LEISHMANIASIS: AN OVERLOOKED PUBLIC HEALTH CONCERN.

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Abstract

Leishmaniasis is a protozoan disease that represents an emergent threat with high morbidity and mortality rates. The disease is endemic in areas of the tropics, subtropics and in the southern Europe. Currently, leishmaniasis has a wider geographical distribution than before and this is mainly attributed to the constant increase of leishmaniasis' risk factors that include migration, environmental changes, deforestation, urbanization, immunosuppression and malnutrition. Thus, leishmaniasis is a potential threat for several areas.
This article reviews on the clinical and epidemiological features of leishmaniasis. Based on disease epidemiology and analyzing the associated risk factors, several prevention measures are being discussed in detail. Taking into consideration the lack of a commercially available vaccine, the lack of access to efficient drug therapy mainly in the developing countries, the limited local resources of the affected countries, it is concluded that elimination of the disease is still a challenge for the international health community.

Keywords: epidemiology, leishmaniasis, prevention, risk factors

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Introduction

Leishmaniasis is a major vector-borne disease caused by obligate intramacrophage protozoa of the genus *Leishmania,* parasites infecting numerous mammal species, including humans. Leishmaniasis is transmitted by the bite of phlebotomine sand flies and its species are widespread on all continents except Antarctica [Bañuls et al, 2007]. In this regard, leishmaniasis is endemic in areas of the tropics, subtropics, and southern Europe. Specifically, is endemic in 88 countries and is the only tropical vector-borne disease that has been endemic to southern Europe for decades [Dujardin et al, 2008]. Currently, leishmaniasis has a wider geographical distribution pattern than before and it is considered to be a growing public health

concern for several countries. The increase in leishmaniasis' worldwide incidence is mainly attributed to the increase of several risk factors that are clearly manmade and include massive migration, deforestation, urbanization, immunosuppression, malnutrition and treatment failure [Desjeux, 2001]. Human made changes to the environment, as well as the population movements, may lead to alterations in the range and density of the vectors and reservoirs and consequently may increase human exposure to infected sand flies.
Undoubtedly, leishmaniasis is a potential threat for the European region and one of the most neglected diseases in developing countries. In this review, the clinical and epidemiological features of leishmaniasis are being described in detail. Moreover, prevention measures and control strategies are being discussed in order to reduce the risk of the disease transmission.

DISEASE TRANSMISSION, CLINICAL AND EPIDEMIOLOGICAL FEATURES.

Leishmaniasis is a protozoan disease caused by a parasite member of the *Leishmania* genus and presents high morbidity and mortality rates. The disease is being transmitted to its vertebrate host by the female infected sand fly. The female needs a blood meal for egg maturation. Hence, like mosquitoes, only the female sand fly is haematophagous.
Leishmania parasites' life cycle is complex. Specifically, these parasites have two basic life cycle stages: one extracellular stage within the invertebrate host (phlebotomine sand fly) and one intracellular stage within a vertebrate host. The parasites exist in two main morphological forms the amastigotes and promastigotes, which are found in vertebrate and invertebrate hosts, respectively [Koutis, 2007]. The invertebrate hosts are small insects of the order Diptera, belonging to the *Phlebotominae* subfamily and only two of the six genera described are of medical importance: *Phlebotomus* of the "Old World" (Africa, Asia, and Europe) and *Lutzomyia* of the "New World" (the Americas) [Killick-Kendrick, 1990; 1999]. Some phlebotomine species -such as *Phlebotomus papatasi* and *P.sergenti*- can support the growth of only those species of Leishmania with which they are infected in nature, whereas other species –such as *Lutzomyia longipalpis* and *P. argentipes* - can develop mature transmissible infections when infected with several Leishmania species [Pimenta et al, 1994; Kamhawi et al., 2000; Rogers et al., 2004; Koutis, 2007]. The potential diseases' reservoir include many different orders of mammals such as rodents, canids, edentates, marsupials, procyonids, primitive ungulates and primates [Lainson et al, 1987]. On the other hand, humans are considered to be accidental hosts of these parasites. The life cycle begins when an infected female sand fly takes a blood meal from the vertebrate host. During the blood meal obtainment, the sand fly introduces its mouthparts into the skin tearing tissues and the salivary gland content is injected together with *Leishmania* promastigotes into the host's skin [Titus et al, 1988; Andrade et al, 2007]. The promastigotes are long, flagellate and extracellular. The promastigotes are then phagocytosed by the host's macrophages and consequently the parasite evolves into amastigote forms –spherical, intracellular forms without flagellum-which they reproduce by binary fission. The multiplication of the parasites occurs inside the macrophages, which are their main targets. The macrophage lyses and the cycle continues when other hosts' phagocytes are being infected [Bañuls et al., 2007]. The establishment of the disease depends on the success of the Leishmania parasite to differentiate into the amastigote form [Bodgan et, 1990; Chang, 1990; Sereno et al, 2005]. It should be mentioned that sanflies' populations are more active from dusk to dawn. Infrequently, sandflies are not being involved in leishmania transmission. It has been established that visceral leishmaniasis –a clinical form of the disease described later on the text- could be directly transmitted via blood (needle sharing, transfusion, transplacental spread) or organ transplantation [Cruz et al, 2002; Pagliano et al, 2005; Morillas-Marquez et al, 2002]. Moreover, it has been shown that cutaneous infection can also be developed in case of

