Failure of Miltefosine in Visceral Leishmaniasis Is Associated With Low Drug Exposure

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Background. Recent reports indicated high miltefosine treatment failure rates for visceral leishmaniasis (VL) on the Indian subcontinent. To further explore the pharmacological factors associated with these treatment failures, a population pharmacokinetic-pharmacodynamic study was performed to examine the relationship between miltefosine drug exposure and treatment failure in a cohort of Nepalese patients with VL.

Methods. Miltefosine steady-state blood concentrations at the end of treatment were analyzed using liquid chromatography tandem mass spectrometry. A population pharmacokinetic-pharmacodynamic analysis was performed using nonlinear mixed-effects modeling and a logistic regression model. Individual estimates of miltefosine exposure were explored for their relationship with treatment failure.

Results. The overall probability of treatment failure was 21%. The time that the blood concentration was >10 times the half maximal effective concentration of miltefosine (median, 30.2 days) was significantly associated with treatment failure: each 1-day decrease in miltefosine exposure was associated with a 1.08-fold (95% confidence interval, 1.01–1.17) increased odds of treatment failure.

Conclusions. Achieving a sufficient exposure to miltefosine is a significant and critical factor for VL treatment success, suggesting an urgent need to evaluate the recently proposed optimal allometric miltefosine dosing regimen. This study establishes the first evidence for a drug exposure-effect relationship for miltefosine in the treatment of VL.

Keywords. leishmania; pharmacokinetics; pharmacometrics; visceral leishmaniasis; pharmacodynamics; exposure-effect relationship; modeling; population PK-PD.

Miltefosine is currently still the only oral effective drug available to treat the neglected tropical parasitic disease visceral leishmaniasis (VL) [1]. Excellent efficacy of miltefosine in the treatment of VL was established in controlled clinical trials more than a decade ago in the Indian state of Bihar, one of the major areas of VL endemicity [2]. Subsequently, miltefosine was introduced as first-line therapy for VL in most of the Indian subcontinent, including Nepal, where the drug was adopted in a multilateral program to eliminate the disease from the subcontinent [3–6].

Recently, the first reports appeared on the efficacy of miltefosine for VL following its rollout in primary health clinics. For both Nepal and India, after 5 and 10 years of use, respectively, disturbingly lower final cure rates were reported than the reported efficacies from previous clinical trials. This mainly concerned high relapse rates during follow-up: in India 7% of infections relapsed within 6 months [7], and in Nepal 11% relapsed within 6 months and 20% overall relapsed within 12 months [8]. To identify the cause of these high failure rates, several factors were further investigated in a Nepalese cohort of patients receiving conventional miltefosine treatment (approximately 2.5 mg/kg body weight/day for 28 days), including parasite susceptibility, patient risk factors, and quality of and exposure to the drug, as described elsewhere [8]. In regard to miltefosine exposure, end of treatment (EOT)

concentrations were studied in a subset of this cohort. Because of the extremely slow biphasic elimination of miltefosine, with an initial half-life of 4.99–7.18 days and a terminal half-life of 35.5 days, concentrations keep accumulating until EOT [9, 10]. Nevertheless, simply comparing the crude mean miltefosine EOT concentration, at which only a subset of patients typically has reached steady-state concentration [9, 10], did not reveal a significant difference between patients with relapse and those who achieved cure [8].

This finding, however, does not reject the hypothesis that low drug exposure leads to treatment failure, because several factors may be obscuring this comparison. For instance, there was high between-subject variability in the time of sampling at EOT. Moreover, the EOT concentration is perhaps not the best proxy value of total miltefosine exposure to relate to its antiparasitic effect, since this neglects the shape of the concentration-time profile. A population model-based approach can be a more powerful way to analyze the pharmacokinetic-pharmacodynamic relationship, and its value is widely recognized in drug development [11–14].

