

## REVISÃO

### OUR PRESENT KNOWLEDGE OF THE ECOLOGY AND CONTROL OF LEISHMANIASIS IN THE AMAZON REGION OF BRAZIL

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Species of *Leishmania* Ross, 1903<sup>49</sup>, are exclusively parasitic protozoa of the Order Kinetoplastida, Family Trypanosomatidae. They are digenetic (heteroxenous) organisms, with promastigotes and paramastigotes (with a single, free flagellum) in the alimentary tract of the insect host, and round-oval amastigotes (no free flagellum) in the macrophages of vertebrate hosts: as far as is known there is no sexual stage in the parasite's lifecycle. The insect hosts appear to be limited to species of the haematophagous phlebotomine "sandflies" (Diptera: Psychodidae: Phlebotominae), and the vertebrate hosts to a variety of mammals. The latter function as reservoirs of infection for the sandfly, and transmission to the next mammalian host is predominantly by injection of the parasite during the bite of the infected insect.

The primitive nature of the promastigote form has led to the widely adopted view that the sandfly vectors of *Leishmania* (or their ancestors) represent the primitive and primary hosts of the parasite: a suggestion which finds support in a definite cycle of development which takes place within the sandfly gut, and sometimes attached to the gut wall. *Leishmania* probably adapted to survive in mammals, after the inoculation of the organism during the evolution of the blood-sucking habit of ancestral sandflies. The mammalian hosts now act not only as a supply of blood for the sandfly, but as a source of infection from the amastigotes located in the skin or blood of these animals.

A large number of mammalian species act as reservoirs of *Leishmania*. The parasites are commonly found in rodents, canids, marsupials, edentates, procyonids, primitive ungulates (hyraxes) and primates; rare infections have even been recorded in bats. No leishmanias have yet been recorded in amphibians or birds (although there are some sandfly species which feed on these animals), and *Leishmania*-like parasites of reptiles (*Sauroleishmania*) have now been removed from the genus *Leishmania*.

Ecological and epidemiological studies over the past 25 years have shown that there are many more leishmanial parasites than we previously suspected, and further evidence is rapidly accumulating to suggest that each species is not, as commonly supposed, randomly distributed in a miscellanea of sandfly and mammalian hosts, but that there are natural barriers limiting certain leishmanias to certain sandfly/mammalian combinations<sup>20 25</sup>.

In the natural mammalian reservoir host, *Leishmania* rarely produces disease, the infection usually remaining benign and inapparent – as is usually the case in an ancient and well balanced host-parasite relationship. In the wrong hosts, however, including man and some of his domestic animals such as the dog and donkeys, infection most commonly produces ugly skin lesions, or a visceral disease which is usually of a fatal nature unless adequately treated. Luckily, many sandfly species appear to be host-restricted, or are at least unattracted to man, and a great many leishmanias (probably the majority) thus remain with no opportunity to infect him. It is quite possible that they would in any case be incapable of establishing themselves in his tissues. Nevertheless, those species of *Leishmania* that we know to infect man are of very great medical importance: so much so that leishmaniasis is ranked among the six most important tropical diseases, with an estimated global incidence of some 400,000 new cases occurring each year<sup>3</sup>. The endemic regions cover a vast area of our planet; throughout most of tropical Africa, India, eastern Asia (but unknown in Australasia), central Asia, the Mediterranean basin, and some neighbouring European countries, and most of Latin America.

#### Leishmaniasis in Brazil

The ecology and epidemiology of the American leishmaniasis, in general, have recently been discussed at length<sup>20</sup>. It is inevitable, therefore, that there will be some repetition in this paper, the object of which is to review and update our present knowledge on the leishmanias and leishmaniasis in the Amazon Region of Brazil: one of the few remaining areas where

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we can still study the ancient sandfly/mammalian life-cycles of these parasites, relatively uncomplicated by man's influence.

Vianna<sup>54</sup> gave the name (his spelling) of *Leishmania brazilienses* to a leishmanial parasite causing disseminated cutaneous leishmaniasis in Além Paraíba, on the borders of Minas Gerais and Rio de Janeiro States, Brazil: a lapsus calami generally overlooked, and later corrected to *L. braziliensis* by Matta<sup>40</sup>.

