Natural History, Clinical Evolution, and the Host-Parasite Interaction in New World Cutaneous Leishmaniasis

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In the New World, human diseases caused by the genus *Leishmania* are widespread, ranging from southern Texas to Northern Argentina and the Caribbean Islands. This review will address the natural history of the *Leishmania* that primarily affect skin and upper respiratory mucosa of humans. This group of diseases will be referred to as American cutaneous leishmaniasis (ACL).

ACL is produced by a group of genetically related species, each of which has characteristic manifestations and areas of endemnicity (Table 1)\(^1\); however, none of the clinical manifestations is unique to a particular species, because there is considerable overlap between clinical spectrums. Likewise, in a given locale several *Leishmania* species may be transmitted.\(^5\) All the listed species can cause simple cutaneous lesions. Mucosal lesions are mostly characteristic of the *Leishmania* subgenus *Viannia*, particularly *L. (V.) braziliensis* and *L. (V.) panamensis*, but have also been caused by *L. (V.) guyanensis*\(^3\) and *L. (L.) amazonensis*.\(^6\) Diffuse cutaneous leishmaniasis (DCL) is a rare form of leishmaniasis caused by *L. (L.) mexicana* in the United States, Mexican, Central American, and Caribbean regions and by *L. (L.) amazonensis* and *L. (L.) venezuelensis* in South America.\(^7,8\) These species more frequently cause simple cutaneous leishmaniasis.\(^9,10\) *L. (L.) mexicana* is also associated with chronic lesions of the external ear (chiclero's ulcer)\(^11\) in the Yucatan peninsula of Mexico.

Despite these well recognized associations between *Leishmania* subspecies and clinical manifestations, anecdotal reports indicate that *Leishmania* species occasionally produce clinical manifestations beyond their recognized spectrum.\(^12-14\) The reasons why a given *Leishmania* will produce differing manifestations in a different host or setting is not completely understood.

Proposed parasite and host determinants of the clinical diversity of New World *Leishmania* are addressed later in this review.

The number of cases of ACL has been estimated to be 59,300 yearly. Fifty-nine million persons reside in areas where the ACL-causing *Leishmania* species are transmitted.\(^15\) In the United States *L. (L.) mexicana* is transmitted only in southern Texas near the border with Mexico. To date, only nine cases have been reported from this area, including one case of DCL.\(^16\) Consequently, nearly all ACL patients that present to physicians practicing in the United States will have acquired their lesions while traveling outside the United States.\(^14,17\) Travelers who have acquired ACL are usually not typical tourists or business travelers. They invariably have had contact with forested and/or rural areas either during activities such as field biological studies, community service, military excursions, or ecotourism. In a series of 59 cases of ACL in U.S. travelers, the diagnosis and treatment were frequently delayed, in part due to physicians' lack of knowledge about ACL.\(^17\) It appears that more physicians, particularly dermatologists and those working in travelers' clinics, need to become familiar with the spectrum of clinical manifestations and course of ACL.

During the past decade we have conducted a series of investigations of *L. (Viannia)* infection in Colombia to describe the natural history of ACL and its clinical manifestations and examine host and parasitic determinants of variations in natural history of these *Leishmania* species. Consequently, this article will first describe in detail clinical evolution of lesions caused by *L. (Viannia)*. We will describe more concisely the cutaneous manifestations of other New World *Leishmania* species. Then the current understanding of the pathogenesis of ACL will be reviewed, including mechanisms of dissemination, and host and parasite determinants of disease expression.

Clinical Relevance of the Natural History of ACL

The clinical course of ACL is complex due to a variable incubation period, subclinical infection, spontaneous
healing, metastatic spread, latency, reactivation, reinfection, and chronic lesions. Members of the \textit{L. (Viannia)} subgenus have a particularly intriguing, highly variable, natural history.\textsuperscript{18} One aspect of the natural history of ACL that is relevant to diagnosis and management is the capacity of \textit{Leishmania} to persist in humans. It has been suggested that \textit{Leishmania} infections persist for life.\textsuperscript{19,20} Therefore, physicians must recognize that ACL lesions may present at any time after infection, including many years after leaving an area of endemic transmission\textsuperscript{21} and may recur. Reactivation of latent \textit{Leishmania} occurs in immunocompetent patients but may be precipitated by an immunosuppressive event, such as administration of drugs and HIV infection.\textsuperscript{22,23} These possibilities should be taken into consideration when evaluating chronic mucosal and skin lesions in immunosuppressed patients. The possibility of recurrence should also be considered in describing prognosis of ACL to patients and planning follow-up care.

The concept that the natural history of ACL varies by \textit{Leishmania} species is pertinent to patient management. In some settings where resources are limited, cost benefit considerations have led to a policy of treating the more severe lesions.\textsuperscript{24} Since the spontaneous healing rate of lesions caused by \textit{L. (L.) mexicana} is high, at least 68%,\textsuperscript{10} and current drug therapy is costly, potentially toxic, and requires parental administration, it may be appropriate to only treat with antimonials when lesions have not healed spontaneously. Furthermore, antimonial therapy appears to modify the clinical course of lesions due to \textit{L. (V.) braziliensis} but not those caused by \textit{L. (L.) mexicana}, so the value of treating the latter with antimonials is questionable.\textsuperscript{25} Unfortunately, only the randomized clinical trials of ACL conducted in Guatemala by Navin and colleagues have been able to evaluate \textit{Leishmania} species as a modifier of treatment efficacy.\textsuperscript{10} Further controlled clinical trials should examine whether the efficacy of drug therapy varies by \textit{Leishmania} species.

### Overview of the Natural History of ACL

Despite the importance of the natural history of ACL to its clinical management, investigations describing natural history are limited, primarily because the necessary prospective studies require following mobile, geographically distant and widely dispersed patients during several years. Also, due to the variation of natural history by species, numerous prospective studies are needed to describe the clinical evolution of each species. Consequently, the current picture of clinical evolution is derived from piecing together information on the three major stages of \textit{Leishmania} infection: infection to initial manifestations; initial lesions to healing; healed lesions to recurrence.