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needle injection of contaminated with leishmania material.

The leishmania parasite is involved in different pathologies that range from the cutaneous to the visceral forms, depending on the species of leishmania and the host immune response [Barral et al, 1991; Liew et al., 1993]. In humans, infection with leishmaniasis parasites could also result in asymptomatic forms. It is well documented that asymptomatic human hosts could contribute in the maintenance of the leishmaniasis foci [Bañuls et al., 2007; Costa 2002; Riera et al, 2004]. It has been shown by several epidemiological studies that the majority human infections by leishmania parasites remain asymptomatic [Badaro et al, 1986; Bucheton et al, 2003]. These asymptomatic human subjects are able whether to clear the infection or they remain asymptomatic carriers for years. Thus, leishmaniasis development depends on several risk factors such as malnutrition, immunosuppression, age, immunological status and genetic factors. Several investigators have reported the negative effect of malnutrition on leishmaniasis clinical course. In this regard, it has been shown that malnutrition in *Leishmania donovani* infected subjects is able to alter the immune response and consequently increases the risk of clinical leishmaniasis [Harrison et al, 1986; Anstead et al, 2001]. Furthermore, it has been observed that there are ethnic differences in the ratio of asymptomatic to symptomatic infections [Bucheton et al, 2003]; this finding suggests that human susceptibility to the clinical expression of leishmaniasis depends on host's genetic risk factors. On the other hand, several studies conducted on animal models and on populations living in endemic areas suggest that allelic variants at specific genetic loci increase the risk of clinical leishmaniasis development [Bradley et al, 1979; Vidal et al., 1995; Bucheton et al, 2003].

The leishmaniases are characterized by a spectrum of clinical manifestations: ulcerative skin lesions which they develop at the site of the sand fly bite (localized cutaneous leishmaniasis); multiple non-ulcerative nodules (diffuse cutaneous leishmaniasis); destructive mucosal inflammation (mucosal leishmaniasis) and disseminated visceral leishmaniasis [Desjeux, 2004]. Cutaneous leishmaniasis is frequently self-healing in the "Old World"; cutaneous infection can remain subclinical or become clinically apparent after a variable incubation period that averages several weeks. The first sign of an infection is a small erythema that develops on the site of the sand fly bite after a variable period of time. The erythema develops into a papule, then into a nodule; the nodule ulcerates over a period of 2 weeks to six months to become a lesion that is characteristic of the cutaneous leishmaniasis [Peters et al, 1987]. The lesions vary in severity (size), clinical appearance and time to cure [Peters et al., 1987; Reithinger et al, 2007]. There is a tendency for lesions to self-cure and the time to cure of the disease onset varies. Spontaneous self healing usually results in lifelong protection from the disease. In the case of cutaneous leishmaniasis, different *Leishmania* species can infect the macrophages in the dermis with variable clinical presentations and prognoses [Arevalo J et al, 1996; Dedet et al, 2003]. In diffuse cutaneous leishmaniasis, the lesions are multiple, long-lasting, disseminated and cannot be spontaneously healed [Bañuls et al, 2007]. This form of leishmaniasis is considered to be a severe public health problem since it has devastating consequences to the patient. In the case of mucosal leishmaniasis, the most affected area is the nasal mucosa although lesions may be found in the lips, mouth, pharynx and larynx [Lessa et al, 2007]. These lesions are not self-healing and are usually seen months or years after the first episode of cutaneous leishmaniasis. It is established that mucosal involvement is usually secondary to cutaneous lesions, although there are some cases where the mucosa is the primary site. The reason why leishmaniasis patients develop mucosal involvement is not full understood. Nevertheless, disease progression, from cutaneous to mucosal lesions, is mediated through the lymphatic system and rarely by direct contact between the mucosa and the cutaneous lesion. Additionally, although

