed to confirm the roles of the various sandflies as vectors of leishmaniasis. Two such surveys took place: one in Douara in July 1987 and the other in Nasrallah and Sidi Bouzid in July and September 1989. The sandflies caught in CDC-light traps were dissected under a magnifying glass and the Leishmania injected into the adipose cushions of the feet of the BALB/c mice. From these lesions (after about one and a half months), we prepared a culture in BHAB medium and conducted an isoenzymatic typing of the strains that were isolated.

**Isoenzymatic Typing of Leishmania Strains**

The isoenzymatic typing of leishmania strains was conducted with the cooperation of L’Instituto di Sanita de Roma (Gramiccia and Gradoni) in accordance with the procedures described by Maazoun et al. (1981).

**Results**

**I. Kala-azar**

a) **Human Statistics:** Sporadic cases of kala-azar, in the form of classical infantile mediterranean kala-azar, have occurred in Tunisia: 77% of cases were less than 5 years old, while only 2.8% of patients were over 14 years. Slightly more males were affected than females. The patients were generally from rural areas and of modest means.

The most common type of habitat is a permanent house or one made out of clumps of earth: poorly ventilated, surrounded by orchards irrigated by a nearby well and sheltered by a high cactus hedge. On the clinical level, fever, splenomegaly, hepatomegaly, wasting, pallor, pancytopenia and hypergammaglobulinemia are the most
common signs of the disease. The mortality rate still ranges from 5% to 8%, according to the health services. Table 1 shows the number of cases of KA reported in Tunisia from its discovery in 1904 until 1989. The most recent surveys (Anderson 1938; Vermeil 1956; Chadli et al. 1968; Rachid et al. 1983) emphasized the stability and grip of the foci in the sub-humid and semi-arid areas in the north of the country³ where the annual temperature is between 16-18°C and the rainfall above 250 mm; most of the cases are found in areas where the rainfall is between 600 and 400 mm per year. After a respite between 1970 and 1975 as a result of the eradication of malaria, which lead to a zero plasmodium rate in 1978, there was not only a new outbreak of kala-azar in the usual foci, e.g., Zaghouan (with an annual incidence of 20.73 cases per 100,000 children between 1982 and 1989, compared with 2.78 between 1958 and 1981) but it also spread to central Tunisia; 41.65% of total cases have occurred during the last eight years, especially in the Governorate of Kairoun, and new foci have appeared in Sidi Bouzid. The South was also affected with cases occurring in the Governorates of Sfax and Tozeur.

b) Canine Leishmaniasis: The results of the surveys carried out in Sidi Bouzid are summarized in Tables 2 and 3.

Table 2 shows the number of dogs tested in the focus during the 1987-88 survey and the number of IFI-positive dogs at a rate at least equal to 1/100. The positive rate varied between 1/100 and 1/3,200. Thirty-five dogs, out of the 380 tested in the focus, were found to be positive, i.e. 9.21%, compared with one dog in 123 in the control areas, i.e. 0.81%. The overall positive rate observed in the regions studied was 7.15%. In a recent study on canine leishmaniasis conducted using IFI and AD tests (Ben Saïd 1991) involving 265 dogs in the Enfidha region (a traditional focus of kala-azar), a plain in the North of the Governorate of Sousse on the eastern Mediterranean coast, it was noted that the figures varied from 2.94% to 16%, depending on the town. These rates are comparable to those found in our study, thereby indicating that the Sidi Bouzid region has become a highly endemic focus of kala-azar.

When we related the number of leishmanial dogs to cases of kala-azar in the focus, we found, however, that only two cases had a leishmanial dog in the home (cases II and V); within a 3 km radius of each case, however, we found between 5 and 13 leishmanial dogs per patient, i.e. about 10 affected dogs for every case of KA (Table 3).