With desperately few taxonomic criteria with which to identify *Leishmania* species, it long remained the custom to refer to all cases of cutaneous and mucocutaneous leishmaniasis in Brazil and neighbouring countries as due to *L. braziliensis*. It was not until 1954, in fact, that a new name appeared, associated with cutaneous leishmaniasis in the Guyanas and north Amazonian Brazil, when Floch<sup>13</sup> referred to *L. tropica guyanensis* as the causal agent of "pian-bois" in those regions, and reserved the name of *L. t. braziliensis* for the parasites associated with all forms of the cutaneous and/or mucocutaneous disease south of the Amazon River. Pessoa<sup>44</sup> correctly amended these names to our present-day *L. braziliensis guyanensis* and *L. b. braziliensis*.

With the exception of "chiclero's ulcer", in the Yucatan region of Central America, it is fairly safe to say, however, that up to the late 1960's few protozoologists (and even fewer dermatologists) accepted the suggestion that any form of dermal leishmaniasis in Brazil – or throughout the Americas, for that matter – might be due to more than one parasite, namely *L. braziliensis*. This was in spite of additional new names given to the parasites associated with "uta" in the Peruvian Andes (*L. peruviana* Velez, 1913<sup>53</sup>), "chiclero's ulcer" (*L. mexicana* Biagi<sup>7</sup> emend. Garnham<sup>17</sup>) and "diffuse cutaneous leishmaniasis" in Venezuela (*L. pifanoi* Medina & Romero<sup>42</sup>, emend. Medina & Romero<sup>43</sup>). Here the situation rested until the late 1960's, when the incrimination of forest rodents as reservoir hosts of *L. mexicana* in Belize<sup>26 27</sup> prompted similar field studies in Brazil.

#### Leishmaniasis in Amazonian Brazil

##### *Leishmania mexicana amazonensis* Lainson & Shaw, 1972<sup>23</sup>.

Working in Pará and Mato Grosso States, Lainson & Shaw<sup>22</sup> noted striking biological differences between a *Leishmania* commonly found in rodents and the parasites most frequently isolated from man. The rodent parasite was shown to be so similar to *L. mexicana* of northern Central America, that it was

given the new sub-specific name of *L. m. amazonensis*.

The sandfly host was shown<sup>21 51</sup> to be *Lutzomyia flaviscutellata* (Mangabeira 1969<sup>39</sup>), a low-flying and essentially nocturnal insect which is luckily not much attracted to man. Human infection is thus uncommon, and largely restricted to nighttime hunters and fishermen, and others penetrating low-lying swamp forest (igapó) or riverine forest (várzea), at night, where the *Lu. flaviscutellata* population is particularly dense. Due to the low-level flight range of this sandfly (it is rarely encountered above 1 metre) the wild reservoir hosts are largely found among terrestrial or semiterrestrial animals, particularly rodents and marsupials: the principal vertebrate host is the echimyid rodent, *Proechimys* spp.

In spite of the relative rarity of human disease due to *L. m. amazonensis*, this parasite is of considerable importance for two reasons. In the first place, human infection not infrequently proceeds to incurable "diffuse cutaneous leishmaniasis" (DCL). Although this condition has been shown to be linked with a deficient cell-mediated immunity in the patient, it has till now been found associated *only* with individuals infected with parasites of the *L. mexicana* complex, and on no occasion have we recorded DCL in the many hundreds of other patients infected with *L. braziliensis* or related parasites in north Brazil. In the second place, both the sandfly and vertebrate hosts of *L. m. amazonensis* are commonly found in a wide variety of habitats; from high primary forest to the low, dense secondary growth which follows deforestation (particularly favoured by rodents). The enzootic has even adapted remarkably well to completely non-indigenous pine and gmelina tree plantations in the Amazon Region of Brazil<sup>46</sup>, suggesting that although destruction of primary forest may cut the incidence of human leishmaniasis due to some parasites, e.g. *L. b. guyanensis*, it may result in an increase in the disease due to others, such as *L. m. amazonensis*. This is a particularly sombre thought in view of this organism's association with DCL.

##### *Leishmania braziliensis guyanensis* Floch, 1954<sup>13</sup>.

The process of unravelling the epidemiology of "pian-bois" has made a fascinating story in which field observations, basic parasitology and entomology, and modern biochemical techniques have all played an equally important part.