Overall, very little is known about the exposure-effect relationship of miltefosine in the treatment of VL [1]. To gain further insight into the possible correlation between various measures of exposure, pharmacokinetic targets, and efficacy of miltefosine and to overcome the limitations of untimely sampling around the EOT, we analyzed the data from the Nepalese cohort, using a combined sequential population pharmacokinetic-pharmacodynamic analysis, with the objective to identify an exposure-effect relationship as a possible explanation for the observed high relapse rate.

METHODS

Patients

The population of patients in this pharmacokinetic-pharmacodynamic study is a subset of the Nepalese cohort treated with miltefosine and studied in the framework of the Kaladrug-R project [8]. This study was conducted between March 2010 and August 2011 in a Nepalese referral hospital, BP Koirala Institute of Health Sciences (BPKIHS). VL patients who met previously described inclusion criteria [8], such as confirmation of VL by detection of *Leishmania* organisms in a bone marrow aspirate, who had given informed consent for the Kaladrug-R study and from whom a blood sample was obtained around EOT (approximately day 28) were eligible for this pharmacokinetic-pharmacodynamic study. Individual fat-free mass was estimated from each patient's weight and height [15]; if height was unavailable (for 3 of 81 patients), fat-free mass was assumed to correspond to 90% of the total body weight of the individual [10]. Patients were followed for a total of 12 months after treatment, with follow-up visits at 3, 6, and 12 months after completion of therapy. They were examined for clinical

signs of relapse, and, if found, bone marrow was reexamined for *Leishmania* parasites to confirm treatment failure. Patients who did not visit for scheduled follow-up at BPKIHS were actively traced in their homes. The research protocol of this prospective study was approved by the ethics committees of the Nepal Health Research Council and the University of Antwerp in Belgium.

Treatment

Patients were treated with miltefosine (Impavido, Paladin Labs, Montreal, Canada), for 28 days, according to the national guidelines: adults (≥12 years of age) with a body weight of >25 kg received 50 mg twice daily (total dose, 100 mg/day), adults with a body weight of ≤25 kg received 50 mg once daily (total dose, 50 mg/day), and children (2–11 years of age) received 2.5 mg/kg body weight/day rounded to the nearest 10 mg. Treatment adherence was monitored, and results were published separately [16]. No major adherence issues were detected and missed doses were recorded and incorporated in our analyses.

Samples and Bioanalysis

A single whole-blood sample was obtained per patient on the occasion of their EOT visit, approximately at day 28 after the start of miltefosine treatment. Samples were kept frozen at a minimal temperature of −20°C both during storage and transport. Miltefosine concentrations were measured using a validated liquid chromatography technique coupled to tandem mass spectrometry (LC-MS/MS) with a limit of detection of 4 ng/mL, as described previously [17].

Population Pharmacokinetic Analysis

All calculations, simulations and estimations were performed on a dual-core desktop computer running NONMEM 7.2 (ICON Development Solutions, Hanover, MD) [18], the R statistical software package (version 2.15.2; available at: http://www.r-project.org/) [19], and Perl speaks NONMEM (PsN, version 3.5.3; available at: http://psn.sourceforge.net) [20, 21]. Pirana (version 2.7.0b; available at: http://www.pirana-software.com) was used to structure the model development work and interpret the output [22].

Nonlinear mixed-effects modeling was performed using a previously developed and extensively evaluated open 2-compartment population pharmacokinetic model of miltefosine, with first-order absorption and elimination from the central compartment as the structural base model [9, 10], using first-order conditional estimation with interaction between between-subject variability and residual error. Clearance (CL/*F*) from and the volume of distribution (V_c/F) of the central compartment were allometrically scaled with a power of 0.75 and 1, respectively, using individual fat-free mass as body size descriptor, as previously established for miltefosine over a wide range of body sizes [10]. Intercompartmental clearance and peripheral