³ Before 1956, the population affected was mainly of Italian and French origin; this observation enabled C. Nicolle to discover the reservoir of kala-azar: the dog which is found in homes of Europeans, but not in those of native people. In addition, the French and Italians were virtually the only people to develop and tend the vineyards; it is interesting to note that during the first half of the century the distribution of KA in the north of the country coincides with the wine-producing region (Map 3).
Table 1: Number of Cases of Kala-Azar (1904-1989)

<table>
<thead>
<tr>
<th>Years</th>
<th>1904-1967⁴</th>
<th>1968-1981⁴</th>
<th>1982-1989</th>
<th>TOTAL</th>
</tr>
</thead>
<tbody>
<tr>
<td>North</td>
<td>259</td>
<td>116</td>
<td>341</td>
<td>716</td>
</tr>
<tr>
<td>Center - Sahel</td>
<td>30</td>
<td>61</td>
<td>215</td>
<td>306</td>
</tr>
<tr>
<td>- Kairouan</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>- Kasserine</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mid-South - Sidi Bouzid</td>
<td>1</td>
<td>10</td>
<td>11</td>
<td></td>
</tr>
<tr>
<td>South - Sfax</td>
<td>0</td>
<td>0</td>
<td>11</td>
<td>11</td>
</tr>
<tr>
<td>- Tozeur</td>
<td>1</td>
<td>1</td>
<td>2</td>
<td>4</td>
</tr>
<tr>
<td>- Gabes</td>
<td>1</td>
<td>0</td>
<td>0</td>
<td>1</td>
</tr>
<tr>
<td>TOTAL</td>
<td></td>
<td></td>
<td></td>
<td>1049</td>
</tr>
</tbody>
</table>

Table 2: Canine Leishmaniasis in Sidi Bouzid

<table>
<thead>
<tr>
<th>Focus: Cases I to IV Saddaghia and Guedara</th>
<th>Control Areas: Adlet, Abadlia, Jemel</th>
<th>Selective killings of leishmanial dogs in 1988</th>
<th>2nd Survey</th>
<th>Control of focus: Saddaghia and Guedara</th>
<th>Cases V and VI</th>
<th>Control area: Mezaraa</th>
</tr>
</thead>
<tbody>
<tr>
<td>Number of dogs tested</td>
<td>380</td>
<td>123</td>
<td></td>
<td>101</td>
<td>42</td>
<td>31</td>
</tr>
<tr>
<td>Number of dogs IFI positive &gt; 1/100</td>
<td>35</td>
<td>1</td>
<td></td>
<td>9</td>
<td>7</td>
<td>0</td>
</tr>
<tr>
<td>%</td>
<td>9.21</td>
<td>0.81</td>
<td>8.91</td>
<td>16.66</td>
<td>0</td>
<td></td>
</tr>
</tbody>
</table>

⁴ See Anderson 1938; Chadli et al. 1968; Laveran 1904; Nicolle 1912; and Vermeil 1956.

⁵ See Ben Rachid et al. 1983.
MAP 3:
GEOGRAPHICAL DISTRIBUTION
OF KALA AZAR IN TUNISIA
(1904-1956)

Wine growing and producing area
Table 3: Number of Leishmanial Dogs Within a 3 km radius of Patients' homes: Survey 1987-88 Sidi Bouzid

<table>
<thead>
<tr>
<th>Case</th>
<th>I</th>
<th>II</th>
<th>III</th>
<th>IV</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>Number of leishmanial dogs</td>
<td>13</td>
<td>6</td>
<td>11</td>
<td>5</td>
<td>35</td>
</tr>
<tr>
<td>Town</td>
<td>Saddaghia</td>
<td>Guedara</td>
<td>Saddaghia</td>
<td>Saddaghia</td>
<td></td>
</tr>
</tbody>
</table>

The second survey conducted in 1990 supplemented the data on the cases of KA not studied in 1987-88: case V in El Ksar located 15 km to the south and case VI in Sidi Salem bordering on the northeast of the focus. Out of the 42 dogs tested in these two towns, seven were positive, i.e. 16.66%. In El Mezaraa, a focus bordering on El Ksar, no dogs were found to be infected.

The control, after a year of transmission from the focus studied in 1987-88 where all the leishmanial dogs were put down, had a virtually unchanged positive rate (Table 2). At the same time four new cases of KA were noted in the region between 1989 and 1990.

c) The Isolation and Typing of Strains: All the strains isolated from patients suffering from infantile kala-azar and leishmanial dogs both inside and outside the focus proved to be identical to the reference strain OMSMHOM/TN/80/IPT1 belonging to L. infantum s. st. Nicolle zymodeme MON 1. No strain of L. major was isolated from the dogs in the Sidi Bouzid focus.

