

American leishmaniasis is a disease of high morbidity and low mortality. Immunization procedures have proved to be completely ineffective. At the present time control appears possible only through the use of protective clothing and repellent agents to prevent contact by the vector.

AMERICAN LEISHMANIASIS

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The leishmaniasis constitute a group of infections of certain animals and man caused by flagellated protozoan parasites of the genus *Leishmania*. These include (1) kalaazar, or visceral leishmaniasis, of man and canine species, caused by *Leishmania donovani*, which is prevalent from the Mediterranean areas westward to Northern China, southward into the Sudan and East Africa, and which occurs also in Brazil and other countries of Latin America; (2) Cutaneous leishmaniasis, caused by *Leishmania tropica*, distributed from the Mediterranean through the Middle East into India; (3) American cutaneous and muco-cutaneous leishmaniasis (*L. braziliensis*), which is found only in Latin America. A fourth may be added, the so-called "lepromatous" leishmaniasis which occurs in Venezuela, Bolivia, and Peru.

Differential Diagnosis

The problem of differentiation between the species of *Leishmania* that parasitize man has occupied the attention of investigators for many years. This is especially true of the ones causing cutaneous disease. Actually, the different species are morphologically indistinguishable one from another, and present day classification is based primarily on the clinical picture and types of lesions which are produced.

Many attempts have been made to differentiate the different *Leishmania* through the use of serological methods but the results have been

conflicting. Noguchi in 1924 published results on the cultivation of *L. braziliensis*, *L. tropica* and *L. donovani* in media containing homologous and heterologous sera and concluded that the three species were serologically distinct. A year later Kligler published results which concurred with those of Noguchi. But, in subsequent years, Fonseca (1932), Chodukin (1935), Da Cunha (1942) and Mohiuddin (1952), reported that because of a common antigen they were unable to separate the various species of *Leishmania* by serological methods. Recent work by Pifano (1960) using a modified agglutination reaction; Hertig, et al. (1962) employing immuno-diffusion techniques, and Adler (1963), by culturing on semi-solid media incorporating with homologous and heterologous sera, have indicated that species differentiation is possible by immunological procedures.

At the present time, then, the organisms causing American cutaneous leishmaniasis are considered to be separate and probably distinct species from those of the old world. Three species have been proposed for the Western hemisphere parasites; *Leishmania braziliensis*, *Leishmania braziliensis pifanoi* and *Leishmania mexicana*. Aside from the fact that they are probably serologically valid species, each one is associated with a rather characteristic clinical and pathological picture and has a somewhat different geographical distribution.

Leishmania mexicana produces lesions which

are generally confined to the pinna of the ear, causing mutilation and destruction of this appendage. These lesions are referred to as "chiclero ulcers" because they occur quite commonly in laborers who collect chicle gum. Its distribution is generally limited to lower Mexico, Guatemala, and Honduras. *Leishmania braziliensis pifanoi* (Medina and Romero, 1957) is associated with a nodular infiltrative type of skin lesion which rarely or never ulcerates and superficially resembles lepromatous leprosy. Parasites are usually abundant in the lesions. The infected individual does not react to intradermal antigens made of the other species of leishmanias and responds very poorly to therapy. This type of leishmaniasis is found in Venezuela, Peru, and Bolivia. The third species, *Leishmania braziliensis*, is much more protean in its pathological manifestations, producing, in addition to a wide variety of cutaneous lesions, the very destructive and often fatal muco-cutaneous lesion referred to as Espundia, in which the soft tissue of the nose, the naso- and oropharynx and larynx are destroyed. This species often invades the superficial lymphatics draining an ulcerated lesion, and in so doing can simulate the sporotrichosis complex of a primary ulcer and associated nodular lymphangitis.

Animal Reservoirs

Epidemiologically the leishmaniasis of the western hemisphere appear to be diseases almost exclusively of rural and forest areas where they probably occur in endemic form in a variety of rodent species and are transmitted by *Phlebotomus* sandflies. Human infection is commonly acquired in association with the jungle, without the previous existence of a human source of infection. This points with considerable certainty to the forest origin of the disease, where the specific parasite can circulate without interruption from one vertebrate reservoir to another by means of the *Phlebotomus* vectors.

Over a period of many years many investigators have attempted to demonstrate a forest animal reservoir of American cutaneous leishmaniasis. Infections in dogs were first reported in Brazil by Pedroso in 1912, and sometime later Mello, also in Brazil, noted an infection in a cat. Subsequently many reports of natural infections in domestic animals appeared in the literature; Mazza in Argentina, Pifano in Venezuela, Medina and Romero in Venezuela, and Herrer in Peru reporting infections in dogs. Herrer (1945) noted a parallel between human infections and those in dogs, stating, however, that the incidence in canines was always lower than in humans. He found no infections in other domestic animals and, likewise, none in wild animals.

Until 1956, all attempts, by many investigators, to demonstrate *Leishmania* in wild animals of the western hemisphere resulted in failure. In that year Johnson, Hertig and Fairchild reported, from Panama, the first successful isolation of *Leishmania* from a wild forest host. In February of that year a *Leishmania* was recovered in culture from the blood of a spiny rat, *Proechimys semispinosus panamensis*. This strain of *Leishmania* was subsequently inoculated into a human volunteer with the production of a typical cutaneous lesion from which the parasites were demonstrated in smears, cultures and tissue sections.

