

Treatment of leishmaniasis in HIV-positive patients

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Although, in southern Europe, there has been considerable experience in the treatment of visceral leishmaniasis (VL) in HIV-positive patients, the optimal therapy has yet to be established. Pentavalent antimony salts, free amphotericin B deoxycholate (ABD) and lipidic formulations of amphotericin B are the drugs most commonly used. Treatment with pentavalent antimonials requires daily injections for 28 days, is not well tolerated and leads to initial clinical cure in only 66% of the co-infected cases. Free ABD has to be given, intravenously, for just as long, has significant toxicity and leads to initial clinical cure in even fewer cases (62%). In a prospective, comparative trial, treatment of co-infected cases with a pentavalent antimonial was found to have similar efficacy and toxicity to treatment with free ABD.

The duration of treatment and the associated toxicity may both be reduced by the use of lipidic formulations of amphotericin B. Anecdotal evidence and the results of non-randomized trials indicate that treatment with liposomal amphotericin B is highly effective. In a comparative trial, amphotericin B lipid complex was found to be not only as effective as a pentavalent antimonial but also better tolerated. At the moment, however, such lipidic formulations have only been tested against VL/HIV cases in Europe, not elsewhere in the world, and they remain very expensive.

However successful the treatment in terms of initial clinical cure, almost all VL/HIV cases develop VL relapses. Although the data available on secondary prophylaxis are limited and often inconclusive, it appears that regular treatment with a pentavalent antimonial drug, liposomal amphotericin B or amphotericin B lipid complex can reduce the incidence of leishmanial relapses in HIV-positive patients with VL.

The development of new regimens, use of new oral drugs (such as miltefosine) and the development of new antileishmanial drugs could all improve the treatment of HIV-related VL in the future.

The best way to treat leishmaniasis in HIV-positive patients is still the subject of considerable controversy. Few HIV-infected cases of cutaneous leishmaniasis (CL) or muco-cutaneous leishmaniasis (MCL) have been reported, and observations on the treatment of such cases of co-infection are largely anecdotal (Alvar *et al.*, 1997). Even for the much more common cases of visceral leishmaniasis (VL) with HIV co-infection, there have been too few comparative studies on the effectiveness of the various treatments possible, both in terms of the drugs and regimes used, for any clinician to make a

well informed decision. Experience in the treatment of leishmaniasis in HIV-positive patients has been more-or-less limited to the countries in the Mediterranean basin. There are insufficient data on the effectiveness of antileishmanial drugs in HIV-positives from other parts of the world to make any valid generalisations about the results of treatment.

As HIV-positive patients usually develop VL when suffering from severe immunodepression and frequently from other serious illnesses, the evaluation of the effectiveness and toxicity of antileishmanial drugs in those co-infected with VL and HIV is particularly difficult. Although 11%–27% of such cases die in the month following the diagnosis of their VL, these deaths are rarely attributed to

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leishmaniasis or to the toxicity of the drugs given to treat the disease (Laguna *et al.*, 1997; López-Vélez *et al.*, 1998; Pintado *et al.*, 2001). The clinical criteria used to assess response to the antileishmanial treatment, so useful in the immunocompetent patient, have less value in the immunodeficient. Only initial cure must be considered since, if the immunodepression continues, the probability of total recovery is almost nil and the patients tend to have several relapses of their VL. The parasitological response to antileishmanial treatment of HIV-positive cases of VL has only been assessed on a very few occasions (Montalbán *et al.*, 1990; Altés *et al.*, 1991; Medrano *et al.*, 1992; Laguna *et al.*, 1997; López-Vélez *et al.*, 1998; Pintado *et al.*, 2001). Finally, other drugs given to HIV-positive patients can increase the toxicity of some antileishmanial drugs.

