Effectiveness and Safety of Amphotericin B Deoxycholate, Amphotericin B Colloidal Dispersion, and Liposomal Amphotericin B as Third-Line Treatments for Cutaneous and Mucocutaneous Leishmaniasis: A Retrospective Study

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Abstract. Cutaneous leishmaniasis (CL) and mucocutaneous leishmaniasis (MCL) are endemic diseases in America, especially in some countries such as Colombia. Among the therapeutic options is amphotericin B (AB). Nevertheless, its lipid-associated formulations have better safety profiles and effectiveness in other diseases, so far with no comparative studies in CL or MCL. We conducted a retrospective descriptive study describing the effectiveness and adverse effects of AB deoxycholate (ABD), AB colloidal dispersion (ABCD), and liposomal AB (LAB) as third-line treatments for CL and MCL. The effectiveness of LAB (88.5%) was greater than those of ABCD (66.6%) and ABD (80.8%). There were also fewer adverse effects in the LAB group (46.2%) than in the ABD (96.1%) and ABCD (80.9%) groups. LAB is an alternative for the treatment of CL and MCL in patients with therapeutic failure to first- and second-line drugs; findings suggest it might be less toxic and more effective than ABD and ABCD.

INTRODUCTION

Leishmaniasis is a disease caused by protozoa of the genus *Leishmania* that generates a wide spectrum of clinical manifestations that depend on the species of *Leishmania* involved and the immune factors of the host.^{1–3}

In the New World, leishmaniasis is found from south Texas to South America, with Colombia being the country with the second most cases of leishmaniasis (7,541 cases in 2015). In the country, cutaneous leishmaniasis (CL) is the most frequent clinical form—98.2%—, followed by the mucocutaneous (mucocutaneous leishmaniasis; MCL)—1.2%—and visceral—0.5%—forms.^{1,4,5}

Most cases of leishmaniasis in Colombia are caused by the species *Leishmania panamensis* and *Leishmania braziliensis*, which are associated with an increased risk of mucosal involvement, with the latter also being associated with an increased risk of therapeutic failure.^{6,7} The recommended treatment in the country is systemic, with the first line being pentavalent antimonials, whose effectiveness is approximately 75%. For cases of therapeutic failure and for some special cases, pentamidine, miltefosine, or amphotericin B (AB) are recommended.^{8,9} However, there is great concern about therapeutic failure with the pentavalent antimonials. The experience in the Old World has shown that this failure can increase in incidence over time, even to the point of the drugs becoming useless, whereas a decrease in sensitivity to antimonials has been described in Colombia, especially in repeated cycles of drug administration and for *L. braziliensis* and *L. panamensis*.^{7,10,11} The second and third cycles of pentavalent antimonials, recommended in some guidelines, are associated with levels of effectiveness of only 59% and 50%, respectively.^{11,12} The effectiveness of pentamidine can reach up to 87%, whereas the response to miltefosine depends on the species, being most frequently associated with incidences of failure in *L. braziliensis* between 12% and 47%.^{7,13,14} The aforementioned circumstances give relevance to the study of

treatment strategies for therapeutic failure, with a particular interest in AB.

AB, a macrolide polyene used in the New World for 50 years in cases of therapeutic failure, binds to the ergosterol of the membrane of the parasite, increasing its permeability, which leads to successful therapy in 95–100% of cases.^{14,16} Nevertheless, it is a drug with two main mechanisms of significant toxicity. First, recognition by Toll-like receptor 2 (TLR 2) triggers the release of inflammatory molecules such as interleukin 1B, leading to fever, arthralgia, headache, and gastrointestinal symptoms associated with the infusion. Second, vasoconstriction of the afferent arteriole and uptake by renal tubular cells trigger hypokalemia, hypomagnesemia, renal tubular acidosis, polyuria, and hyperazotemia.¹⁶ However, in lipid-associated formulations, deoxycholate is replaced by lipids that surround the AB monomers, reducing their uptake by susceptible tissues, thus reducing toxicity, mainly renal.¹⁶

