The Importance of Examining the Leishmania Parasite

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Abstracts

There are more than 20 different species of Leishmania (protozoan genus) that can cause human leishmaniasis. The most affected regions of the world are eastern and western hemisphere and the affected countries could be Mexico, Central and South America, southern Europe, Africa, Middle East and some parts of Asian continent. It is comparable to malaria and may cause death if not attended in time. Years back World Health Organization had declared leishmaniasis infection as NTD which means 'Neglected Tropical Disease'. But still alone in Saudi Arabia 40,000 cases are detected every year. This present study evaluates the genesis, symptoms, diagnosis and treatment of Leishmaniasis, also the importance of its detection will also be discussed.

Keywords: Leishmania, parasite, leishmaniasis, sand fly, treatment.

1. Introduction

Human beings use to suffer a number of parasitic diseases and leishmaniasis is one of them, looking at the effect of the same it is comparable to malaria and may cause death if not attended in time. Years back World Health Organization had declared leishmaniasis infection as NTD which means 'Neglected Tropical Disease'. Leishmaniasis is present in almost all parts of the world, only exceptions are Australia and Antarctica as the tropical conditions are not favorable for the growth of the parasite. But other than that, most of the developed and developing nations are affected by the same. However, the respective reasons for the spread of the parasite were the war between countries, human migration, changes in the habitat of sand flies and evne other related environmental changes including the climate change.

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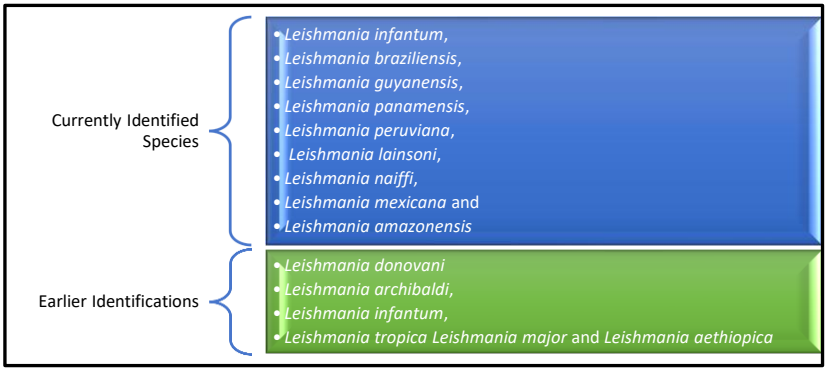
the affected countries could be Mexico, Central and South America, southern Europe, Africa, Middle East and some parts of Asian continent.



Source: Melissa et al (2019)

Figure 1: Identified Species of Leishmania

However, it should be acknowledged and kept in mind that the classification of Leishmania is problematic due to the use of different genetic markers in social studies. There have been repeated calls for the classification of this genus, but this has not yet been done. Leishmania infection is transmitted to humans through the bite of the Sandfly or female sandfly. Although rare, transmission through needle sharing, body transmission and sexual transmission have also been documented. The symptoms of leishmaniasis include two main types: mucocutaneous leishmaniasis, disseminated cutaneous leishmaniasis, disseminated leishmaniasis, regenerative leishmaniasis and cutaneous leishmaniasis such as post-kala-azar cutaneous leishmaniasis and kala-azar or visceral leishmaniasis. Although many overlaps, exceptions, hybrids and combinations exist, each event can be attributed to a species of Leishmania. It has also been found that Leishmania has mosaic aneuploidy, affecting genetic diversity not only within the species but also within the isolate. Leishmania symptoms can also be affected by the presence of sand fly saliva and cosmetics, and by host resistance.

WHO listed Leishmanian infection as NTD in 2007, but then again, the identification of the patients never stopped as in 2018 alone 2 lakhs of patients infected by Leishmania were identified and most of them were from the rural background, moreover 1 billion of people were under threat of getting infected. As a matter of fact, the cure or treatment of Leishmania is not so easy because of the financial constraint at times and some of the times due to geographical constraints. Due to the above given reasons the mortality rates are not reported as they should have been as in Bangladesh the fatality rate was around 2%, in Pakistan it was 13% and some of the middle eastern countries the rate was around 19%, then in middle east alone more than 40,000 cases were reported every succeeding year. As per the reports of WHO there is a decrease of cases as well i.e. the respective decrease was around 12% for all of the world.

