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Current situation of Cutaneous Leishmaniasis and challenges of drug resistance: a narrative review

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Abstract

Leishmaniasis is a parasitic disease caused by a flagellated parasite of the genus Leishmania. It is a disease widely distributed in the world. It is primarily a zoonotic infection affecting men in a secondary way and transmission occurs through the bite of an infected sandflies. The disease has different clinical forms, but it can present as American Tegumentary Leishmaniasis (ATL) and as Visceral Leishmaniasis (LV) or kala-azar, the latter being the most severe form of the disease. American Tegumentary Leishmaniasis has two main clinical forms, Cutaneous Leishmaniasis and Mucous Leishmaniasis and within this classification they still present a wide variety of clinical manifestations. There are more than twenty species that cause the disease and the clinical presentation is strongly associated with factors related to this parasite-host relationship and the type of immune response triggered. The treatment of leishmaniasis has undergone a great deal of variation over the years and since 1940 the

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standard treatment has been intramuscularly administered antimonials. However, its toxicity requires attention in the follow-up of the treatment and the cases of resistance to the drug have increased, ceasing to be used as a drug of choice in some regions of the world.

Keywords: Leishmaniasis, epidemiology, treatment, drug resistance.

INTRODUCTION

American Tegumentary Leishmaniasis (ATL) is a non-contagious vector-borne disease, caused by a mandatory flagellated and intracellular protozoan, which belongs to the family Trypasonomatidae, order Kinetoplastida, *Leishmania* genus. There are more than twenty species capable of causing disease in humans and more than 30 species of mosquitoes vectors of Leishmaniasis (DESJEUX, 2004; SUNDAR; CHAKRAVARTY; MEENA, 2019). All species of *Leishmania* are transmitted by the bite of the infected female of dipterans of the subfamily Phlebotominae, belonging to the *Lutzomyia* genus in the New World, and Phlebotomus in the Old World (GONTIJO; CARVALHO, 2003).

Therefore, it is primarily a zoonotic infection, initially affecting other animals, and man may be secondarily involved (BRASIL, 2017). Most cases of cutaneous leishmaniasis in the Old World are localized and tend to spontaneously regress or progress to healing in a few weeks or months. In the New World, due to the different species of *Leishmania*, it presents a higher risk of developing the secondary mucous form (MOKNI, 2019).

The clinical presentation of Leishmaniasis can take two forms, Tegumentary Leishmaniasis (TL) and Visceral Leishmaniasis (VL). Leishmaniasis is present in 98 countries and three territories on five continents. It is estimated that in the world there are 200 to 400 thousand cases of VL and 0.7 to 1.2 million cases of Cutaneous Leishmaniasis (CL) annually. It is estimated that more than 90% of cases of VL occur in six countries: India, Bangladesh, Sudan, South Sudan, Brazil and Ethiopia. And about 70-75% of reported cases of CL come from ten countries: Afghanistan, Algeria, Colombia, Brazil, Iran,

Syria, Ethiopia, North Sudan, Costa Rica and Peru (ALVAR et al., 2012; WHO, 2010).

Among parasitic diseases, leishmaniasis represents the second leading cause of mortality worldwide, behind only malaria and is the third most common cause of morbidity in terms of years of life lost due to disability (in English, DALY´s) behind malaria and schistosomiasis. With the increase in the occurrence of cases in countries considered non-endemic, an effect of greater global mobility, it is important to know the disease and realize the relevance that it has (DIAZ; PONTE-SUCRE, 2018; KARIMKHANI, et al., 2016). Therefore, it is important to understand the different characteristics of the disease, the treatment options and their current situation in order to study new mechanisms to combat Leishmaniasis.

The aim of this study was to provide a comprehensive review of the literature, followed by a critical analysis of current challenges. In this context, our main objetive in this review was describes general characteristics associated with ATL, mainly in its cutaneous form, as well as its epidemiology, the methodologies currently used in the diagnosis and treatment of leishmaniasis and the current challenges to the problem, such as the development of drug resistance.

METHODOLOGY

This literature review addresses the main information about Leishmaniasis and the current challenges related to the disease, diagnosis and treatment. Bibliographic consultations were performed using Scopus, Science Direct, Google Scholar, CAPES and Pubmed Journals. The period of publications was from 1986 to 2020, and the keywords used were: “Leishmaniasis”, “treatment”, “treatment failure” and “drug resistance”. In the analysis of the initial list, data not related to the purpose of this review were excluded.

