REVIEW

Leishmaniasis

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Epidemiology, disease patterns, immunology, diagnosis, treatment and control measures of leishmaniasis are described. Various issues relating to leishmaniasis are highlighted: the relative lack of importance given to this disease is compared with other infections, climate change and its possible effect on extension of endemicity of this infection, and new diagnostic tests that are helping better diagnosis, especially in resource-poor areas. Other important aspects discussed include the potential for newer oral treatment to change the way this disease is managed; leishmania–HIV coinfection and groups at risk; and the development of an effective vaccine.

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Infectious diseases steal the headlines on a regular basis and are ranked high among other major news items, such as natural disasters, conflict situations and terrorism. Emerging infectious diseases with wide-threatening potential, such as SARS and pandemic influenza, are usually the ones that get best coverage and, consequently, better funding. Diseases with high prevalence, such as HIV/AIDS and malaria (table 1), should bring better financial returns from investing in research on new treatments and developing vaccinations than similar investment in research on infections, with lower prevalence. Leishmaniasis is one such infection, which rarely shares this limelight and thus largely remains a neglected disease.[2 3]

Despite this, several issues regarding leishmaniasis merit discussion:

- Resistance to conventional drug treatment has developed in certain areas of the world, necessitating a change of preferred agents
- Rapid, less invasive diagnostic procedures have been developed, which are most useful in poorly resourced parts of the world
- Despite advances in the understanding of the immunology of the disease and the unravelling of the *Leishmania* genome, a vaccine has not been developed yet
- The different extent of disease in different people emphasises the complex immunology of leishmaniasis, brought to the fore more recently with the advent of HIV–leishmania coinfection and its difficult eradication in this scenario (box 1).

This review article is based on information from bibliographic research, Pubmed searches on leishmaniasis, review articles and papers in their reference lists, and from the authors' personal archives.

EPIDEMIOLOGY

Leishmaniasis is endemic in more than 60 countries worldwide,[2] including southern Europe, North Africa, the Middle East, Central and South America, and the Indian subcontinent. It is not endemic in SouthEast Asia and Australia.[4] The burden of cutaneous leishmaniasis disease (90% of cases) is borne by Afghanistan, Pakistan, Syria, Saudi Arabia, Algeria, Iran, Brazil and Peru and for visceral leishmaniasis by India, Bangladesh, Nepal, Sudan and Brazil[1] Recently, the number of reported cases and geographical areas have increased,[5] and this has sparked concern regarding the contribution that global warming might have on this observation.[6 7]

One of the causative organisms of leishmaniasis, *Leishmania donovani*, was first described in 1903 by Leishman and Donovan almost simultaneously.[4] *Leishmania* is a protozoon that is able to infect animals, humans and sandflies. There are at least 20 species of *Leishmania*. Each may cause disease specific to the species and the host response. Organism prevalence differs by geographical distribution. Thus, disease patterns differ by geographical area (table 2[4 8–10]).

The reservoirs of the disease are animals, such as canines and rodents (zoonotic cycle), and, in countries such as Sudan, humans (anthroponotic cycle).[11] The sandfly is the vector of the disease and ingests the organism, as an amastigote, into its digestive tract when feeding on an infected animal or person. The amastigote develops into a promastigote in the sandfly's digestive tract and is then injected into the susceptible host at the next feed. The promastigote then infects macrophages and develops into amastigotes (fig 1).

About 70 different species of sandfly can transmit leishmaniasis,[12] *L lutzomyia* in the Americas and *L phlebotomous* elsewhere.[10] The sandfly characteristically feeds at dusk, and, being a weak flier, tends to remain close to its breeding area, not too high from the ground. Different species have different feeding and resting patterns. These different characteristics are important in formulating control strategies.

Table 1 shows the incidence and prevalence of leishmaniasis. Infection is more common in men than in women, but this may reflect increased exposure to sandflies. Although disease occurs irrespective of age, children aged 1–4 years are

Abbreviations: PCR, polymerase chain reaction; PKDL, post-kala azar dermal leishmaniasis; Sb(V), pentavalent antiminy; Th, T helper; TNFα, tumour necrosis factor α; INFγ, interferon γ

Box 1: Current issues in leishmania infection

- Leishmania is given less importance than other more prevalent infectious diseases.
- Climate change may be responsible for the extension of endemicity of leishmaniasis to previously non-endemic countries.
- New diagnostic tests should improve diagnosis, especially on the field, in resource-poor areas.
- Leishmania infection may relapse in patients with HIV and is difficult to eradicate, if at all, in this setting. There have been calls for leishmania infection to be officially recognised as an AIDS-defining illness.
- Anthroponotic transmission may occur in injecting drug users who share needles.
- The leishmania genome has been unravelled, but an effective vaccine is not yet available.