PENTAVALENT ANTIMONIALS

The pentavalent antimonials are the most frequently used drugs in the treatment of VL in HIV-positive patients. The results of several retrospective studies indicate a high level of clinical response to such drugs (Berenguer *et al.*, 1989; Montalbán *et al.*, 1990; Altés *et al.*, 1991; Medrano *et al.*, 1992; López-Vélez *et al.*, 1998). However, the drug doses and criteria used to identify clinical recovery were not uniform, the observation of clinical cure was made difficult by the co-existence of other, AIDS-related diseases, and clinical improvement does not necessarily mean that there has been parasitological recovery (Laguna *et al.*, 1994); in all of these series a high percentage of patients relapsed in the year following the end of treatment. Three-week treatments with pentavalent antimonials appear less effective than those lasting 4 weeks (Rosenthal *et al.*, 1995; Laguna *et al.*, 1997). In the early 1990s, no one knew very much about the frequency of initial recovery or the toxicity associated with antimonial use in those co-infected with

HIV, and no one had much experience with the use of other antileishmanial drugs in the treatment of HIV-associated VL. Laguna *et al.* (1999) therefore carried out a multi-centre, prospective, randomized and open study of HIV-positive Spanish patients who were suffering from their first episode of VL. In this study, the effectiveness and toxicity of 4 weeks' treatment with meglumine antimoniate (at 20 mg Sb^v/kg.day) were compared with those of treatment with amphotericin B deoxycholate (at 0.7 mg/kg.day) for 4 weeks. Intention-to-treat analysis of the data showed that only 29 [65.9%, with a 95% confidence interval (CI) of 50%–79%] of the 44 patients treated with the antimonials achieved initial parasitological recovery, although another 10 of the 44 failed to complete treatment (five because of toxicity and five because of death). Twenty-nine (85%; CI = 68%–94%) out of the 34 patients who completed antimonial treatment showed a clearing of parasites from their bone-marrow (Laguna *et al.*, 1999). As the patients did not receive secondary prophylaxis, many relapsed; the probability of relapse at 12 months was estimated to be about 70%.

The results of most of the relevant studies highlight the particular difficulties that exist in the antimonial treatment of VL in patients infected with HIV, both in terms of the poor effectiveness and toxicity. In those with HIV, antimonials cause more toxicity than in HIV-negative patients (Pintado *et al.*, 2001), with clinical pancreatitis, myocarditis and renal insufficiency all being reported (Berenguer *et al.*, 1989; Montalbán *et al.*, 1990; Altés *et al.*, 1991; Medrano *et al.*, 1992; Laguna *et al.*, 1997, 1999; López-Vélez *et al.*, 1998; Delgado *et al.*, 1999; Pintado *et al.*, 2001). Biochemical pancreatitis is a very frequent adverse effect in both the immunodeficient and immunocompetent. Most HIV-infected patients show a slight but asymptomatic increase in serum concentrations of amylase and lipase. Antimonial toxicity leads to 11%–28% of HIV-positive patients discontinuing treatment, mainly because of clinical pancreatitis (Laguna *et al.*,

1997, 1999; Delgado *et al.*, 1999; Pintado *et al.*, 2001). Electrocardiographic changes, such as a small increase in the Q-Tc interval and alterations in the T wave, are detected in up to 14% of HIV-infected patients (Laguna *et al.*, 1999). The true significance of these slight electrocardiographic alterations is not known and treatment need only be stopped when the Q-Tc interval rises above 500 ms or serious arrhythmias appear (Herwaldt and Berman, 1992; Laguna *et al.*, 1999).

AMPHOTERICIN B DEOXYCHOLATE

Although, amphotericin B deoxycholate (AB) has been found to be one of the most active anti-*Leishmania* drugs in experimental VL, its clinical use has been limited by difficulties in its administration and its toxicity. Only in areas where VL cases show a high level of resistance to antimonials has AB been much used, with great effectiveness (Murray, 2000). There has been little use of AB in HIV-positive patients with VL (Rosenthal *et al.*, 1995; López-Vélez *et al.*, 1998; Pintado *et al.*, 2001). Though the results obtained so far are generally encouraging, only 28 (62.2%; CI = 47%–76%) of the 45 patients treated with AB in the comparative study by Laguna *et al.* (1999) recovered, giving a recovery 'rate' similar to that seen with meglumine antimoniate. Many (33%) of the patients treated with AB failed to complete treatment (because of toxicity, failure to comply or death) but, as with the antimonial, most (93%; CI = 76%–98%) of those who did complete treatment achieved an initial parasitological recovery. The frequency of relapse with 12 months of completing treatment was similar for AB and meglumine antimoniate (Laguna *et al.*, 1999).