The CL and MCL studies that have evaluated the role of the lipid-associated formulations of AB are mostly case reports and case series that have been performed primarily on travelers as the first or second lines of management, where an effectiveness of between 50% and 93% has been described. In these studies, the dosage and administration protocols have been variable, with a total average dose ranging between 7.5 mg/kg and 36 mg/kg, administered on different schedules.^{15,17,18}

MATERIALS AND METHODS

A retrospective descriptive study (opportunity sampling) was conducted with patients diagnosed with CL and/or MCL who were treated at the Central Military Hospital (Hospital Militar Central [HMC]) in Bogota, Colombia, between January 2010 and March 2016. The patients included in the study were diagnosed by direct smear, polymerase chain reaction, and/or anatomopathological study and presented failure with schedules and doses appropriate to the first two lines of treatment (pentavalent antimonials and miltefosine and/or pentamidine, respectively). Following the first two lines of treatment failure, the patients received treatment according to the protocol established by the Infectious Diseases

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Department of the hospital (see Table 1), which comprises AB in any of the three chemical forms: AB deoxycholate (ABD)—Fungizone® (ABD; Ben Venue Laboratories, Inc. for Bristol-Myers Squibb Company, Bedford, OH), AB colloidal dispersion—Amphocil® (ABCD; InterMune Inc., San Francisco, CA), or liposomal AB—Ambisome® (LAB; Gilead Sciences, Inc., Foster City, CA). The follow-up of the protocol execution lasted for at least 6 months after administration of the drug.

For analyzing the data, the following descriptors were considered: location of the lesions, diagnosis, total AB dose in mg, total dose in mg/kg, dose in mg/kg/day, adverse effects by the system, pretreatment creatinine, highest creatinine during treatment and posttreatment, time to cure, and therapeutic failure. We reviewed 165 histories of patients diagnosed with CL or MCL who received AB, of which 90 patients were excluded (see Figure 1).

This study was funded and authorized by the Ethics Committee of the Central Military Hospital (HMC) in Bogota, Colombia.

RESULTS

General characteristics. All the patients were men, with an average age of 27.2 years, which was similar among the treatment groups. The median time of evolution of the disease was 64 weeks. The median size of the lesions was 5 cm², and 52.1% of the patients had a single lesion. Differences between groups are shown in Table 2.

The skin lesions were located predominantly on the head and neck (49.3%) and on the upper extremities (31.5%). The location of mucosal lesions was mostly nasal (94.11%), wherein six of them with perforation of the nasal septum. The extent of the mucosal lesions was measured directly and/or by nasal fibroscopy.

There was comorbidity in 12.3% of the patients, including arterial hypertension, gouty arthritis, and low idiopathic CD4, and 15% of them received concomitant drugs, mainly antihypertensives.

All patients received at least one treatment cycle with N-methylglucamine, with 45.1% receiving two or three cycles. No patient was treated with sodium stibogluconate.

Treatment with pentamidine as the second-line therapy was received by 93.1% (68) of the patients. Of these, 13 received both miltefosine and pentamidine and the rest received only miltefosine as a second-line treatment.

Treatment protocol. The protocols established for different ABs are summarized in Table 1. ABD was received by 26 (35.6%) patients, ABCD by 21 (28.7%), and LAB by 26 (35.6%).

Regarding the administration route of the drug, the LAB and ABCD chemical formulations were administered through a

peripherally inserted central catheter (PICC), whereas that of ABD was administered through a central venous catheter. The treatment protocols that the patients received are summarized in Table 3, as is the adherence to the protocols established in our institution.

Effectiveness. Effectiveness was defined as the total cicatrization of the cutaneous or mucosal lesion for up to 6 months after treatment. The effectiveness of LAB (88.4%) was greater than those of ABCD (66.6%) and ABD (80.7%).

Therapeutic failure was observed in 19.2%⁵ of the patients who received ABD, 11.5%³ of those who received LAB, and 33.3%⁷ of those who received ABCD.

Based on the form of leishmaniasis, the cure rates were 78.5% for CL and 82.3% for MCL; in this group, the patients who presented perforation of the nasal septum had a lower cure rate (50%).

The group with three or more lesions had a higher cure rate (100%) than did the group with one to two lesions (76.6%).