The *Leishmania* sp. parasite

The species of the *Leishmania* genus are all morphologically similar, presenting two main stages of development in their life cycle: the amastigote and promastigote forms. The amastigote form is intracellular, not flagellated, immobile, which resides in the reticuloendothelial cells of the vertebrate host and is 2 to 6 μm in

length. The promastigote form is flagellated and extracellular. It is presented in an elongated form, approximately 20 μm long and replicates freely in the digestive tract of the sandfly or in culture. They are digenetic organisms that have kinetoplast, glucosome, mitochondrial DNA. A unique feature of these parasites is the ability to change the number of copies of individual genes or groups of genes, chromosomes and even the entire genome. This genetic plasticity allows the parasite to amplify the number of copies of specific genes (DIAZ; PONTE-SUCRE, 2018; MOKNI, 2019).

Epidemiology

In the Americas, 18 countries are considered endemic for cutaneous and mucosal leishmaniasis. According to the Pan American Health Organization (PAHO) in 2017, 49.959 cases were reported by 17 endemic countries, with French Guiana reporting these data directly to France. Of these data, more than 70% of them were reported by four countries: Brazil, Costa Rica, Mexico and Ecuador. The most severe form, LV is considered endemic in 12 countries in the Americas. Nine countries reported more than four thousand cases of LV in 2017, with 97% of cases registered in Brazil (WHO, 2019).

The CL is divided according to its geographical distribution into CL from the Old World and the New World and different species of the disease in these two regions. Five species cause CL in the Old World, namely*, L. donovani*, *L. aethiopica*, *L. infantum, L. major and L. tropica* that occur in southern Europe, southwest Asia, Central Asia and Africa (GONTIJO; CARVALHO, 2003; SUNDAR; CHAKRAVARTY; MEENA, 2019).

In the New World, TL can be caused by different phylogenetically distinct species. And although a particular species or subgenus is commonly associated with certain clinical manifestations, they are not unique to a species. TL can be caused by different species of the subgenus *Leishmania (L) and Viannia (V)*, they are*: Leishmania (V) braziliensis*, *L. (V) guyanensis*, *L. (V) panamensis*, *L. (V) peruviana*, *L. (V) lainsoni*, *L. (V) lindenberg*, *L. (V) shawi*, *L. (L) Mexican*, *L. (L) amazonensis* and atypically *L. (L) infantum*, which despite being associated VL, there are cases of TL in Central America in endemic areas of VL (GRIMALDI; TESH, 1993; WHO, 2010; WHO, 2019).

Leishmaniasis have different transmission patterns with different vectors, reservoirs and agents, which makes it difficult to control the disease. In humans, after being bitten by the infected vector mosquito, ATL has an incubation period of two to three months, on average, and can vary from 10 days to two years (BRASIL, 2017; GONTIJO; CARVALHO, 2003). Thereafter, the infection may progress asymptomatically or progress to CL, progressing like an erythematous papule progressing slowly to nodules, plaques and ulcers with raised, hardened edges and coarse-grained tissue (BRASIL, 2017; GONTIJO; CARVALHO, 2003).

Clinical classification

There are different proposals for clinical classification for leishmaniasis. In the most widely adopted classification, ATL presents different clinical forms, including inapparent and lymphatic infection. Classically, it manifests itself in two clinical forms: as Cutaneous Leishmaniasis (CL), the most common, which can be disabling when presenting with multiple lesions and Mucous Leishmaniasis (LM) that in most cases occurs after skin lesions. These two forms still have a broad clinical spectrum, with different clinical manifestations. Thus, leishmaniasis can be classified according to the group to which they belong. In the group of cutaneous involvement or Cutaneous Leishmaniasis we have Localized Cutaneous Leishmaniasis (LCL), Diffuse Cutaneous Leishmaniasis (DCL), Disseminated Cutaneous Leishmaniasis, Cutaneous relapse CL (CRL). The mucosal involvement group or Mucous Leishmaniasis has the late mucous form, the mucous form without previous skin lesion, concomitant mucous form, contiguous mucous form and the primary mucous form. And finally, the group of lymph node involvement with Lymph Leishmaniasis (BRASIL, 2017; MARZOCHI; MARZOCHI, 1994; MOKNI, 2019).

