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Population pharmacokinetic modeling of benznidazole in Brazilian patients with chronic Chagas disease

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ABSTRACT

The present study aimed to establish a population pharmacokinetic (PopPK) modeling of benznidazole (BZD) in Brazilian patients with chronic Chagas disease. This was part of a Brazilian prospective cohort study with eight patients diagnosed with Chagas disease during the beginning of BZD treatment up to the 60th day. On the 15th day of treatment, a blood sampling was collected and analyzed. A one-compartment PK model was developed using Pmetrics. Patients with an average age of 50.3 (SD: 6.2) years old, 6 female patients and 2 males, 70.2 kg (14.2), receiving a 5 mg/Kg/day dose were included. PK parameters estimated for CL, V and Ka were 6.27 L/h, 38.97 L and 1.66 h-1, respectively. This is the first study to establish a population pharmacokinetic modeling of BZD in Brazilian patients with chronic Chagas disease. Therefore, further studies are needed to obtain the complete characterization of BZD pharmacokinetics.

KEYWORDS: Chagas disease. Benznidazole. Pharmacokinetic.

INTRODUCTION

Chagas disease (CD) is caused by the protozoan *Trypanosoma cruzi*. It is endemic in 21 countries in the Americas and affects approximately six million people worldwide, with an annual incidence of 30,000 new cases and 12,000 deaths per year[1]. In Brazil, it was estimated at least one million people were infected with *T. cruzi* in 2021. Despite a reduction on the incidence of acute cases, in the last 15 years there has been shown a systematic occurrence of these cases, mainly in the Amazon region. In 2020, 146 new cases were confirmed, with a mortality rate of 2% (3/146).

The disease is characterized by an acute and a chronic phase, and approximately 30 to 40% of chronically infected individuals develop overt clinical forms, such as cardiac and digestive ones[2]. There are two drugs available to treat the disease, nifurtimox and benznidazole (BZD); the last is the drug of choice in Brazil[3]. Although BZD has been commercially available for over 50 years, few studies on its pharmacokinetics are found in the literature. Among the published studies, limited data is available using population pharmacokinetic models[4-7].

The well-proven effectiveness of BZD on the acute phase has been contrasting with the uncertainty on its use for the chronic phase until recently, when novel data have emerged and showed encouraging findings on parasitological outcomes, as well as in preventing the evolution towards the cardiac spectrum of disease, lower cardiovascular events and mortality[8], especially in the early chronic phase

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of the disease. However, BZD did not appear to reduce the clinical deterioration of heart function for those presenting with Chagas cardiomyopathy[9]. Besides these findings, the drug presents a widespan of adverse effects. About half of patients will complain of adverse drug reactions during the 60 days of treatment, leading to nearly one out of four withdrawal treatments, mainly due to dermatological-related effects[10]. Recently, shorter and reduced new dosing schemas have been proposed to improve patients' tolerability and drug accessibility with no significant adverse effect on parasitological and clinical outcomes, although there were no pharmacokinetic results[11,12]. Furthermore, despite the efforts made to discover new drugs against the disease, it is believed that there will be no new drugs on the market in the coming years[13].

Pharmacokinetic studies on BZD published to date have been carried out in populations of different nationalities. It is known that ethnicity can lead to a distinct pharmacokinetic profile. Therefore, it is essential to understand the pharmacokinetic profile of BZD in different populations living in countries in which the disease is endemic, including the Brazilian one[7]. The present study aimed to establish a population pharmacokinetic (PopPK) modeling of BZD in Brazilian patients with chronic CD.

PATIENTS, MATERIALS AND METHODS

This was part of a prospective cohort study in Brazilian patients with chronic CD during the beginning of a standard BZD treatment up to the 60th day. The patients were recruited at the Instituto de Infectologia Emilio Ribas (IIER) and Hospital das Clinicas, Faculty of Medicine, University of São Paulo (HCFMUSP), during the medical appointments. In the 15th day of treatment, small volumes of whole blood samples were collected as dried blood spots (DBS). A bioanalytical method for drug quantification was developed and validated, using the dried blood spot technique-based liquid chromatography-tandem mass spectrometry method, according to Bedor *et al.*[14]. Patients infected with HIV, presenting with renal or liver impairment, pregnancy or lactation were considered ineligible; patients with no drug compliance were excluded. Patients receiving an oral dose of 5 mg/kg/day for 60 days were included. This project was submitted and approved by the Research Ethics Committee of the Instituto de Infectologia Emilio Ribas (CAAE Nº 90818718.0000.061).