volume of distribution were not scaled allometrically for mechanistic reasons described previously [10]. Given the sparseness of the Nepalese data set, estimation of population pharmacokinetic parameters was performed by combining it with all prior obtained miltefosine pharmacokinetic data (from an adult European study [9], an adult Indian study [23], and a pediatric Indian study [24]). To enable handling of whole-blood concentrations (as collected in the Nepalese cohort) in combination with plasma concentrations (as collected in the other studies), a fixed correction factor (f_c) of 0.25 was introduced to convert the predicted plasma concentrations ($C_{pl,pred}$) to whole blood concentrations ($C_{wb,pred}$) [1]. Between-subject variability in CL/F, V_c/F, and absorption rate (k_a) were modeled with an exponential error. Residual error (including within-subject variability) was modeled with a proportional error model, using a study-specific estimate for the Nepalese data, since it was obtained from a distinct study, with a different population (inclusion and exclusion criteria), and measured in a different laboratory. The validity of using a study-specific residual variability was evaluated (data not shown). Absolute oral bioavailability was unknown for miltefosine; therefore, parameters were reported relative to bioavailability (eg, CL/F and V_c/F). Model adjustments were evaluated for their goodness of fit. Model evaluation was guided by the objective function value (OFV; equal to minus twice the log likelihood) and by graphical goodness-of-fit assessment through a visual predictive check (using PsN and Xpose).

When data are sparse and less informative on individual parameters, it is expected that the empirical Bayes estimate will be shrunk toward the population mean. Shrinkage in empirical Bayes estimates of between-subject variability of parameter j (η_j) was calculated for the Nepalese data as follows [25]:

$$\text{Shrinkage}_{\eta j} = 1 - \frac{\text{std}(\eta_{i,j})}{\omega_j}, \qquad (1)$$

where std($\eta_{i,j}$) is the SD of the distribution of individual estimates of between-subject variability for parameter j for i individuals, and ω_j is the population model estimate of the SD in η_j.

Individual estimates of drug exposure were calculated in NONMEM, using a differential equation solver and the individual population pharmacokinetic model parameter estimates. Among other parameters calculate include the area under the concentration-time curve from day 0 through EOT (AUC_{0-EOT}), the AUC from 0 to infinity ($AUC_{0-\infty}$), and the period that the miltefosine blood concentration was either above the mean half maximal effective concentration (EC_{50}; $T > EC_{50}$) or >10 times the mean EC_{50} ($T > 10 \times EC_{50}$), which were determined with intracellular drug susceptibility testing of available clinical *Leishmania* isolates from this particular Nepalese cohort (the fixed mean EC_{50} was used: 4.4 μM or approximately 1.79 μg/mL) [26].

Pharmacokinetic-Pharmacodynamic Analysis

The pharmacokinetic-pharmacodynamic relationship between miltefosine exposure and final treatment outcome was explored with various individual drug exposure estimates from the population pharmacokinetic analysis. Patients were excluded from the pharmacodynamic analysis if they died from a cause probably unrelated to VL before the end of follow-up or if they experienced a treatment switch because of severe adverse events during treatment. Failure was defined as no initial cure at EOT or as relapse (ie, initial cure at EOT but with reappearance of clinical symptoms and/or signs along with confirmation of *Leishmania* infection by detection of *Leishmania* organisms in a bone marrow aspirate smear during follow-up). The probability of failure for the ith individual (p_i) was modeled with linear logistic regression performed on the dichotomous treatment outcome data (0 = cure and 1 = failure) with NONMEM, using the Laplacian estimation method, and with the conditional and likelihood options. The logit of p_i ($Logit_i$) was defined as follows:

$$Logit_i = \theta_1 + \theta_2 \cdot (\text{MIL}_i - \text{MIL}_\mu), \qquad (2)$$

where θ_1 (see equation 3) and θ_2 are the fixed-effect parameters defining intercept and slope, respectively, and MIL_i is a covariate corresponding with an individual estimate of miltefosine exposure (ie, C_{EOT}, AUC_{0-EOT}, $AUC_{0-\infty}$, $T > EC_{50}$, or $T > 10 \times EC_{50}$) centered around its respective population mean value (MIL_μ). If increasing miltefosine exposure reduces the probability of treatment failure, θ_2 should be negative. The intercept θ_1 was defined as follows to estimate the baseline probability (BASE) of an outcome with a value of 1:

$$\theta_1 = \ln\left(\frac{\text{BASE}}{1 - \text{BASE}}\right) \qquad (3)$$

Finally, the individual estimate of probability p_i was calculated as follows:

$$p_i = \frac{e^{Logit_i}}{1 + e^{Logit_i}}, \qquad (4)$$

for which an outcome of 1 corresponds to a prediction equal to p_i and an outcome of 0 corresponds to a prediction of $1-p_i$. The likelihood ratio test was used to assess the improvement of fit and influence of miltefosine exposure covariates (MIL) on the probability of treatment failure when compared to the model with the miltefosine exposure covariates excluded. A P value of .05 corresponds with a ΔOFV decrease of 3.84 ($\alpha = 0.05$; χ^2, 1 df). For graphical presentation of the observed probability versus the model-estimated probability, observations were

Table 1. Demographic and Baseline Characteristics of Patients

Parameter	Value
Patients enrolled, no.	81
Sex	
Male	50
Female	31
Age, y	20 (2–65)
Aged <12 y	20
Body weight, kg	40 (8–56)
Height, cm[a]	147 (75–172)
Body mass index[a]	17.9 (10.8–25.8)
Miltefosine treatment duration, d	29 (7–36)
Daily miltefosine dosage, mg/kg/d	
Overall	2.4 (1.7–4.0)
Children (age <12 y)	2.5 (1.7–3.0)
Adults (age ≥12 y)	2.3 (1.8–4.0)
Included in the pharmacodynamic analysis	76
Treatment outcome	
Failure	16
Cure	60

Data are no. of patients or median value (range).

[a] Height and, thus, body mass index (defined as the weight in kilograms divided by the height in meters squared) was unavailable for 3 patients.

binned in groups of equal size to obtain an observed probability per bin.

RESULTS

Patients

Eighty-one patients were enrolled in this pharmacokinetic study; baseline demographic and clinical characteristics and outcome can be found in Table 1. The majority of patients (62%) were male. The included patients were relatively young; 25% were children <12 years of age. Five patients were excluded from the pharmacodynamic analysis: 2 patients were lost to follow-up because of untimely death unrelated to VL, and 3 patients had a treatment switch because of severe adverse events during their miltefosine regimen.

Population Pharmacokinetic Analysis

The measured miltefosine EOT concentrations of the Nepalese VL patients are shown in Figure 1. Miltefosine EOT concentrations were significantly lower in children, compared with concentrations in adults (Figure 2), although daily miltefosine doses (in milligrams/kilogram of body weight) were comparable (Table 1). This indicates, as demonstrated previously, that children are less exposed to miltefosine when they receive a similar
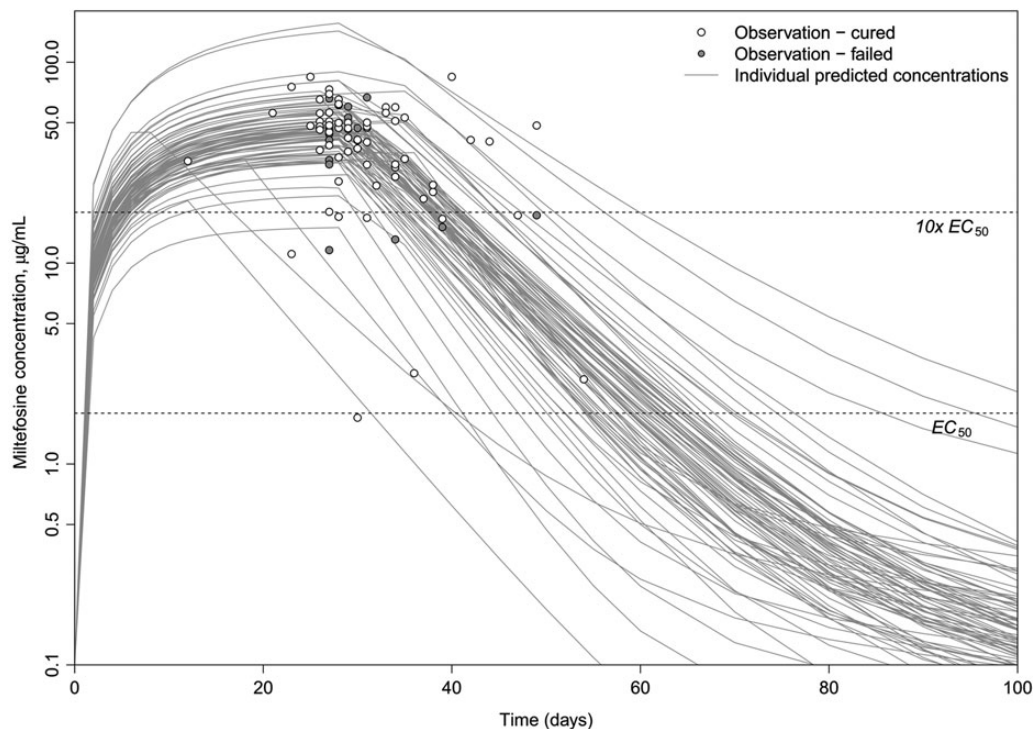