particularly at risk of infection in Mediterranean regions, and childhood infection may account for more than half of all cases in some of these countries.[13] Untreated visceral leishmaniasis carries a mortality of 75–95%, whereas cutaneous leishmaniasis can disseminate to affect the mucosa, resulting in death from secondary infection.[8]

DISEASE PATTERNS

Three main types of disease patterns occur: visceral, cutaneous and mucocutaneous leishmaniasis. The type of disease expressed depends both on the type of *Leishmania* species and on the zymodeme (electrophoretic isoenzyme pattern) expressed on that species. Thus, one zymodeme may cause visceral leishmaniasis whereas another zymodeme of the same species may cause cutaneous leishmaniasis.[14]

Visceral leishmaniasis

The incubation period varies from 3 to 8 months[15 16] (range from 10 days[17] to 34 months[18]). Symptoms include fever, weight loss, hepatosplenomegaly (usually spleen much larger than the liver), lymphadenopathy, pancytopenia and hypergammaglobulinaemia.[11] Skin pigmentation may be a feature (kala azar: black disease). It may be asymptomatic and self-resolving but usually runs a chronic course and may be fatal without treatment, or despite treatment.[19] Death usually occurs because of severe secondary bacterial infections in advanced disease.

Some cases of visceral leishmaniasis present atypically and cases have been reported that affect the lungs, pleura, oral mucosa, larynx, oesophagus, stomach, small intestine, skin and bone marrow.[10]

Table 1 Global epidemiology of different infectious diseases

Disease	Annual incidence	Mortality (no of deaths/year)	Prevalence (total no of infected people)
HIV/AIDS	5.6 million	2.6 million	34 million
Malaria	300 million	1–2.7 million	NA
Tuberculosis	7.8 million	1.8 million	1.7 billion
Visceral leishmaniasis	500 000	80 000	12 million
Cutaneous leishmaniasis	1.5–2 million	NA	NA

NA, not available.
Sources: http://www.who.int; http://www.cdc.gov; and http://www.hopkins-id.edu[1].

Variations of visceral leishmaniasis

Post-kala azar dermal leishmaniasis (PKDL) develops after resolution of visceral leishmaniasis. The interval to development of PKDL is variable and PKDL occurs in a small percentage of patients in Africa and India.[10] This is usually due to infection by the *L donovani* sensu stricto cluster.[20] The skin lesions are macular, maculopapular or nodular, and usually spread from the perioral area to other areas of the body (table 3).

Cutaneous leishmaniasis

Cutaneous leishmaniasis starts as a papule at the site of a sandfly bite, which then increases in size, crusts (fig 2) and

Table 2 Disease patterns and organisms prevalent in different geographical locations[4 8 9 10]

Disease patterns	Old-World organisms	New-World organisms
Visceral leishmaniasis	*L donovani* (India, Kenya); *L infantum* (Southern Europe and North Africa); *L tropica*	*L chagasi; L amazonensis*
Post-kala azar dermal leishmaniasis	*L donovani* sensu stricto	
Viscerotropic leishmaniasis	*L tropica*	
Cutaneous leishmaniasis	*L tropica; L major; L aethiopica; L infantum; L donovani*	*L mexicana* species complex; *L mexicana; L amazonensis; L venezuelensis Viannia* subgenus; *L (V) braziliensis; L (V) panamensis; L (V) guyanensis; L (V) peruviana; L* major-like organisms *L chagasi*
Mucosal leishmaniasis	*Viannia* subgenus; *L (V) braziliensis; L (V) panamensis; L (V) guyanensis; L amazonensis*	
Leishmaniasis recidivans	*L tropica; L major*	
Diffuse cutaneous leishmaniasis	*L aethiopica*	*L mexicana* species complex

L, *Leishmania;* V, *Viannia* subgenus.
Leishmanias were classified into the subgenera *Leishmania* sensu stricto (Old and New World) and *Viannia* (New World) by Lainson and Shaw.[9]

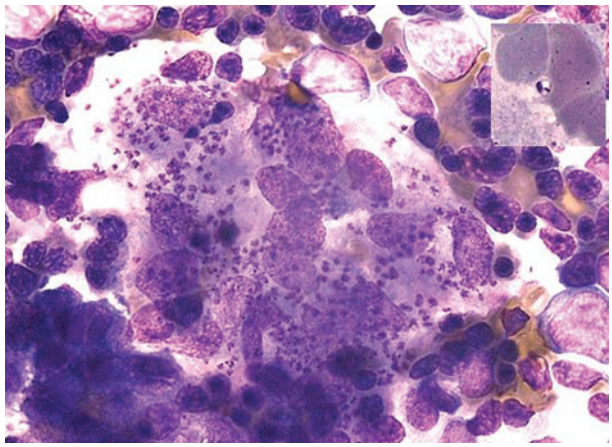

Figure 1 Abundant amastigotes of *Leishmania* in a bone marrow aspirate. Inset shows the amastigote with its nucleus and kinetoplast (specialised mitochondria containing DNA). Photograph courtesy of Dr Alicia Grochowska, Consultant Haematologist, St Luke's Hospital, Malta.