The spectrum of natural history of an infectious agent can be characterized rather simply by three terms: infectivity, pathogenicity, and virulence (Fig 1). Infectivity describes the relative ease with which contact with a microbe results in infection. Pathogenicity describes the degree to which an infection leads to clinical manifestations, rather than asymptomatic infections. Pathogenicity can be estimated by measuring the number of persons who develop lesions among those who are newly infected. Among the agents causing ACL, the pathogenicity appears to vary by species and locale. Virulence describes the degree to which a clinically manifest infection progresses to more severe or prolonged manifestations (Fig 2). A description of the virulence of the parasites causing ACL should include the frequency of very large lesions, multiple lesions, chronic lesions, metastasis, and subsequent reactivation. Because different mechanisms may contribute to pathogenicity and virulence, their distinction will fur-

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Table 1. Geographical Distribution and Clinical Manifestation of \textit{Leishmania} Species Most Commonly Associated with American Cutaneous Leishmaniasis

<table>
<thead>
<tr>
<th>Parasite</th>
<th>Clinical Manifestation</th>
<th>Country</th>
</tr>
</thead>
<tbody>
<tr>
<td>\textit{L. (V.) braziliensis}</td>
<td>Cutaneous, Mucocutaneous</td>
<td>Argentina, Belize, Bolivia, Brazil, Colombia, Costa Rica, French Guiana, Guatemala, Honduras, Mexico, Panama, Paraguay, Peru, Venezuela</td>
</tr>
<tr>
<td>\textit{L. (V.) panamensis}</td>
<td>Cutaneous, Mucocutaneous</td>
<td>Colombia, Costa Rica, Ecuador, Honduras, Nicaragua, Panama, Venezuela</td>
</tr>
<tr>
<td>\textit{L. (V.) guyanensis}</td>
<td>Cutaneous, Mucocutaneous (rare)</td>
<td>Brazil, Colombia, French Guiana, Guyana, Peru, Surinam</td>
</tr>
<tr>
<td>\textit{L. (L.) peruviana}</td>
<td>Cutaneous</td>
<td>Peru</td>
</tr>
<tr>
<td>\textit{L. (L.) mexicana}</td>
<td>Cutaneous, Diffuse cutaneous leishmaniasis</td>
<td>Belize, Colombia, Costa Rica, Dominican Republic, Guatemala, Honduras, Panama, United States (Texas)</td>
</tr>
<tr>
<td>\textit{L. (L.) amazonensis}</td>
<td>Cutaneous</td>
<td>Bolivian, Brazil, Colombia, Ecuador, French Guiana, Panama, Peru, Venezuela</td>
</tr>
</tbody>
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\textsuperscript{Adapted from Escobar et al.}
Figure 2. Estimation of pathogenicity and virulence based on the relative numbers of clinical apparent cases and mild, moderate, severe, and fatal cases.

Figure 1. Conceptual model, natural history of Leishmania (Viannia) infection in residents of endemic area. Each box represents potential stage in clinical progression; transitions are indicated by solid vertical line (steams) and are identified by encircled numbers. Nodes (●) indicate where branches may lead to similar outcomes. Arrows pointing to encircled numbers indicate that clinical course is repeated at beginning at numbered transition (from Wiegle et al.,4 Journal Infectious Diseases, with permission).

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ther our description of the complex natural history of ACL. For example, L. peruviana, the agent of Uta in the Peruvian Andean region, appears to be highly pathogenic but of relatively low virulence.26 In contrast, L. (Viannia) infections in rural Tumaco, Colombia, are relatively less pathogenic, but more virulent.18,27

We have found that a conceptual model of the natural history of L. (Viannia) infection furthers our ability to examine the details of acquisition and progression of this infection. In the first step, 1, a naive host, lacking history of contact, infection, or sensitization must come into contact with the Leishmania parasite, usually by contact with an infective sandfly vector. This contact may or may not lead to an infection, 2, replication of the parasite in human cells. Once the infection is established in macrophages and tissue histiocytes it may progress to a clinically manifest lesion or remain asymptomatic, 3. The course of primary lesions, 4, ranges from rapid, spontaneous healing to chronic lesions. Given time, even chronic lesions may heal. 5, however the healing of chronic lesions is accelerated by therapy, such as antimonials. Eventually, a quiescent phase is reached, 6, through varying pathways. In this quiescent phase all lesions are healed, defined as complete re-epithelization and the absence of signs of inflammation in all lesions. However, persons in the quiescent phase vary in their potential for subsequent ACL lesions. At this point, two factors will determine whether a person in the quiescent phase has the potential to develop subsequent lesions: (1) whether persistent, latent Leishmania remain in intracellular sanctuaries and (2) whether the host has acquired a protective immune response that will resist subsequent contacts with Leishmania. The four possible combinations of these factors are represented by the four boxes below step 6 as shown in Figure 1. Only those who have both rid their body of Leishmania and acquired a protective immune response are certain to remain free of ACL, indicated by the furthest left box in the model: “cured” parasite-free and immune. Those persons whose acquired immune response to Leishmania is not protective will remain susceptible to exogenous Leishmania infection. Depending on their exposure to this organism they may become exogenously reinfected, 2. Quiescent hosts who harbor latent parasites may progress over time in three ways, 7. Some may eventually eliminate the latent Leishmania and become “cured and parasite-free.” The infection may reactivate in others, leading once again to clinically apparent lesions, 4. Others may maintain a persistent asymptomatic infection throughout their life. The clinical evolution from the stage of chronic infections, 5, is actually more complex and dynamic than depicted in the model. While the initial lesion is healing, satellite lesions and/or nearby subcutaneous nodules may develop. These nodules may enlarge and necrose centrally, becoming new ulcers. Likewise as the central
lesion heals the satellite lesions often enlarge. Less often new lesions develop distant from the initial lesion. Over time these cycles may repeat. During the same time interval some lesions may repeatedly heal and reactivate while other lesions remain continuously active or continuously healed. This dynamic situation is consistent with the concept that a very localized immune response, which is not capable of controlling *Leishmania* replication in other skin sites, lymphatics, or blood macrophages, may contribute to the healing of skin lesions.

