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Chapter 2

Visceral Leishmaniasis

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Abstract

Clinically, leishmaniasis is of three types—visceral leishmaniasis (VL) or kala-azar, cutaneous leishmaniasis (CL) and mucocutaneous leishmaniasis (MCL). Post-kala-azar dermal leishmaniasis (PKDL) is considered as a complication of VL. VL is characterized by fever, anemia and splenomegaly in a VL-endemic area (malaria excluded). A subject with such symptoms should be subjected to an rK39 strip test. Confirmation of diagnosis is made by demonstration of the parasite (*Leishmania donovani*) from samples obtained by aspiration of bone marrow or iliac crest puncture. Miltefosine, stibogluconate, amphotericin B, liposomal amphotericin B and paromomycin are effective available anti-leishmaniasis drugs. Vector (*Phleblotomus argentipes*) control for reduction of transmission and early diagnosis and complete treatment are essential elements of case management. There is no effective vaccine against VL. This review on VL aims at providing state-art knowledge on epidemiology, diagnosis and case-management and vaccine development.

Keywords: leishmaniasis, PKDL, rK39 strip test, kala-azar vaccine

1. Introduction

The distinct clinical forms of leishmaniasis are visceral leishmaniasis (VL), cutaneous leishmaniasis (CL) and mucocutaneous leishmaniasis (MCL). PKDL is considered to be a complication of VL. Kala-azar is a neglected tropical disease (NTD). It affects the poorest of the poor living in endemic areas. Post-kala-azar dermal leishmaniasis (PKDL) is associated with stigma. Fortunately, it is not difficult to diagnose the disease and several drugs are available for treatment of the disease.

1.1. VL

VL is prevalent in 88 countries, and there is an estimated 2 million new cases per year, of which 5,00,000 are VL and 15,00,000 are CL. The disease burden is calculated at 23,57,000 disability-adjusted life years, a significant ranking among communicable diseases [1]. More than 147 million people living in the Southeast Asia Region are at risk. In this region, the highest disease burden is seen in the northern part of the state of Bihar, India. A large number of VL cases are seen in the international cross-border areas between the countries. In view of this, cross-border collaboration is crucial for elimination of the disease [2]. VL prevails mostly among poor people in marginalized communities. It attacks the internal organs and can be fatal if left untreated as it affects the vital organs of the body. Symptoms include irregular bouts of fever, weight loss, enlargements of the spleen and liver and anemia. All patients diagnosed to be VL require prompt and complete specific medical treatment; otherwise, the patient may die.

1.2. PKDL

PKDL is considered an important long-lasting complication of kala-azar. This is seen in about 1–15% in the Indian subcontinent and in about 60% of treated, partially treated and untreated or active cases of kala-azar in Sudan [3]. The lesions generally appear from 1 to 15–20 years of kala-azar. The typical lesions are most prominently seen in the face (macular, papular and nodular). In contrast to the Indian subcontinent, the nodular lesions generally ulcerate as they grow in Sudan. The macular lesions are sometimes confused with leprosy. Diagnosis is confirmed by demonstration of *Leishmania donovani* from the tissue obtained from the lesions. A polymerase chain reaction (PCR) test to detect the DNA is highly reliable and can be performed in special laboratories. Treatment is long-term use of sodium stibogluconate (sometimes in combination with rifampicin) [4]. A recent study showed that a 12-week course of miltefosine is safe and effective in the treatment of PKDL [5]. Another study showed that miltefosine 50 mg 3 times daily for 60 days or 50 mg twice daily for 90 days has been shown to be effective [6]. In 2008, Berman remarked that 'Miltefosine is effective and can be recommended for visceral disease in India and in Ethiopia, and for cutaneous disease in Colombia and Bolivia. For unusual forms of disease that require long periods of treatment such as diffuse CL, oral miltefosine is probably the treatment of choice'. In 2006, Simon and Engel remarked that 'Miltefosine is active against most *Leishmania* species, including those that cause CL'. It has been demonstrated that in 2015 the efficacy of miltefosine has declined. Paromomycin has been shown to be effective against PKDL [7]. Amphotericin B is effective in the treatment of PKDL and several courses, where gaps between the two courses are required.

1.3. CL

CL and MCL are the most common manifestations of leishmaniasis. This is also known as oriental sore. MCL is seen in the tropics and subtropics. The variety of leishmaniasis causes disfigurement. CL is caused by *L. tropica* and *L. major*. *L. braziliensis* causes MCL transmitted by *Phleblotomus argentipes*, sand fly. A clinical diagnosis should be supported by a PCR test [8].