In the forests of French Guyana, Floch<sup>14</sup> noted that one particular species of sandfly gathered in large numbers on the trunks of the larger trees, from where it avidly attacked man when disturbed. The insect was

identified (erroneously, as it transpired) as *Lutzomyia anduzei* (Rozeboom), and Floch intuitively felt that this sandfly might be the vector of "pian-bois". Some years later, in neighbouring Surinam, Wijers & Linger<sup>58</sup> also noted this phenomenon, again identified the fly as *Lu. anduzei*, and demonstrated promastigotes in 12 specimens among an unspecified number they dissected. Flagellates from two flies were inoculated into a hamster, but the animal failed to become infected.

Armed with this knowledge, Lainson et al<sup>29</sup> studied an area of forest near Monte Dourado in Pará State, Brazil, north of the Amazon River. They dissected 55 specimens of "*Lu. anduzei*", found 4 to be heavily infected, and on this occasion the parasite was established in hamsters and later in blood-agar culture medium. During these studies suspicions were aroused that the sandfly concerned was not in fact *Lu. anduzei* Rozeboom, but a closely related and previously undescribed species, which Ward & Fraiha<sup>55</sup> subsequently named *Lu. umbratilis*. Additional studies in the same and other areas of north Brazil, and in French Guyana, provided further evidence incriminating this sandfly as the major vector of *L. b. guyanensis*<sup>4 37 38</sup> and, following a very detailed study of the ecology of the sandfly fauna in the Monte Dourado area over a number of years<sup>47</sup>, it was concluded that "only the females of *Lu. umbratilis* have the habits... and abundance... consistent with a major vector to man of *L. b. guyanensis*".

In 1980 the author's laboratory<sup>28 34</sup> turned its attention to a search for the wild animal reservoir of the parasite and, in view of the vector's habits, concentrated principally on arboreal animals captured in primary forest of the Monte Dourado area. The organism was finally isolated from 27 out of 59 (46%) two-toed sloths (*Choloepus didactylus*) and 6 out of 27 (22.2%) lesser anteaters (*Tamandua tetradactyla*), with rare infections occasionally found in terrestrial or semi-terrestrial animals such as rodents and marsupials: a high rate of infection has also been found in *C. didactylus* in French Guyana<sup>18</sup>. Finally, to complete the epidemiological picture, biological and biochemical (enzyme profiles) comparison has failed to detect any differences between the parasites from *Lu. umbratilis*, the wild animal reservoirs and man.

In the forest canopy, then, there is a constant cycle of transmission maintained by the sandfly *Lu. umbratilis*, at night, among arboreal animals, particularly the sloth *C. didactylus* and the anteater *T. tetradactyla*. Gravid sandflies descend to the forest floor to oviposit, and then migrate back to the canopy for further blood-meals on their arboreal hosts. The migrations result in unusually large concentrations of

female *Lu. umbratilis* on the bases of the larger trees (those whose upper branches form part of the canopy), particularly in the early hours of daylight, and many of these will be infected. These flies are normally disinclined to feed on man, but when disturbed by his activities (felling of trees, or by the simple process of leaning against a tree) they will attack him avidly. Thus, during a daytime capture of sandflies from tree-trunks (for about one hour, at 8.00 am), two field-workers collected 77 specimens attacking their arms: of these, 72 (92.5%) were *Lu. umbratilis*, of which 16 were heavily infected. It is small wonder that the two men, between them, developed 13 lesions due to *L. b. guyanensis* on their wrists and forearms<sup>37</sup>. Perhaps the most overwhelming evidence to incriminate a sandfly vector that one could wish for!

As most forest-workers engaged in tree-felling operate only during the day, we feel that most transmission to man takes place at ground level, in the daylight hours and particularly in the early morning. This is not to say that some transmission to man does not occur at night, and indeed we have personal evidence that it does, but on a small scale, for the following reasons. Firstly, there is relatively little human activity at night in the forest (mainly occasional hunters) and, secondly, at night most of the female *Lu. umbratilis* population is actively seeking blood among (reservoir) hosts high up in the forest canopy.