As with the antimonials, AB induces a higher level of toxicity in patients infected with HIV than in their HIV-negative counterparts. In the study by Laguna *et al.* (1999), up to 60% of patients showed some adverse effect during treatment with AB, especially anaemia, nephrotoxicity and hypopotassemia; 40%

of the patients had <8 g haemoglobin/dl and 36% showed renal toxicity, although only 20% required the suspension of their treatment.

LIPIDIC AMPHOTERICIN B

In an attempt to reduce the toxicity associated with AB treatment, the drug has been combined with phospholipids which, on dispersing in water, spontaneously form vesicular structures, some of them spherical, composed of drug solution surrounded by one or more layers of phospholipids. Such lipidic preparations allowed a great advance in the treatment of VL during the 1990s. Three commercial preparations of lipidic AB have been used: amphotericin B lipidic complex (ABLC; Abelcet[®]; Liposome Company, Princeton, NJ); liposomal amphotericin B (ABL; AmBisome[®]; NeXstar, San Dimas, CA); and amphotericin B cholesterol dispersion (ABCD; Amphotec[®]; Sequus Pharmaceuticals, Menlo Park, CA). Although these lipidic AB formulations have different pharmacodynamic characteristics, all are taken up by tissue macrophages, especially those of the liver and spleen. Once taken inside a macrophage by endocytosis, the cell's phospholipases break open the liposome, freeing the AB. In this way, the AB is targeted at the host cells of the leishmanial amastigote (Janknegt *et al.*, 1992).

In experimental VL, such lipidic amphotericins have been found to be superior to antimonials and free AB, allowing greater doses of AB and higher frequencies of recovery yet giving less toxicity. Comparative studies in experimental VL with similar doses of ABCD, ABLC and ABL have proved that every preparation of lipidic AB has a different efficacy against *Leishmania* and that the intensity of parasite suppression is greater in the liver than in the spleen (Mullen *et al.*, 1997).

Liposomal Amphotericin B

ABL has been tested in immunocompetent patients from Europe, Brazil, Kenya and

India. This preparation, in a total dose of at least 20mg/kg given in five or more doses of 3–4 mg/kg, has proved itself to be very effective in European patients with VL (Davidson *et al.*, 1994, 1996). In India and Kenya, respective total doses of 6 mg/kg (2 mg/kg.day on days 1, 5 and 10) and 14 mg/kg (2 mg/kg.day on days 1–6 and 10) cured all the patients treated (Berman *et al.*, 1998). In Brazil, however, a total dose of 20 mg/kg (2 mg/kg.day on days 1–10) only cured 83% of those treated (Berman *et al.*, 1998). It seems, therefore, that the relatively low effective doses of ABL used in India would not be useful in Europe, Africa or Brazil. In India, even a single dose of 5 mg ABL/kg cured 91% of the patients given it (Murray, 2000). Adverse effects following ABL treatment are rare and usually mild (Meyerhoff, 1999).

Treatment with ABL has also been used on European patients with VL/HIV co-infection, almost always after the patients have developed relapses. The doses and lengths of treatment have varied, although total doses have usually been at least 20 mg/kg (Lazanas *et al.*, 1993; López *et al.*, 1993; Torre-Cisneros *et al.*, 1993; Davidson *et al.*, 1994; Laguna *et al.*, 1995). Only two prospective, non-comparative studies have evaluated the effectiveness and toxicity of ABL on Italian patients dually infected with HIV and VL. In the first of these studies, seven patients were treated with 100 mg ABL/day for 21 days (giving a total dose of 29–38.9 mg/kg); the five patients who showed initial parasitological recovery all had post-treatment relapses (Davidson *et al.*, 1994). In the second study, in an attempt to reduce the number of relapses, and knowing that AB from ABL has a long tissue half-life, Russo *et al.* (1996) used high and intermittent doses (4 mg/kg.day on days 1–5, 10, 17, 24, 31 and 38) to treat 10 HIV-positive patients. This regimen appeared similar in efficacy to the lower dosage used by Davidson *et al.* (1994) and did not manage to prevent relapses. Studies comparing the efficacy of

ABL with that of other antileishmanial drugs have not been carried out on HIV-positive cases of VL.