eventually ulcerates. It may take 3–18 months to heal in >90% of cases.[10] The incubation period lasts from 2 weeks to several months and cases up to 3 years have been reported in Old-World cutaneous leishmaniasis.[17 21] In New-World cutaneous leishmaniasis, the incubation period is usually 2–8 weeks.[22]

Variations of cutaneous leishmaniasis

Leishmaniasis recidivans is characterised by tuberculoid lesions developing around scars of healed cutaneous ulcers, disclosing a low parasite count on biopsy. Infections tend to be resistant to treatment.

In diffuse cutaneous leishmaniasis, dissemination of skin lesions rarely occurs over the face and hands and feet, disclosing high parasite numbers owing to poor cell-mediated immune response. This is more common in the New-World *Leishmania* but also occurs with *L aethiopica* in East Africa.[23]

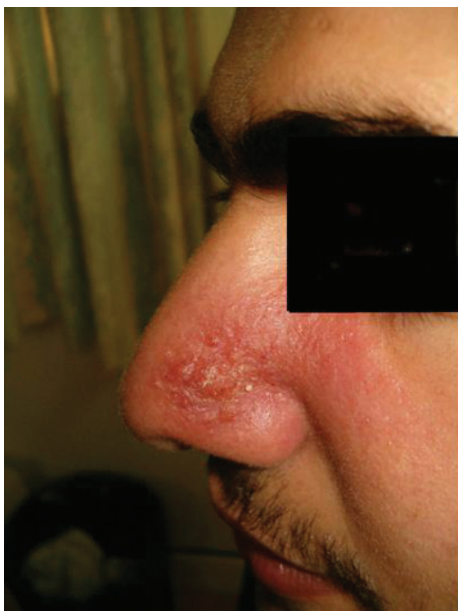

Figure 2 Cutaneous leishmaniasis at crusting stage in a patient from Malta (informed consent was obtained from the patient to publish his photograph).

Box 2: The immune response

T helper (TH) 1 immune response

- Th CD4 cells producing interleukin (IL)2, IL3 interferon γ
- Will promote immune responses that are primarily cell mediated or inflammatory by activating cytotoxic T cells, natural killer cells and macrophages
- Associated with disease resolution in leishmaniasis

Th2 immune response

- Th2 cells produce IL4, IL5, IL6 and IL10, which favour induction of antibody responses by B cells
- Associated with disease progression in leishmaniasis

Mucocutaneous leishmaniasis

The incubation period is 1–3 months, but mucocutaneous leishmaniasis may occur many years after the initial cutaneous ulcer has healed. Mucosal involvement occurs in South American cases of cutaneous leishmaniasis (espundia) affecting the nose, oral cavity and pharynx. This causes difficulty in eating and an increased risk for secondary infection, which carries considerable mortality.

IMMUNOLOGY

The immune response (box 2) to Leishmania infection is cell mediated. The organism is exclusively intracellular, mainly inside macrophages as a replicating amastigote. The outcome of infection will depend on whether the host mounts primarily a T helper (Th)1 or Th2 response.[24] Studies on animals suggest that the same parasite epitope can induce a Th1 response in animals with resolving infection or a Th2 response in others with disease progression.[25] Other animal studies showed that Th1 and natural killer cells produce interferon γ (INFγ), which mediate resistance, whereas interleukin (IL)4-producing Th2 cells conferred susceptibility to infection.[26] Human studies have also shown that IL4, a component of the Th2 response, may also be associated with disease progression.[27]

In the Th1 response, promastigotes attach to reticuloendothelial cells and Th CD4 cells produce IL2, IL3 and interfon γ (INF γ), which activate macrophages. IL12 and tumour necrosis factor (TNF) are also important in this type of response. The promastigotes are then phagocytosed by the activated macrophages into vacuoles, which then fuse with lysosomes.

Host genetics inevitably influences the type of immune response. Studies on mice and humans have shown that genes such as those coding for natural resistance-associated macrophage protein 1, TNF or the major histocompatibility complex are thought to have a major role in the outcome of infection.[28] The parasite itself can affect the macrophage and dendritic cell responses. Specific gene loci such as the *A2* gene can code for products which promote *L donovani* infectivity.[12] Thus, the interplay between the host-determined delayed-type hypersensitivity, antigen-specific T cell reactivity and cytokine secretion on the one hand, and the type and virulence of the particular infecting *Leishmania* species on the other hand determines what type of disease expression develops in the host.