Figure 1. Individual miltefosine concentration-time profiles. Observed concentrations (circles) are plotted over the individual model-based predicted concentration-time curves for the 81 subjects in our data set. The broken lines represent 1 times (lower line) and 10 times (upper line) the mean in vitro half maximal effective concentration (EC$_{50}$) of miltefosine for the clinical *Leishmania* isolates tested for drug susceptibility in the Nepalese cohort.
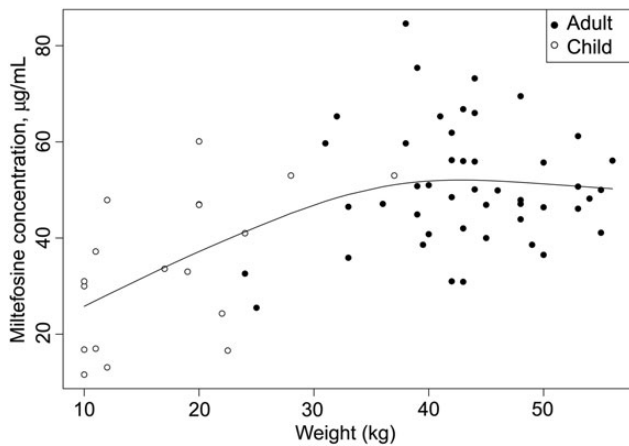
Figure 2. Observed miltefosine end-of-treatment (EOT) concentrations among children and adults, by body weight. Adults are individuals aged ≥12 years, and children are individuals aged <12 years. The solid line shows a fitted polynomial smoothed regression line. For comparability, only observed concentrations within 7 days of EOT were included here.

Table 2. Population Pharmacokinetic Model Estimates

Primary Parameter	Estimate	Precision, %[a]	Between-Subject Variability, %
Absorption rate, /d	9.6	Fixed	19.4
Central clearance, L/d[b]	3.69	3.4	35.1[c]
Intercompartmental clearance, L/d	0.0316	16.6	Not estimated
Central volume of distribution, L[b]	38.5	4.5	31.6[c]
Peripheral volume of distribution, L	1.69	8.6	Not estimated
Residual variability, %[d]	24.5	36.4	Not applicable

Secondary parameters, derived from the individual model-based estimates, were initial half-life (median, 6.26 days [range, 4.18–9.27 days]) and terminal half-life (median, 48.9 days [range, 48.6–51.0 days]).

[a] Relative standard error.

[b] Estimate is given for a standardized fat-free mass of 53 kg.