A population pharmacokinetic model was developed using the Pmetrics version 1.9.7 (Laboratory of Applied Pharmacokinetics and Bioinformatics, Los Angeles, CA, USA) in RStudio version 1.3.1056. As a result of sampling limitation, a one compartment structural model was constructed using the nonparametric adaptive grid (NPAG) algorithms. The elimination of BZD from the central compartment was modeled as a linear process. Residual error was modelled as gamma * (1 + 0.1*concentration), value = 5. Available clinical covariates were assessed for biological plausibility and subsequently evaluated in a covariate analysis by applying stepwise linear, log, polynomial and power regression for the continuous variables. Covariates were correlated with pharmacokinetic parameters and linear model was used for categorical variables. Selected covariates tested on the structural model parameters included gender, age, weight, body mass index, smoker and alcohol consumption.

Model evaluation was performed by diagnostic plots and statistical examination for comparison and selection of models. The first screening was conducted by visual evaluation for each run, the goodness of fit and the determination coefficient of the linear regression considered observed and predicted plots values (R-squared closer to 1, intercept closer to 0, slope closer to 1, lowest mean bias (as weighted predicted error) and imprecision as {[SD*(weighted predicted error)] 2}. Secondly, methods were compared by the log-likelihood ratio test (-2*LL) for the nested model, Akaike information criterion (AIC) and Bayesian information criterion (BIC). Potential covariates were separately entered into the model and statistically tested; if inclusion of the covariate improved the -2*LL, AIC or BIC values and the goodness-of-fit plots, then the covariate was retained in the final model. Finally, to evaluate the internal consistency of the model predictions with observations, we assessed normalized prediction distribution errors (NPDE) and the posterior predictive check graphically. The proportion of observations between 5th and 95th simulated percentiles above 90% were considered adequate.

RESULTS

Eight patients were included, with an average age of 50.25 years old (SD: 6.22), 6/8 patients were female, the average weight was 70.16 kg (SD: 14.20) and they were receiving an oral dose of BZD of 5 mg/kg/day for 60 days. Two people refused to answer about their education and, among the others, 2 patients had completed the elementary school and 4 had completed the high school. Five participants reported that they have never smoked or drunk alcohol (Table 1).

PK parameters estimated for CL, V and Ka were 6.27 L/h, 38.97 L and 1.66 h-1, respectively (Table 2). The inclusion of weight normalized to 72.8 kg as a covariate on clearance significantly reduced the log-likelihood ratio

Table 1 - Sociodemographic data of patients with chronic Chagas Disease during treatment with benznidazole (n=8).

Variable	Frequency	Relative frequency (%)
Sex		
Male	2	25.0
Female	6	75.0
Age (years)		
40-50	4	50.0
51-60	4	50.0
Ethnicity		
Black	1	12.5
Mixed ethnicity	7	87.5
Education level		
Elementary school	2	25.0
High school	4	50.0
Refused to answer	2	25.0
Health problems		
Yes	5	62.5
No	3	37.5
Taking medicine other than BZD		
Yes	6	75.0
No	2	25.0

Table 2 - Estimates of BZD pharmacokinetic parameters for the final covariate model.

Parameter	Mean (SD)	Median	% CV
CL (L/h)	6.27 (0.09)	6.32	1.4
V (L)	38.97 (8.33)	37.66	21.4
Ka (h^{-1})	1.66 (0.03)	1.67	2.0

CL = clearance; V = volume of distribution of the central compartment; Ka = constant rate for benznidazole absorption; SD = standard deviation; CV = coefficient of variation.

(Δ-2LL: -82.65; ΔAIC: -73.35; ΔBIC: -80.59) and improved the model fit as assessed by goodness-of-fit plots, with a population predicted plot correlation coefficient (r2) of 0.797, slope 1.07 (95% CI 0.53 to 1.6). The observed versus predicted diagnostic plots and visual predictive check plots (n = 1,000) are presented in Figure 1.