Amphotericin B Lipidic Complex

Infusions of ABLC have been tested clinically in several open studies, with immunocompetent Indian cases of VL who had no response or who had relapsed after receiving a pentavalent antimonial. The results indicated that doses of 2 or 3 mg/kg.day for 5 days cured 80% and 100% of patients, respectively (Sundar and Murray, 1996; Sundar *et al.*, 1997). The only significant toxicity observed was the frequent occurrence of shivers and fever during infusion. These side-effects lasted for < 1 h, could not be prevented with paracetamol, and had disappeared in half of the patients by the fifth dose administered. It remains unclear if these encouraging results are reproducible in other parts of the world since, as has been observed with other lipidic AB preparations, the effective doses of ABLC for treatment of VL seem to vary from one country to another.

Experience of ABLC for the treatment of VL in the HIV-infected is limited to a single, open, randomized, prospective and multi-centre pilot trial (Laguna *et al.*, 2003), in which Spanish patients suffering from their first episodes of VL were treated with ABLC (3 mg/kg.day for 5 or 10 days) or meglumine antimoniate (20 mg Sb^v/kg.day for 28 days). Overall, 57 patients were included in the study: 18 on ABLC for 5 days, 20 on ABLC for 10 days, and 19 on meglumine antimoniate. In an intention-to-treat analysis, a similar, generally low level of effectiveness was found for the three groups (with 33%, 42% and 37% of those treated showing initial response, respectively). The antimonial appeared the most toxic, 10 of the 19 patients given meglumine antimoniate stopping treatment early because of serious side-effects. These preliminary results indicate that the total dose of ABLC needed

to cure VL in HIV-positive patients in the Mediterranean area is >30 mg/kg. Further studies are necessary to confirm these observations and to test other doses.

OTHER TREATMENTS

There have been numerous observations on other treatments for VL in patients co-infected with HIV but these have been made in uncontrolled studies and many are merely anecdotal, weakening their scientific significance. A few cases have been treated with pentamidine (Montalbán *et al.*, 1990; López-Vélez *et al.*, 1998) and there are unconfirmed reports of the use of paromomycin or azoles, either alone or combined with antimonials (Alvar *et al.*, 1997).

The combination of Sb^v with allopurinol is well tolerated in HIV-positive patients (Laguna *et al.*, 1994; Delgado *et al.*, 1999) and, as seen with treatments based only on Sb^v , the highest frequency of recovery required treatment for at least 4 weeks. There is, however, no good evidence to show that this combination is any better than Sb^v alone (Dellamonica *et al.*, 1989; Mora *et al.*, 1990; Herwaldt and Berman, 1992; Laguna *et al.*, 1994; Rosenthal *et al.*, 1995).

HIV infection is known to produce a sharp drop in the host's secretion of interferon- γ (IFN- γ) and the combination of a pentavalent antimonial with IFN- γ appears to have a synergistic effect in the treatment of VL in immunocompetent patients (Górgolas *et al.*, 1994). The combined treatment of HIV-positive VL cases with an Sb^v drug and IFN- γ therefore seems an attractive idea. Only a few co-infected cases have yet been treated with such a combination, however, and the results have been inconclusive (Lortholary *et al.*, 1990; Torre-Cisneros *et al.*, 1993; Górgolas *et al.*, 1994). Worryingly, in two patients with VL and Kaposi's sarcoma, treatment with IFN- γ induced a rapid progression of the tumour (Albrecht *et al.*, 1994). In the early 1990s, a Spanish, multi-centre study investigating the potential

benefits of adding IFN- γ to a pentavalent antimonial, in the treatment of VL in HIV-positives, was suspended after the results of an interim analysis indicated that the combination was no better than the antimonial alone (unpubl. obs.)