**Clinical Presentation and Evolution of ACL**

**Brazilienis Complex (Viannia) Subgenus**

**Primary Infection**

Humans usually acquire *Leishmania* infection when female sandflies take blood meals required for sandfly reproduction. During the sandfly bite, infective stage, "metacyclic" promastigotes are preferentially released and injected along with a potent vasodilator. 

*L. (Viannia)* infection can also be acquired by accidental injection of laboratory-grown *Leishmania*. The parasites causing visceral leishmaniasis (VL) can be transmitted parentally and vertically. Since parasitemia occurs less often in ACL caused by *L. (Viannia)* than in VL, parental and vertical transmission of *L. (Viannia)* are biologically plausible, but probably rare events.

After promastigotes are injected, specific receptors on tissue histiocytes permit their entry into these cells. Intracellularly, promastigotes transform into amastigotes, become rounded, and lose their flagella. Amastigotes replicate in the parasitophorous vacuole of the macrophage by binary fission. The sandfly bite produces only a small red macule which often goes unnoticed and disappears rapidly. During the incubation period, which lasts from 2 weeks to many years, a series of cellular events occur, culminating in the first cutaneous manifestation, a papule or small nodule which may itch but is usually painless.

This nodule is produced by a dermal mass containing *Leishmania*-laden vacuolated macrophages and a lymphocytic infiltrate. Multiple nearby simultaneous infective sandfly bites produce lesions of similar size and duration on the same extremity.

**Histopathology**

Early descriptions of the histopathology of ACL classified lesions along a spectrum similar to that developed for leprosy, ranging from the "lepromatous" extreme with numerous amastigotes to the "tuberculoid" extreme characterized by organized granulomas and sparse amastigotes. This classification system has not been reproduced in studies of *L. (Viannia)* models because the same lesions often contain elements of both granulocytic necrosis and macrophages, only some of which are organized into granulomas. The epidermis undergoes an intense hyperplasia in the areas that are not ulcerated and denuded of the upper epithelium. The most consistent histological association is that lesions of longer duration have fewer amastigotes. However, in a study of 221 lesions caused by *L. (Viannia)*, the majority of lesions contained abundant lymphocytes (92%), abundant histiocytes (92%), and granulomas (78%). Fewer lesions contained amastigotes (40%), eosinophils (8%), giant cells (22%), epithelioid cells (16%), areas of necrosis (34%). Persons who had scars typical of an earlier *Leishmania* lesion had a greater frequency of a "healing" pattern of histology, that is, fewer amastigotes and a greater frequency of giant cells and epithelioid cells than did those who lacked such scars (Table 2).

![Figure 3. Progression of an acute lesion caused by *L. (Viannia)*. (1) Early nodule, (2) expanded nodule, (3) ulcerated nodule, and (4) characteristic ulcer with a raised, indurated border.](image)

| Table 2. Comparison of the Presence of Amastigotes in the Biopsies of Active Dermal Leishmaniasis in Patients With and Without Scars Indicative of Prior Leishmaniasis |
|---------------------------------|--------|--------|--------|
| **Amastigotes in Biopsy**       | **No** | **Yes**|
| **Previous Lesion (Typical Scar)** | **n/N** | **%** | **n/N** | **%** |
| Absent                          | 68/129 | 35     | 18/26  | 69*    |
| Present                         | 124/129| 65     | 8/26   | 31     |

* p < 0.001. Chi squared test. 
Adapted from Gutierrez et al. 16
ongoing parasite spread and primary infection (more frequent amastigotes, less frequent giant cells and epithelioid cells) than did those without adenopathy.

In lesions that have healed, complete re-epithelialization occurs, accompanied by fibrosis and a marked reduction in inflammatory cell infiltrates. Relative to active lesions, necroses and epithelial changes are rare. In healed lesions the remaining infiltrates consist of perivascular lymphocytes, and occasional organized granulomas. However, re-epithelialization and a "healed" histological pattern are not a guarantee of elimination of Leishmania.37,45

Typical Scars
Classical lesions of L. (Viannia) usually produce scars that are somewhat unique. The scar is often smoother and therefore more reflective than the surrounding skin. Its surface is slightly depressed relative to the level of the surrounding skin. Pigment changes are often observed. Although skin scars of any type were very common in Tumaco, a rural area of Colombia in which L. (Viannia) is endemic, only typical scars were strongly associated with a positive leishmanin skin test. Therefore, the presence of a typical scar may be useful in the clinical diagnosis of past ACL, especially in persons who have long resided in an endemic area.

We found that among persons who were leishmanin skin test positive, the presence of a typical scar was a strong predictor of subsequent new active lesions during the next 3 years. Among persons who were skin test positive and had no active lesion when first examined, 0.2% of those who lacked a typical scar developed new lesions per year, whereas 2.3% of those who had typical scar developed a new lesion each year, an elevenfold increase. Very few of the persons with scars had received antimonial treatment for their initial lesions. Evidently, something about L. (Viannia) infection that produced lesions severe enough to leave a lasting scar also increased the risk of a clinically apparent recurrence. One explanation may be that the nature of the host's immune response determines the susceptibility to clinically apparent infections in this setting where the circulating parasites usually produce asymptomatic infections. This variation in host susceptibility may not be observed in a setting where the circulating Leishmania is highly pathogenic. The low pathogenicity of the L. (Viannia) transmitted in the Tumaco area has allowed us to examine host variations in susceptibility, as discussed later.