Cases of unapparent infection do not present a current or previous cutaneous lesion of TL. Detection is done by indirect methods through positivity in the Montenegro skin test, by detecting specific antibodies or by detecting the parasite's DNA using molecular methods. However, the use of serological methods to detect these cases is controversial, since no serological method has been validated for this purpose, mainly due to the risk of cross reactions.

Asymptomatic cases have no indication of specific treatment (BRASIL, 2017).

Localized Cutaneous Leishmaniasis (LCL) is the most common form of ATL, with a tendency to spontaneous cure and has a good response to treatment. It has a single lesion or some lesions that are usually ulcerative and close to the inoculation site. After the bite of the infected sandfly and the incubation period, which can vary from 10 days to three months, a small, low, pruritic inflammatory papule appears that progresses slowly to a nodule. The nodule has a rounded or oval shape and progresses to a central ulcer, with a raised, hardened border and satellite papules may appear around the central lesion. The localized form may have regional lymphadenopathy and nodular lymphangitis. In the northern region of Brazil, multiple injuries are mainly caused by *L.* (*V.*) *guyanensis*. No estado do Amazonas cinco espécies estão relacionadas a infecção: *L.* (*L.*) *amazonensis*, *L.*(*V.*) *braziliensis*, *L.*(*V.*) *naiffi*, *L.* (*V.*) *lansoni* e a espécie mais prevalente *L.* (*V.*) *guyanensis* (FIGUEIRA et al., 2014; GONTIJO; CARVALHO, 2003; MOKNI, 2019; FRANCESCONI et al., 2018).

There are many morphological variations described in the literature due to the remarkable polymorphism of the lesions, but this variation does not always have any relevance in the clinic. Among these there are impetigoid, lichenoid, tuberculous or lupoid, nodular, vegetative, erysipeloid, sporotrichoid, psoriasiform, mycetomatous, periungual, eczematous, ectimatoid, zosteriform, annular, palmoplantar, cicatricial, squamous cell carcinoma *like*. This wide variation is generally related to the injury site, the species involved, the time of evolution and the type of inflammatory reaction triggered (GONTIJO; CARVALHO, 2003; MOKNI, 2019).

The Diffuse Cutaneous Leishmaniasis (DCL) is a rare and severe clinical form, presenting multiple non-ulcerated lesions and formation of plaques and nodules that cover large skin extensions. And it is characterized as a long-term disease due to immunosuppression caused by the parasite. In Brazil, this clinical form is associated with the species *L.* (*L.*) *amazonensis*. Most cases of CDL arise from an infection acquired in childhood, which due to the immaturity of the immune system, the parasite multiplies without control leading to an increase in the number of lesions and wide

distribution in the skin extension (BRASIL, 2017). In CDL, treatment is difficult or ineffective due to specific immunodeficiency and is characterized by frequent relapses. The Intradermorretion of Montenegro (IDRM) is negative and serology positive, this is due to the predominance of the secretory type response to the detriment of the cellular response, maintaining high levels of circulating antibodies (BRASIL, 2017; MARZOCHI; MARZOCHI, 1994).

Disseminated CL progresses with numerous lesions (more than ten lesions), usually small and ulcerative in two or more non-contiguous body segments, distant from the inoculation site. The species most commonly associated with this form is *L. braziliensis*, but also Were reported cases of *L. guyanensis*, *L. panamensis* e *L. amazonensis*. Skin relapse CL (CRL) leads to active nodular lesions around or within the scar of a previous lesion of CL. The therapeutic response is usually less than that of the primary lesion. Lymphatic Leishmaniasis is characterized by an increase in lymph nodes and other structures of the lymphatic system. The term ascending nodular lymphangitis refers to inflammation of the lymph cord and lymph nodes in the path from the injury site to the regional drainage lymph nodes. This form is also called sporotrichoid, similarly to sporotrichosis, but does not show a tendency to fistulization (BRASIL, 2017; BITTENCOURT, 1993; DESJEUX, 2004; MARZOCHI; MARZOCHI, 1994).

The other form of TL, Mucous Leishmaniasis (ML), has a chronic evolution and greater destructive potential. If not treated properly, it can leave severe deformities and sequelae located in the mucous membranes of the upper airways, such as the mucous membranes of the oral, nasal, pharynx and larynx. There may also be a concomitant involvement with the cutaneous lesion, called cutaneomucosal leishmaniasis. Visceral Leishmaniasis (VL) is a systemic manifestation that usually leads to death when not treated (BRASIL, 2017; DESJEUX, 2004; SUNDAR; CHAKRAVARTY; MEENA, 2019).