TREATMENT OF RELAPSES

After their first episode of VL, patients co-infected with HIV tend to have further episodes, even after achieving an initial parasitological recovery. These new episodes are usually recrudescences resulting from the inability of the host's immune system to control the leishmanial infection. The characterization of *Leishmania* isolates collected from the same patients during different episodes of VL indicates that very few of the relapses are the result of post-treatment re-infection (Morales *et al.*, 2001).

Relapses are often treated with the same drug used against the initial episode of VL (López-Vélez *et al.*, 1998; Pintado *et al.*, 2001). If the drug used is an antimonial, however, the re-treatments may be markedly less effective than the first treatment because of the appearance of resistance in the leishmanial parasites (Alvar *et al.*, 1987; Lortholary *et al.*, 1990). The use of a drug to treat relapses that is different to the one used against the initial episode has not been adequately explored. There have been numerous reports showing that liposomal amphotericin B is highly effective and not very toxic when used to treat relapses following Sb^v treatment, although such re-treatment does not prevent further relapses (Davidson *et al.*, 1994; Laguna *et al.*, 1995; Russo *et al.*, 1996).

SECONDARY PROPHYLAXIS

Many of the opportunistic infections that appear in HIV-positive patients show a relapsing course but there are highly effective primary or secondary prophylactic treatments

available for most of them. Pentamidine every 3 or 4 weeks, ABL in fortnightly doses, allopurinol or itraconazol have all been used as prophylaxis against VL episodes in cases of VL/HIV co-infection. Although none of these treatments has clearly been shown to be effective, there are some data available that indicate that secondary prophylaxis could be useful in the prevention of VL relapses in HIV-positive patients. Thus in an open, retrospective but non-randomized study, it was observed that monthly treatment with a pentavalent antimonial seemed to prevent relapses (Ribera *et al.*, 1996). In another study, also retrospective, it was shown that patients treated monthly with a pentavalent antimonial or liposomal amphotericin B were significantly less likely to relapse than patients who did not receive such prophylaxis (Pintado *et al.*, 2001). Finally in the only open, prospective and randomized clinical assay done to date, patients treated with ABLC (3 mg/kg every 21 days) were compared with patients not receiving prophylaxis. After a year of follow-up, 50% of those receiving prophylaxis but 78% of the other patients had relapsed, indicating that ABLC could be effective as secondary prophylaxis against VL in HIV-positive patients (R. López-Vélez and S. Videla, unpubl. obs).

CONCLUSIONS AND FUTURE TREATMENTS

The response to treatment of VL in HIV-infected patients is worse than that in immunocompetent patients. Treatment with pentavalent antimonial drugs or amphotericin B deoxycholate for a period of 4 weeks produces a high level of toxicity and fewer than 70% of the patients who start treatment achieve an initial parasitological cure. These drugs require prolonged treatment, usually involving hospitalization and strict clinical and analytical control to detect

toxicities early on. Liposomal amphotericin B may offer better effectiveness and much less toxicity and can be administered to outpatients and in short cycles. However, use of this drug in HIV-positives has only been explored in a few studies and with a relatively small number of patients. There has been even less experience with the use of lipidic amphotericin B complex, although this formulation seems better tolerated and at least as effective as the antimonials.

One major drawback with all the lipidic amphotericins is their relatively high cost, although adequate pharmaco-economic studies, which evaluate not only the direct cost of each of the available antileishmanial drugs but also the associated hospital costs, have not been carried out. Lipidic amphotericins are expensive to buy but, as they can be administered in the outpatient section of a hospital and as part of a regimen lasting <2 weeks, they may be relatively cheap to use. The introduction of highly effective antiretroviral treatment (HAART) has meant a profound change in the epidemiology of VL/HIV co-infection, with a sharp drop in the annual number of new cases of VL (Tumbarello *et al.*, 2000; López-Vélez *et al.*, 2001). Nevertheless, cases of VL and CL in HIV-positives will continue to occur, especially in those areas where the HIV pandemic is spreading into new areas where leishmaniasis is already endemic.

The development of new regimens based on already-tested drugs, experimentation with oral drugs such as miltefosine (Jha *et al.*, 1999), and the development of new antileishmanial drugs will hopefully improve the future treatment of leishmaniasis in HIV-positive patients.

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