mucosal leishmaniasis can be caused by *Leishmania panamensis, L. amazonensis, L. major L. tropica* and *L.infantum*, it seems that *Leishmania braziliensis* is responsible for the majority of mucosal leishmaniasis' cases [Saravia et al, 1985; Barral et al, 1991; Kharfi et al, 2003; Morsy et al, 1995; Aliaga et al, 2003]. Visceral leishmaniasis –also known as Kala-zar- is the most severe form and is a systemic disease that is fatal if left untreated. Visceral leishmaniasis s caused by *L. donovani* (in East Africa and in India) and *L. infantum* (in Europe, Latin America and North Africa). Visceral leishmaniasis' patients present symptoms and signs of persistent systemic infection including undulating fever, fatigue, weakness, lost of appetite and weight loss; furthermore as the disease progresses is manifested by lymph node, spleen, liver enlargement, lymphadenopathies, and anaemia. After recovery, patients may develop a chronic cutaneous form called post-kala-zar dermal leishmaniasis that usually appears within two years after apparent clinical cure of the visceral infection [Salotra et al, 2006]. The post-kala-zar dermal leishmaniasis is characterized by skin lesions that can be of various types and initially are most prominent on the facial area.

Leishmaniasis includes two major epidemiological entities: zoonotic which includes animal reservoir hosts in the transmission cycle of the disease, and anthroponotic in which humans are considered to be the sole source of infection for the sandfly vector. From a point of view of leishmania control, priority has been given to anthroponotic foci, since these foci have been proven to be the source of visceral leishmaniasis' epidemics [Desjeux, 2004; Seaman et al, 1996]. Leishmaniasis has a worldwide geographic distribution and is endemic in 88 countries (66 in the Old and 22 in the New World). Specifically, leishmaniasis is endemic from northern Argentina to southern Texas (not in Uruguay, Chile or Canada), in southern Europe, Asia (not southeastern Asia), the middle east and Africa (particularly east and North Africa). Leishmania is not endemic in Australia or Oceania [Desjeux, 1996; Herwaldt, 1999; Koutis, 2007]. As mentioned earlier, in the case of Europe, leishmaniasis is endemic in all southern countries. Particularly, in southern Europe most of the reported cases are due to zoonotic visceral leishmaniasis, which is the most dangerous form with a high fatality rate when untreated; cutaneous leishmaniasis is also present. Approximately, a total of 700 autochthonous new cases are reported annually for southern European countries. However, if Turkey is included the reported cases per year are increased in 3950 [Dujardin et al, 2008]. However, autochthonous leishmaniasis appears not to be limited in the southern Europe, since some indigenous visceral leishmaniasis cases have been already reported in northern Italy and southern Germany [Maroli et al, 2008; Bodgan et al, 2001].

The estimated annual incidence is 1-1.5 million cases of cutaneous leishmaniasis; in the Old World over 90% of annual cases occur in Afghanistan, Algeria, Iran, Iraq, Saudi Arabia, and Syria, whereas in the New World the majority of cases occur in Brazil and Peru. On the other hand, the estimated annual number of new cases of visceral leishmaniasis is about 500.000; over 90% of worldwide cases are in Bangladesh, northeastern India (particularly in Bihar State), Nepal and Sudan (Old World), and in northeastern Brazil (New World). The estimated number of people at risk of infection is about 350 million [Herwaldt, 1999; Desjeux, 1996; WHO, 1990]. However, there is no doubt that the number of cases occurring around the world is considerably greater then the officially reported due to the fact that numerous cases are undiagnosed, misdiagnosed or unreported, especially when patients have no access to medical facilities; additionally, leishmaniasis is not always a notifiable disease and this could contribute to the underestimation of the new cases' number [Desjeux, 2004].

