Transactions of the Royal Society of Tropical Medicine and Hygiene

journal homepage: http://www.elsevier.com/locate/trstmh

Mini-review

Immunological stimulation for the treatment of leishmaniasis: a modality worthy of serious consideration

Ahmed Mudawi Musa [a], Sassan Noazin [b], E.A.G. Khalil [a], Farrokh Modabber [a,c,d,*]

[a] *Institute of Endemic Diseases, University of Khartoum, Khartoum, Sudan*
[b] *World Health Organization, 1211 Geneva 27, Switzerland*
[c] *London School of Hygiene and Tropical Medicine, Keppel Street, London WCIE 7HT, UK*
[d] *Drugs for Neglected Diseases initiative (DNDi) 15 Ch. Louis-Dunant, Geneva 1202, Switzerland*

ARTICLE INFO

Article history:
Received 22 February 2009
Received in revised form 21 July 2009
Accepted 21 July 2009
Available online 26 August 2009

Keywords:
Leishmaniasis
Therapy
Immunochemotherapy
Immunotherapy
Immunocompromised
Immunostimulation

ABSTRACT

Instead of relying on drugs to reduce the parasite burden of leishmaniasis, and waiting for the effector immune response to develop in time to control the parasites, immunotherapy in conjunction with chemotherapy can rapidly induce the effector immune response. With a safe and potent drug plus an affordable therapeutic vaccine (immunostimulant), which remains to be developed, a single visit by patients with visceral or cutaneous leishmaniasis might be sufficient to induce a quick and lasting recovery. Drug toxicity and the emergence of resistance could also be dramatically reduced compared with present long-term monotherapy. Immunotherapy could be an effective addition to chemotherapy for leishmaniasis.

Leishmaniasis is caused by the *Leishmania* genus of protozoan parasites, and manifests in various forms: self-healing or chronic cutaneous leishmaniasis [CL, or post-kala-azar dermal leishmaniasis (PKDL)]; debilitating mucosal leishmaniasis (ML); and fatal (if not treated) visceral leishmaniasis (VL). The natural history of leishmaniasis is depicted in Figure 1 in a simplified schematic model applicable to VL and some forms of CL, with the assumption that there is a direct relationship between parasite load and disease. In an immunocompromised host the disease returns upon stopping treatment (Figure 1A). In cured or asymptomatically infected individuals, fulminating disease appears after immunosuppressive drugs or HIV infection. However, strong immunity is developed following successful recovery in immunocompetent individuals (Figure 1B). Hence the protective immune response is an important part of recovery from leishmaniasis.

The available treatment options are far from satisfactory as they are either expensive (amphotericin B) or toxic (antimonials), or resistant parasites have either emerged or are imminent with monotherapy (miltefosine and paromomycin). One solution is to combine these drugs to allow shorter, less toxic and more affordable treatment. This approach is being addressed by WHO, the Drugs for Neglected Diseases *initiative* and their endemic country partners. Another approach is immunochemotherapy, whereby a low-dose or short course of an effective drug (possibly one dose of amphotericin B) is given with one injection of a vaccine or immunomodulator to quickly induce the effector immune response (Figure 1C).

Immunotherapy was pioneered and used in thousands of CL and a few ML patients by Convit et al. in Venezuela.[1] Machado-Pinto et al. showed a highly significant cure rate for CL using Mayrink's vaccine (Biobras, Montes Claros, Brazil) plus a low dose of antimonials, with far fewer side effects of myalgia, severe pain at the site of injection, nausea, vomiting, headache, joint pain, etc. than associated with full doses of antimonials.[2] This vaccine has also been

* Corresponding author. Tel.: +41 79 815 4318.
E-mail address: fmodabber@dndi.org (F. Modabber).

0035-9203/$ – see front matter © 2009 Royal Society of Tropical Medicine and Hygiene. Published by Elsevier Ltd. All rights reserved.
doi:10.1016/j.trstmh.2009.07.026

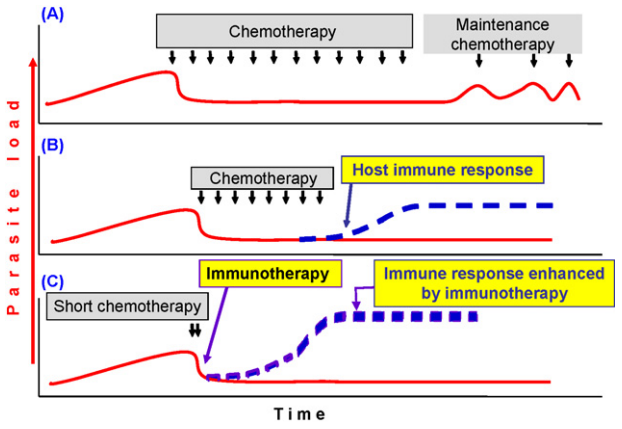

Figure 1. Schematic representation of chemotherapy plus immunotherapy in leishmaniasis. (A) Immunocompromised hosts. Chemotherapy reduces parasite load and induces short-term cure, but in the absence of an effector immune response, patients ultimately relapse and require maintenance treatment. (B) Immunocompetent patients. Treatment induces initial cure and the host's immune response maintains parasites under control. Long-lasting immunity is maintained as long as the host remains immunocompetent. (C) Immunochemotherapy in immunocompetent patients or immunocompromised patients with a certain minimum immunocompetence (after antiviral therapy in HIV-*Leishmania* coinfected individuals). Immunotherapy just after chemotherapy would induce an immune response sooner than in B above, reducing the need for long-term chemotherapy. This would reduce drug-related toxicity and the emergence of resistant parasites. Solid red line: parasite load; broken blue line: effector immune response; small black arrows: duration of chemotherapy. The more prominent broken blue line in part C indicates the response induced by immunotherapy in contrast to that naturally induced by the host, shown in part B.