The strain MHOM/TN/87/KA, 364, isolated from a 21 month-old child suffering from KA, from Barnoussa in the Governorate of Kef, deserves special mention. Its culture in NNN was easy, unlike the strain isolated from cutaneous lesions. Its biochemical typing showed that it was a variant of L. infantum zymodeme Mon 24. This zymodeme has been isolated from Oriental Ulcer in northern Algeria (Belazzoug et al. 1985), Italy and Tunisia. This very same zymodeme was isolated in Italy in an adult suffering from AIDS-related visceral leishmaniasis.

An inventory of the sandflies found in the Sidi Bouzid region was compiled by Hellal and colleagues in 1985. Apart from the genus Sergentomyia, four species of sandfly were present: P. papatasi (84.6%), P. perniciosus (13.2%), P. longicuspis (0.5%) and P. perfiliewi (0.04%). In order to isolate the leishmania in the sandfly, a second study was conducted in September 1989. It proved possible to isolate L. major from P. papatasi, but not L. infantum from the suspected vector, P. perniciosus.
The role of *P. perniciosus* as the vector of *L. infantum* Zym. Mon 1 has already been proven in Italy and more recently in Algeria (Izri et al. n.d.).

d) **Ecology of the KA Focus in Sidi Bouzid:** From preliminary studies on the ecological changes that have taken place in this region during the past few years, we were able to note the following:

- A significant increase in water resources development projects, especially surface wells. The density of wells per km² is significantly greater in the focus areas than it is throughout the governorate. As a whole, there were 0.28 wells per km² in 1976 and 0.98 wells per km² in 1987. In the focus areas, the number of wells rose from 3.53 per km² in 1976 to 13.23 per km² in 1987.

- Changes in the agricultural landscape. Irrigated farming has increased considerably since 1970. It revolves around market gardening, fodder, cereals and tree crops. The focus areas appear to specialize in market gardening and fodder crops with very few tree crops. Market gardening increased 55-fold throughout the governorate between 1974 and 1981.

We can conclude from this that irrigation and the changes in the use of farmland have compensated for the region's aridity, while helping to create a microclimate that is sufficiently humid for the survival and growth of the vector of kala-azar, *Phlebotomus perniciosus*, usually much more numerous in the humid areas of the North. This new focus was created when a large number of vectors and sick dogs were introduced from endemic areas at the same time.

II. **Zoonotic Cutaneous Leishmaniasis (ZCL)**

a) **Human Statistics:** Zoonotic cutaneous leishmaniasis (ZCL) seems to have always been widespread in southern Tunisia but absent in the northern and central parts of the country (Rachid et al. 1983). It results in multiple, wet cutaneous lesions on the face and limbs; they take about 3 to 6 months to heal and leave permanent scars.

An epidemic broke out in 1982 in the El Khobna Basin (Governorate of Kairouan) near the recently completed Sidi Saad dam. The disease then spread to ten governorates and nearly 30,000 cases have been recorded to date (Map 4). Epidemiological studies have enabled the identification of the parasite in humans as *Leishmania major* Mon 23 (Ben-Ismail, Gramiccia, et al. 1987).

b) **The Douara Focus Rodent Study:** It was possible to study all the elements of the ZCL cycle in the Douara focus (Ben-Ismail, Ben Rachid, et al. 1987). One-hundred and twenty-six rodents were caught during this study. They belonged to the following species: *Psammomys obesus* (Cretzschmar, 1828): 96 specimens; *Meriones shawi* (Duvernoy, 1842): 14; *Meriones lybycus* (Lichtenstein, 1910): 6; *Gerbillus* (G.) *campestris* (Le
Vaillant, 1857): 6; Gerbillus aureus (Selzer, 1956): 2; Gerbillus nanus (Blanford, 1875): 1; and Mus musculus (L.): 1.

The locations where the rodents were caught are indicated on Fig. (2). Nineteen rodents were found to be carriers with leishmanial lesions on their ears; this was confirmed by a direct examination of dermal smears. Of these 19, 15 of the 96 P. obesus (15.6%), 2 of the 14 M. shawi (14.3%) and 2 of the 6 M. libycus were carriers (Table 4).