[c] Between-subject variabilities of clearance and volume of the central compartment were correlated by 87%.

[d] Residual variability specifically for the Nepalese subset of data; separate residual variabilities were estimated for the other subsets (unreported here).

body weight–based dose, compared with adults [10]. The previously developed population pharmacokinetic model for miltefosine fitted the sparse Nepalese miltefosine EOT concentrations adequately, and pharmacokinetic parameters and the associated variabilities could be estimated with high precision for the sparse data set when combined with prior pharmacokinetic data sets (from Europe and India). The population pharmacokinetic parameter estimates are shown in Table 2. The study-specific residual error for the Nepalese miltefosine data was a modest 24.5% (relative standard error [RSE], 36.4%). Appropriateness of estimating pharmacokinetic parameters using only a single EOT concentration was positively evaluated by comparing estimates from the full rich Dutch miltefosine data set with estimates from a subset of that same data set with only a single EOT sample/patient (data not shown). The individual predicted concentration-time curves are shown in Figure 1, together with observed concentrations. Shrinkage of empirical Bayes estimates of between-subject variability was evaluated specifically for the Nepalese data set and amounted 14.5% and 34.5% for CL/F and V_c/F, respectively, which is modest given the sparseness of the data set.

Various measures of miltefosine exposure were estimated for all enrolled subjects with the population pharmacokinetic model based on individual dosing, parameter estimates, and predicted plasma concentrations (Table 3).

Pharmacokinetic-Pharmacodynamic Analysis

The observed probability of miltefosine treatment failure in the Nepalese VL patients was 21%. Of these patients for whom miltefosine treatment failed, 37.5% had an age of <12 years, compared with 25% in the full data set. Correlations between the estimated miltefosine exposure values and the observed probability of treatment failure were graphically explored by binning the exposure values in 3 groups of equal size and plotting mean exposure values versus the observed probability of treatment failure within each bin. A linear correlation between the various miltefosine exposure values (C_{EOT}, AUC_{0-EOT}, $AUC_{0-\infty}$, $T > EC_{50}$, and $T > 10\times EC_{50}$) and the probability of treatment failure could be observed.

A base pharmacodynamic logistic regression model was developed that accurately estimated the observed population probability of treatment failure (base probability, 0.211 [RSE, 22.2%]). Miltefosine exposure values were introduced as covariates in the logistic regression model (equation 2). Use of the observed concentrations led to a worse fit of the model, compared with the model-based predicted C_{EOT} (ΔOFV, 4.52). All included model-based measures of miltefosine exposure led to a decrease of OFV and, thus, to an improved fit of the model, but only inclusion of the exposure covariate $T > 10\times EC_{50}$ resulted in a significantly better fit of the model to the therapy outcome data (ΔOFV, −5.19, corresponding to a P value of .02 [χ^2, 1 df]), compared with the base model. The mean $T > 10\times EC_{50}$ in our population was 30.2 days. The final estimates (RSE) of intercept BASE and slope θ_2 correlated to the centered effect of $T > 10\times EC_{50}$ were 0.195 (24%) and −0.08 (48%), respectively. This corresponds with an increased odds ratio for treatment failure of 1.08 (95% confidence interval, 1.01–1.17) for each 1-day decrease in exposure to a concentration of $> 10\times EC_{50}$. The mean model predicted probability of failure as a function of the achieved drug exposure ($T > 10\times EC_{50}$) is depicted in Figure 3, together with a 90% confidence interval and

Table 3. Miltefosine Exposure Estimates Derived From the Population Pharmacokinetic Model

Measure of Exposure[a]	Abbreviation	Unit	Median (Range)[a]
End of treatment concentration	C_{EOT}	µg/mL	35.3 (11.6–120)
Area under the curve from 0 to the end of treatment	AUC_{0-EOT}	µg/mL · d	724 (265–2260)
Area under the curve from 0 to infinity	$AUC_{0-\infty}$	µg/mL · d	1140 (340–4200)
Time that concentration is greater than the EC_{50}	$T > EC_{50}$	d	57.4 (38.5–99.5)
Time that concentration is >10 times the EC_{50}	$T > 10\times EC_{50}$	d	30.6 (0–54.3)

Abbreviation: EC_{50}, half maximal effective concentration.

