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The effect of breed, sex, and drug concentration on the pharmacokinetic profile of ivermectin in cattle

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Abstract

Ivermectin (IVM) is one of the most widely used antiparasitic drugs worldwide and has become the drug of choice for anthelmintic and tick treatment in beef cattle production. It is known that pharmacokinetic parameters are fundamental to the rational use of a drug and food safety and these parameters are influenced by different factors. The aim of this study was to evaluate the pharmacokinetic profile of IVM in *Bos indicus*, *Bos taurus*, and crossbreed cattle (*B. indicus* × *B. taurus*) kept under same field conditions and the possible impacts of sex and IVM formulation (1% and 3.15%). It was observed that IVM concentration was significantly affected by breed. The plasma concentrations of IVM, AUC, C_{max}, and $t_{1/2\beta}$ were significantly higher in *B. indicus* compared to *B. taurus*. Crossbreed animals showed an intermediate profile between European and Indian cattle. No alteration in pharmacokinetics parameters was detected when comparing different gender. Concerning the pharmacokinetic data of IVM formulation, it was verified that T_{max}, AUC, and $t_{1/2\beta}$ were higher in 3.15% IVM animals than those from 1% IVM formulation. The results clearly indicated that the IVM plasma concentrations in *B. indicus* were higher than that in *B. taurus*.

KEYWORDS

antiparasitic drug, bovine, livestock, veterinary pharmacology, withdrawal period

1 | INTRODUCTION

Ivermectin (IVM) is classified in the avermectin group of compounds, which are macrolide antibiotics derived from the fermentation products of the actinomycete *Streptomyces avermitilis*. IVM is probably one of the most widely used antiparasitic drugs worldwide and has become the drug of choice for anthelmintic and tick treatment in beef cattle production (Ballweber & Baeten, 2012; Campbell & Benz, 1984).

The pharmacokinetic parameters of IVM vary extensively and can influence the concentration of the drug in the plasma. Thus, factors such as species, route of administration, drug formulation, body weight, body condition, and amount and type of nutrition are some of the many factors that can all influence the plasma concentration of the drug (Górniak, 2014; Jerzsele, 2012). In addition, pharmacokinetic parameters are fundamental to the rational use of a drug, and when considering food safety, these parameters reveal the residue concentrations in edible tissue and the withdrawal time from a food animal (Palermo-Neto & Righi, 2014).

Considering the same animal species, in a previous pharmacokinetic study conducted in two different breeds of cattle, yak (*Bos grunniens*) and gobra (*Bos indicus*), the IVM plasma concentrations were lower in both of these breeds than in *Bos taurus* (Dupuy et al., 2003; Ndong, Ba, & Sane, 2005). However, the comparison of the pharmacokinetic results obtained from Dupuy's and Ndong´s studies with those of the *B. taurus* cattle was done from data obtained

in the literature (Lanusse et al., 1997; Lifschitz et al., 1999; Toutain, Campan, Galtier, & Alvinieri, 1988).

Brazil possesses the largest commercial cattle herd, with over 215 million heads of cattle. Since 2004, the country has been the world's largest exporter of beef, and it has been estimated that 80% of Brazilian herds are of zebu origin (CFMV, 2019). Considering that IVM is the most widely used antiparasitic in Brazil (Le Gall, Klafke, & Torres, 2018), the main purpose of the present study was to compare the pharmacokinetic profile of IVM in *B. indicus* (i.e., Tabapuã), *B. taurus* (i.e., Aberdeen Angus), and crossbreed animals *B. indicus* (i.e., Nelore) × *B. taurus* (i.e., Aberdeen Angus) kept under same field conditions. Additionally, we also evaluated the possible impacts of sex and IVM formulation, 1% IVM versus 3.15% IVM, on the pharmacokinetic parameters of this drug in cattle.

2 | MATERIALS AND METHODS

The procedures were approved by the Animal Care Committee of School of Veterinary Medicine and Animal Sciences, University of São Paulo (FMVZ-USP; protocol number 6510200217, February 15, 2019), and all animal care and handling were performed by experienced personnel under veterinary supervision.

2.1 | Drugs

The following drugs were used: 1% IVM (IVM 1%; Ivomec® injectable; Merial Animal Health Ltda.) and 3.15% IVM (IVM 3.15%; Ivomec Gold® injectable, Merial Animal Health Ltda.). The treatment was subcutaneously (SC) administered in the shoulder area at a concentration of 0.2 mg/kg or 0.63 mg/kg body weight for IVM 1% and IVM 3.15%, respectively, as indicated by the drug labels.

2.2 | Animals, feeding, and production system

The study was divided into three trials to evaluate the IVM pharmacokinetics under the following conditions: (a) different bovine breeds (*B. taurus*, *B. indicus*, and *B. Taurus* × *B. indicus*); (b) different bovine sexes (male, castrated males, and female); and (c) drug concentration (IVM1% and IVM 3.15%).