Currently, leishmaniasis has undoubtedly a wider geographical distribution than before; it is now reported in areas that previously were not endemic such as western Upper Nile in southern Sudan. Moreover, in several areas of the world there is an increase in the number of cases. This disturbing increase is not only attributed to environmental changes –which increase exposure to sandfly

vectors- but also to the increase of individual risk factors, which facilitate the evolution from infection to disease manifestation [Desjeux, 2001;]. In this regard, migration from rural to urban areas has lead to the urbanization of visceral leishmaniasis in Brazil and in countries of South America. Specifically, in the case of northeastern Brazil, several climatic and socioeconomic changes have lead to massive and unplanned migration from rural locations to main cities such as Fortaleza, Natal, Joao Pessoa, Sao Luis and Salvador do Bahia. These population movements have lead to the dramatically increase of the number of visceral leishmaniasis' cases and this increase has been attributed to the presence of domestic animal reservoirs in these areas. In particular, a possible association between human infection and the presence of dogs in around human dwellings was observed [Cunha et al, 1995]. The same phenomenon of visceral leishmaniasis' urbanization has been also been observed in Colombia and Venezuela [Aguilar et al, 1998]. In East African countries, the movement of seasonal workers, and the accumulation of refugees in areas where visceral leishmaniasis is endemic have played a major role in the dissemination of the disease and the rise of epidemics. The temporary migration of people from non-endemic to endemic areas is a risk factor for the disease dissemination, since these people can import the disease upon their return to a non-immune population. Specifically, a severe epidemic has occurred in Sudan with 100.000 deaths among a population of less than one million [Seaman et al, 1996]. Environmental factors also play an important role in the disease dissemination and maintenance; for example, the presence of cowsheds and organic debris nearby houses facilitates sandfly breeding sites. Moreover, the presence of ponds and the high sub-soil water level keep the level of humidity high. As a result of high humidity levels is the increase survival of the sandfly vectors [Thakur, 2000]. Additionally, the presence of cracks and crevices in the walls of houses made of mud and dried grass provide resting places for adult sandflies. Furthermore, malnutrition is another risk factor for severe visceral leishmaniasis' development. It has been shown in Brazil that children suffering malnutrition were 9 times more likely to develop visceral leishmaniasis [Cref, 1987]. The increase in leishmaniasis' cases worldwide is widely associated with the spread of HIV infection. . Immunosuppression is one of the major factors responsible for increased susceptibility to primary Leishmania infection or to reactivation of a silent infection. Leishmania and HIV coinfections have been reported in 35 out of 88 countries in which leishmaniasis is endemic and are a growing concern in Brazil, eastern Africa and the Indian subcontinent, where both diseases overlap geographically [Cruz et al, 2006; Koutis, 2007]. It should be mentioned that *Leishmania spp* resistance in some regions to antimonial drugs (the first-line drugs in the majority of developing countries) could be a novel risk factor for the disease's incidence increase [Croft et al, 2006].

CONTROL STRATEGIES & PREVENTION MEASURES

Since anti-leishmanial vaccines are still being developed, the current control strategies for leishmaniasis rely on case management (case detection and treatment), vector and reservoir control. Attention has been mainly focused on prevention strategies of visceral leishmaniasis, the form with the highest fatality rate. Nevertheless, prevention strategies should be also considered for cutaneous leishmaniasis, which is also a major burden for certain areas, with serious psychosocial effects [Reithinger et al, 2005; Koutis, 2007].

Case management that includes early diagnosis and treatment is essential for both individual patients and for the community. However, case management is difficult to be conducted and inefficient if feasible; this practice is mainly restricted by several factors such as the lack of access to affordable, active drugs, the incorrect prescribing and the poor compliance. Nevertheless, for visceral leishmaniasis, serological diagnosis is mainly based on

ELISA, IFA and Direct agglutination test; parasitological diagnosis can also be conducted using spleen, bone marrow and lymph nodes aspirates. Usually, for visceral leishmaniasis treatment first line drugs are being used such as the pentavalent antimonials; in developed countries second line drugs are being used (such as amphotericin B), but unfortunately the use of these drugs is being restricted to the developed areas due to their high cost. Diagnosis for cutaneous leishmaniasis is being relied on skin smears and treatment is based on pentavalent antimonials.