In conclusion, Arias & Naiff<sup>5</sup> noted an interesting variation in the ecology of "pian-bois" near Manaus, Amazonas State, where forest was being cut for the development of a new residential area. Five out of 7 common opossums (*Didelphis marsupialis*) were found infected with *L. b. guyanensis* and it was suggested that this high infection rate was due to the disappearance of the primary edentate hosts following man's destruction of their habitat, and a resulting increase in the number of opossums, which are notorious scavengers and attracted by man's presence. An analysis of blood from 975 engorged *Lu. umbratilis* from the area in question, however, showed that 64% had fed on two-toed sloths and only 1% on opossums<sup>9</sup>, clearly invalidating this hypothesis. An alternative explanation<sup>20</sup> has been suggested, namely that sloths still remained the source of infection, and that opossums represented a "dead-end" host in which "The infection rate (had) simply built up to a high level in an unnaturally large, forest-fringe population of this species which had resulted from this animal's attraction to the area by man's refuse". It is significant that rare or no infections have been observed in large numbers of *D. marsupialis* examined in areas of undisturbed, primary forest<sup>6 18 34</sup>.

*Leishmania braziliensis braziliensis* Vianna, 1911<sup>54</sup> emend. Matta, 1916<sup>40</sup>.

infected flies was recorded during a number of visits to this region<sup>30</sup>.

Elucidating the ecology and epidemiology of leishmaniasis due to this parasite has proved a much more difficult proposition, and the situation remains confused both in the Amazon Region and in relation to human leishmaniasis caused by *L. b. braziliensis* sensu lato in other geographic regions of Brazil and elsewhere in South and Central America. There are a number of reasons for this. Firstly, as is unfortunately the case with most recognized species of protozoal parasites, there is no type material of Vianna's parasite or, by modern standards, an adequate description with which we can directly compare our present day isolates of "*L. b. braziliensis* sensu lato". Even the assumed linkage of the organism with the clinical condition of mucocutaneous leishmaniasis, or "espundia", does not help us, for Vianna's patient was described as suffering from disseminated cutaneous lesions. It in fact remains distinctly possible that the infection was not due to what we are now calling *L. b. braziliensis*, but to a parasite of the *mexicana* complex! All that it is possible to do at the moment is to continue stockpiling and comparing all available isolates of *Leishmania* from man, wild mammals and sandflies, in given geographic areas of Latin America, until we can designate a neotype which can serve as a firm basis with which to sort out the considerable number of peripylarian leishmaniasis at present being uncovered. Another problem confronting us is the difficulty in handling many of these isolates in the laboratory, with consistently poor growth of such parasites in all manner of *in vitro* culture media and in the hamster<sup>19 24</sup>.

Studies on the epidemiology of *L. b. braziliensis* s. l. in the Amazon Region of Brazil have largely been limited to Mato Grosso and Pará, principally in the range of hills known as the Serra dos Carajás, in the densely forested southern part of the latter State. Here, a high incidence of cutaneous leishmaniasis suffered by a mining company, in the early 1970's, prompted a survey of the sandfly fauna<sup>57</sup>, resulting in the relatively speedy pinpointing of the local vector<sup>33</sup>.

The sandfly in question, *Psychodopygus wellcomei* Fraiha et al, is a member of the *squamiventris* group, notable for its inclusion of a number of species or subspecies with morphologically indistinguishable female forms: identification of these at present rests on the morphology of the males. *Ps. wellcomei* is an extremely anthropophilic sandfly, attacking man not only at night but also during the day, particularly in the early hours and in overcast weather. A total of 16

Our early work in the Carajás area was carried out high up on the hills, at about 500-700 m: the only forest accessible at that time, by air. Recent road construction, however, has now enabled similar studies to be made down to the foothills, with a most interesting finding. The breeding-out of gravid females in the laboratory (Dr. P. D. Ready, unpublished observations) showed that our higher altitude population of "*Ps. wellcomei*" was in fact comprised of 95% true *wellcomei* and 5% of *Ps. complexus* (Mangabeira), another member of the *squamiventris* group. Below about 300 m the position slowly reversed, with an increasing proportion of *Ps. complexus* as one descends the hills. We feel that this is, however, unlikely to invalidate our earlier incrimination of *Ps. wellcomei* as the major vector, as all our observations were made above 500 m: it is significant, too, that parasites isolated from man in the lowlands appear to be different from that in the higher altitude, *wellcomei* habitat. More recently, Dr. L. Ryan (unpublished observations) has continued this study, in the same area, and encountered a further 35 supposed *Ps. wellcomei* with flagellate infections. To date, 14 of these are considered as due to *L. b. braziliensis* s. l. on monoclonal antibody serology (Shaw & Lainson, unpublished observations). Attempts to raise offspring from the eggs of these flies have unfortunately failed but, while the possibility that some may have been *Ps. complexus* cannot be entirely ruled out, the numerical superiority of *Ps. wellcomei* at the capture site makes it most likely that all or most were this species. Recent (unpublished) results on cuticular hydrocarbon analysis in collaboration with Prof. David Molyneux's laboratory, at Salford University, indicates the technique to have great promise in differentiating sandfly species and may help solve this problem in future field studies in the Carajás.