Spontaneous Healing
Although it has long been recognized that some lesions caused by Leishmania heal spontaneously, the frequency and early time course of healing was described recently by following the placebo arms of three randomized clinical trials in Panama, Ecuador, and Guatemala. The rate of spontaneous healing was highest in Ecuador, where 75% of patients healed in 6 weeks and in Guatemala among patients with lesions caused by L. (L.) mexicana, where 68% healed in 14 weeks. However, only 8% of persons with L. (V.) braziliensis lesions in Guatemala healed in 14 weeks and none of the 11 persons with L. (V.) panamensis healed during the 6 weeks of follow-up in Panama. This wide variation in spontaneous healing rates indicates that the natural history of ACL varies by species and geographic location and that the apparent efficacy of drug treatment should be judged relative to the course of disease in untreated patients.

Chronic Cutaneous Lesions
Patients with chronic lesions have an increased morbidity not only because of the length of their illness but because chronic lesions tend to be larger and more diverse in their clinical manifestations. Because chronic lesions may no longer possess the fairly unique clinical picture of acute lesions, they are easily confused with other chronic skin diseases, explaining why terms such as psoriasiform, eczematous, varicelliform, verrucous, keloidal, chromomycoid, and carcinomatike were coined to describe the atypical manifestations of chronic ACL. Therefore, leishmaniasis should be considered in the differential diagnosis of any chronic skin lesions in patients who have visited or resided in endemic areas. Unfortunately, conventional diagnostic methods for ACL, such as microscopy, culture, and histopathology, are less sensitive in chronic lesions due to the low density of amastigotes. During the last decade we have diagnosed leishmaniasis parasitologically in numerous patients who had been diagnosed and treated for other diseases. In general these patients had chronic and atypical forms of ACL. On the other hand, we have established a different diagnoses in many patients who were referred to us for evaluation of leishmaniasis. With the help of dermatologists and pathologists (Table 3), during a 3-year period 17 diagnoses other than leishmaniasis were established in 72 such cases. Of these, the
most frequent were sporotrichosis and bacterial skin infections.51

Leishmaniasis recidiva cutis (LRC), also known as lupoid leishmaniasis and leishmaniasis recidivans, is a form of chronic cutaneous leishmaniasis associated with \textit{L. tropic} infection in the Old World, that has rarely been reported in the New World.49,52 It contains inflammatory papules and nodules on the periphery or inside of a classical cutaneous leishmaniasis lesion. They may appear before the classical ulcer has healed or after complete re-epithelization has occurred. These patients have vigorous cellular immune responses but low antibody titers. The histopathology is one of a well-defined granuloma, with few amastigotes and no necrosis. Although we have not systematically looked for cases of LRC, it is our impression that it is relatively frequent among our cases of chronic and recurrent ACL, and is probably more common than reported in the New World.

Chronic cutaneous lesions are most common in settings where access to antimonial therapy has been limited or in reference centers to which persons with chronic lesions are willing to travel. During our first year of case detection for ACL in rural Tumaco, 46\% of cases had chronic cutaneous lesions, defined as lesions of 6 months or longer duration.27 Now, 12 years later, chronic cases are very rare in this setting. Obviously, the chronic lesions that were prevalent when the case detection began had been accumulating over many decades. Once chronic cases were treated, new cases emerged either from new infections or reactivation of latent infections.45 We have found that lesions caused by \textit{L. (V.) braziliensis} are more frequently chronic than lesions caused by \textit{L. (V.) panamensis} (Fig 4).

Recurrence of ACL

The term recurrence of ACL describes the onset of active lesions after a time of quiescence, that is, after an interval in which all previous lesions have re-epithelized. The frequency of recurrences among persons with some evidence of previous ACL in three population-based studies in Brazil, Colombia, and Peru were 2.7\% of 369 cases, 2.0 per 100 person years, and 2.9 per 100 person years, respectively.18,26,53 but may have been influenced by whether the earlier lesions had been treated and the methods of surveillance. Recurrences may be due to reactivation of latent parasites at the same skin site as earlier lesions, or at other metastatic locations, or due to exogenous reinfections acquired from a new exposure to infected sandflies. Reactivations occur most often at or near the same locations of an earlier lesion and usually occur within a year after healing of the initial lesion.26,45,53 The frequency of reactivation appears to be determined by the species of \textit{Leishmania}, the treatment received for earlier lesions, and the immunological competence of the host. Reactivation of healed lesions due to \textit{L. (Viannia)} appear to be less frequent in those treated with 20 mg/kg/d or more of antimonium for 20 days.38,54 In a total of 59 patients whose acute lesions healed after they received this dose intravenously in the form of sodium stibogluconate (Pentostam®), only one relapsed during 3 to 12 months of followup.25,35,55 These patients were all soldiers who were not at risk for reinfection, so the one relapse was

\textbf{Figure 4.} Comparison of \textit{L. (V.) braziliensis} and \textit{L. (V.) panamensis} with respect to the reported duration of cutaneous lesions at time of diagnosis. For each species the percent of patients who report each duration is depicted in a histogram, as are the mean and median duration in months (from Saravia et al., 1996, Journal of Infectious Diseases, with permission).
thought to be due to a reactivation. Hopefully this low reactivation rate will be reproduced in patients with more long-standing lesions and with the more widely available antimonial, meglumine antimoniate (Glucantime®), which is administered intramuscularly.

Between 1983 and 1990 we followed 498 parasitologically diagnosed patients with ACL due to L. (Viannia) for up to 4 years to understand the pattern and determinants of recurrences following what was at that time a standard treatment regimen of Glucantime. By comparing the initial Leishmania isolate with the isolate from recurrent lesions from 24 patients in terms of enzyme phenotype and genotype, we determined that half of these recurrences were likely to be due to new infections. As would be expected, the 12 lesions that were considered to be due to reinfections occurred longer after the initial lesions and more often distant from the initial lesions than was the case for the lesions due to reactivations. During the entire 4 years of follow-up, 16% of the cases recurred (Fig 5). The highest rate of recurrence was in the first year following healing of the earlier lesion, after which time the rate of recurrence was relatively constant.