Diagnostic methods

The diagnosis of ATL generally requires the association of epidemiological, clinical and laboratory elements due to the complexity of the clinical spectrum of leishmaniasis and the diseases

that make a differential diagnosis with TL (GONTIJO; CARVALHO, 2003; OPS, 2019). The diagnosis of certainty of TL is made in the detection of the parasite or its products in tissues or biological fluids of the host, through direct and indirect diagnostic methods. Direct methods are those that allow the detection of the parasite in the sample. Examples of direct methods are direct parasitological examination, culture, histopathological examination and Polymerase Chain Reaction (PCR). When it is not possible to visualize or isolate the parasite, the diagnosis is made with clinical data accompanied by an indirect diagnostic test. Indirect tests are those that allow the detection of specific antibodies against species of *Leishmania*, mainly those of the IgG type. Examples are, indirect immunofluorescence (IIF), indirect enzyme immunoassay (ELISA) and IDRM (BRASIL, 2017; OPS, 2019; ESPIR et al., 2016).

In Brazil, the Ministry of Health recommends that the suggestive diagnosis should be accompanied by direct research of the parasite and/or IDRM. However, IDRM is in disuse due to low production, lack of trained professionals in health services to identify parasites in smears and lack of distribution of the antigen, the product essential for its execution. After these, if a conclusive diagnosis is not obtained, other diagnostic methods must be performed, such as serological, parasitological and molecular exams (BRASIL, 2017).

Laboratory diagnostic methods vary according to the limitations of each technique and vary with the experience of the health professional, quality of equipment, inputs, time of disease progression and the different types of etiological agents that cause leishmaniasis (BRASIL, 2017). Serological tests for the diagnosis of CL, for example, are of limited use because they have low sensitivity and varying specificity, but are very useful in the diagnosis of CL (OPS, 2019).

The direct parasitological exam is the exam of first choice because it is a faster, cheaper and easier to perform technique. In this technique, it is possible to identify the amastigote forms, but it is not possible to identify the species (BRASIL, 2017).

Tissue samples can be subjected to histopathological analysis. TL presents a typical histopathological picture of diffuse ulcerated granulomatous dermatitis. In most cases, the granulomas observed

are typified as "tuberculoids", with associated lymphoplasmacytic inflammatory infiltrate and, occasionally, necrosis. It is also characteristic of TL, malformed granulomas, characterized as poorly defined aggregates of activated macrophages, also called "Montenegro glades". In an extensive study conducted in Brazil, five histopathological patterns were identified: 1) Exudative cell reaction; 2) Exudative and necrotic reaction; 3) Exudative and necrotic-granulomatous reaction; 4) Exudative and granulomatous reaction and 5) Exudative and tuberculoid reaction (BRASIL, 2017; GONTIJO; CARVALHO, 2003; MAGALHÃES et al., 1986; SILVEIRA et al., 2008).

The TL is considered to be a chronic inflammatory disease, histiolinfoplasmocyte, with or without tissue necrosis and granulomatous reaction (MAGALHÃES et al., 1986; SILVEIRA et al., 2008). The diagnosis of certainty occurs in the visualization of amastigote forms of the parasite, characterized by being rounded or oval, with nucleus and kinetoplast. However, visualization of the parasite is not always obtained, making histopathological findings, in this case, only suggestive of Leishmaniasis. This technique also does not allow identification at the species level (GONTIJO; CARVALHO, 2003).

The isolation in in vitro culture is a method of confirming the presence of the etiological agent of the disease, in addition to enabling subsequent identification at the species level. Here, the cutaneous or mucosal fragments obtained in the biopsy of the lesion's edge are inoculated in Novy and McNeal's blood agar culture medium modified by Nicolle (NNN) and *Liver Infusion Triptose* (LIT) between 24ºC and 26ºC. Promastigote forms can be found from the fifth day. However, you must wait a month before releasing a negative result (BRASIL, 2017; GONTIJO; CARVALHO, 2003).

In vivo isolation, despite high sensitivity among other parasitological methods, is little used because it is more complex and expensive. The method consists of inoculation via Intradermal, the muzzle and / or hind legs of hamster (*Mesocricetus auratus*) of the material obtained by biopsy, already crushed in sterile saline. The animals are observed for a period of three to six months. The appearance of the first tissue lesions is usually noticed after the first month. Among the disadvantages of the technique are the long time

until the result is released and the limitation of the insulation effectiveness, which is quite varied depending on the species of *Leishmania* (BRASIL, 2017; GONTIJO; CARVALHO, 2003).