Table 4:

<table>
<thead>
<tr>
<th>SPECIES</th>
<th>TOTAL</th>
<th>INFECTED</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>No.</td>
<td>%</td>
</tr>
<tr>
<td>Psammomys obesus</td>
<td>96</td>
<td>15</td>
</tr>
<tr>
<td>Meriones shawi</td>
<td>14</td>
<td>2</td>
</tr>
<tr>
<td>Meriones libycus</td>
<td>6</td>
<td>2</td>
</tr>
</tbody>
</table>

A study of the antibodies in the P. obesus, found negative in the parasitological examination, enabled us to detect an extra 36% IFI-positive.

The lesions found in the Psammomys were often difficult to detect: hair loss and darkening of the skin and by palpation: infiltration and small nodules. On the other hand, those found in the Meriones were much clearer, oedematous and destructive often on both sides.

During another study in July 1989, most of the Psammomys caught had leishmanial lesions; 20/20 at Nasrallah and 8/10 in the same focus of Douara, i.e. 80% to 100% were affected.

It therefore seems that P. obesus is the natural reservoir of L. major. A diurnal, prolific rodent that devours the Chenopodiaceae, it is not really a migratory animal. The greatest incidence of the disease in humans is found near its biotopes. Its distribution does not spread to the north, while in central Tunisia its distribution is exactly the same as that for ZCL. The Meriones, however, are nocturnal and migratory rodents that eat grain and are responsible for spreading the disease.

Entomological studies have identified the Phlebotomus papatasi as being the vector of ZCL (Ben-Ismail, Ben Rachid, et al. 1987). This species, whose anthropophilic is well known, seems to prefer a biotope of rodent burrows, where the darkness, humidity and temperature (25°C) are ideal. P. papatasi can be found in the immediate proximity of the parasite reservoir whose cycle it maintains.

All the strains isolated in this focus from the rodents P. obesus (9 strains), M. shawi (1 strain), M. lybicus (1 strain), and from humans (4 strains) are identical to the reference strain MHOM/MA/81/LEM265: L. major Mon 25.
GOVERNORATES
BY ZCL
(1982-1990)
30712 CASES
DOUARA ZOONOTIC CUTANEOUS LEISHMANIASIS FOCUS, TUNISIA

FIGURE 2

- Houses
- Oasis
- Cultivations
- Chénopods
- Jujube-tree
- Oueds
c) The Ecology of ZCL

The construction of dams and the implementation of agricultural development programs have an impact on the agents in the ZCL cycle and transmission. According to local inhabitants, the outbreak of the epidemic in Sidi Saad was preceded by an increase in the rodent population. This can perhaps be explained by the interruption, as a result of the construction of the dam, in the flooding that occurred in the basin and routinely decimated a high proportion of these rodents every year. The enrichment of the area's ground water also helped the existing Chenopodiaceae flourish, thereby increasing the food source of the Psammomys. Another factor was the cultivation in these regions of vast quantities of Atriplex, a plant grown for sheep fodder, but also enjoyed by the Psammomys (Bernard 1971). In addition to these facts, the true rivals of the Psammomys, the camel and sheep, suffered various fates.

The camel is a multi-purpose beast used especially for ploughing and its water consumption can range from 20 to 30 litres a day; the high water content of chenopodiaceae, therefore, made an ideal food. Hence, the camel's presence provided, along with the flooding, a natural way of controlling the Psammomys. Since the camel is an encumbrance in modern farming, it is now rare in Kairouannais and central Tunisia in general. It can, however, be found in the sub-Saharan areas where it is a tourist attraction and a means of transportation in the desert.

The Tunisian sheep, itself a victim of flooding and poor reproduction, cannot seriously compete with the Psammomys. Given the lack of modern rearing methods, its very presence, in the current situation, is a factor in desertification and the atriplex crop, intended for its consumption, instead encourages the proliferation of its formidable rival.

III: Sporadic Cutaneous Leishmaniasis (SCL) of the North

This form of leishmaniasis affects the north of the country sporadically (Map 2). Its distribution pattern is the same as that for kala-azar, except that it stops at the mountain range of the Tunisian Ridge (Table 4). The annual incidence of this form can be found in Table 5.