Lymphatic Spread

Clinically apparent spread of L. (Viannia) can involve two components of the lymphatic system: the localized lymphatic chain and the regional lymph nodes draining the area of the skin lesions. A well-recognized entity of nodular lymphangitis also known as sporotrichoid leishmaniasis, Pian Bois and Bush Yaws, involves the local lymphatic channels proximal to a skin lesion in an extremity. Most classically a lesion of the hand or forearm is followed by a streak of erythema and a cord of small beadlike nodules marching up the long axis of the arm. Leishmania can be cultured from these nodules, proof that this lymphangitis is due to Leishmania rather than a bacterial superinfection. The nodular lymphangitis of ACL is clinically very similar to that caused by Sporothrix schenckii, both of which produce painless ulcers and nodules. Other common causes of nodular lymphangitis, including Nocardia brasiliensis, Mycobacterium marium, Mycobacterium chelonae, and Francisella tularensis, are usually more tender but should be considered in the differential diagnosis of sporotrichoid leishmaniasis.

The high frequency and relevance of regional lymphadenopathy in ACL has been only recently recognized. This bubonic form of leishmaniasis appears to be more common than nodular lymphangitis and may comprise 77% of cases of L. (V.) braziliensis in some settings. Both nodular lymphangitis and regional lymphadenopathy may appear together. In hamster models of L. (Viannia) infection, spread to regional lymphatics occurred as early as 5 days after inoculation, preceding the clinical appearance of lesions at the site of inoculation. In these models Leishmania spread to the spleen, bone marrow, and liver as early as 30 days after inoculation, and was recovered from these organs as late as 9 months postinfection. Similarly, in humans with L. (V.) braziliensis infection, lymphadenopathy can precede the presentation of cutaneous lesions. In a recent study of 169 cases of ACL with lymphadenopathy, the lymphadenopathy preceded the onset of skin lesions in 69.2% of the cases by an average of 2 weeks. Adenopathy was associated with a less frequent history of past ACL, consistent with the concept that spread to lymph nodes usually occurs early in the course of a primary infection, analogous to the pathogenesis of primary pulmonary tuberculosis. Patients with adenopathy had more frequent fever, hepatomegaly and splenomegaly, suggesting that the hamster model of L. (Viannia) metastasis may replicate this spectrum of the clinical evolution in humans.

Involvement of the lymphatic system by L. (Viannia) has several implications for clinical management. When adenopathy accompanies skin lesions, the lymph node may be a preferable site for culture of Leishmania. Both nodular lymphangitis and adenopathy will regress as the skin lesion is treated with antimonials and do not require excision or drainage of the involved nodes. When lymphadenopathy precedes the onset of skin lesions, leishmaniasis must be considered in the differential diagnoses of adenopathy along with tuberculosis, mycosis, toxoplasmosis, cat scratch disease, plague, and lymphoma. If Leishmania is established as the cause of the adenopathy, then antimonial therapy appears to be indicated since most patients who present with only adenopathy will develop skin lesions if not treated.

Although the sporotrichoid form of leishmaniasis has been classically associated with L. (V.) viannensis, it
has been observed in other members of the L. (Viannia) subgenus. Adenopathy associated with ACT in Brazil was caused by L. (V.) braziliensis. In Colombia we have found that lymphadenopathy occurs with a similar frequency in patients with lesions due to L. (V.) braziliensis (30 out of 75, or 40%), L. (V.) panamensis (144 out of 360, or 40%), and L. (V.) guyanensis (5 out of 14, or 36%).

Hematogenous Spread and Mucosal Lesions
Most patients with ACL due to L. (Viannia) present with single cutaneous lesions. Among those who present with multiple lesions, the secondary lesions are usually on the same extremity as the primary lesion. However, secondary lesions at distant body sites occur in about 33% of those with multiple lesions or about 8% of patients overall, some of which are probably due to dissemination and others to multiple bites. In unusual reports of disseminated cutaneous leishmaniasis, with 20 or more lesions due to L. (Viannia), hematogenous spread is assumed. Cases who develop lesions at distant sites from their healed primary lesions after leaving the endemic area provide indisputable evidence for both latency and hematogenous metastasis. Furthermore, L. (Viannia) has been recovered from the blood of patients with lesions due to L. (V.) braziliensis. In Colombia we have suggested that facial lesions may occasionally spread to the mucosa directly and through the lymphatics.

Mexicana Complex, L. (Leishmania) Subgenus
Diffuse cutaneous leishmaniasis (DCL) is a rare form of ACL caused by members of the Mexicana complex, which presents initially as a macule, papule, or nodule, usually on the face or extremities, then progresses over many years to numerous nodules and plaques which may coalesce, especially over the face and ears to produce the classical “leonine” faces. The lesions, which are painless and usually nonulcerating, contain large numbers of amastigotes within vacuolated macrophages, but a limited lymphocytic infiltrate. This lack of inflammatory reaction is consistent with these patients’ lack of Leishmania-specific cellular immune response. Other characteristic features of DCL include absence of visceral involvement, a poor response to antimonalies, frequent relapses, and widespread dissemination with rare involvement of the anterior nares. DCL has been most frequently reported from Brazil, Venezuela, Mexico, and the Dominican Republic.

L. (L.) mexicana usually produces cutaneous ulcers that are smaller and have more abundant amastigotes as compared to lesions caused by L. (V.) braziliensis, and often heal spontaneously. Relative to L. (V.) braziliensis, this species more often produces lesions on the face and ears, including characteristic chronic lesions of the ear and ear cartilage, called “chicleros ulcer.” Cutaneous lesions due to this species have been reported from the United States, Mexico, Belize, and Guatemala.