Some cases are believed to harbour *Leishmania* for indefinite periods before the disease is expressed, suggesting latent infection.[29] Parasites have also been detected in lymph nodes after clinical cure.[30] This is probably the basis of recrudescence of leishmaniasis, which can occur even decades after the initial infection, if cell-mediated immunity is disturbed. It is thought that "sterile" immunity—that is,

Table 3 Disease patterns in leishmaniasis

Disease type	Incubation period	Clinical features
Visceral leishmaniasis	3–8 months (range 10 days to 34 months)	Fever, weight loss, hepatosplenomegaly, lymphadenopathy, pancytopenia and hypergammaglobulinaemia skin pigmentation
Post-kala azar dermal leishmaniasis	Variable; develops after resolution of visceral leishmaniasis	Skin lesions around mouth and other parts of body
Cutaneous leishmaniasis	2 weeks to several months (rarely up to 3 years)	Papule at the site of a sandfly bite increases in size, crusts and ulcerates
Leishmaniasis recidivans		Tuberculoid lesions develop around scars of healed cutaneous ulcers; low parasite count on biopsy
Diffuse cutaneous leishmaniasis		Rare, dissemination of skin lesions occurs over face, hands and feet; high parasite numbers due to poor cell-mediated immune response
Mucocutaneous leishmaniasis	1–3 months (may occur many years after the initial cutaneous ulcer has healed)	Mainly in South America, affects the nose, oral cavity and pharynx, resulting in difficulty in eating

Table 4 Diagnostic methods

Diagnostic test	Comments
Visceral leishmaniasis	
Microscopical detection of amastigotes in smears of tissue aspirates or biopsy samples	Bone marrow aspirates or biopsy sensitivity 55–97%; lymph node aspirate smears (sensitivity 60%) or biopsy specimens; splenic aspirates (sensitivity>97%); risk of life-threatening haemorrhage
Tissue culture	On special media like Novy, McNeal or Nicolle or inoculated into animals such as hamsters
Leishmania antibody (DAT)	Sensitivity of 72%, specificity of 94%; some cross-reactions in leprosy, Chagas disease, malaria and schistosomiasis; in HIV, may be false negative
Anti-K39 antibody in blood droplet	Sensitivity of 90–100% in patients with symptoms; useful in clinical management in resource-poor areas
Leishmania DNA detection in tissue aspirates and peripheral blood by PCR	Sensitivity 70–93% in peripheral blood
Detection of *Leishmania* antigen and antibody in the urine	High sensitivity and specificity
Cutaneous leishmaniasis	
Microscopical examination of skin scrapings or biopsy specimens taken from the edge of lesions*	Rapid and low-cost; limited sensitivity especially in chronic lesions
Cultures of the lesions	More sensitive than microscopy; may become contaminated; different species have different growth requirements
Antibody detection	Poorly sensitive; in American cutaneous leishmaniasis there have been reports of cross-reactivity
Montenegro (leishmanin) skin test	Unable to distinguish between current and past infection; reports of false positivity in other skin infections

DAT, direct agglutination test; PCR, polymerase chain reaction.
*A practical guide to diagnostic techniques in CL can be found at http://www.thelancet.com/journals/lancet/article/PIIS0140673698101782/fulltext#abstract

complete eradication of the organism—probably rarely develops in visceral infection.

DIAGNOSIS

Visceral leishmaniasis

Diagnosis of visceral leishmaniasis is usually based on microscopic detection of amastigotes in smears of tissue aspirates or biopsy samples. An aspirate or biopsy of the bone marrow is frequently the tissue of choice, with sensitivities in the 55–97% range (fig 1). Lymph node aspirate smears (sensitivity 60%) or biopsy, and splenic aspirates (sensitivity >97%)[12] may also be taken for diagnosis, although the splenic aspirates may give rise to life-threatening haemorrhage (table 4).

Sometimes the parasite can be cultured from microscopy-negative tissue samples on special media such as Novy, McNeal or Nicolle medium or inoculated into animals such as hamsters.

Leishmania antibody (direct agglutination test) may be detected with a sensitivity of 72% and a specificity of 94%.[11] Some cross-reactions in leprosy, Chagas disease, malaria and schistosomiasis may occur. In HIV, antibodies to *Leishmania* may become undetectable.