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6 The Sidi Saad dam was built between 1978 and 1982 to protect the Kairouan plain against the flooding of the wadis (Zeroud, Marguellil and Nebhana) that periodically devastated the region, thereby preventing any development. In 1969, for example, the huge plain was flooded with an incredible amount of water with a flow of 17,000 m³/s over a front more than 20km wide. This flooding destroyed bridges, roads, railways, power lines and homes and killed people and animals (Ministry of Equipment).
Table 5: Annual Incidence of SCL

<table>
<thead>
<tr>
<th>YEAR</th>
<th>Number of SCL Cases</th>
</tr>
</thead>
<tbody>
<tr>
<td>1976</td>
<td>17</td>
</tr>
<tr>
<td>1977</td>
<td>13</td>
</tr>
<tr>
<td>1978</td>
<td>13</td>
</tr>
<tr>
<td>1979</td>
<td>17</td>
</tr>
<tr>
<td>1980</td>
<td>10</td>
</tr>
<tr>
<td>1981</td>
<td>7</td>
</tr>
<tr>
<td>1982</td>
<td>5</td>
</tr>
<tr>
<td>1983</td>
<td>12</td>
</tr>
<tr>
<td>1984</td>
<td>13</td>
</tr>
<tr>
<td>1985</td>
<td>31</td>
</tr>
<tr>
<td>1986</td>
<td>8</td>
</tr>
<tr>
<td>1987</td>
<td>7</td>
</tr>
<tr>
<td>1988</td>
<td>26</td>
</tr>
<tr>
<td>1989</td>
<td>15</td>
</tr>
<tr>
<td>Total</td>
<td>194</td>
</tr>
</tbody>
</table>

SCL occurs in 90% of cases as a single ulcer on the face which rarely lasts longer than 30 months. The most common lesions are small crusty ulcers, surrounded by an erythematous reaction, quite often the colour of red wine (Chaffaï et al. 1988). On a parasitological level, the main characteristic of the promastigotes is that they are difficult to maintain in culture; this has for a long time prevented their biochemical characterization. The only strain that it has proved possible to type is identical in all aspects to L. infantum zymodeme Mon 24, already known in Algeria and Italy (Belazzoug et al. 1985; Ben Rachid et al. 1983). The recent application of specific DNA probes on amastigotes removed directly from lesions of eight patients made it possible to identify the causative agent of SCL as L. infantum. Italian studies indicate that P. perfiliewi is probably the vector of this form.

In the Sidi Thabet region, where both KA and SCL are found, 177 P. perniciosus were captured in September 1988, but only one female P. papatasi. The seasonal fluctuations of the various species still need to be clarified.

IV. Anthroponotic Cutaneous Leishmaniasis (ACL)

Anthroponotic cutaneous leishmaniasis (ACL) developed in the micro-foci located in the southeast between Toujène and Tataouine (Chaffaï et al. 1988). Its current incidence is less than 10 cases per year. This area has not been affected by ZCL.

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7 Statistics from the Department of Parasitology, Faculty of Medicine, Tunis.
As a rule only one ACL lesion is found (70%) on the face or limbs. The salient feature of ACL is the severity of the ulcers, which can last up to six years in some individuals. The lesions are dry and vegetated without a crust. Ulcerated forms that spread have been observed (Chaffai et al. 1988). Unlike SCL, ACL causes high levels of serumal antibodies to form, just as ZCL does.

On the parasitological level, culture is easy. It has proved possible to type only human strains, thereby identifying the causative agent as L. tropics MON. 8 (syn.: L. Killicki) (Rioux et al. 1986). The ACL found in Tunisia is different from the L. tropica forms found throughout the world because of its non-urban distribution and the occurrence of isolated and sporadic cases, which casts doubt upon its anthroponotic nature.

In the Tatahouine region, five species of sandfly have been catalogued: (July-August 1987):

- **P. papatasi** 67 specimens
- **P. sergenti** 32 specimens
- **P. alexandri** 17 specimens
- **P. chabaudi** 9 specimens
- **P. longicuspis** 3 specimens

*Phlebotomus sergenti*, common in the areas of transmission, is the most likely vector of ACL (Rioux et al. 1980) in Tunisia.