Lesions caused by L. (L.) amazonensis usually present as a single ulcerated lesion that is very responsive to treatment and may heal spontaneously. However, the diversity of clinical presentation of this species appears to vary by setting, perhaps due to differences in the virulence of the local variants, or referral patterns. In 62 cases reported from Amazonian Brazil, nearly all had relatively recent, single lesions with no dissemination to other skin sites, mucosa, or viscera. In contrast, a more diverse spectrum was observed among 40 cases from Bahia, Brazil, including 11 cases of visceral leishmaniasis, 4 cases of postkalazar dermal leishmaniasis (PKDL), 5 cases of ML, and 1 case of DCL. Among the 19 cases with cutaneous ulcers, 5 were considered to have disseminated due to the presence of 5 or more lesions. Similar to L. (Viannia), L. (L.) amazonensis may disseminate hematogenously despite evidence of a Leishmania-specific cellular immune response. Such disseminated cases have a clinical presentation and natural history that is distinct from that of anergic DCL, a much rarer disease.
Dermal Lesions Caused by Viscerotropic Leishmania

Leishmania that cause visceral leishmaniasis in the New World, L. chagasi/infantum, can also produce cutaneous disease. Cutaneous lesions caused by L. chagasi are usually nodular, but occasionally ulcerate. Lesions caused by L. chagasi are not easily distinguished from lesions caused by the more prevalent dermatotrophic Leishmania. Co-infection with HIV can result in unusual cutaneous presentations of L. infantum/chagasi infection. Exposure to endemic areas of transmission of L. infantum/chagasi should raise suspicion that cutaneous lesions may be leishmaniasis. The proportion of infections that result in visceral or cutaneous disease is unknown, but does vary from one geographic region to another. For example, L. chagasi infection has only been associated with cutaneous disease in Costa Rica, while other countries in Central America report both visceral leishmaniasis and cutaneous lesions caused by L. chagasi. In Brazil, visceral disease appears to be the predominant outcome of infection with this parasite. Because the point of entry of infection is the skin, it is possible that cutaneous manifestations occur frequently but are of minimal consequence and resolve quickly, and thereby go undetected by the health system and the patient. In areas where Leishmania that normally cause ACL are concomitantly transmitted, the distinction of etiologic agents may not be routinely ascertained and result in an underestimate of the cutaneous pathology attributable to L. chagasi.

Relevance of Natural History of ACL to Its Control

Understanding the natural history of infections caused by the members of the L. (Viannia), braziliensis-complex and L. (Leishmania), mexicana-complex has implications for control programs for residents of endemic areas and travelers to endemic areas. Persons traveling to Leishmania-endemic areas should be advised to use personal protective measures, such as clothing and use of DEET, bed nets, and avoidance of outdoor activities during the time of maximal sandfly biting. Travelers should be educated to recognize lesions that may be due to Leishmania and to seek prompt diagnostic and treatment services.

Control of ACL in residents of areas with ongoing Leishmania transmission is more challenging and must be tailored to the local ecology of the zoonotic cycle, epidemiology, and natural history of the local dermatotropic species and available health resources. For instance, in settings where ACL morbidity is high due to prevalent lesions and reactions rather than newly acquired infections, diagnosis and treatment services may be more appropriate than control measures aimed at preventing new infections. The four key control strategies of vaccines, vector control, diagnoses/surveillance, and treatment of lesions have made encouraging progress. Although prevention of all Leishmania infections by vaccines or vector control is an appealing goal, it may not be feasible or appropriate for all areas of Leishmania transmission. Likewise, the benefit of detecting and treating all cases should consider the likelihood of the lesions progressing to the more severe forms of ACL, that is ML, DCL, and RCL. Therefore, further studies of the natural history of ACL are needed to make informed policy decisions regarding the application of existing control measures and those forthcoming. This is especially important with respect to areas and species in which little is known about the frequency and time course of progression from infection to the various disease forms.

Host-Parasite Interaction and the Outcome of Infection

Dissemination and Pathogenesis of Cutaneous Leishmaniasis

Dissemination of parasites from the site of inoculation of infective promastigotes is effected by mononuclear phagocytic cells. The first step in the process of dissemination involves phagocytic Langerhans cells in the epidermis, which migrate to the dermis and then to draining lymph nodes in response to proinflammatory cytokines. Early in experimental Leishmania infection, 24–96 hours postinoculation, Langerhans cells migrating from the epidermis are the only cells with demonstrable Leishmania in draining regional lymph nodes. Uptake of parasites evidently occurs in the dermis during the transit of Langerhans cells to the lymph nodes. These migratory antigen-presenting cells, besides being a vehicle of Leishmania dissemination, are potent activators of lymphocytes in draining lymph nodes. Later in infection, macrophages located in lymph nodes are also found to express parasite antigens and presumably harbor amastigotes.

Macrophages and blood monocytes can disseminate parasites through the vascular circulatory system and its reticulendothelial compartments. Leishmania have been observed in peripheral blood and cultured from peripheral blood spleen and bone marrow of patients with different forms of leishmaniasis caused by species normally associated with cutaneous disease. Although more frequently associated with visceral leishmaniasis, parasitemia has been further demonstrated in cutaneous leishmaniasis by molecular hybridization and amplification of the ribosomal DNA repeat unit of L. braziliensis in peripheral blood leukocytes.