[a] Values were calculated using the individual model-based estimates and are based on the individual (actual) dose administered.

the observed probability of the binned $T > 10\times EC_{50}$ values as derived from our data set.

DISCUSSION

This study establishes that the observed high frequency of miltefosine treatment failure in Nepalese VL patients is significantly associated with achieved drug exposure (ie, $T > 10\times EC_{50}$ values). Miltefosine is an essential oral drug in the treatment of the neglected tropical disease VL, but the recently reported decaying efficacy rates under the current conventional miltefosine dose regimen may seriously threaten its future use. To our knowledge, this study is the first to investigate the exposure-effect relationship of any antileishmanial drug.

Figure 3. Probability of treatment failure versus miltefosine exposure. The solid line represents the logistic model predicted probability of treatment failure, and the gray area denotes the 90% confidence interval. The transparent bars indicate the interval of the observed time that the miltefosine concentration is >10 times the half maximal effective concentration ($T > 10\times EC_{50}$) is covered by the bins, with approximately 25 observations in each bin, whereas the filled circles on top of the bins indicate the mean observed data-based probabilities of treatment failure per bin at the mean $T > 10\times EC_{50}$ of the bin.

Previously we described the body size–related differences in miltefosine pharmacokinetics between adults and children [10]. Consequently, we proposed a revised allometric miltefosine dosage regimen to achieve equivalent exposure in children as compared to adults. Nevertheless, in the Nepalese cohort described here, miltefosine was used according to the conventional miltefosine treatment guidelines (2.5 mg/kg/day), both in adults and children. Similar to our previous findings, administration of this dose led to lower miltefosine exposure in children, compared with adults, with significantly lower EOT concentrations. Moreover, these observations were corroborated by the finding that an age of <12 years was the only risk factor found to be correlated with treatment failure [8]. This emphasizes the need to evaluate safety and efficacy of the proposed allometric miltefosine regimen [10].

The proportion of miltefosine treatment failures in this Nepalese cohort of VL patients was significant. In the subset of patients who were enrolled in this pharmacokinetic study, the overall failure rate was 21% (the failure rate among children was 33%), which is much higher than the rates previously reported in the region, which do normally not exceed 5%–10% [7]. This observed higher failure rate might be caused by the study design, since patients were followed up for a 12-month period instead of the conventional 6-month period. On the other hand, the emergence of miltefosine-resistant *Leishmania* clones is anticipated in this region, where anthroponotic transmission is the main route of transmission. Nevertheless, clinical isolates obtained from this cohort did not show in vitro resistance to miltefosine [8]. Also it may be argued that these late treatment failures might be reinfections. Although reinfections cannot be completely excluded, the available results of genetic profiling and fingerprinting studies do not support this [8]. Additionally, no difference was found between pre- and posttreatment clinical isolates regarding in vitro drug susceptibility [26]. Moreover, reinfection within a period of 12 months is highly unlikely based on infection dynamics models [27].