Among the immunological tests, the Montenegro Intradermoreaction test (IDRM) is based on the delayed cell hypersensitivity response, which can be negative in the first four to six weeks after the appearance of the first skin lesion (BRASIL, 2017). This test does not allow to differentiate a current disease from a previous one, as most of the time it remains positive after clinical cure. It also does not differentiate between disease and parasite exposure without disease (BRASIL, 2017; GONTIJO; CARVALHO, 2003).

The technique consists of applying 0.1 mL of standardized antigen to the flexor surface of the forearm, with a concentration of 40 mg of protein nitrogen per ml. If after 48 or 72 hours a hardness of five mm or more is detected, the result is considered positive (GONTIJO; CARVALHO, 2003). A positive IDRM, in endemic areas, can be interpreted as previous disease, previous application of IDRM antigen, infection, allergy to the test diluent or cross-reaction with other diseases. Thus, in these regions the positivity of the test varies from 20 to 30% (BRASIL, 2017).

Among the serological tests available for the diagnosis of TL, the most used are indirect immunofluorescence (IIF), immunoblotting (Western Blot) and the immunoenzymatic test (ELISA). Serological tests have varied sensitivity and specificity, depending on the technique adopted and the clinical form analyzed (GOMES et al., 2014; GONTIJO; CARVALHO, 2003). Among these, the indirect immunofluorescence reaction is the most used. This technique, despite its good sensitivity, presents cross reactions with other diseases, especially with Chagas and Calazar's disease. The IIF result in TL varies with the parasite's reduced antigenicity or low levels of circulating antibodies. Despite generally presenting negative results in diffuse cutaneous form, its sensitivity was estimated at 71% in cutaneous forms and 100% in mucous form (GONTIJO; CARVALHO, 2003). As the levels of anti-*Leishmania* antibodies in TL do not remain high after treatment, a positive result usually indicates a current infection (ZERPA; PADRON-NIEVES; PONTE-SUCRE, 2018).

Presenting a sensitivity of up to 100% in the diagnosis of LT, Western Blot is a sensitive technique, useful in identifying asymptomatic patients and is also much less invasive than the parasitological methods that require a skin biopsy. However, when compared to direct examination, for example, it is a more expensive technique and requires a larger laboratory structure to be performed (GOMES et al., 2014; POMARES et al., 2012; SZARGIKI et al., 2009).

The serological tests are therefore of limited application in the diagnosis of leishmaniasis. In the literature, its use is indicated to complement the diagnosis made through clinical findings and other exams (GOMES et al., 2014; POMARES et al., 2012; ROMERO et al., 2005).

The polymerase chain reaction (PRC) is the most used technique for the detection of parasitic DNA in the sample and allows molecular typing without going through a previous culture step. It is more sensitive than traditional techniques, however attention is needed when choosing primers, as this can directly influence the sensitivity of the technique. Real-time Polymerase Chain Reaction (qPCR) has also been useful in the diagnosis and monitoring of the therapeutic response as it performs quantitative analysis of the protozoa in ATL lesions (MOKNI, 2019; NEFFATI et al., 2011).

Treatment

Currently, there is a regional variation in the treatment of ATL, based on the experience of the local scenario since different species respond differently to the available therapeutic options. In the Old World, most patients with CL can be treated topically. In the New World, these patients are treated systemically, since endemic species present in the region usually cause metastasis and can evolve to more severe forms, such as ML (SUNDAR; CHAKRAVARTY; MEENA, 2019).

Pentavalent antimonials

In use since the 1920, antimonials have long been used as a drug of first choice and remain the main drugs used to treat different forms of leishmaniasis. Antimonials were first used by the Brazilian doctor Gaspar Viana, in 1912, describing the curative action of trivalent antimony (Sb^{III}) or emetic tartar, obtaining satisfactory results (VIANNA, 1912).

From 1940 on, pentavalent antimonials (Sb^v), as well as aromatic diamidines (pentamidine) started to be used in the treatment of leishmaniasis. Pentavalent antimonials, such as Pentostam® and Glucantime®, are available in two forms, such as sodium stibogluconate (SSG) and meglumine antimoniate (MA), respectively (PONTE-SUCRE; PADRON-NIEVES, 2018; SUNDAR; CHAKRAVARTY; MEENA, 2019). Currently, the WHO recommends, in the Americas, the administration of pentavalent antimonials, intramuscularly or intravenously, at a dose of 10 to 20 mg/kg/day, in a single dose for 20 or 30 days (OPS, 2019; SUNDAR; CHAKRAVARTY; MEENA, 2019).