Immunochromatographic strip testing of blood from a finger prick for leishmanial anti-K39 antibody has been used successfully in field serodiagnosis,[31] with a sensitivity of 90–100% in patients showing symptoms. This test is useful for clinical management in resource-poor areas.[2]

Leishmania DNA can also be detected in tissue aspirates and peripheral blood using polymerase chain reaction (PCR), with some series giving a sensitivity of 70–93% in peripheral blood.[11] High sensitivities to the level of one parasite have been recorded.[32] Newer methods with high sensitivity and specificity include the detection of *Leishmania* antigen and antibody in urine.[33 34]

Cutaneous leishmaniasis

The diagnosis is usually based on microscopical examination of skin scrapings or biopsy specimens usually taken from the edge of lesions. This is rapid and low-cost but has limited sensitivity, especially in chronic lesions.[35]

Cultures of the lesions, although more sensitive, may become contaminated by bacterial and fungal elements in the biopsy specimen itself. Also, different species have different growth requirements. *Leishmania* species may be identified using isoenzyme electrophoresis, but this is lengthy, expensive and necessitates cultivation of parasites on a large scale. Monoclonal antibodies can also be used for identification of species in cultured strains, but a direct analysis of clinical specimens is better achieved using PCR. PCR is rapid, with high specificity and sensitivity. Detection and genetic characterisation of *Leishmania* can also be obtained simultaneously.[35] One study on American cutaneous leishmaniasis yielded a PCR sensitivity of 100%.[36]

Antibody detection is poorly sensitive owing to the lack of significant antibody production in cutaneous leishmaniasis. Moreover, in American cutaneous leishmaniasis there have been reports of cross-reactivity of leishmanial antigens with antibodies induced by other kinetoplastids such as *Trypanosoma cruzi*.[37]

Other available means of diagnosing cutaneous leishmaniasis include the Montenegro (leishmanin) skin test, which detects specific cutaneous delayed-type hypersensitivity. It involves intradermal injection of *L mexicana* antigen, and monitoring for a local reaction.[38] Limitations of this test include its inability to distinguish between current and past infection, as well as reports of false positivity in other skin infections.[39]

TREATMENT

Visceral leishmaniasis

Treatment is largely based on pentavalent antimonials. Increasing resistance to antimonials is a major problem, and this is most evident in north Bihar, India, where the failure rate to this treatment is >50%.[12 40] Pentavalent antimony (Sb(V)) can take the form of sodium stibogluconate (100 mg Sb(V)/ml) or meglumine antimonate (85 mg Sb(V)/ml). These can be given intravenously or intramuscularly with equal efficacy. It is usually given at a dose of 20 mg Sb(V)/kg for 28 days, depending on the species and the clinical syndrome. Recently, a randomised trial on US military personnel showed a shorter 10-day course to be equally effective.[41] A maximum dose of 850 mg daily has been recommended[42] to minimise side effects such as arrhythmias. Some experts, however, believe that this might predispose patients to resistance, and so they advocate higher doses. In some resistant patients with both visceral leishmaniasis or cutaneous leishmaniasis, INFγ was added successfully to Sb(V) to induce remission.[43]

Amphotericin B is an effective antibiotic used in Sb(V)-resistant patients. It is toxic and needs to be given for a prolonged period on an inpatient basis. The alternative is using the liposomal form, which is highly effective and less toxic, although up to now prohibitively expensive. The trend in southern Europe is shifting towards using liposomal amphotericin B as the preferred treatment, even though the response rate is still around 90% for antimonials. However, a recent trend in increasing resistance to Sb(V) in this area has been recorded, possibly attributed to using meglumine antimonate to treat infected dogs.[44] The effectiveness of short courses of this liposomal amphotericin B is resulting in improved cost benefits.[40 45] Studies on treatment using lower doses of this agent are also showing promise to improve cost-effective treatment in resource-poor areas with high-level antimonial resistance.[46]

Miltefosine has been a breakthrough because it is the first effective orally active drug against leishmaniasis. Studies on treatment with this drug for 3 or 4 weeks have shown a cure rate between 95% and 100%.[47 48] It has also compared very well (cure rate of 94%) with amphotericin B (cure rate 97%) at 6 months of follow-up. It has the added benefit of a very good safety profile.[49] The potential of this drug for treatment of large numbers of patients as outpatients in resource-poor areas is high, although concerns about compliance and eventual resistance have been expressed.[12]

Other drugs used in treating leishmaniasis have also been effective. Pentamidine is another agent that could be used in treatment-resistant patients with visceral leishmaniasis.[50] Its use is limited by its substantial toxicity, thus requiring close inpatient monitoring.[51] Paromomycin (aminosidine) has been used effectively in resistant cases in northern Bihar, India.[52] Pending commercial availability, this treatment should offer cost savings, although issues of potential toxicity like nephrotoxicity or ototoxicity may need further evaluation.[53] Sitamaquine is another oral agent currently in phase II studies in India.[12] This drug has been associated with a 50–67% cure rate.[54 55] The imidazole and triazole drugs are not recommended for use in those with visceral leishmaniasis.