Regarding the kg/day dose, no differences in effectiveness could be established between the 3 mg/kg/day or 5 mg/kg/day doses in the two groups that received lipid-associated formulations because only four patients received this last dose. In the ABD group, patients who received < 0.7 mg/kg/day, 0.7–1 mg/kg/day, and > 1 mg/kg/day had cure rates of 91.6%, 90.9%, and 0%, respectively.

Regarding the total dose/kg, P10 (10th percentile) and P90 (90th percentile) were determined for each chemical formulation. In the ABD group (P10 = 520 mg and P90 = 1,509 mg), 50% of those who received P10 and 100% of those who received P90 were cured. In the ABCD group (P10 = 1,190 mg and P90 = 2,268 mg), 40% of those who received P10 and 100% of those who received P90 were cured. In the LAB group (P10 = 1,226 mg and P90 = 2,366 mg), 50% of those who received the P10 dose and the P90 dose were cured.

Adverse effects. Overall, 54/73 (72.6%) of the patients presented adverse effects, of which 25/26 (96.1%) were from the ABD group, 17/21 (80.9%) were from the ABCD group, and 12/26 (46.1%) were from the LAB group.

The most frequent adverse effects were electrolyte alterations in 36.9%, fever in 35.6%, asthenia/chills in 23.2%, vomiting in 19.1%, headache in 19.1%, nausea in 17%, and elevation of transaminases in 16.4% of the patients. In the ABD group, 30.7% of the patients presented bacteremia, mostly associated with a central venous catheter, requiring temporary suspension of ABD.

Hepatic alterations were present in 16.4% of the patients, consisting of 42.3% of the ABD group and 3.8% of the LAB group. The latter had previously received ABCD.

Regarding alterations in renal function, the median creatinine in the total population was 0.96 ± 0.15 mg/dL. The differences between the baseline and highest creatinine levels

TABLE 1
Institutional protocols for different types of amphotericin

(n), variable, rate (%), unless otherwise specified	Amphotericin		
	ABD	ABCD	LAB
Daily dose (mg/kg)	0.7–1	3 or 5	3 or 5
Total dose (mg/kg)	25 (22–28)	21 or 35	21 or 35
Days of treatment (days)	Consecutive days	1, 2, 3, 4, 5, 14, and 21	1, 2, 3, 4, 5, 14, and 21
Number of doses	25 (25–30)	7	7

ABD = amphotericin B deoxycholate; ABCD = amphotericin B colloidal dispersion; LAB = liposomal amphotericin B.

FIGURE 1. Flowchart of included and excluded patients. AB = amphotericin B; ABD = amphotericin B deoxycholate; ABCD = amphotericin B colloidal dispersion; LAB = liposomal amphotericin B.

were 0.41 mg/dL for the ABD group, 0.05 mg/dL for the ABCD group, and 0.16 mg/dL for the LAB group. Two patients in the LAB group had a 1-fold increase in the baseline creatinine level, whereas in the ABD group, one patient had a 1-fold increase in the baseline creatinine level and one patient had a 2-fold increase in the baseline creatinine level. The creatinine values returned to levels similar to the baseline at the end of the treatment. An increase of ≥ 0.3 mg/dL with respect to the baseline creatinine level (included within the Acute Kidney Injury Network criteria for acute renal failure) was observed in 18 patients, of which 13 belonged to the ABD group and five to the LAB group.

Of the patients who received a single, two, or three previous treatments with N-methylglucamine, 70%, 75%, and 100%, respectively, presented adverse effects. Likewise, 93.3% of patients who had previously received any chemical formulation of AB presented adverse effects.

The major results are summarized in Table 4.

DISCUSSION

The purpose of this study was to perform a retrospective descriptive study describing the effectiveness and adverse effects of ABD, ABCD, and LAB as third-line treatments for the New World CL and MCL, a context in which no previous study has been conducted. Our findings cannot be interpreted as it was a randomized, controlled trial; thus, comparison of

different treatments must be made cautiously. We nonetheless tentatively conclude that LAB at a dose of 3–5 mg/kg/day administered in seven doses on days 1–5, 14, and 21 is an alternative treatment for patients with CL and/or MCL who had presented therapeutic failure to the first, second, and/or third lines of treatment (defined according to international and national management guidelines),^{8,9} apparently with better effectiveness and safety profiles than ABD and ABCD; this was statistically evidenced in adverse effects.