Intravenous Sb^v treatment for leishmaniasis is long-lasting, causes pain at the injection site, and can produce more serious adverse effects, from cardiac arrhythmias, abdominal pain, skin rashes, changes in liver and kidney function, pancreatitis, etc. Adverse reactions contribute for many to abandon treatment, totally or partially. This favors the development of drug resistance and the selection of more resistant parasites (PONTE-SUCRE; PADRON-NIEVES, 2018). Studies have shown a failure to respond to treatment of up to 30% in endemic areas and this resistance has paved the way for the use of new drugs, mainly in the Indian Subcontinent (ISC). Still, Sbvs are still in use as a drug of choice in Latin America and East Africa (AREVALO et al., 2007; SUNDAR; CHAKRAVARTY; MEENA, 2019).

Amphotericin B

An antimicrobial in the polyene class, Amphotericin B (AmB) targets the membrane of promastigotes and amastigotes and is administered mainly intravenously and is approved for use during pregnancy. Used in the last 70 years as intravenous deoxycholate (d-AmB) in the systemic treatment of fungal infections and since 1960 in the treatment of different forms of leishmaniasis, it is a very toxic medicine. Its nephrotoxicity is the main adverse effect associated with the use of d-AmB. Other known effects are acute infusion reactions, anemia, neutropenia, liver disorders and thrombocytopenia. Thus, d-AmB has been progressively replaced by less toxic liposomal formulations (L-AmB) (FALCI; PASQUALOTTO, 2015; MOSIMANN et al., 2018).

In order to improve bioavailability orally or topically and seek a cheaper, more efficient and safer drug, new bioavailability systems for amphotericin B have been created. The forms tested in recent years include liposomes, solid lipid nanoparticles, nanoparticles based on polymers and proteins, nanocapsules, nanococleates, complexes with the inclusion of cyclodextrins, dendrimers, emulsions, drug conjugates and microneedles. However, due to the complexity of these new formulations and the required safety and efficacy prerequisites, we do not yet have a nanoformulation available for clinical use (LANZA et al., 2019).

As for the effectiveness of using L-AmB in the treatment of VL, 95 to 100% efficacy, caused by *L. donovani* and *L. infantum*, has been reported in the Indian Subcontinent (ISC) and southern Europe, respectively (SUNDAR et al., 2008). The effectiveness of L-AmB contributed to this medication becoming the main therapy against leishmaniasis in regions that are not responsive to antimonials. However, there is a geographical variation in the effectiveness of this drug, with low efficacy against VL in East Africa being reported. It is also unclear whether this variation is related to factors linked to the host or the parasite, or to a combination of both (LANZA et al., 2019; PURKAIT et al., 2012).

Few studies report the effectiveness of L-AmB in the treatment of CL and ML in the Old and New World, with some documented case reports. Today the clinical importance of amphotericin B is also threatened by the emergence of new cases of acquired resistance to the drug. Studies seek to understand the mechanisms that have led to resistance. Changes in membrane composition, in ABC transporters with MDR-1 overexpression and changes in thiol metabolic pathways, are some identified mechanisms that have been shown to influence the acquisition of amphotericin B resistance in clinical isolates of *L. donovani* (LANZA et al., 2019; PURKAIT et al., 2012).

And despite the improved risk of toxicity, L-AmB still has a high cost, which in turn ends up restricting its use, especially in emerging countries and environments with limited resources, such as Brazilian public hospitals (FALCI; PASQUALOTTO, 2015).

Pentamidine

Another drug used is marketed in its lyophilized form of pentamidine isothionate. Pentamidine is an aromatic diamidine which comes in the form of two salts, pentamidine isothionate and mesylate. Isothionate is more widely used, as mesylate has greater pancreatic toxicity. This drug accumulates in the parasite's mitochondria and inhibits mitochondrial topoisomerase II, binding to sites rich in adenine-thymine (AT) sequences in the minor groove of mitochondrial DNA (in the kinetoplast), inhibiting the transcription process (BRASIL, 2017; NEVES et al., 2011; SOARES-BEZERRA; LEON; GENESTRA, 2004). In Brazil, the cure rate was higher than 90%. It is especially reserved for the use of *L. guyanensis* and *L. panamensis*. Among the possible adverse effects, rhabdomyolysis is frequently observed, but usually without renal complications. And the occurrence of transient diabetes can also be observed (CORREIA et al., 1996; MOKNI, 2019).