used in some cases of HIV-*Leishmania* coinfection in Brazil. Musa et al. used alum-autoclaved *L. major* plus BCG (Razi Institute, Iran) in a preliminary trial involving 30 refractory PKDL patients, and showed immunochemotherapy to be superior to chemotherapy (final cure rates 100% vs. 40%, $P < 0.004$).[3] Chronic cases (>6 months) are difficult to treat with drugs alone, and are considered to be an important reservoir of infection. Badaro and colleagues used a mixture of defined recombinant antigens and an adjuvant (granulocyte-macrophage colony-stimulating factor) plus antimony to treat refractory ML patients successfully in a preliminary trial.[4] Combining IFN-γ and antimony were shown to be effective in the treatment of some VL and diffuse CL patients in Brazil.[5] No vaccine is available, but these observations are proof of the principle that immunostimulation is a valid approach in need of further investigation, and may be applicable to most forms of leishmaniasis, including HIV-*Leishmania* coinfection under certain conditions.

The mechanism involved is not fully understood. However, in PKDL lesions there is an influx of αβ T cells, but diminution of CD1a (Langerhans) cells. It seems that even in the presence of effector Th1 cells, parasites are not killed and lesions persist. Even IFN-γ treatment does not cure all patients with leishmaniasis. This may be because of down-regulation of B7-1 and up-regulation of B7-2 by IL-10, which leads to a predominantly Th2 response.[6] It should be noted that a minority of PKDL patients attained cure without showing a pure Th1 response.

As vaccination has a long-lasting effect and most drugs work quickly, it may be possible to introduce immunotherapy at the onset of chemotherapy. However, the best time must be studied in different forms of the disease, and in combination with different drugs.

Immunochemotherapy would have advantages over secondary chemoprophylaxis (maintenance therapy) for patients with HIV-*Leishmania* coinfection, once antiretroviral treatment had boosted their immune response to a certain level.

In all studies except Badaro's,[4] first-generation vaccines (FGV; killed promastigotes of different *Leishmania* spp.) were used. These are inexpensive (less than US$1.0/dose), but they do not meet the current requirements for new vaccines, and except for Venezuela, limited batches were produced for trials and they are not available commercially. Nevertheless, the experience gained from testing them in humans has paved the way for novel approaches to treatment. Despite failing to induce adequate prophylactic immunity in healthy individuals, FGVs were able to enhance recovery; possibly because, as shown in mice, different effector mechanisms may be involved in prophylaxis (CD4, Th1) and recovery (CD8).

With well-defined second-generation vaccines being developed, and well-characterized adjuvants now available or being tested for other diseases, notably cancers, immunochemotherapy should be considered as an alternative modality for the treatment of leishmaniasis, including HIV-*Leishmania* coinfection, PKDL and chronic refractory CL, the forms that are difficult to treat with available drugs.

Authors' contributions: AMM, SN, EAGK and FM undertook all the duties of authorship. FM and AMM are guarantors of the paper.

Funding: None.

Conflicts of interest: None declared.

Ethical approval: Not required.

References

1. Convit J, Ulrich M, Zerpa O, Borges R, Aranzazu N, Valera M, et al. Immunotherapy of American cutaneous leishmaniasis in Venezuela during the period 1990-99. *Trans R Soc Trop Med Hyg* 2003;**97**:469–72.
2. Machado-Pinto J, Pinto J, da Costa CA, Genaro O, Marques MJ, Modabber F, et al. Immunochemotherapy for cutaneous leishmaniasis: a controlled trial using killed *Leishmania (Leishmania) amazonensis* vaccine plus antimonial. *Int J Dermatol* 2002;**41**:73–8.
3. Musa AM, Khalil EAG, Mahgoub FA, Elgawi SH, Modabber F, Elkadaru AE, et al. Immunochemotherapy of persistent post-kala-azar dermal leishmaniasis: a novel approach to treatment. *Trans R Soc Trop Med Hyg* 2008;**102**:58–63.
4. Badaro R, Lobo I, Munos A, Netto EM, Modabber F, Campos-Neto A, et al. Immunotherapy for drug-refractory mucosal leishmaniasis. *J Infect Dis* 2006;**194**:1151–9.
5. Badaro R, Johnson Jr WD. The role of interferon-gamma in the treatment of visceral and diffuse cutaneous leishmaniasis. *J Infect Dis* 1993;**167**(Suppl 1):S13–7.
6. Ismail A, Khalil EAG, Musa AM, Elhassan IM, Ibrahim ME, Theander TG, et al. The pathogenesis of post kala-azar dermal leishmaniasis from the field to the molecule: does ultraviolet light (UVB) radiation play a role? *Med Hypotheses* 2006;**6**:993–9.