In several countries, AB is used as a second- or third-line drug for the treatment of tegumentary or visceral leishmaniasis once the drugs considered to be first line (pentavalent antimonials, pentamidine, and miltefosine) have failed. The effectiveness levels of pentavalent antimonials, pentamidine, and miltefosine are 50–80%, 87%, and 53–88%, respectively, and depend on the species of the parasite, the geographical acquisition area, the management schedule, and the immunological condition of the host.^{7–12} Therefore, the number of patients who require treatment with presentations of AB is considerable.

ABD was the first AB developed. It requires daily application and is associated with adverse renal, hepatic, and electrolyte effects, among others. AB in its lipid-associated formulation was developed to decrease toxicity, especially renal, and to increase effectiveness. These improvements are achieved because the lipid association decreases the amount of medication free in the plasma and, therefore, its uptake by

TABLE 2
Demographics and characteristics of current disease of CL and MCL

Variable, median (IQR), unless otherwise specified	Amphotericin, (n)			Total (73)	P-value
	ABD† (26)	ABCD‡ (21)	LAB§ (26)		
Age in years $\pm$ SD	28.5 $\pm$ 4.8	26 $\pm$ 3.5	26.5 $\pm$ 4.2	27.2 $\pm$ 4.3	0.24
CL	17 (65.4 %)	19 (90.5 %)	20 (76.9 %)	56 (76.7 %)	0.17
MCL	9 (34.6 %)	2 (9.5 %)	6 (23.1 %)	17 (23.3 %)	0.17
Size of the lesion (cm ²)	5.5 (2–11.9)	3 (1.2–12)	6.2 (3–12)	6 (2–12)	0.29
Time of evolution of the disease (weeks)	68 (48–112)	52 (40–96)	56 (40–144)	58 (42–110)	0.96

ABD = amphotericin B deoxycholate; ABCD = amphotericin B colloidal dispersion; IQR = interquartile range; LAB = liposomal amphotericin B.
Statistical analysis used: Pearson's chi-squared test

TABLE 3
Treatment and adherence to institutional protocols of AB

Variables, median (range), unless otherwise stated	Amphotericin, (n)		
	ABD (26)	ABCD (21)	LAB(26)
Treatment protocol			
Dosage (mg/kg/day)	0.70	3	3
Total dose (mg)	1,490 (350–1,650)	1,470 (1,050–2,485)	1,585.5 (1,197–2,485)
Total dose (mg/Kg)	21.4 (5.8–25.8)	21 (19.4–26.4)	21 (19.3–34.3)
Days of treatment	33.5 (5–57)	23 (19–30)	21 (16–29)
Number of doses	30 (5–48)	7	7
Scheme adherence			
Total dose (%)	12.6	100	100
Treatment days (%)	14.2	72.3	72.3
Daily dose (%)	42.3	100	100

ABD = amphotericin B deoxycholate; ABCD = amphotericin B colloidal dispersion; LAB = liposomal amphotericin B.

susceptible renal cells. The formulation also favors phagocytosis by macrophages and their distribution in the reticuloendothelial system, which increases the concentration of the drug in the phagosome, where the parasite is located.^{14–16,19–22}

For the present study, identification of the *Leishmania* species for each patient was not performed. However, Pérez JE et al.⁷ published a series of patients who received medication in the HMC for CL. In the said publication, the species were identified by hsp70 PCR–restriction fragment length polymorphism, finding that 95.4% of the patients were infected by *L. braziliensis* and 2.3% by *L. guyanensis* even though the predominant parasite species in the country is *L. panamensis*. Therefore, it is most likely that most patients in our study were infected by *L. braziliensis*.⁷

The effectiveness of LAB was greater than those of ABCD and ABD, although statistical significance was not established possibly because of sample size. The greater effectiveness of LAB could be attributed to pharmacokinetic and pharmacodynamic advantages, as shown in murine models where the concentration of LAB has shown increasing intralesional concentration of the drug and subsequent reduction of the parasitic load.²³

Similarly to what has been reported by other authors, we observed no difference in the probability of cure among patients who presented CL and MCL, except for those who presented perforation of the nasal septum.