In the following year (1957), the same group isolated *Leishmania* from 21 of 200 *Proechimys semispinosus* by means of blood culture. The rodents were collected from six endemic foci. In spite of intensive search of smears and section, they were unable to demonstrate parasite in any tissue site. In 1958 not one of 130 *Proechimys* was found positive, but an isolation was obtained from one of 14 *Hoplomys gymnurus*. Since then no positive cultures have been obtained from either of these two rodents, and moreover intensive search for *Leishmania* in hundreds of wild-caught animals of some 35 different species has proved fruitless.

Following on the work of the Panama group, Forattini in 1960 reported his studies on 928 wild-caught animals in Brazil. Of the total, 881 were rodent species representing nine genera and 47 were from other species of mammals. In the rodent group three isolations of *Leishmania* were obtained by blood culture, one from *Kannabateomys amblyonyx* (tree rat), a second from *Cuniculus paca* (spotted agouti), and a third from *Dasyprocta azarae* (agouti). The *Dasyprocta azarae* show an extensive lesion of the skin about the nose from which *Leishmania* were isolated. Later in the same year Alonar, et al. (1960) reported one successful isolation of *Leishmania* in blood culture from *Rattus rattus alexandrinus*. In their studies 39 wild rodents of four genera were negative. The positive isolation came from a group of 153 domestic rats of two genera.

While the search for wild animal reservoirs of leishmaniasis continued, no further successful attempts were reported until 1962 when Lainson and Strangways-Dixon in British Honduras found *Leishmania* in six of 13 *Atotylomys* sp., one of seven *Peromyscus* sp., and three of 44 *Heteromys* sp. The lesions were restricted, in all cases, to small whitish areas on the tail which were found to contain *Leishmania* in variable numbers. The organisms were inoculated into hamsters and mice, and dermal lesions identical to those produced by human strains were ob-

tained. A human volunteer was inoculated with the culture forms from a *Peromyscus* sp. and a typical lesion developed at the inoculation site. They examined 350 wild animals of other species with negative results. Unofficial reports from the Belem area of Brazil apparently confirm the findings of Lainson and Strangways-Dixon in that rodents exhibiting the same type of lesions, containing *Leishmania*, have recently been found.

From the evidence just presented it can be concluded, with some caution, that rodents can serve as animal reservoir hosts for the different species of *Leishmania* in Latin America, but whether they will eventually prove to be the true reservoir host is problematical. In none of the published reports on the finding of infected animals in the Western hemisphere has there been any mention of an association with *Phlebotomus*, the presumed vector, and no transmission studies have been carried out. These two rather fundamental problems in the epidemiology of leishmaniasis remain essentially unsolved.

Vectors and Transmission

It has been generally assumed that *Phlebotomus* sandflies are the most likely vectors of cutaneous leishmaniasis in Latin America. This is known to be the case with the old world leishmaniasis where transmission has actually been accomplished through the bite of infected flies.

The earliest investigation on the experimental transmission of leishmaniasis in the Western hemisphere was made by Aragao in 1922 during a study of an epidemic of the disease which appeared in a forested area near Rio de Janeiro. This investigator allowed wild-caught specimens of *Phlebotomus intermedius* to feed on the border of human lesions, and demonstrated the motile leptomonad stage of the parasite in the stomach of the flies three days later. These were inoculated into the skin of a dog's nose and a lesion was produced.

In 1940 Pifano found that five of 72 *Phlebotomus panamensis*, which had been caught feeding on the border of an ulcer, were infected with flagellates which he presumed were *Leishmania braziliensis*. He later encountered one *Phlebotomus longipalpis* and two *Phlebotomus migonei* naturally infected with flagellates.

Pessoa and Pestana (1940) also found natural infections of leptomonad flagellates in *Phlebotomus migonei* caught in endemic areas of the State of Sao Paulo, Brazil.

Continho and Barreto, in 1943, examined a total of 11,393 *Phlebotomus* from an endemic area in Brazil and found 26 infected with leptomonad flagellates. Three species of sandflies were represented: *P. whitmani*, 4940; *P. migonei*,

3742; *P. pessoai*, 2711. The over-all infection rate was 0.22%.

Forrattini and Dos Santos (1952 and 1954) in the course of a study in Sao Paulo found one *P. whitmani* infected with flagellates in 104 specimens and no infections in 678 *P. intermedius*.