Sandflies are highly susceptible to insecticides and they are indeed susceptible to the same insecticides as Anopheles mosquitoes, the malaria vector. It is encouraging that even though sandflies possess the essential biochemical mechanisms for resistance development to various insecticides, the reports on resistance are few The only insecticide resistance for sandflies is for the organochlorine DDT (dichloro-diphenyl-trichloroethane) in India [Alexander et al, 2003; Davies et al, 2003]. In this regard, in 1950s', visceral leishmaniasis had almost disappeared from Indian subcontinent after the implementation of large-scale DDT spaying campaigns; however, the disease re-emerged after the cessation of these spraying programs. This has been mainly attributed to resistance development of *Phlebotomus argentipes* to DDT [Singh et al, 2001]. Sandfly control is now mostly dependent on pyrethroids. House spraying is focused on the control of endophilic sandflies (that rest mostly indoors after feeding). House spraying with the pyrethroid lambdacyhalothrin resulted in a risk reduction of cutaneous leishmaniasis by 60% and 54% in Kabul and in the Peruvian Andes, respectively [Reyburn et al, 2000; Davies et al, 2000]. However, residual spraying is much more effective in urban situations when every house and animal shelter is treated than in rural areas where relatively few dispersed houses are sprayed and the sandflies represent a small proportion of the vector population. Several studies have shown that pyrethroid-treated bednets provide 50-65% protection against leishmaniasis [Alexander et al, 2003; Reyburn et al, 2000; Alten et al, 2003; Jalouk et al, 2007]. The synthetic pyrethroids used for nets' treatment combine the properties of low to moderate mammalian toxicity, low volatility and high insecticidal activity [Alexander et al, 2003]. The use of insect repellents (such as DEET, Diethyl-toluamide) or protective clothing has been also being suggested as a prophylactic measure against leishmaniasis. The above are being suggested for people who are at risk for leishmania transmission such as travelers and soldiers in manoeuvres or hunters. DEET was effective for over 4 hours [Alexander et al, 2003]. Repellents applied to clothing rather than skin has been proposed as an alternative approach to personal protection against sandfly vectors. However, several studies have concluded that impregnated clothing to protect humans from sandfly vectors may be impractical [Alexander et al, 2003].

Where leishmaniasis is primarily zoonotic (Latin America, Mediterranean region, central and southeast Asia) leishmania transmission to humans may be reduced by targeting the animal reservoir. In this case, screening and treatment of domestic animals, especially of dogs has been proposed; Screening of dogs may allow culling without delay [Handman, 2001]; nevertheless, a recent study showed poor specificity of the diagnostic assay (75%) used for diagnosing the infected dogs [Reithinger et al, 2002]. On the other hand, treating infected dogs is not an effective control strategy as relapses are frequent and dogs can regain infectivity weeks after treatment, even if they are clinically cured [Alvar et al, 1994]. Besides, the widespread veterinary use of leishmaniasis drugs can lead to parasite' resistance. On the other hand, killing of the seropositive animals has also been proposed, but the efficiency and acceptability of this control method is increasingly debated [Tesh, 1995]. Several canine vaccine candidates are under study and one of them has been recently registered in Brazil for veterinary use against symptomatic canine leishmaniasis with 80% efficacy [Borja-Cabrera et al, 2002, 2004]. Treating dogs with topical

insecticide lotions can significantly reduce sandfly bites on dogs and consequently protect them from infection [Reithinger et al, 2001]; In this regard, insecticidal effect is long termed and it requires often regular reapplications; another control method of protecting dogs from the disease is to fit them with collars impregnated with insecticides, such as deltamethrin. The use of treated collars reduced sandfly bloodfeeding by up to 90% and reduced the risk of L. infantum incidence rates in domestic dogs in southern Italy [Maroli et al, 2001].

In conclusion, since there is no antileishmanial vaccine available control measures are dependent on personal protection from sandfly bites and on reservoir or vector control. Leishmaniasis control remains a difficult issue and eradication of the disease is even more difficult. The current leishmaniasis control programs have largely failed mainly because of the insufficient regional health delivery systems and due to the limited local resources. In this regard, WHO has classified leishmaniasis as an emerging and uncontrolled disease (belonging to category 1 of the diseases). There is also hope that the first leishmaniasis vaccine will become available within a decade. Until an efficient vaccine becomes commercially available, the identification of risk factors could greatly help in designing prevention strategies.

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