Scarce cases of AB-resistant *Leishmania* have been described, in which changes in the sterol content of the membrane of the parasite have been documented, probably

because of variations in the sterol methyltransferase enzyme, losing the site of action of any AB.^{24,25} It is probable that the cases of therapeutic failure in our study are rather due to immunological or idiosyncratic conditions of the patient or pharmacodynamic variables^{23,26} and factors, which are also related to therapeutic failure of other antimicrobial agents.⁷

For treatment with the lipid-associated ABs, patients received 3 mg/kg/dose. However, a percentage of them presented therapeutic failure after which they received the drug at a dose of 5 mg/kg/dose, obtaining a cure rate of 75%. This is not surprising, considering the reports of other authors, where a successful treatment by repeating the dose of the drug was observed.¹⁵

Patients who presented more and larger lesions were more likely to be cured; these variables were more imperative in the LAB group. Nonetheless, these findings do not coincide with those described in other studies.^{9,22} On the other hand, patients with a lower kg/day dose (< 1 mg/kg/day) in the ABD group had a higher cure rate and a lower prevalence of adverse effects; this could be due to an increase in the number of days of exposure with no need of suspension.

The dose corresponding to P10 of the total dose for each group was calculated to identify a minimum effective dose. In all chemical forms, patients treated with P10 of the total dose had a lower probability of cure (40–50%). By contrast, 100% of those patients who received P90 of the total dose of ABD and ABCD were cured.

Cure was diagnosed after up to 3 months of follow-up. Usually, after 45 days, failure is diagnosed, which implies the presence of late responders. The time to present therapeutic failure in the current study was up to 6 months of follow-up,

TABLE 4
Important outcomes summarized

Variable, rate (%), unless otherwise specified	Amphotericin (n)								P-value
	ABD (26)		ABCD (21)		LAB (26)		Total (73)		
	CL (n = 17)	MCL (n = 9)	CL (n = 19)	MCL (n = 2)	CL (n = 20)	MCL (n = 6)	CL (n = 56)	MCL (N = 17)	
Failure	5/26 (19.2)		7/21 (33.3)		3/26 (11.5)		15/73 (20.5)		0.18
CL MCL	3/17 (17.6)	7/9 (77.7)	6/19 (31.6)	1/2 (50)	3/2015	0/6 (0)	12/56 (21.4)	8/17 (47.1)	*
Success	21/26 (80.8)		14/21 (66.6)		23/26 (88.5)		58/73 (79.5)		0.18
CL MCL	14/17 (82.4)	2/9 (22.3)	13/19 (68.4)	1/2 (50)	17/20 (85)	6/6 (100)	44/56 (78.6)	9/17 (52.9)	*
Adverse effects	17/18† (94.4)		17/21 (80.9)		12/26 (46.2)		46/65 (70.8)		0.001

ABD = amphotericin B deoxycholate; ABCD = amphotericin B colloidal dispersion; LAB = liposomal amphotericin B.

* Separate analyses for CL and MCL, all *P* values > 0.05.

† Data without patients with bacteremia due to possible association with prolonged use of a central venous catheter.

Statistical analysis used: Pearson's chi-squared test.

emphasizing the importance of follow-up for at least 6 months given the presence of late failure.

There was a statistical difference between the groups regarding adverse effects. The prevalence of adverse effects in the LAB group was lesser than that of the ABCD and ABD groups. Although statistical significance was not established, the most frequent symptoms associated with infusion were fever, asthenia, chills, vomiting, and headache. Similar data have been reported in other pathologies, where the adverse effects of immunological origin associated with infusion are higher in ABCD and are similar to those evidenced in ABD infusions.²⁰ LAB, whose charge is negative and forms smaller liposomes, is recognized by TLR 4 and not by TLR 2. The difference in this recognition may attenuate the response of macrophages and, thus, reduce the effects associated with infusion.²⁷

Creatinine levels (including elevation ≥ 0.3 mg/dL) showed a greater difference between the baseline and highest levels during treatment in the ABD group; nephrotoxicity was shown in one patient in this group. It has been described that the frequency of nephrotoxicity in patients receiving ABD is at least 3-fold higher than when using LAB.¹⁶