Secondary lesions, whether contemporaneous with primary lesions, have provided insight into the mechanisms of metastatic disease. Mucosal disease and secondary lesions outside of the route of lymphatic drainage from the primary lesion support hematoge-
The immune response to Leishmania is a major determinant of the outcome of infection. Severe disease forms are instructive in this regard. The refractory diffuse form of cutaneous leishmaniasis, which in the New World is essentially restricted to the mexicana complex, is the result of induced antigen-specific immune paralysis. Successful treatment is accompanied by the recovery of cell-mediated responsiveness to Leishmania. Experimental human infections conducted by Convit et al. in Venezuela established the host response as the pivotal determinant of disease expression in infections with Leishmania of the mexicana species complex. Inoculation of volunteers with organisms isolated from the typically nonulcerating cutaneous lesions of DCL patients, who are characteristically anergic to intradermal challenge with leishmanin, invariably resulted in self-limiting, simple cutaneous lesions and skin test conversion. In vitro analyses of the immune response to Leishmania antigens has subsequently documented antigen-specific suppression of lymphocyte responses by monocytes from patients with DCL. These experiments and the rare sporadic occurrence of diffuse leishmaniasis among more frequent cases of simple cutaneous leishmaniasis highlight the contribution of the human host to the outcome of infection with organisms of the mexicana complex. Chronic cutaneous and mucosal disease caused by Leishmania of the Viannia subgenus are characterized by pronounced hypersensitivity to Leishmania antigens at the tissue and systemic levels. In contrast with the antigen-specific anergy in nonhealing DCL, clinical observations of patients infected by L. (Viannia) support an association between antigen-specific hypersensitivity and chronic, tissue-destructive disease presentations. Hence, nonhealing disease can result from either antigen-specific anergy or hyper-reactivity.

More recently, cutaneous hypersensitivity to leishmanin and previous, scarring cutaneous leishmaniasis were found to be determinants of new clinically apparent disease episodes among endemically exposed individuals. Chronic lesions may, therefore, provide a marker for susceptibility to disease in humans analogous to the nonhealing BALB/c/L. major model of cutaneous leishmaniasis.

Prospective population-based epidemiological investigations have revealed information on the asymptomatic and mild disease outcomes of infection that are not detected by passive case detection (Fig 6). More severe and nonresolving disease presentation generally constitute a larger proportion of passively detected cases. One important finding has been the recognition of the occurrence of subclinical infections in endemic areas. Differences in host susceptibility are likely to underlie the occurrence and frequency of subclinical infection as well as the spectrum of disease severity. The bases of natural host susceptibility are not fully understood. However, the target cell of infection, the mononuclear phagocyte, is a critical determinant of the course of in-
Infection. Recent in vitro studies of macrophages differentiated from peripheral blood monocytes from individuals who had either experienced subclinical infection or chronic disease due to L. panamensis, showed the cells from the latter to be more susceptible to infection by promastigotes as well as to support the intracellular survival of amastigotes. Consistent with these observations, messenger RNA of proinflammatory cytokines that down-regulate microbicidal functions of macrophages is increased in chronic lesions caused by L. mexicana. As suggested by experimental models of cutaneous leishmaniasis, the local expression of these and other cytokines may influence both the course of infection and disease in the human host.

Immunosuppression Alters the Spectrum of Infection and Disease

Further evidence of the host contribution to the outcome of infection derives from the unusual and frequently devastating and refractory disease presentations associated with immunosuppression. Reactivation of prior infection and exacerbation of disease occur in HIV-positive individuals. Other illnesses and immunosuppressive conditions such as diabetes, malnutrition, tuberculosis, radiation therapy, and transplantation-related immunosuppression have also been associated with disease progression.

Genetic Bases for Distinct Host Response

The response to Leishmania infection in naturally exposed human populations is heterogeneous. Whether the differences in response phenotype are genetically determined has not been established. However, genetic analyses of the outcome of Leishmania infection in inbred and congenic strains of mice have provided evidence that susceptibility and resistance phenotypes are indeed associated with genotype and that hemopoietic cells transfer the phenotype. Both the leishmanicidal compartment of subpopulations of phagocytic mononuclear cells and the Th1 or Th2 lymphocyte response profile correlate with the resistant and susceptible phenotypes in genetically defined strains of mice. The genes involved in macrophage activation and natural immunity, and lymphocyte-dependent resolution of disease, however, are distinct. Major and minor histocompatibility complex genes have been linked to some extent to the outcome of experimental cutaneous leishmaniasis.

Immunogenetic investigations of susceptibility to cutaneous leishmaniasis in humans have generally focused on possible associations with major histocompatibility antigens, which would presumably have bearing on lymphocyte-dependent immunity. A family study of localized cutaneous leishmaniasis (LCL) in Venezuela yielded evidence of associations between HLA-BW22 and DQw3 antigens and this presentation of American cutaneous leishmaniasis. The HLA class II specificity DQw3 was also found to be more frequent among Brazilian patients with mucocutaneous disease than healthy controls without a history of leishmaniasis. To strengthen the evidence for a link between genotype frequency and disease susceptibility, the history of exposure and occurrence of subclinical infection in the healthy comparison group, as well as the nonuniformity of ACL, will need to be considered in the design and analysis of future studies. For example, mucocutaneous disease may be a primary or secondary manifestation, the result of contiguous spread or metastasis, self-limiting or progressive and mutilating, and is caused by a variety of species of Leishmania. Since the bases of susceptibility may differ according to the antigenic make-up of the etiologic agent and the mechanism of pathogenesis, these distinctions, as well as the heterogeneity of the immune response (antibody and cell-mediated) are potentially important variables in the classification of cases for immunogenetic studies.

Acquired Resistance

Recurrent leishmaniasis is informative with respect to acquired resistance as well as susceptibility. Evidence of prior cutaneous leishmaniasis is commonly observed in patients with mucosal leishmaniasis. Likewise, patients with active cutaneous lesions who reside in endemic areas of transmission of Leishmania of the Vianna subgenus often have scars characteristic of previous dermal leishmaniasis. Regardless of the mechanism of recurrent disease, reactivation, or reinfection, it is clear that recovery from natural infection with Leishmania of the braziliensis complex does not always con-
fer resistance. Nevertheless, most individuals who have presented with American cutaneous leishmaniasis experienced one episode of disease (Fig 5). Indeed, the relative infrequency of recurrent leishmaniasis provides evidence of acquired resistance. Histopathological evaluation of recurrent lesions revealed a lower parasite burden in recurrent lesions in comparison with that observed in lesions of patients without prior history of disease, suggesting the acquisition of partial resistance. The decreasing incidence of disease in relation to incidence of infection with age (Fig 7) is also consistent with the acquisition of resistance following infection.