1.4. Diagnosis

Clinical features like prolonged fever (>14 days), anaemia and splenomegaly (malaria excluded) in a kala-azar-endemic area will constitute a suspect case. This can be supported by an rK39 strip test. Accurate VL diagnosis till 1990 required parasitological confirmation by microscopy or culture of the blood, bone marrow, lymph nodes or spleen. Splenic aspirate was sometimes fatal and this prompted development of rapid diagnostic tests (RDTs). An RK39 test had a sensitivity of 72.1% and a specificity of 76.9% and DAT sensitivity was 62.8% and its specificity was 69.2%, using initial diagnosis (confirmed on clinical and serological basis) as reference in both cases. Both rK39 and DAT have the potential in diagnosing VL using urine [9]. A polymerase chain reaction test (PCR) to detect the DNA is highly reliable and can be performed in special laboratories.

2. Management

2.1. Management of VL

2.1.1. Miltefosine

This is the first ever oral drug developed against VL. Initially the drug was used in the treatment of skin metastases from breast cancer. Subsequently, it was found to be effective in vitro against the *Leishmania donovani* parasite [9]. Phase I–Phase III clinical trials conducted in India demonstrated that the drug was effective against VL to the extent of ~95% [10–12]. Most of the side-effects included nausea, vomiting, abdominal pain, diarrhea and fever [11]. These side-effects occur during the first week of treatment. A Phase IV trial was conducted involving 13 centres in Bihar (India). This pivotal trial clearly demonstrated that the drug can be dispensed in the kala-azar elimination programme in the first condition [13]. A few major side-effects that occurred affected the kidney, liver and bone marrow. The dose of the drug is 50 mg 2 times daily after food to avoid gastric irritation for 28 days. In children, the dose is 2.5 mg/kg daily for 28 days [12]. In summer time, these patients are usually dehydrated and they should be rehydrated with ORS or IV fluids depending upon the degree of dehydration. Anemia is very common in such patients. Blood transfusion may be required and should be given. If these simple measures are taken, the patients tolerate the drug better. Miltefosine is teratogenic and should not be given to pregnant mothers.

2.1.2. Paromomycin

This drug belongs to the group of aminoglycosides. Unlike miltefosine, this drug is administered by intramuscular injections. A full course comprises daily injections (11 mg/kg/day) for 21 days [14, 15]. Although paromomycin is an aminoglycoside, it does not exhibit much ototoxicity or nephrotoxicity. It is safe in pregnancy. The most common adverse effects associated with paromomycin are abdominal cramps, diarrhea, heartburn, nausea and vomiting. Long-term use of paromomycin increases the risk for bacterial or fungal infection. Signs of

overgrowth include white patches in the oral cavities. Other less common adverse events include myasthenia gravis, kidney damage, enterocolitis, malabsorption syndrome, eosinophilia, headache, hearing loss, ringing in the ear, itching, severe dizziness and pancreatitis.

2.1.3. Amphotericin B

This is an anti-fungal drug and has substantial activity against *Leishmania donovani*. When resistance to stibogluconate becomes high, amphotericin B became the first-line drug in many places. The dose is 15 alternate-day infusions of 1 mg/kg over 30 days (total dose, 15 mg/kg) or daily treatment with 1 mg/kg for 20 days (total dose, 20 mg/kg). The most common side-effects are chill, rigor and fever. Injection of antihistamine alleviates the symptoms. The drug exhibits nephrotoxicity and ototoxicity [16–18].

2.1.4. Liposomal amphotericin B

Liposomal amphotericin B is safer than amphotericin B and safest among all anti-VL drugs. It is given by intravenous infusion. The dose is 5 mg/kg × 3 days or 3 mg/ kg × 5 days. A single dose of 10 mg/kg has shown a cure rate of more than 95%. Liposomal amphotericin B replaced miltefosine as the first-line drug in the kala-azar elimination programme [19–21]. However, it is felt that this decision to switch over from miltefosine to liposomal amphotericin B could have been delayed as the programme was going on smoothly using miltefosine as the first-line drug.

2.1.5. Sodium stibogluconate

For more than last 6 decades, sodium stibogluconate was the effective drug treatment for visceral leishmaniasis and PKDL. The dose is 20 mg/kg/day given by intramuscular injection for 30 days [22]. Afterwards, the parasites developed resistance to the drug and it became ineffective. In order to overcome the drug resistance, the dose of the drug was increased but the cardiotoxicity of the drug increased. In Bihar, India, currently, the drug resistance is to the tune of 60% [23]. However, the drug is still used in places where the parasites are sensitive to the drug [24]. It can be given by both intramuscular and intravenous routes. The intramuscular injections are painful.

2.1.6. Urea stibamine

Urea stibamine was an effective anti-leishmanial drug. Since the developer did not keep any record of the compound, the drug had its natural death [25].