Pentoxifylline

The Pentoxifylline is another drug with a potential leishmanicide that belongs to the class of peripheral vasodilators. It acts as a TNF-α modulated agent and is recommended by WHO and PAHO as an adjunct in the treatment of ATL. In Brazil it is recommended as an adjunct in association with meglumine antimoniate, reducing its toxicity and without indication as a single therapy. It has shown good results, decreasing the treatment time until the cure outcome in LMA. Although useful in the treatment of ML, it has shown no additional benefit in the treatment of CL in combination therapy with Sb^{v}, not being indicated in the treatment of CL. A tablet with 400 mg of Pentoxifylline is administered orally two or three times a day for 30 days. Its use is contraindicated in pregnancy and children (BRASIL, 2017; BRITO et al., 2017).

Paromomycin

Paramomycin, a drug that belongs to the aminoglycoside family, is an aminoglycoside antibiotic extracted from cultures of *Streptomyces rimosus* var. *paromomicinus*, identical to the aminosidine produced by *Streptomyces chrestomyceticus* (SUNDAR et al., 2007). It has proven leishmanicidal activity and is currently an alternative drug to the treatment of leishmaniasis and has been used in parenteral and

topical administration. In ISC, it is used intramuscularly at a dose of 12, 16 or 20 mg of sulfate/ kg/21 days, showing good efficacy in the treatment of VL. However, the long period of intramuscular administration is a major challenge for correct adherence to treatment. In some countries they are used in association with pentavalent antimonials. As for its effectiveness, it presents controversial results depending on the clinical form of leishmaniasis and the route of administration of the paramomycin (SUNDAR et al., 2007; SUNDAR; CHAKRAVARTY; MEENA, 2019).

In Brazil, its systemic use was more than 90% effective in treating primary skin lesions caused by *L. (V.) braziliensis*. And in India it presented results not inferior to the treatment of VL when compared to the treatment with amphotericin B. However, it presented low efficacy in the cases of mucocutaneous leishmaniasis. The Paramomycin resistance is not known, as it has not yet been used extensively. However, on *in vitro* trials with two *L. aethiopica* isolates from relapsed CL patients, they were 3 to 5 times less sensitive to the drug after treatment. Few studies suggest that this resistance is related to decreased drug uptake, due to decreased synthesis of cytoplasmic proteins responsible for the initial binding to the cell surface and consequent reduction of drug accumulation (CHAKRAVARTY; SUNDAR, 2010; CORREIA et al., 1996; PONTE-SUCRE et al., 2017).

Miltefosine

Miltefosine belongs to the class of alkylphosphocholines, which are phosphocholine esters of long chain aliphatic alcohols, and was initially developed for the treatment of cancer. In addition to acting against cancer cells, miltefosine has proven activity against many species of parasites, some pathogenic bacteria and fungi (DORLO et al., 2012). The exact mechanism of molecular action of miltefosine against *Leishmania* is not yet fully understood, but it involves changes in phospholipid biosynthesis, changes in phospholipase activity, inhibition of cytochrome c oxidase activity, mitochondrial depolarization, intracellular ATP levels and cell death by apoptosis (DORLO et al., 2012; PONTE-SUCRE et al., 2017).

It was registered in India in 2002 as the first oral medication available against leishmaniasis, with cure rates above 94% in the

treatment of VL. Thereafter, it replaced the use of pentavalent antimonial (SSG) as the treatment of choice at ISC. However, only ten years after its adoption at the ISC, its effectiveness was reduced, presenting after 12 months of follow-up, relapse rates of 10% and 20% in India and Nepal, respectively (RIJAL et al., 2013; SUNDAR et al., 2008; SUNDAR; CHAKRAVARTY; MEENA, 2019).

Because it has a long elimination half-life, which allows subtherapeutic levels to remain for a few weeks after standard treatment, it was predicted that mechanisms of resistance to miltefosine would soon appear in regions where it was widely used (PONTE-SUCRE et al., 2017; SUNDAR; OLLIARO, 2007). With increasing reports of non-responsive or reduced efficacy of miltefosine in India and Nepal, a recent study confirmed in laboratory two clinical isolates resistant to miltefosine. In these findings, it was observed that the resistance mechanism was associated with a lower internationalization of miltefosine caused by mutations in the gene that encodes the protein responsible for the entry of miltefosine in the cell, LdMT, a member of the P4-ATPase subfamily. When this protein is overexpressed, internalization in the drug increases up to 20 times and also increases susceptibility in promastigotes (GAMARRO; SANCHEZ-CANETE; CASTANYS, 2013; SUNDAR; CHAKRAVARTY; MEENA, 2019; SRIVASTAVA et al., 2017).