The most careful and complete study on the natural infections of leptomonad flagellates in *Phlebotomus* was reported by Johnson, McConnell and Hertig in 1963. In their search for vectors of leishmaniasis in Panama they dissected and examined for leptomonad flagellates over 5,000 wild-caught female *Phlebotomus*. They found natural infections in 416 out of a total of 4,861 females of six of the seven man-biting species (*P. gomezi*, *P. panamensis*, *P. sanguinarius*, *P. shannoni*, *P. trapidoi* and *P. ylephiletor*). Infected sandflies were found in each of five endemic areas and in two areas in which no human cases had been reported. The over-all infection rate was 8.5%. The lowest infection rate was 1.9% in *P. panamensis*, and the highest 15.4% in *P. trapidoi*. They also reported a higher infection rate, 10.6%, in the specimens collected during the rainy season (June through December) than in those collected during the dry season, 4.1%. Although the source of the leptomonad infections was unknown, the vertical distribution of the species in simultaneous ground-level and tree platform catches, together with differential infection rates, suggested that the source may have been arboreal animals. In culture the morphology of the leptomonads was similar to *L. braziliensis*, and lesions produced in the hamster by inoculation of two of the sandfly strains were similar to those produced by Panamanian strains of *L. braziliensis* isolated from human lesions.

Actual transmission of leishmaniasis by the bite of *Phlebotomus* sandflies has been reported only twice in Latin American literature.

In 1962 Coelho and Falcao, working at the Centro de Pesquisas in Belo Horizonte, Brazil, reported two experiments in which hamsters were infected with *L. mexicana* by the bite of experimentally infected sandflies. In the first of these experiments a hamster was exposed to the bite of 27 *P. longipalpis* five days after an infective meal. Twelve or 44.4% of these sandflies were found infected and seven had organisms in the anterior cardia or the pharynx. The exposed hamster developed palpable subcutaneous nodules some five months later. In the second experiment a clean hamster was exposed to the bite of 20 *P. renei* seven days after feeding on an infected hamster. Eighteen of these sandflies or 90% were shown to be infected, and in two the flagellates were located in the anterior cardia and pharynx. The hamster developed lesions which

were demonstrable at about five months. In both hamsters parasites were recovered from the lesions.

Lainson and Strangways-Dixon (1962), in British Honduras, successfully infected one human volunteer by the bite of *P. paraensis* four days after it fed on an infected hamster. The lesion in the human volunteer developed in 17 days. Attempts to transmit the disease by *P. glephiletor* and *P. geniculatus* were unsuccessful, although infected individuals were used in the feeding experiments.

These transmission experiments, although in need of confirmation, appear to strengthen the hypothesis that the American leishmaniasis are transmitted by *Phlebotomus* sandflies.

Human Infections

It can be stated almost categorically that human infection with *Leishmania* in the Western hemisphere is acquired only in association with forests. There is one exception to this statement. In the so-called "Uta" zone of Peru forests are nonexistent. This region occupies the highland of the country at altitudes of 1,200 to 2,800 meters. It is a rocky desert type of landscape where cactus replaces the tropical vegetation of the low lands. Leishmaniasis in this area is equally distributed between inhabitants of the villages and the country population. It is primarily a cutaneous infection and only rarely are the muco-cutaneous lesions manifested.

In the remainder of Central and South America infections are seen only in individuals who, in the pursuit of their daily occupations, are required to visit or work in tropical forests: farmers, loggers and woodcutters, road building personnel, homesteaders, land development personnel, scientists, hunters and missionaries; even the occasional tourist may be included here.

American leishmaniasis is a disease of high morbidity and low mortality. The populations at greatest risk are the peasant farmers and homesteaders who by reason of their economic status will resort to home treatment or administration of "curanderas" before seeking medical aid and when finally seen by a doctor are very often temporarily crippled by their disease. Those that are fortunate (or perhaps unfortunate) enough to affect a cure of their cutaneous lesions will often develop in time (three to 15 years) the insidious destructive muco-cutaneous lesion

which produces permanent disability and may even lead to death.

It is generally accepted that the muco-cutaneous disease is probably a late manifestation of the cutaneous form caused by *L. braziliensis* and reflects improper early treatment. Clinical experience in Panama indicates that in a great majority of cutaneous cases the parasite is not confined to the visible skin lesions but invades widely along the superficial lymphatics. Adler has demonstrated that in such cases, parasites can occasionally be found in the apparently normal mucosa and cartilage of the nasal passages where they can presumably remain dormant for long periods of time (up to 15 years), and then under the influence of unknown stimuli become active and produce widespread, destructive lesions which can even extend into the upper trachea.

It is not difficult to understand the very real need for providing adequate treatment for all cases of cutaneous leishmaniasis in areas in which the muco-cutaneous complication also occurs.

Control

The basic objective in the prevention and/or control of human arthropod-borne disease is, of course, protection against the introduction of the parasite into the host. This can ordinarily be accomplished in a number of ways:

1. Use protective clothing and bed nets.
2. Make the host unattractive to the vector by use of repellent agents.
3. Destroy the insect vectors through insecticides, destruction of breeding areas, etc.
4. Destroy the animal reservoirs.
5. Immunize the human host.

In human leishmaniasis the first two methods offer the only reasonable possibilities at the present time. The difficulties of destroying the vectors or animal reservoirs are obvious and need little comment. Hertig points out that *Phlebotomus* sandflies are very susceptible to insecticides, particularly DDT, and under certain circumstances its use would be effective in the immediate vicinity of forest dwellings. Its utilization in the "Uta" zone of Peru proved to be relatively effective.

Immunization procedures have proved to be completely ineffective in the control of American leishmaniasis.