In this study we investigated in depth the relationship between measures of miltefosine exposure and the probability of treatment failure. The only identified risk factor was the estimate of the time that plasma concentrations were >10 times
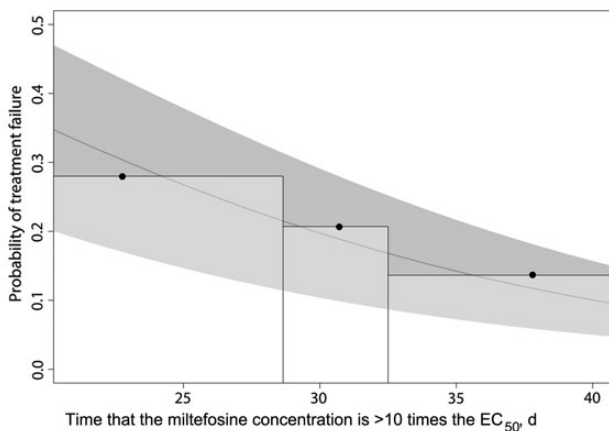
the mean in vitro EC_{50} established in this cohort. Other measures of exposure, such as various AUCs, were not found to be significantly associated with treatment failure. This may indicate that the mechanism of action of miltefosine is defined by a time-dependent killing effect, rather than by a concentration-dependent effect. This time dependency is supported by preclinical data and assumed mechanisms of action of miltefosine (ie, apoptosis, immunomodulation, and membrane lipid metabolism) [1, 28, 29]. In turn this may explain why the duration of miltefosine treatment was found to be important during the limited dose-finding studies in VL [30]. A longer treatment duration would lead to a longer attainment of a threshold concentration, implying that miltefosine treatment duration is of critical importance for treatment success. On the other hand, the administered daily dose should be high enough to be able to reach that threshold concentration and to reach it as fast as possible. In this context, it is important to find the optimal threshold concentration that needs to be attained for a period for miltefosine to exert its antileishmanial effect. The value of $10\times EC_{50}$ (17.9 µg/mL) corresponds with the highest level of miltefosine-resistance of *Leishmania* in vitro (40 µM or 16.3 µg/mL) [1]. This relatively high systemic concentration to be attained may indicate that miltefosine concentrations at the site of infection (ie, spleen, bone marrow, or liver) are lower, although this does not follow from animal distribution studies, or that there remain sanctuary sites, where the *Leishmania* parasites reside, with less-than-optimal miltefosine penetration. Clinical relevancy of in vitro *Leishmania* susceptibility testing has been doubted before [31], and also in this cohort no association between in vitro susceptibility of isolates and treatment outcome was found [8]. All these issues deserve further consideration and evaluation, for instance by measuring target site-specific pharmacokinetics.

The mean estimated $T > 10\times EC_{50}$ was 30.2 days. This value was associated with a failure rate of 19.5%. A decrease of the $T > 10\times EC_{50}$ by 1 day was associated with a 1.08-fold increased odds of treatment failure. Two patients with an estimated $T > 10\times EC_{50}$ of 0 days had a probability of treatment failure equal to the base probability (73%), and both patients experienced relapse. In the absence of placebo-controlled trials for VL, this base probability of treatment failure of 73% might be compared to historic observations that VL is inevitably fatal if left untreated [32]. The use of dichotomous outcome data (ie, cure vs failure) may not be optimal to fully characterize an exposure-response relationship. In future trials, the time until relapse should be monitored more accurately to get an impression about whether drug exposure can be correlated to time until relapse, as is the case for other parasitic diseases, such as malaria. More emphasis should be put on the evaluation of pharmacodynamic markers, such as quantitative measurements of parasite load by polymerase chain reaction, to enable a more precise characterization of the exposure-response relationship of antileishmanial drugs, which,

in turn, would allow for a better prediction of possible relapse cases and, ultimately, optimal treatment protocols.

In conclusion, this study is the first to explore the pharmacokinetic-pharmacodynamic relationship between miltefosine exposure and VL treatment failure. Although the reasons behind treatment failure in VL are probably far from singular, drug exposure is one of them. Pharmacokinetics studies are therefore particularly needed now that increasing failure rates for the conventional miltefosine treatment regimen are being reported. Again, we established that children are less exposed to miltefosine than adults under the current conventional 2.5 mg/kg body weight dosing regimen. Combined with the finding that being a child (age <12 years) was a significant clinical risk factor for treatment failure, the introduction and clinical evaluation of the previously proposed allometric miltefosine dosing regimen is urgently indicated. This is the first step toward the definition of pharmacokinetic-pharmacodynamic targets to be attained for miltefosine in the treatment of VL.

Notes

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