In PKDL, treatment is indicated only for those who have severe and prolonged disease. Sb(V) (2-month course usually sufficient) and liposomal amphotericin B are both effective (table 5).[20 56]

Cutaneous leishmaniasis

In deciding the best mode of treatment of cutaneous leishmaniasis, some facts need to be considered. Old-World cutaneous leishmaniasis is not a life-threatening disease and >90% of patients heal spontaneously within 3–18 months.

The outcome of cutaneous leishmaniasis in the New World depends on the infecting species and may vary from benign to more severe manifestations. It is thus important to try to identify the infecting species either by determining the endemic species of the specific geographical area or by diagnostic procedures. This can throw light on the prognosis and management options.

Treatment of cutaneous leishmaniasis will accelerate the cure and reduce scarring. This is especially important at cosmetically important sites. The options in the treatment of cutaneous leishmaniasis include local and systemic treatment. Criteria in favour of local treatment[56] include,

- Old-World cutaneous leishmaniasis,
- small single lesions,
- no risk of development of mucocutaneous leishmaniasis,
- no lymph node metastases and *L mexicana* lesions.

New-World lesions except *L mexicana*, mucosal or lymph node involvement and lesions refractory to local treatment would be indications for systemic treatment.

Local treatment of cutaneous leishmaniasis

Physical modes of treatment including cryotherapy have been used with success ranging from 77% to 100% at 4 weeks.[57 58] Local infrared heat lamps have also produced good results, although invariably this treatment is accompanied by skin bulla formation.[59]

Paromomycin (aminosidine) ointment is produced in two different formulations. When combined with methylbenzethonium chloride, it gives a cure rate of 74–85%, which is more effective than the cure rates when combined with urea.[60] However, paromomycin–methylbenzethonium produces more severe local inflammatory reaction than paromomycin–urea.[61]

Intralesional infiltration of the dermis and base of the lesion with Sb(V) may be carried out. This is a relatively painful procedure that needs to be performed regularly every 1–2 weeks for 3–8 times. The cure rate with this procedure is about 75%.[12] If this is not effective, systemic treatment should be considered.

Imiquimod, a topical immunomodulator, has been successfully used in combination with meglumine antimonate in patients resistant to meglumine alone.[62]

Systemic treatment of cutaneous leishmaniasis

Systemic treatment with antimonials in general requires a 20-day course. *L major*, *L tropica* and *L mexicana* usually respond to a 10-day course.[12] Pentamidine has been used as the preferred treatment in *L guyanensis* infection, but studies with *L panamensis*, *L brasiliensis* and *L mexicana* have also given a cure rate of 96%, comparable to 91% with meglumine antimonate.[63 64]

Of the imidazole/triazole drugs, oral fluconazole has been found useful in *L major* infections, with a cure rate of 79%.[65] Ketoconazole has been studied in *L braziliensis panamensis* infections, with efficacy (74%) similar to that of stibogluconate (68%).[66]

Oral miltefosine has been studied in Colombia and Guatemala, with short-term cure rates of 91% in areas where *L (V) panamensis* is common. In areas where *L (V) braziliensis* and *L mexicana mexicana* are common, the cure rate fell to 53%.[67]

Liposomal amphotericin B has not been extensively studied in cutaneous leishmaniasis, but isolated reports of its use in resistant patients show its effectiveness.[68 69] Similarly, systemic aminosidine has not been commonly used, and a study comparing it with antimonials for cutaneous leishmaniasis caused by *L braziliensis* was not in favour of its use in this form.[70] Allopurinol has shown some improvement in antimonial activity, but its use alone has not shown any statistically significant benefit.[71]

Mucocutaneous leishmaniasis

Treatment of mucocutaneous leishmaniasis with antimonials is unsatisfactory, especially in severe disease.[72] Amphotericin B[73] and more recently liposomal amphotericin B have been used successfully in difficult cases.[74] Steroids may have to be used in patients in whom respiratory compromise is possible.