Patients who received ABD presented electrolyte abnormalities and transaminase elevations more frequently. Most patients received the drug by central venous catheter in the ABD group, 30.7% of them presented catheter-associated bacteremia that might be explained by the long period of use of an invasive central access device (more than 2 weeks). The risk of presenting catheter-associated bacteremia was considered greater when ABD was used because the duration of use of this device is at least 20 days versus five for amphotericin with a lipid-associated presentation. The use of a PICC has been recommended, although it is not clear that PICC use is associated with a lower risk of infection.²⁸

Patients who had previously received a greater number of treatment schedules with N-methylglucamine, any treatment with AB, and a larger total dose (35 mg/kg) of lipid-associated formulations had higher percentages of adverse effects.

This research has some limitations, including that the *Leishmania* species were not identified; the optimal follow-up of ML after treatment is 12 months, but this information was not available; retrospective design, sample size, loss of population due to failure to meet the inclusion criteria, convenience sampling, and the inclusion of only male population, who were mostly healthy, young, and in the military; and limitations inherent these characteristics that did not allow other statistical analysis. There is little information in the literature regarding this topic; therefore, despite the limitations of our work, the results are useful for patient management and constitute a basis for future studies, whose design can confirm the proposed hypotheses and answer cost-effectiveness questions.

Received June 19, 2018. Accepted for publication September 27, 2019.

Published online December 9, 2019.

Acknowledgments: We thank Sandra Liliana Rubiano, for her important contribution to the research and Sandra Carolina Ordóñez and Jaiber Gutierrez, for their contribution. We also thank the Department of Dermatology and Internal Medicine of the Central Military Hospital for their collaboration and support to the research and the patients.

Financial support: This research was financially supported by the Central Military Hospital, Bogotá, Colombia.

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REFERENCES

1. Von Stebut E, 2007. Immunology of cutaneous leishmaniasis: the role of mast cells, phagocytes and dendritic cells for protective immunity. *Eur J Dermatol* 17: 115–122.
2. Tiuman TS, Santos AO, Ueda-Nakamura T, Filho BPD, Nakamura CV, 2011. Recent advances in leishmaniasis treatment. *Int J Infect Dis* 15: e525–e532.
3. Pearson RD, Sousa AQ, 1996. Clinical spectrum of leishmaniasis. *Clin Infect Dis* 22: 1–13.
4. Kevric I, Cappel MA, Keeling JH, 2015. New World and Old World leishmania infections. *Dermatol Clin* 33: 579–593.
5. World Health Organization, 2012. *Control of Leishmaniasis: Report of a Meeting of the WHO Expert Committee on the Control of Leishmaniasis* [in Spanish]. March 22–26, 2010, Geneva, Switzerland: WHO, (Technical Report Series), 200.
6. Arevalo J et al., 2007. Influence of *Leishmania* (Viannia) species on the response to antimonial treatment in patients with American tegumentary leishmaniasis. *J Infect Dis* 195: 1846–1851.
7. Perez-Franco JE, Cruz-Barrera ML, Robayo ML, Lopez MC, Daza CD, Bedoya A, Mariño ML, Saavedra CH, Echeverry MC, 2016. Clinical and parasitological features of patients with American cutaneous leishmaniasis that did not respond to treatment with meglumine antimoniate. *PLoS Negl Trop Dis* 10: e0004739.
8. Pan American Health Organization PAHO/WHO, Colombia Ministry of Social Protection, 2010. *Comprehensive Care Guide for Leishmaniasis 2010* [in Spanish] [Internet]. Available at: <http://www.vigepi.com.co/3salpub/5guias/guias/420g%20lei.pdf>. Accessed May 18, 2015.
9. World Health Organization/PanAmerican Health Organization, 2013. *Leishmaniasis in the Americas. Recommendations for the Treatment*; 2013. Geneva, Switzerland: WHO. Available at: https://www.who.int/neglected_diseases/resources/978-92-75-31752-5/en/.
10. Saravia NG, Fernández Olga DY, Muvdi Sandra VL, 2011. Susceptibility of clinical strains of *Leishmania* to pentavalent antimony and miltefosine: challenges and opportunities. *Biomédica* 31 (Suppl 3): 3–315.
11. Croft SL, Sundar S, Fairlamb AH, 2006. Drug resistance in leishmaniasis. *Clin Microbiol Rev* 19: 111–126.
12. Rojas R, Valderrama L, Valderrama M, Varona MX, Ouellette M, Saravia NG, 2006. Resistance to antimony and treatment failure in human *Leishmania* (Viannia) infection. *J Infect Dis* 193: 1375–1383.
13. Tuon FF, Amato VS, Graf ME, Siqueira AM, Nicodemo AC, Neto VA, 2008. Treatment of New World cutaneous leishmaniasis—a systematic review with a meta-analysis. *Int J Dermatol* 47: 109–124.
14. Sundar S, Chakravarty J, 2013. Leishmaniasis: an update of current pharmacotherapy. *Expert Opin Pharmacother* 14: 53–63.
15. Wortmann G, Zapor M, Ressler R, Fraser S, Hartzell J, Pierson J, Weintrob A, Magill A, 2010. Liposomal amphotericin B for treatment of cutaneous leishmaniasis. *Am J Trop Med Hyg* 83: 1028–1033.
16. Hamill RJ, 2013. Amphotericin B formulations: a comparative review of efficacy and toxicity. *Drugs* 73: 919–934.
17. Balasegaram M, Ritmeijer K, Lima MA, Burza S, Ortiz Genovese G, Milani B, Gaspari S, Potet J, Chappuis F, 2012. Liposomal amphotericin B as a treatment for human leishmaniasis. *Expert Opin Emerg Drugs* 17: 493–510.
18. Cunha MA, Leao ACQ, de Cassia Soler R, Lindoso JAL, 2015. Efficacy and safety of liposomal amphotericin B for the treatment of mucosal leishmaniasis from the new world: a retrospective study. *Am J Trop Med Hyg* 93: 1214–1218.
19. Pan American Health Organization, 2017. *Epidemiological report of the Americas. Leishmaniasis*. Washington, DC: Pan American Health Organization. Available at: <http://iris.paho.org/xmllui/handle/123456789/34856?locale-attribute=es>.