Sandflies are the carriers of Leishmania and they are found all across the world, the fiercest condition lies in the fact that the life cycle if sand flies keep on going for whole year, irrespective

of the season, this is for the sandflies in tropical areas, but in sub-tropical areas the lifecycle of the sand flies takes place in summer season only. The reason for the growth of sand flies is that they are active in the night only and in the day time they are not visible to their predators.

The total number of cases recorded in Saudi Arabia, majority of the same can be attributed to frequent travel and immigration. Then there are some of the cases where infection increased due to climate change, increasing temperature, etc.

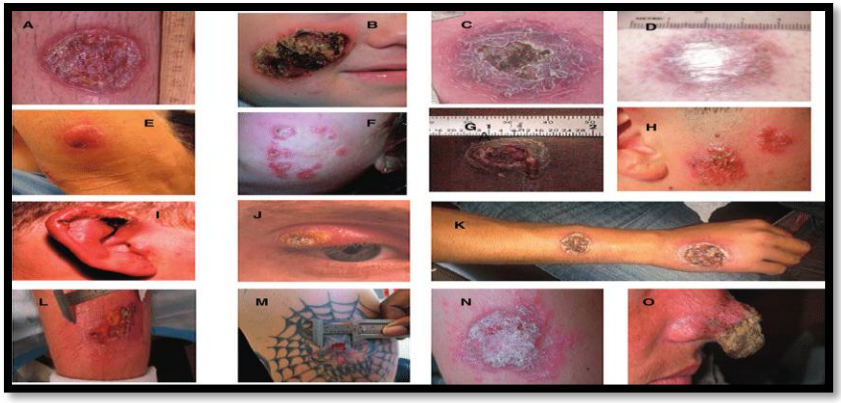
Life Cycle:

As stated above the female sand fly is the carrier of parasite and it is active in the night time i.e. ready to squander the parasite into humans and other species of animals. The parasite of Leishmania has two different phases of life cycle i.e. promastigote and amastigote. In the first stage a flagellum is developed in the intestine of the sand fly and while coming in contact with human blood stream the parasite is injected in skin of the host and the mononuclear cells of the host transform it into amastigote form. The amastigotes multiply and develop within the reticuloendothelial system of the host, causing either the asymptomatic or symptomatic forms of the disease based on underlying host and parasite species' factors. The amastigotes can travel hematogenous and lymphatically to cause mucosal and visceral disease.

The spread of Leishmania is driven primarily by symptoms and post-kala-azar cutaneous leishmaniasis, as asymptomatic cases are not expected to be transmitted through sand. In some areas, people need to control the life cycle of Lactobacillus tropicalis and Lactobacillus donovoni, which is characteristic. However, animals can live long lives and not show any signs or symptoms of the disease. Dogs, rodents, marsupials, monkeys and edentulous animals are susceptible. Dogs are the most important animal reservoir of L. infantis. Less commonly, leishmaniasis can be transmitted through organ transplantation, blood transfusion, injectable drugs or body piercing.

Symptoms:

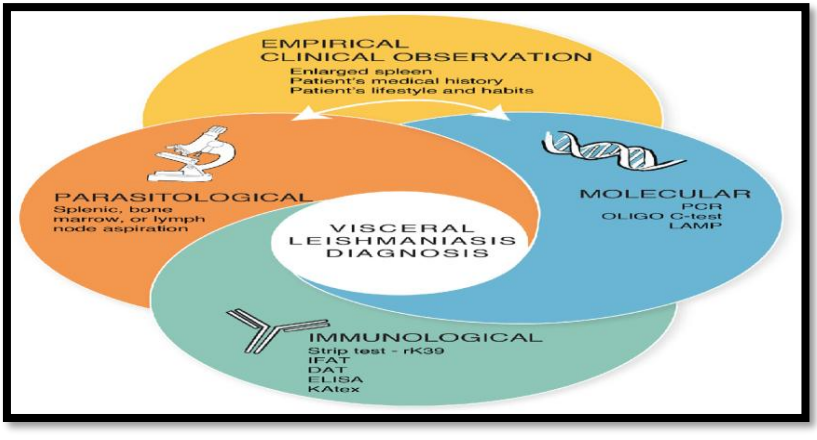
Although leishmaniasis can present with a variety of clinical manifestations, the three main phenotypic diseases are cutaneous leishmaniasis, mucosal leishmaniasis, and visceral leishmaniasis. These categories can be further subdivided into clinical diseases including American ML, New World VL, CL, Old World CL, post-Kala-Azar CL, Leishmania-relapsing, disseminated CL, and published CL. Skin diseases can also present in many forms. In some individuals, the infection may remain asymptomatic or subclinical, but it can also present acutely, subacutely, or chronically. CL disease usually occurs at the site of the sand bite (usually the right side of the face and flank) and occurs alone without purulent papules, although multiple lesions may occur. Over weeks or months, the papules develop into painless lesions with borders that may heal spontaneously or scar and worsen over several months. However, many atypical cutaneous findings occur, including nodular, sporotrichous, disseminated, psoriasiform, verrucous, zosteriform, eczematous, and/or erysipelas-like. Small bacteria outside the plaque/ulcer. All patients with leishmaniasis should be evaluated for evidence of mucosal disease by nasopharyngeal examination. Recurrent leishmaniasis presents as satellite lesions around old scars and is often confused with cutaneous tuberculosis.



Source: Aronson et al (2016)
Figure 2: Symptoms of leishmaniasis

Diagnosis:

As a matter of fact, the diagnosis of leishmaniasis is a difficult task but can be detected with parasitic and immunologic confirmation. The equipment and process is somewhat technical and need resources of distinct kind, this can be one of the reason that in some of the countries still the detection of leishmaniasis is not possible or may be a critical task to be performed. The nearest possible detections are being made on the basis of patient's history, epidemiology, clinical symptoms, and signs on physical examination; after working on the same further process are being performed at the level of testing, etc. Then on the other hand some of the obvious symptoms can be prolonged fevers, fatigue, weight loss, anemia, leucopenia, and hepatosplenomegaly in a patient from an endemic region such as Middle east or South African countries.



Source: Bengtson et al (2020)
Figure 3: Diagnosis of leishmaniasis

Given that treatment can be toxic and can vary greatly depending on the type of diagnosis, the diagnosis of leishmaniasis is inconclusive and confirmation requires tissue and/or molecular testing (e.g. polymerase chain reaction) for evidence of amastigotes. Since direct evidence is preferred, collection of appropriate specimens for clinical examination, culture and PCR is essential. Although PCR-based methods are more sensitive than culture or analysis, these methods are often not standardized and their availability is limited to treatments in large hospitals or centers. Tissue can be collected by scraping, biopsy or aspiration. For CL, clean tissue should be collected for PCR and culture, and biopsy should be taken from the smaller margins of disease for histological examination. Sensitivity for VL varies by tissue collection site; spleen biopsy yields the highest rate of positive samples (>90% sensitivity), but this technique carries some risk of life-threatening bleeding. Therefore, the preferred primary site is the bone marrow, although it has a lower incidence (50%-80%); other possible tissue sites include the liver, large lymph nodes, and whole blood, especially as an anti-inflammatory.