**Control Measures**

The control measures can be divided into two categories: (i) general measures and (ii) specific measures. Apart from the development of more detailed research on leishmaniasis and related subjects, general measures would include the following.

- **a) Training** and introduction to leishmaniasis for the medical and paramedical staff of the regions concerned. Numerous courses were given in Sidi Bouzid and Mahdia that included an exchange of information about experiences and problems that had been encountered. A nationwide course took place in 1988 for the doctors and auxiliaries of the Public Health Department.

- **b) Incorporation of the control of the leishmaniasis as a public health problem** in the operations of the Basic Health Care Division (BHCD) for KA and ZCL. By means of its outposts in the most remote areas and its close contact with the population, the BHCD is able to define its courses of action and, through health education, make people aware of the problems. With regards to KA this would require:
  - monitoring and early detection of human cases in the field so that action can be taken quickly to reduce fatal effects;
  - assuming responsibility for certain survey activities
- carrying out a census of the dog population so that an extensive study can be conducted;  
- testing dogs; and  
- making people more aware of the clinical signs of sick dogs and promoting the social acceptability of surveys and slaughtering of dogs.

Similarly because ZCL is spreading rapidly over a wide area, community participation is especially needed at various levels:

- at school, the teacher is the first to identify cutaneous lesions. By confirming the leishmanial origin of these lesions, it is possible to identify the areas affected, especially those recently surveyed.  
- the village people actively participate in the manual capture of the Psammomys by pointing out the various foci of the rodents and even monitoring the equipment used in the field for capturing them.

c) Increase government awareness by improving communications between the Ministry of Public Health and the other ministries involved. For example, with the Ministry of Agriculture this would involve:

- developing an integrated control program against the three diseases that have ravaged the country - rabies, hydatid cyst and visceral leishmaniasis, especially active control measures against the Meriones:  
- rational use of the land in risk areas by taking account of public health problems in the priorities of clearing, reafforestation, and erosion prevention.  
- strengthening outpost veterinary facilities so that they are comparable to those of the BHCD in human medicine.

With the Ministry of the Interior, the campaign against stray dogs must be improved by making the registration of the birth of dogs compulsory and introducing a vaccination book. With national education and information authorities appropriate action must be take to popularization new control measures in schools and the media.

In addition to these general measures, specific control measures for leishmaniasis would include:

a) Controlling the canine reservoir: Selective killing of leishmanial dogs was the method we adopted in this study. Killing was carried out in 1987 and 1988 and the control of dogs in the areas affected in 1990. The findings appear in Table 2. According to these findings, there has been no change either in the incidence of KA or in that of canine leishmaniasis.

On the face of it, this measure must be considered a failure in the short term. The dog census itself might be responsible for this failure. We were, in theory, able to
survey those dogs with owners, but in approximately 40% of cases, the owners refused to respond. Carefully planned health education and legislation making it compulsory to register dogs are therefore essential; these measures will also help in controlling rabies and hydatid.

The number of stray dogs was only estimated. Since the exact number is not known, the incidence of leishmaniasis in stray dogs cannot be determined. It is therefore essential that stray dogs be put down.

Since it is impossible to conduct a large-scale survey, the selective killing method might ultimately be dangerous because it affects only those dogs with owners, in other words, those registered as having been vaccinated against rabies. Their killing might result in an imbalance in favour of dogs that have not been controlled or vaccinated.

The mass killing of dogs cannot be considered except in the case of an epidemic. Combined with treating the sick and spraying insecticides in the home, it has had some success in China. A resurgence of kala-azar occurred in that country, however, between 1968-75 following the relaxation of these measures. The ecological impact of the mass killing of dogs has yet to be evaluated.

Another available option is to treat infected dogs. Dogs are usually treated with pentamidine (lomidine). It is reserved for dogs held in great affection by their owners; since this treatment only clears up the condition temporarily rather than curing it, it must be repeated every year. Such treatment is used in towns. It is inconceivable and unrealistic (cost, awareness) on an individual level in rural areas. In Tunisia, meglumine antimoniate (glucantime) is primarily reserved for human treatment. The use of glucantime for treating dogs in Italy has resulted in canine strains becoming resistant to antimony, thereby making the dog a permanent reservoir of leishmania.

b) Improving living conditions and providing balanced foods: Improving living conditions raises the problem of operational costs. Such improvement would enable the rational use of insecticides and the control of other possible reservoirs such as Rattus rattus, which has been found to be a carrier of leishmania in Yugoslavia, Italy and Spain.