2.1.7. Anti-fungal agents

Anti-fungal agents like ketoconazole and anti-tuberculosis drugs were not found effective in the treatment of VL. However, both ketoconazole and pentostam were more effective than placebo against *L. braziliensis* panamensis cutaneous leishmaniasis. Oral ketoconazole is comparable in efficacy to this parenteral pentostam regimen and can be recommended as initial treatment for this disease. Sitamaquine is undergoing clinical trial for VL treatment and initial

results are encouraging [26]. Sitamaquine interacts with phospholipids and accumulates rapidly in the *Leishmania*. An advantage of sitamaquine is its short elimination half-life, preventing a rapid emergence of resistance.

2.1.8. Combination therapy

In view of the drug resistance and toxicity [27], it was expected that using the combination of two drugs will prevent or delay appearance of drug resistance, minimize toxicity, enhance efficacy and shorten duration of therapy. A safety and efficacy trial of combinations were conducted in Bangladesh [28].

1. Liposomal amphotericin B alone or a combination of:
2. single dose of Liposomal amphotericin B (day 1) with miltefosine (day 2–8)
3. single dose of liposomal amphotericin B (day 1) with paromomycin (day 2–11)
4. combinations of miltefosine with paromomycin (day 1–10)

All the combinations were non-inferior to the standard treatment with liposomal amphotericin B in usual doses. The combination therapy efficacy of all the regimens was ~95%. It was recommended that combination therapy would be an alternative to liposomal amphotericin B (10 mg/kg) in the context of kala-azar elimination programme in the Indian subcontinent.

3. Vector control

Vector control is of paramount importance in combating VL. When malaria control programme was carried out in India, as a collateral benefit, the incidence of kala-azar came down. In India, generally DDT is used for vector control but in Bangladesh and Nepal, synthetic pyrithroids are used. The exact role of long-lasting net (LLN) or long-lasting impregnated nets (LLIN) is not completely clear [29].

4. Elimination of kala-azar from Southeast Asian region

Keeping in view the high disease burden in India, Nepal and Bangladesh and availability of effective tools to diagnose the disease in the field situation and the effective and safe drug to treat the disease in an outpatient setting, the three countries embarked on eliminating the dreaded disease. In 2005, a memorandum of understanding was signed by India, Nepal and Bangladesh under the auspices of World Health Organization to cooperate and collaborate with each other to eliminate the disease from their respective countries. The target of elimination was less than 1 case per 10,000 people in an endemic area. Three countries of the WHO's Southeast Asian Region —Bangladesh, India and Nepal—are poised to eliminate VL (kala-azar) as a public health problem. The number of cases have reduced by 53%, from a high of 1,82,000 cases during 2005–2008 to 85,000 cases during 2011–2014. The 10,209 new cases reported in 2014 represent a 75%

decrease from 2005 when the kala-azar elimination programme was launched. In fact, Nepal has already achieved elimination and sustained for 2 years. Bangladesh is approaching fast towards elimination and India is expected to catch up [30–32].

5. Kala-azar vaccine

Conceptually, it is ideal to have a vaccine which will provide long-lasting immunity and simultaneously protect against VL and CL. Vaccines against VL and CL should be cost-effective. Currently, there are no effective and safe vaccines against VL but several are in various stages of developments. These candidate vaccines should be able to elicit balanced T_H1- and T_H2-mediated immune response. In view of safety concerns, live-oral vaccines are no longer recommended. The approach now is to insert a suicidal killed vector directly into the leishmania genome.

Research into first-generation vaccines based on whole-cell, killed leishmania parasites demonstrated that killed parasites showed efficacy as both therapeutic and prophylactic vaccines. Numerous preparations of killed parasites were tested. Although they showed good safety profiles, no first-generation vaccine using killed parasites has been demonstrated having sufficient efficacy as a prophylactic vaccine. The second generation of vaccines exploits the subunit, recombinant protein approach utilizing to augment the immune response. Third-generation vaccines derived from antigen-encoding DNA plasmids including heterologous prime-boost *Leishmania* vaccine have been examined for control and prevention of visceral leishmaniasis [33–35]. Vaccines based on recombinant protein and antigen-encoding DNA plasmids have given promising results.

6. HIV/kala-azar

Kala-azar patients may acquire HIV infection. Both the diseases lower the immunity of the person and opportunistic infections supervene. These patients easily acquire cryptosporidial infection and they respond to paromomycin. Tuberculosis is also common in these patients. Treatment of the three diseases requires a large number of drugs and may cause drug-drug interactions [36].

7. Concluding remarks

VL is potentially a life-threatening disease affecting the poorest of the poor in several regions of the world. There was a dearth of drugs, diagnostics and vector control methods until recently. The active collaboration of the scientists of the three countries joined by several other national and international agencies was the culmination of reliable diagnostics, drugs and vector control methods to diagnose and treat the disease. The third arm is vector control which is

essentially to interrupt transmission of the parasite. The WHO HQ and WHO regional office of the Southeast Asian Region extended formidable technical support to the programme. The role played by international agencies is unforgettable.

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