Table 1: Diagnostic methods of leishmaniasis

Disease pattern	Direct vs indirect	Diagnostic method	Comments
Cutaneous leishmaniasis	Direct	Biopsy, scraping, aspirate	Sensitivity dependent on expertise of pathologist and quality of specimen. Obtain from edge of ulcer and base
		Microscopy	Giemsa stained microscopy
		Culture	Amastigote typically 2–4 μm in diameter, round to oval in shape with nucleus and kinetoplast
		Histology	Special media, as organism is fastidious it can take weeks to become positive.
		PCR	Most sensitive, identifies species which is helpful in excluding ML associated species. PCR is also helpful in cases with low parasite burden.
	Indirect	CL Detect	Immunochromatographic assay for the rapid detection of *Leishmania* species antigen in ulcerative skin lesions Sensitivity 96%, specificity 90%
		Serologic tests (see below)	Not recommended for diagnosis of CL
Visceral leishmaniasis	Direct	Splenic aspirate (parasite isolation, culture, histology, and PCR per above)	Most sensitive (93–99%) compared to bone marrow and lymph node aspirate for diagnosing VL but risk of splenic hemorrhage
		Bone marrow aspirate	Bone marrow sensitivity (52–85%) sensitivity. Safer to perform than splenic aspirate
		LN Aspirate	Lymph node aspirate sensitivity (52%–58%)
		Peripheral blood	Assess blood for buffy coat, in vitro culture, and molecular analyses. Helpful in diagnosis for immunocompromised and HIV
	Indirect	Serological tests:	Cannot distinguish active from prior infection. Not helpful for CL. Often non-reactive in immunocompromised hosts.
		Rapid Diagnostic Test (rK-39)*	Detect specific antibody against antigen present in *L. donovani, chagasi-infantum* Results available in 20–25 min Easy to perform, quick and cheap- particularly helpful in underserved areas Sensitivity varies depending on region and parasite species Can cross react with other infections—for example Chagas disease
		Direct Agglutination Test (DAT)*	Uses whole organisms to look for antibody. Gives antibody tires ranging from 1:100 up to 1: 151200. Sensitive (>95%) and specific (>85%) test when performed correctly Needs well trained technician to perform over 2-3 days

Source: Aronson et al. CID, PAHO, Burza et al., Berman et al.

Treatment:

In general, treatment of leishmaniasis should be tailored to the patient, the type of leishmaniasis, and the subtype of the parasite. PCR testing for type identification is particularly useful in cultures of patients with skin infections who may be at risk for ML. Treatment of cutaneous leishmaniasis depends on presentation, host, and type. In immunocompromised individuals, CL caused by L. mexicana and E. coli without evidence of mucosal infection usually resolves spontaneously and does not require treatment. However, local treatment is recommended for simple lesions that do not resolve spontaneously, whereas treatment is recommended for

complex CL. Complex CL, defined as >5 lesions; species associated with mucosal disease. A variety of treatments are available (see Table 3), including pentavalent antimony (meglumine antimonate and antimony sodium gluconate), amphotericin (deoxycholate and liposomal formulations), pentamidine, and oral therapy (such as fluconazole and miltefosine). Pentavalent antimony is considered the standard of care for CL in Latin America. . Heat therapy is more commonly used in South America and is associated with significant pain relief and healing. Although there is no optimal treatment, experts recommend individualized treatment for patients based on published clinical reports of the location and nature of symptoms, immune system, concomitant diseases, pregnancy planning, adverse drug reactions, and animal species and geographic information. . Unlike CL, mucosal leishmaniasis does not resolve spontaneously and can lead to severe damage, destruction, and death (due to pneumonia or respiratory tract infection). Therefore, early treatment is essential after the diagnosis of nasopharynx is established. Bad breath. Similar to patients with complex CL, patients with ML should be treated individually (using the drugs previously described for CL). Individuals with symptoms and confirmed VL need early treatment. Pentavalent antimony was previously considered the standard of care, but there are concerns that Leishmania species may develop resistance. IDSA recommends liposomal amphotericin B for the treatment of VL in immunocompetent patients and miltefosine should be considered in nonpregnant, nonlactating patients. Pentavalent antimony may be used as an alternative agent at sites with known weak patterns in patients with leishmaniasis resistant to liposomal amphotericin B or miltefosine.

2. Conclusion:

Leishmaniasis is a complex medical condition that is difficult to diagnose and treat. Advances in vaccine development, diagnosis, education, and treatment can prevent serious illness and death from this disease. Recommendations include the use of isolation controls, internal controls, Leishmania control standards, repeated testing, and participation in external controls. Molecular methods are still expensive and require expertise compared to other diagnostic methods, and studies are needed to make PCR platforms more widely and effectively used, especially in regions where leishmaniasis is endemic.

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