For some time the known distribution of *Ps. wellcomei* was restricted to the Serra dos Carajás, Pará, but this sandfly has now been shown to be a predominant species in similar ranges of forested hills in the northeastern State of Ceará<sup>48</sup>. In this respect it is significant that parasites isolated from man in Ceará and Bahia have proved to be indistinguishable from *L. b. braziliensis* s. l. in the Serra dos Carajás<sup>1041</sup> (Shaw & Lainson, unpublished observations) and it is likely that the three localities form part of the same enzootic, which may be much more extensive than previously thought.

Although a very limited number of wild rodents have been found infected with *L. b. braziliensis* – like parasites in Mato Grosso and São Paulo States<sup>15 16 22</sup>, we have been singularly unsuccessful in detecting infection in a wide range of mammals (principally terrestrial rodents, and a variety of marsupials) in our Serra dos Carajás study area, and the search continues. The vector, *Ps. wellcomei* has been shown to be essentially a forest-floor species, however, and we remain convinced that the reservoir host of the parasite is terrestrial in habits, or at least comes frequently to the ground.

The existence of biochemically and serologically very similar or indistinguishable leishmanias of man in geographically widely separated parts of Brazil and in other parts of South and Central America, has been discussed elsewhere<sup>20 24 52</sup>. Some of these localities are so distant, or so ecologically different (particularly in terms of the phlebotomine and/or mammalian fauna) that in spite of the previously discussed evidence for the host-specificity in the parasite/sandfly/mammal combinations, it is difficult to conclude other than that *L. b. braziliensis* has different sandfly/mammalian hosts in these geographic regions. This conclusion must be viewed with caution, however: while it is relatively easy to say that two organisms are *different*, it is much more difficult to say they are the *same*.

Finally, during the above-mentioned epidemiological studies in the Amazon Region, we have uncovered what are undoubtedly new leishmanial parasites<sup>25</sup>, two of them in man. Most, however, have been found in a variety of wild mammal and sandfly species, and there are doubtless more to be discovered: some of these parasites can certainly be established in the hamster, but whether or not they can infect man remains to be seen.

*Leishmania donovani chagasi* Cunha & Chagas,  
1937<sup>11</sup>

Visceral leishmaniasis in the Amazon Region has until recently been regarded as a rather rare, sporadic disease of man<sup>20</sup>, largely restricted to the coastal villages of Pará State: the epidemiology has remained obscure, although the fox, *Cerdocyon thous*, has been incriminated as a natural host and, as infection in this animal is of an inapparent nature, it has been suggested that "... present-day endemic areas elsewhere in South America may have originated from ancient sylvatic enzootic involving *C. thous* or related canids".

As throughout the rest of the geographic range of American visceral leishmaniasis, the vector has long been assumed to be the sandfly *Lutzomyia longipalpis* (Lutz & Neiva), although this conclusion has been based entirely on the (usually) domestic and peridomestic abundance of this insect in the better known endemic regions of the Americas, the rare records of unidentified promastigotes in dissected females and experimental transmission of the parasite to hamsters by the bites of laboratory - bred *Lu. longipalpis*<sup>31</sup>.

Specimens of *Lu. longipalpis* from Pará have shown slight morphological differences from those in the major endemic areas of Bahia and Ceará States in northeast Brazil<sup>32 39 56</sup> in that the males of the northeastern form have two pairs of white spots on the 3rd and 4th abdominal tergites, while the males of the Pará form have only one pair of spots, on the 4th tergite. Ward et al<sup>56</sup>, found the two forms to be sexually isolated: they suggested that the less anthropophilic habits of the Amazonian form and the more highly developed domestic habits of the northeastern form might account for the low and high endemicity of human visceral leishmaniasis in these regions, respectively.