Table 1. Dose, route administration and clinical adverse effects according to drug used for the treatment of cutaneus leishmaniasis.

Drug	Dose	Route of administration	Treatment time	Clinical adverse effects
Pentavalent antimonials (*SSG* and MA)	10 to 20 mg/kg/day, in a single dose	Intramuscularly or Intravenously	20 or 30 days	Cardiac arrhythmias, abdominal pain, skin rashes, changes in liver and kidney function, pancreatitis.
Amphotericin B Deoxycholate (d-AmB)	0.7 to 1.0 mg/kg/day, daily or in alternate doses.	Intravenously	daily (15 to 20 days) or in alternate doses (30 to 40 days)	Fever, headache, nausea, vomiting, anorexia, tremors, chills, phlebitis, cyanosis, hypotension, hypokalemia, hypomagnesaemia, impaired kidney function, anemia and behavioral disorders.
Amphotericin B lipossomal (L-Amb)	2 to 5 mg/kg/day, until reaching a total dose of 25 to 40 mg / kg.	Intravenously	10 days	Nausea, vomiting, headache.
Pentamidine	3 to 4 mg/kg/day, in alternate days,	Intramuscularly		Pain, induration and abscesses (if Intramuscularly), nausea, vomiting, dizziness, adynamia, myalgia, headache, hypotension, lipothymia, syncope, hypoglycemia and hyperglycemia.
Pentoxifylline	One 400 mg tablet, three times a day, for 30 days.	Orally	30 days	Flush (facial flushing with a feeling of heat) and gastrointestinal disorders, such

				as feeling of gastric pressure, fullness, nausea, vomiting or diarrhea.
Paramomycin	12, 16 or 20 mg of sulfate/ kg/day	Intramuscularly	21 days	
Miltefosine	2.5 mg/kg/day, maximum dose of 150 mg/day orally	Orally	28 days	Nausea, vomiting, abdominal pain and diarrhea

Fonte: BRASIL, 2017; CHAKRAVARTY; SUNDAR, 2019; SUNDAR; CHAKRAVARTY; MEENA, 2019.

Prevention and control

Despite efforts to develop and analyze an effective vaccine, no vaccine candidate has emerged from clinical trials. Improvements in general sanitary and hygiene conditions help to reduce the vector population. In some regions preventive measures to eliminate the mosquito vector of malaria with the application of chemical insecticides helped to eliminate leishmaniasis in these places, returning with the interruption of this activity. Individual protection measures such as the use of repellents, clothes and mosquito nets with permethrin impregnation to reduce the contact of the transmitting mosquito and the human population at home also have some effectiveness, reducing the risk of disease transmission. Until today, disease prevention and control measures are associated with vector control measures and human reservoirs, the latter in relation to timely diagnosis and adequate treatment, in addition to strategies appropriate to each transmission pattern (BRASIL, 2017; MOKNI, 2019; OLIVEIRA; DUTHIE; PEREIRA, 2019).

CONCLUSION

Despite its relevance, leishmaniasis has been one of the most important neglected tropical diseases for decades. And even with the efforts of the scientific community aimed at the development of new drugs and vaccines, bringing good advances in this direction, there is no prospect of reaching the ideals of treatment in the near future. The first and second choice medications are for systemic treatments and are associated with toxicities that can limit the completion of a complete course of the therapeutic regimen. In addition, they are for parenteral administration, which makes patient compliance difficult.

The standard treatment remains old, based on the administration of pentavalent antimonials. Attention is also given to cases of resistance to drugs used in the treatment of leishmaniasis.

With increasingly frequent reports of resistance, it became urgently necessary to develop new drugs with less toxicity, non-parenterals and easy adherence to treatment. The combination of approaches that seek a better understanding of the action potential of emerging drugs, from preclinical trials to more advanced phases and those that aim to better understand the parasite-host relationship can accelerate the process of developing new leishmanicidal drugs.

CONFLICTS OF INTEREST

The authors declare no conflicts of interest.

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