In all experiments, the cattle were maintained under semiconfined conditions in a production system on pastures (*Brachiaria* sp.) and received supplemental commercial concentrate with 18% crude protein (0.5% body weight) and corn silage (5% body weight). A commercial mineral supplement (Tortuga) and fresh water were offered ad libitum. The animals underwent a 30-day adaptation period before the trial began and were not treated with IVM or any other macrocyclic lactone for a minimum of 120 days prior to study. All animals used in this study were sourced from farms within a radius of 50 km.

Aiming to certify the animal's health, five days before the pharmacokinetic assay and immediately after the sample collections, the animals were weighed and clinically evaluated. Blood samples were collected via jugular venipuncture for hematological and biochemical evaluation. The following biomarkers were determined: red blood cells (RBCs), hemoglobin (Hb), hematocrit (Hct), mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC), leukocytes, eosinophils, and monocytes. Commercial kits (CELM®, Barueri) were used to determine the concentrations of albumin, total protein, cholesterol, glucose, aspartate aminotransferase (AST), gamma-glutamyl transferase (GGT), alkaline phosphatase (ALP), urea, and creatinine. Feces were collected for coproparasitological evaluation by the McMaster egg counting technique before the experimental period.

2.3 | Treatment and sampling

Trial one (breed comparison): Twenty-one male bovines were assigned to the following three groups according to breed: Tabapuã (n = 9), Aberdeen Angus (n = 7), and Nelore × Angus (n = 5); the animals ranged in age from 18 to 24 months and weighed 490.2 ± 12.2, 463.0 ± 13.5, and 280.7 ± 13.8 kg, respectively. The experimental period began in February 2018 and ended in April 2018. The Tabapuã cattle were located at the University of São Paulo (USP) Experimental Station, Pirassununga, Sao Paulo state (SP), Brazil (S21°58′, W47°27′). The Nelore × Angus cattle were located at Pirapora, a commercial farm in Tambau, SP, Brazil (S21°71′, W47°25′). The Aberdeen Angus cattle were located at Cardinal, a commercial farm in Mococa, SP, Brazil (S21°48′, W47°08′). Trial two (sex comparison): Twenty-two Nelore × Angus bovines were assigned to the following three groups according to sex: females (n = 10), males (n = 7), and castrated males (n = 5); the animals ranged in age from 18 to 24 months and weighed 353.2 ± 12.2, 463.0 ± 13.5 and 370.7 ± 13.5 kg, respectively. The experimental period began in December 2017 and ended in April 2018. The animals were located at Pirapora, a commercial farm in Tambau, SP, Brazil (S21°71′, W47°25′). Trial three (drug concentration): Nineteen female Nelore bovines were assigned to two groups according to drug concentration, IVM 1% (n = 10) and IVM 3.15% (n = 9), and the bovines ranged in age from 18 to 24 months and weighed 328.5 ± 7.3. The experimental period began in December 2017 and ended in May 2018. The females were located at Pinheirinho, a commercial farm in Analândia SP, Brazil (S22°05′, W47°65′).

Blood samples were collected from all bovines via jugular venipuncture, using heparinized tubes, 15 min before dosing (day 0), and after all animals were weighed and treated with IVM. From the bovines in trials 1 and 2, blood samples were collected at 1, 3, 7, 11, 14, 18, 21, 25, 28, 32, 35, 39, and 42 days after IVM dosage. For trial 3, samples were collected from the animals in the IVM 1% group on exactly the same days as those bovines from trials 1 and 2. From animals in the IVM 3.15% group, samples of blood were collected on

the same days as mentioned before and also at 49, 56, 63, 70, 77, 84, 91, 98, 105, 112, 119, 126, 133, 140, 144, and 147 days after a single IVM administration. IVM concentrations were determined in plasma samples, which were frozen to −80°C until the time of analysis.

2.4 | Analytical procedures

Plasma aliquots of 300 µl were used. The extraction was carried out applying precipitation method with acetonitrile. In order to obtain the optimized condition for the extraction and removal of matrix interferences, aliquots of acetonitrile were added sequentially followed by vigorous vortexing for about 15 s. Aliquots of 100 µl were added in three steps followed by an aliquot of 300 µl. Each addition followed by vortexing. To obtain the extract, centrifugation was performed at 12,000 *g* for 10 min at a temperature of 5°C. A volume of about 700 µl was transferred to vials and subjected to analysis by LC-MS/MS.

Ivermectin standard with purity of 94.4% was purchased from Dr. Ehrenstorfer. The plasma concentration of IVM (22,23 dehydro-avermectin B1a) was analyzed by liquid chromatography coupled to mass spectrometry (LC-MS/MS) using an Agilent 1260 LC system coupled to an ABSciex API 5000 triple quadrupole mass spectrometer. The chromatographic separation was carried out on a C18 Luna column (3.0 µm, 50 mm × 2 mm; Phenomenex) with a guard column (5.0 µm, 4 mm × 3 mm; Phenomenex). The mobile phase gradient elution was water (A) and acetonitrile (B), both containing 5 mM ammonium acetate and 0.1% acetic acid. The flow rate was 500 µl/min. The gradient started with 80% of A, which decreased to 0% in 1 min and was maintained until 3.5 min. Then, the elution was returned to the initial condition at 4 min and maintained until 6 min. The column temperature was maintained at 40°C. The injection volume was 10 µl.