20. Botero MC, Puentes-Herrera M, Cortés JA, 2014. Formas lipídicas de anfotericina [Lipid forms of amphotericin]. *Rev Chil Infectol* 31: 518–527.
21. Elmahallawy EK, Sampedro Martinez A, Rodriguez-Granger J, Hoyos-Mallecot Y, Agil A, Navarro Mari JM, Gutierrez Fernandez J, 2014. Diagnosis of leishmaniasis. *J Infect Dev Ctries* 8: 961–972.
22. Patel R, 2000. Amphotericin B colloidal dispersion. *Expert Opin Pharmacother* 1: 475–488.
23. Wijnant G-J, Van Bocxlaer K, Yardley V, Harris A, Murdan S, Croft SL, 2018. AmBisome® treatment of murine cutaneous leishmaniasis: relation between skin pharmacokinetics and efficacy. *Antimicrob Agents Chemother* 62: e02009–e02017.
24. Purkait B et al., 2012. Mechanism of amphotericin B resistance in clinical isolates of *Leishmania donovani*. *Antimicrob Agents Chemother* 56: 1031–1041.
25. Al-Mohammed HI, Chance ML, Bates PA, 2005. Production and characterization of stable amphotericin-resistant amastigotes and promastigotes of *Leishmania mexicana*. *Antimicrob Agents Chemother* 49: 3274–3280.
26. Guery R et al., 2017. Liposomal amphotericin B in travelers with cutaneous and muco-cutaneous leishmaniasis: not a panacea. *PLoS Negl Trop Dis* 20: e0006094.
27. Ponte-Sucre A, Díaz E, Padrón-Nieves M, 2013. *Drug Resistance in Leishmania Parasites: Consequences, Molecular Mechanisms and Possible Treatments*. Wien, Austria: Springer-Verlag. Available at: <http://www.springer.com/us/book/9783709102381>. Accessed February 12, 2018.
28. O'Grady NP et al., 2011. Guidelines for the prevention of intravascular catheter-related infections. *Am J Infect Control* 39 (Suppl 1): S1–S34.