During field studies in a small focus of the disease on the Island of Marajó<sup>32 50</sup> we dissected 1,500 females of *Lu. longipalpis* caught in and around the houses of patients with visceral leishmaniasis, and found heavy promastigote infections in 8 (0.5%). Enzyme characterization of the flagellates from one fly showed the parasite to be indistinguishable from *L. d. chagasi* from man and foxes in the same locality, and from man in endemic areas in Ceará and Bahia: parasites from another fly produced fulminating visceral leishmaniasis when inoculated into a hamster<sup>36</sup>. In the same Marajó focus, up to the time of writing this paper, *L. d. chagasi* has been isolated from the viscera of 5 out of 17 (29.4%) specimens of the fox, *C. thous*. Superficially, all the infected animals appeared to be in perfect health.

Up to 1982, a total of 35 cases of autochthonous, human visceral leishmaniasis had been recorded in Amazonian Brazil, all in Pará State. Most of these were restricted to small coastal villages, but an apparently isolated focus of three cases<sup>2</sup> was registered in the small, riverside town of Santarém, about 730 km inland from the coast. After a seemingly quiescent period of some twenty years, a few new cases began to appear in Santarém, in 1982 - 1983, with an alarming surge of 67 confirmed cases during the period January - September,

1984: all but 4 of these were in children below the age of 15 years.

Epidemiological studies were commenced in Santarém in February, 1984<sup>35 36</sup> and quickly revealed a very dense population of *Lu. longipalpis* in those suburbs from which most of the human cases of visceral leishmaniasis were registered. Thus, in one night it was possible to collect 3,324 specimens of this sandfly (706 females) in the back-yard of one house, using a combination of a single CDC light-trap, placed over a brooding chicken, and direct aspiration of flies from a chicken-house in which an infected dog was sleeping. A further 15 engorged flies were caught in another light-trap placed in the childrens' sleeping quarters. Of these children, one had been treated for visceral leishmaniasis some months previously, and the other two were shown to be infected during the present investigation.

A total of 491 of the *Lu. longipalpis* captured on this occasion were dissected, and 35 (7.14%) were found to be infected with promastigotes. The flagellates from 27 of the sandflies were inoculated into hamsters, by the intra-peritoneal route, and 16 of these animals developed typical, fulminating visceral leishmaniasis from 3-6 months later. In addition, transmission of the parasite by the bites of naturally infected *Lu. longipalpis* was obtained on four occasions, by placing anaesthetised hamsters in cages containing variable numbers of these wild-caught sandflies. Finally, 9 out of 16 dogs examined in or near the study area showed abundant to massive numbers of amastigotes in skin-scrapings, and a subsequent serological survey (IFAT) of 416 others, from the same suburb, showed a positivity of 56%.

Isolates from man, dogs and the infected *Lu. longipalpis* have been compared, biochemically, by starch-gel electrophoresis using the enzymes ASAT, ALAT, PGM, GPI, MDH, and MPI<sup>8</sup> and found to be indistinguishable. From these results and the epidemiological observations, therefore, there remains no reasonable doubt as to the important and probably unique role of *Lu. longipalpis* as the vector of *L. d. chagasi* in Amazonian Brazil, and presumably elsewhere in the geographic range of the parasite. The very high infection-rate in the Santarém focus of visceral leishmaniasis was doubtless due to the large population of sick dogs concentrated in overcrowded conditions.

## PREVENTION AND CONTROL

Methods of avoiding cutaneous leishmaniasis on the part of individual persons occasionally penetrat-

ing tropical rain-forest are at present limited to the use of insect repellents, protective clothing or netting, and avoidance of danger areas at night: none of these are very effective. In hot, humid conditions the effectiveness of insect repellent is quickly neutralized by profuse sweating, and the author has on many occasions watched sandflies feeding on exposed skin which had been sprayed only some 10 minutes earlier. Long sleeves, gloves (!) and netting to protect the face might be feasible for a motionless person, but for active forest-workers they are insupportable absurdities. Curtailment of nighttime activities in areas of high risk will doubtless diminish chances of infection, but it will be recalled that a number of anthrophilic sandflies, including at least two major vectors, also feed in the daytime under certain circumstances. Awareness of the possibilities of acquiring infection, prompt diagnosis and medical treatment, go a long way in avoiding an unpleasant and protracted disease. There are *no* available vaccines against any form of leishmaniasis on the market, and no chemoprophylactic drugs. Treatment of the disease is still largely dependant on prolonged courses of injections with antimony derivatives, which are costly and of variable toxicity.