Table 5 Summary of recommended treatment regimens[12 56]

Disease pattern	Drug	Dose	Comments (level of evidence, when available, in brackets)
Visceral leishmaniasis	Miltefosine	2.5 mg/kg/day PO (od or bd) ×28 days	(A)
	Liposomal amphotericin B	2 mg/kg/day IV ×5 days	(A)
	Pentavalent antimony (as stibogluconate or meglumine antimonate)	20 mg/kg/day IM or IV ×28 days	(Some experts advise not exceeding 850 mg od,[42] not effective in northern Bihar, India
	As with VL		
Mucocutaneous leishmaniasis	Amphotericin B	1 mg/kg IV qod ×20–30 doses	This may be better than antimonials in mucosal disease
	Observation alone		*L major* (A) and *L mexicana* (B)
	Pentavalent antimony (as stibogluconate or meglumine antimonate)	20 mg/kg/day IM or IV ×20 days (×10 days in *L major*, *L tropica* and *L mexicana)*	(Some experts advise not exceeding 850 mg od)
	Fluconazole	200 mg PO od	*L major* (A)
	Ketoconazole	600 mg PO od ×28 days	*L panamensis* (A) *L mexicana* (A)
Cutaneous leishmaniasis	Miltefosine	2.5 mg/kg PO od ×28 days	*L panamensis* (A)
	Pentavalent antimony	Intralesional: 1 ml per lesion qod ×8–15 times	*L major* every 1–2 weeks ×3–8 times (A) *L tropica* weekly × 8–11 times (C)
	Pentamidine	2–4 mg/kg od or every 2 days IV ×15 doses	*L panamensis* and *L braziliensis* *L guyanensis* (D)
	Paromomycin	Topical bd	*L major* ×4 weeks (A) *L mexicana* ×20 days (B)

bd, twice daily; IM, intramuscular; IV, intravenous; od, once daily; PO, orally; qod, every other day.
Grades of recommendations based on best available evidence: (A) randomised, controlled trial in representative collective;(B) randomised, controlled trial in partially representative (small patient number, different species included) collective, cohort trial or case–control study in representative collective; (C) series of cases in partially representative (small patient number, different species included) collective, informal expert opinion, other information; (D) cohort trial or case–control study in partially representative collective, series of cases in representative collective.

Leishmania–HIV coinfection

The possibility needs to be kept in mind in patients with HIV with typical symptoms of visceral leishmaniasis, such as pyrexia, pancytopenia and hepatosplenomegaly. However, we should note that splenomegaly may be absent in HIV.[75] Uncommon sites of infection are more frequent, such as the gastrointestinal tract or the upper respiratory tract. Diagnosis is reached as for patients without HIV infection, except that the *Leishmania* antibody test (direct agglutination test) is often negative (42.6%).[76 77] The highest prevalence of coinfection occurs in south western Europe, mostly in Spain.[76] The main risk group is the injecting drug users, and an anthroponotic cycle has been suggested where *Leishmania* present in used syringes[78] are inoculated intravenously.

The importance of cell-mediated immunity in controlling Leishmaniasis in the long term is best shown in *Leishmania*–HIV coinfection. When visceral leishmaniasis occurs in a known HIV-positive patient or when a person with a history of visceral leishmaniasis acquires HIV, there is a high risk that the leishmania infection becomes intractable. Even after appropriate treatment of visceral leishmaniasis, *Leishmania*–HIV is associated with a high relapse rate of 52% between 1 month and 3 years.[76] Visceral leishmaniasis in HIV infection is being proposed for inclusion in the Centers for Disease Control and Prevention clinical category C for the definition of AIDS as an indicator disease.[79]

Although treatment of coinfection has not been adequately studied, Sb(V) is still used widely. The relapse rate does not seem to be affected by the type of treatment given according to a head-to-head study of meglumine antimonate and amphotericin B.[80] Comparison between meglumine antimonate and liposomal amphotericin has been carried out only in smaller studies and no differences were found.[81] One study suggests a role for oral miltefosine when the above treatments have failed in people with coinfection.[82]

Secondary prophylaxis prevents relapses and improves survival.[79 83] Both Sb(V) given once every 28 days[84] and liposomal amphotericin B given every 21 days may be used[83]; no differences in efficacy have been borne out yet. Secondary prophylaxis should be continued at CD4 counts $<200/\mu l$. It may be safe to stop secondary prophylaxis at CD4 counts $>350/\mu l$ and possibly even $>200/\mu l$ while on effective antiretroviral treatment.[84] Antiretroviral treatment has been effective in decreasing relapses of visceral leishmaniasis.[85]

CONTROL

The control of leishmaniasis depends on the prevalent local epidemiological characteristics. Thus, in areas where sandflies are mostly endophilic (rest mostly indoors after feeding), spraying houses with insecticide is effective in reducing the odds of contracting cutaneous leishmaniasis.[86 87] Treated and untreated bed nets have been used effectively in environments where the sandflies are endophagic (feed mainly indoors).[88] In areas where the disease is transmitted anthroponotically, the aim of nets has been also to prevent transmission from infected patients with cutaneous leishmaniasis. Other control strategies, including spraying insecticide in houses[87] insecticide-impregnated curtains and insecticide-treated dogs and dog collars, have been used with good effect.[89]

In Brazil, dogs are culled if they test positive for *Leishmania*, but this strategy has not been effective.[90] In south western Europe, where leishmaniasis is closely linked with injecting drug use and HIV, control of HIV has yielded positive results.[91] Needle-exchange interventions should have similar results.