Individuals who experience subclinical infection may differ from those who experience disease, particularly chronic disease, with respect to natural and acquired resistance. Parasite determinants notwithstanding, distinct frequencies of subclinical and clinically apparent infection in different foci reflect differences in both natural and acquired resistance among individuals in a similarly exposed population. For example, the attack rate is high, yet subclinical infections also occur in situations of occupational exposure or colonization of endemic areas by previously unexposed populations.

Experimental challenge infections in volunteers also provide evidence of acquired resistance following natural or experimental infection. Published experimental challenge infections with Leishmania that cause ACL have been few and involved a total of seven subjects. Nevertheless, infection with L. mexicana protected against disease upon homologous challenge in four out of five subjects. Infection with L. mexicana did not protect against heterologous challenge with L. braziliensis, while infection with L. braziliensis protected against homologous challenge. Although the generalizability of these limited experiences is unknown, the results provide direct evidence of acquired resistance following clinically apparent infection.

The cellular and molecular bases of susceptibility and resistance to leishmaniasis in humans have not yet been defined. Despite the limited understanding of the mechanisms involved, immunotherapeutic interventions have demonstrated the reversibility of clinical susceptibility. Both the hyperreactive and hyporeactive expressions of dermal leishmaniasis can be ameliorated either transitorily or permanently by immunotherapy.

Parasite Determinants of Disease Expression

Clinical and Epidemiology Evidence of Parasite Determinants of Disease

Just as unusual clinical presentations of infection with a particular Leishmania species serve to illustrate the influence of the host response on disease expression, the finding of a particular disease form to be predominantly associated with a species or taxonomic group of Leishmania indicates the existence of parasite determinants of disease. Diffuse cutaneous leishmaniasis, for example, has only been observed in patients with infections by parasites pertaining to the Leishmania subgenus, specifically, L. mexicana, L. venezuelensis, and L. amazonensis (mexicana complex) in the New World and L. amazonica in the Old World. Similarly, visceral leishmaniasis is principally caused by Leishmania of the donovani complex, L. donovani, L. infantum/chagasi, though infections involving the bone marrow and/or spleen and atypical visceral disease can be caused by dermatotrophic species L. tropica and L. amazonensis. The self-healing nature of localized cutaneous lesions caused by L. mexicana as compared with the chronic and often progressive lesions produced by L. braziliensis in the same population of soldiers further substantiates the existence of intrinsic biological differences among Leishmania.

Population-based studies of incident infection and disease allow the pathogenicity of the Leishmania being transmitted in a particular focus to be evaluated. Based upon the definition of pathogenicity as no.diseased/no.infected, L. peruviana with 9.3 incident cases and 11.2 incident infections/100 person years would be considered more pathogenic than L. panamensis with 0.47 incident cases and 6.6 incident infections/100 person years.

Experimental Evidence of Parasite Determinants of Disease

The propensity of the various taxa to cause distinct disease forms is even more clearly evident in genetically defined mice in which different Leishmania species produce distinguishable and characteristic patterns of disease in the same genetically homogeneous strain of mice. While BALB/c mice develop progressive disease when infected with L. major and L. mexicana, lesions fail to develop when this highly susceptible strain is infected with L. braziliensis. Leishmania of the mexicana and braziliensis complexes also differ
radically with respect to disease expression in the hamster. Likewise, the severity of primary lesions and the frequency of metastases in the hamster differs among species and strains of the braziliensis complex (Fig 8).

Experimental studies have established that the infectivity of Leishmania major differs according to growth phase as well as the length of passage in culture. Metacyclogenesis has since been shown to accelerate during the stationary phase and to correlate with infectivity. However, Leishmania of the Viannia subgenus do not show strict growth-phase-dependent infectivity; furthermore, the growth kinetics and point of maximum infectivity differ among species. Differences in infectivity are an important consideration when experimentally analyzing pathogenicity and/or virulence since some expressions of disease are linked to the dose and route of the infective inoculum. Metastasis in the hamster model on the other hand, was found to be a dose-independent trait of particular strains of L. guyanensis and L. pannensis.

Little is known about virulence factors or the molecular basis of parasite pathogenicity. Infectivity, on the other hand, has been analyzed in exquisite detail both at the biochemical and molecular genetic levels. Several lines of evidence support the participation of lipophosphoglycan (LPG) in infectivity and intracellular survival. Mutants lacking LPG are unable to establish infection and LPG evidently subverts macrophage activation, thereby propitiating parasite survival and replication. Stage-specific changes in the structure of LPG are consistent with the participation of this dominant surface glycoconjugate in the relationship of Leishmania with its invertebrate and vertebrate hosts. LPG structure and composition vary among the Leishmania taxa that have been examined; differences in the constitution of LPG may influence the course of infection and disease that characterizes particular species or taxonomic groups of parasites. Among New World Leishmania that cause dermal disease, only the lipophosphoglycan of L. mexicana has been characterized.

The major surface glycoprotein antigen, gp63, may also participate in infectivity since the expression of gp63 correlates with resistance to complement mediated lysis and with infectivity. Carbohydrate moieties on this metalloprotease, and LPG act as ligands with macrophage receptors that mediate phagocytosis such as complement and mannose/fucose receptors, thereby promoting entry into the host cell. Whether gp63 is causally linked to infectivity, that is, that it is necessary and sufficient, is not known. The relationship of gp63 and other parasite molecules with disease expression (i.e., pathogenicity and virulence) remains to be determined.

Advances in recombinant genetics of Leishmania together with appropriate experimental models of pathogenicity and virulence should reveal the molecular bases of parasite determinants of disease. Nevertheless, because the host response is tightly linked to the outcome of infection, it is conceivable that the parasite molecules that elicit particular cytokine repertoires and the genes that code and/or regulate their expression, will constitute virulence factors.

Figure 8. Frequency of cutaneous metastasis in the golden hamster when experimentally infected with different strains of L. pannensis and L. guyanensis (from Martinez et al., with permission).
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