DISCUSSION

To our knowledge, this is the first study that established a population pharmacokinetic modeling of BZD in Brazilian

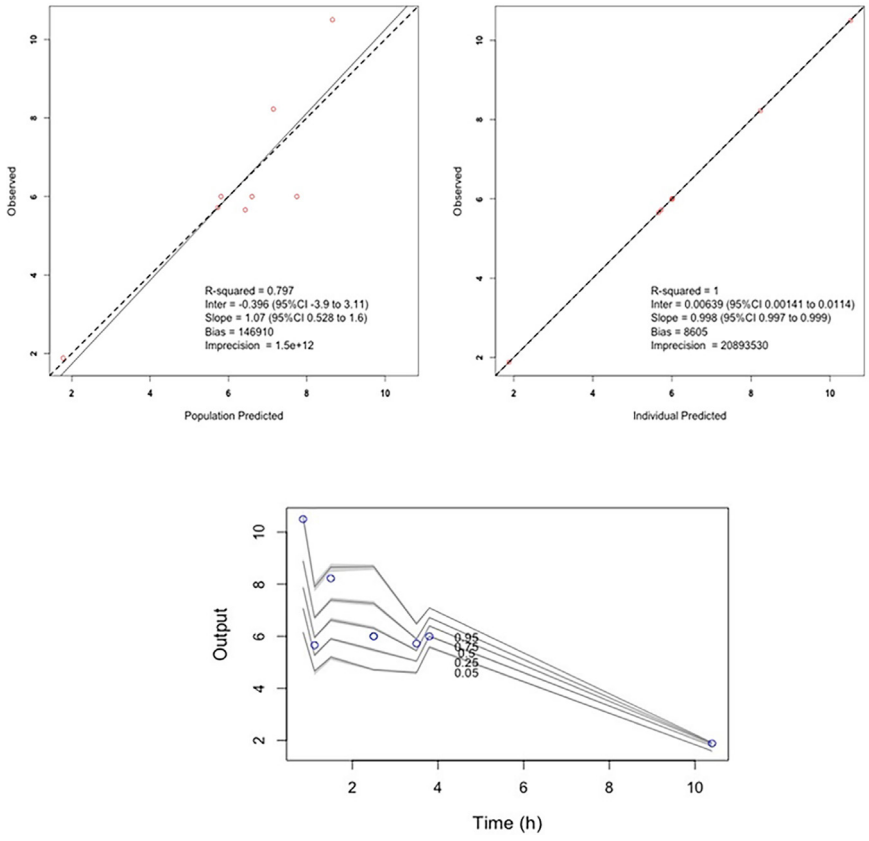

Figure 1 - Internal validation. Observed versus individual-predicted (top left) and population-predicted (top right) concentration diagnostic plots, visual predictive check. The figure presents the correlation between observed data (patients' plasma concentrations) and predicted (model estimated) concentrations. The VPC compare the distribution of simulated observations with the distribution of measured observations in the population, 90% prediction intervals generated from the posterior predictions encompass 90% of the observations.

patients with chronic CD. Molina *et al.*[13] and Soy *et al.*[7] had evaluated the drug pharmacokinetic in Hispanic Latin American healthy volunteers and South American patients with CD, respectively. A PK model of an open compartment with first-order absorption and elimination best fits the present study's data in agreement with previous studies[7]. The absorption rate was also similar in both studies with respect to the normalization according to the total body weight. Otherwise, pharmacokinetics parameters are diverging. Our studies presented higher CL and lower V values, similar to values reported in healthy volunteers[7,13].

Although the study conducted by Soy *et al.*[7] presented mainly a Bolivian population from a neighbor country, the ancestrality of the populations is entirely different and can influence the pharmacokinetic of BZD, leading to differences. It is well-known the ethnicity may influence the cytochrome isozyme expression[15]. The Brazilian population presents mainly a trihybrid origin composed of Africans (31 to 36%), Europeans (45 to 55%), and native Americans (8 to 15%)[16]. On the other hand, in Bolivians, the native American represented 77% to 86%, European 13% to 21% and African less than 2%[17]. Furthermore, the data used by Soy *et al.*[7] were from plasma concentrations, while in our study we used whole blood concentrations. Therefore, additional clinical trials in the area are needed to obtain reliable answers and characterize BZD pharmacokinetics.

The main limitation of this study is the small number of selected patients, making it difficult to test more complex models such as the one with two compartments. Due to COVID-19, medical appointments had to be suspended, and the research was interrupted. However, due to the scarcity of studies in the area, this work represents a first step towards the characterization of the pharmacokinetics of BZD in Brazilian patients.

CONCLUSION

In conclusion, this is the first study to establish a population pharmacokinetic modeling of BZD in Brazilian patients with chronic CD. As observed, pharmacokinetic studies of BZD in humans are still scarce and those available show divergent results, making it difficult to define the drug's PK parameters correctly. Thus, additional studies are needed to allow for a better characterization of BZD pharmacokinetics, and the identification of factors that may contribute to its variability in order to ensure safety and efficacy of the treatment.

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