In India, where anthroponotic transmission is important, effective treatment of patients, especially those with PKDL (who may act as long-term reservoirs), has been found to be effective in controlling transmission when combined with vector control.[3]

Prevention of leishmaniasis with an effective vaccine has, to date, not materialised. The closest effective alternative to vaccination follows from the traditional ''leishmanisation'' technique adopted in the Middle East and eastern Europe. This involved the encouragement of sandfly bites on traditionally unexposed areas of skin such as the buttocks. The resulting lesion would heal spontaneously, in the process providing immunity against cutaneous leishmaniasis in less acceptable areas such as the face. Early studies had recommended using *L tropica* inoculations to induce immunity.[92] This technique has been further developed in Iran, where scientists have produced standardised and stable *L major* populations, which can produce more consistent and acceptable iatrogenically induced lesions.[89]

The safety and efficacy of live attenuated and killed vaccines have been debated, and shifts in favour of development of each have been recorded in recent years. Killed vaccines were favoured in the 1990s because of safety problems with live attenuated vaccines, although recent advances in manipulation of the *Leishmania* genome may make the development of a live attenuated vaccine more feasible.[93]

Work on recombinant DNA-derived antigen vaccines and protein or peptide-based vaccines is a newer approach, and the *Leishmania* Genome Project (www.sanger.ac.uk/Projects/L_major/) is useful in this regard. The realisation that CD8 cells are as important as CD4 cells in inducing resistance[94] and maintaining immunity has led to a shift in vaccine research along these lines.[95] Most vaccine research is targeted against cutaneous leishmaniasis, and any effectiveness against visceral leishmaniasis is still unclear. Work on a vaccine against human visceral leishmaniasis has been less successful[96] but should be boosted after any success on a cutaneous leishmaniasis vaccine. Work on a canine visceral leishmaniasis vaccine seems to be more advanced.[97]

CONCLUSION

Leishmaniasis remains a problematic infection requiring either potentially toxic treatments, or less toxic but expensive drugs. However, the availability of newer oral agents may change the way this disease is managed. A relapse may occur, especially in situations where immunosuppression is present, and secondary prophylaxis needs to be given in this setting. The combination of *Leishmania*, HIV and anthroponotic transmission between injecting drug users heralds a potential

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for higher incidence rates in endemic countries with severe drug misuse problems. In the absence of an effective vaccine, and with extension of endemicity, possibly owing to climate change, these problems may become worse.

MULTIPLE CHOICE QUESTIONS (TRUE/FALSE (T/F); ANSWERS AT END OF REFERENCES)

1. Epidemiology

A. *Leishmania* is endemic in Australasia.
B. *Leishmania* can be transmitted through infected syringes.
C. The epidemiology of *Leishmania* is tightly knit with the epidemiology of the sandfly.
D. The sandfly usually bites during the day.
E. The number of countries where *Leishmania* is endemic is set to increase because of global warming.

2. Visceral leishmaniasis

A. Is characterised by splenomegaly and pancytopenia.
B. Carries a low mortality if not treated.
C. The organism is characteristically extracellular.
D. Fatalities are usually the result of secondary bacterial infections.
E. Almost always responds to antimony products.

3. Immunology

A. An effective vaccine is available.
B. The body's response to infection is T-cell mediated.
C. The disease manifestation depends on *Leishmania* species and host response.
D. Immunosuppression predisposes to *Leishmania* infection.
E. *Leishmania* organisms can remain dormant for years in the reticulo-endothelial system.

4. Treatment

A. Liposomal amphotericin is effective but very expensive.
B. There are no oral medications to effetively treat *Leishmania*.
C. Oral miltofisine is very effective in the treatment of Kala azar.
D. Cutaneous *Leishmania* can be treated both by physical and pharmacological means.
E. *Leishmania* in the immunosuppressed usually necessiates secondary prophylaxis.

5. Diagnosis

A. Identification of amastigotes in tissue smears or histology are commonly used methods of diagnosis.
B. Antibody towards K39 antigen is proving useful especially in field diagnosis of visceral leishmaniasis.
C. Tissue cultures are used routinely.
D. Splenic aspirates are very sensitive and usually safe.
E. *Leishmania* PCR is useless on blood samples.

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ANSWERS

1. (A) F, (B) T, (C) T, (D) F, (E) T; **2**. (A) T, (B) F, (C) F, (D) T, (E) F; **3**. (A) F, (B) T, (C) T, (D) T, (E) T; **4.** (A) T, (B) F, (C) T, (D) T, (E) T; **5.** (A) T, (B) T, (C) F, (D) F, (E) F.