Prevention and/or control of sylvatic leishmaniasis among gangs of labourers, topographers and other forestry workers (principally during forest clearance) can be effective on a small scale: men in forest encampments can to some extent be protected by placing them in adequate clearings, spraying the bases of the larger trees with insecticides (e. g. in "pian-bois" areas)<sup>14 45</sup>, and by prohibiting nighttime hunting.

Visceral leishmaniasis in the major endemic areas of the northeast (Ceará and Bahia) has been well controlled (but *not* eliminated) by the destruction of infected dogs, regular insecticide spraying of houses and animal shelters, and the treatment of patients. In areas with abundant adjacent forest or woodlands, however, such as the Island of Marajó, there is clearly a sylvatic cycle among foxes and *Lu. longipalpis*, but because of the sparsity of both human and canine populations, domestic or peridomestic transmission is sporadic and rare. Although the antimalarial spraying programmes keep the number of *Lu. longipalpis* in houses to a very low level, it is not customary to spray the chicken-houses and, unfortunately, the chicken is an ideal maintenance host for this insect, which accumulates in large numbers in the primitive shelters housing these birds. Marauding foxes commonly enter villages in search of prey, bringing infection to this peridomestic *Lu. longipalpis* population, a small proportion of which becomes infected. These infected flies occasionally transmit to

dogs, and more rarely to man. Canine infection remains much more common than that of man, simply because the sandfly vector is much more highly attracted to dogs.

When, as in Santarém, the dog, human and sandfly populations are all increased to a high level in the very overcrowded suburbs, introduction of infection may result in an explosive outbreak of the human disease, simply due to a "spill-over" from the vast reservoir of infection in the large number of sick dogs. Once again, foxes are probably the original source of infection, in adjacent woodland, where *Lu. longipalpis* has been taken in light-traps. Thus, although periodic control of visceral leishmaniasis may be possible in such situations, the sylvatic cycle renders its total eradication most unlikely.

While the dog serves as a major reservoir of infection for man in the epidemiology of visceral leishmaniasis, there is no firm evidence that it plays any role in the epidemiology of the known cutaneous diseases in the Amazon Region, as recently suggested by Aguilar et al<sup>1</sup>. The number of dogs present in endemic forested regions, with their sparse human populations, is small; and even though occasional dogs may become infected when on hunting expeditions with their owners, they are, as a source of infection for sandflies, but a drop in the ocean compared with the vast and constant reservoir of infection in the wild animals, and not worthy of consideration in the overall epidemiology. The author has seen no infected dog during his 20 years of epidemiological studies on the cutaneous disease in Pará, and it should be remembered that the great majority of human infections are found among labour forces, or military personnel undertaking jungle-warfare training, neither of whom are usually accompanied by dogs. In the Serra dos Carajás, where human infection is so common, dogs are actually prohibited in the newly colonized areas (principally an anti-rabies measure).

Donkeys, which are a common means of transport to forest plantations in certain parts of Venezuela, have frequently been found with skin ulcers due to *Leishmania*, presumably acquired when passing through enzootic woodlands. Aguilar et al<sup>1</sup>, suggest that these animals might also be considered as alternative reservoir-hosts for human infection in the Amazon Region of Brazil, "...where there is a tendency of the local workers to search for the infection mostly in wild animals". The same arguments apply and, in the author's experience, donkeys are a rare sight indeed in Amazonian forests. The possible role of dogs, equines, or even man, as

sources of infection for sandflies might, it is true, be of some epidemiological importance if domestic or peridomestic transmission can be proven; and if it can be shown that any one of these three "victim" hosts are capable of acting as a significant source of infection for an anthropophilic sandfly species, commonly found in or around houses, and which is capable of transmitting the various aetiologic agents of cutaneous leishmaniasis discussed above. We have looked in vain for such a fly in the major endemic areas we have studied in the Amazon Region.

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