Higher quality, better balanced diet falls within the general context of social and economic progress. KA is still a disease caused by poverty and malnutrition.

c) The value of control measures against ZCL: Control measures against ZCL result from an ecological analysis of the foci. Among the programs planned by the Tunisian government within the general context of land development or traditionally undertaken by the people, special attention is given to land that can actively contribute to controlling ZCL.
The campaign against the jujube tree, which is the traditional habitat of the Meriones. A project under way in Kairouannais is designed to clear the jujube trees from fertile and unused land so that crop-bearing species (almond, olive and pistachio trees) may be planted.

Another strategy has been the reafforestation of salty and skeletal soils. This land, which is uninhabited and not suitable for farming, remains a vast expanse where the Chenopodiaceae grow and the Psammomys actively multiply. Two types of plantation have been established there: acacia and cactus.

Acacia is regarded as a magic tree, since it is capable of forcing its roots several metres deep into the ground to draw the water and nitrogen it needs. This process, while successfully stabilizing the soil and regreening desert areas, benefits humans by providing a fodder crop for cattle. The land planted with acacia becomes shady and is no longer conducive to the growth of chenopodiaceae, which are a source of food for the Psammomys and thus for the parasite.

On the other hand, the cactus plantations that we visited had become foci for the Meriones. The rodents make their burrows at the foot of this spiny tree, as with the jujube, to protect themselves from predators.

The method used by the inhabitants of Sfax to till their land is also of interest in rodent control. The inhabitants of Sfax practice dry farming, which is indispensable in this country where there is so little rain, but is possible thanks to the thin soil and humid air. Olive trees are planted 20-24 metres apart. The land is worked using tractors with 7-metre wide blades which have teeth that are able to dig to a depth of 20-30 cm. Tilling is carried out six or seven times a year to reduce water evaporation, aerate the soil and destroy weeds. This tilled land has remained ZCL-free, unlike the so-called transit areas, uncultivated grazing land in the beds of the Wadi where the rodents breed.

These ecological control methods, which have already been tried and tested in some countries (e.g. the Soviet Union), make it possible to avoid using insecticides and pesticides on sandflies and rodents spread out over a wide area and therefore difficult to reach.

d) The use of pesticides: This method is particularly recommended in Meriones control because of the considerable damage they cause to grain crops. It is used by the Ministry of Agriculture in accordance with a procedure codified as follows: grains of wheat poisoned with zinc phosphide (1 kg of zinc phosphide, 2 litres of commercial oil for 100 kg of wheat).

Trials have been conducted with Klerat (an anticoagulant containing 0.05 grams of brodifacoum per kilogram) to get rid of the Psammomys. It is difficult to say whether it is effective since the results are contradictory and the operational costs considerable.
However, one has to ask the question "Is the use of insecticides justifiable?". Observations regarding P. papatasi (Ben-Ismail, Ben Rachid et al. 1987), the vector of ZCL, have clearly demonstrated that the use of insecticides in or around the home affects only a quarter of the females; the remaining three-quarters are found in rodent burrows. Chemical measures against the burrows are difficult to imagine given the extent of the area involved. The only feasible approach is to destroy the burrows.

The use of insecticides remains, however, possible against other forms of leishmaniasis, whose incidence has decreased significantly during the campaign against malaria (1968-72) (Ben Rachid et al. 1983). But in the absence of an epidemic and with only sporadic cases of KA, SCL and ACL, this use can only be on an individual basis and in the home, which is costly for people whose resources are generally limited.

e) Individual measures: Treatment is the only feasible option. Glucantine (meglumine antimoniate) is used to treat all forms of the leishmaniases; for KA and serious forms of cutaneous leishmaniases, treatment lasts 15-20 days involving hospitalization and for simple cutaneous leishmaniases, "minute" treatment by intralesional infiltration. Current practice is not to treat benign forms of ZCL, which usually develop rapidly and against which people can be immunized. The use of local antiseptics will prevent additional infection. The use of mosquito nets and insect repellents is often beyond the reach of people with modest incomes.

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