Mass spectrometer resolution in multiple reactions monitoring (MRM) was unitary, and the dwell time applied was 200 ms for all transitions. The mass spectrometer was operated in positive electrospray ionization mode.

Nitrogen was used as the nebulizer gas, curtain gas, heater gas, and collision gas. The collision gas (CAD) was set at 6 psi, and the nebulizer gas (GS1) and dryer Gas (GS2) were set at 55 psi. The curtain gas and temperature were set at 30 psi and 500°C, respectively. The electrospray capillary voltage was set at 4.5 kV.

Peak concentrations (C_{max}) of IVM in blood and the time of the peak concentration (T_{max}) were obtained directly from the experimental data without interpolation. The area under the concentration–time curve (AUC) was calculated by the linear trapezoidal method. All pharmacokinetic parameters were calculated as described by Shargel and Yu (1993) using the PK Solutions 2.0 (Summit Research Services).

2.5 | Statistical analysis

The plasma concentrations of IVM were analyzed using a mixed linear model (Proc Mixed) with the treatment (breed, sex, or drug concentration); the animals were nested within treatments, and repeated measurements of the IVM concentrations were taken over time using SAS software (version 9.2; SAS Institute, 2008). The animals were considered a random factor in the model.

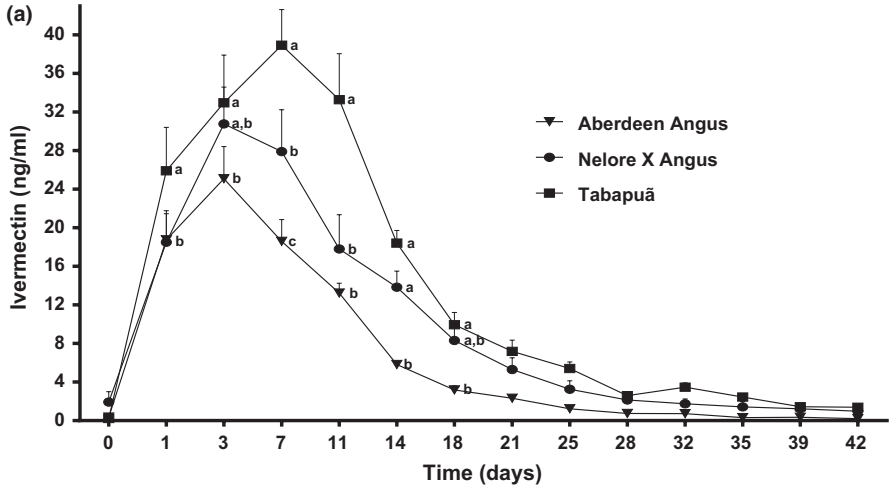
For pharmacokinetic data from the breed or sex comparison, the Bartlett test was used to determine whether the data were normally and homogeneously distributed. A one-way ANOVA with Tukey's post hoc test was used to compare the groups for each variable using SAS software. Pharmacokinetic data from drug concentration comparison were tested using *t* tests (version 9.2; SAS Institute, 2008). In all cases, the probability of significant differences was set at $\alpha = 0.05$. Data are reported as the mean ± standard error of the mean (*SEM*).

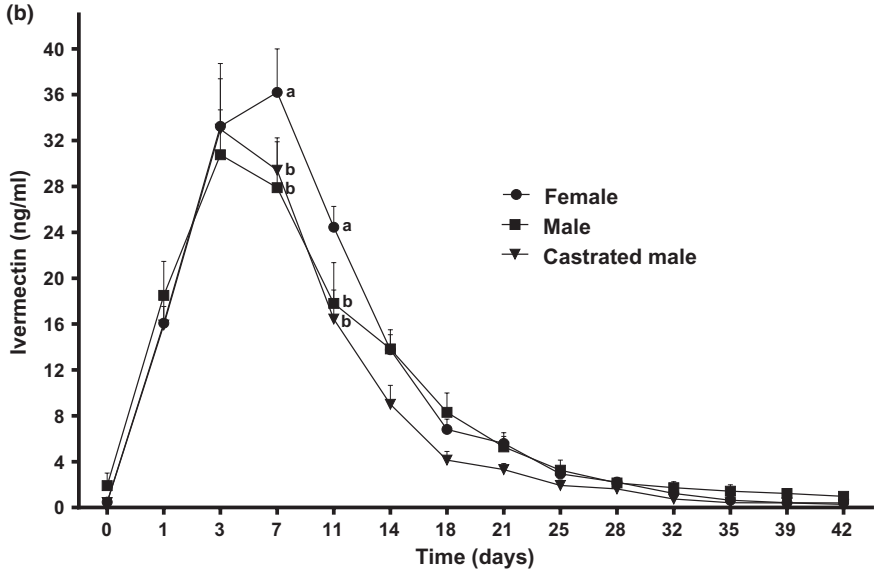
3 | RESULTS

The analytical procedures used to quantify IVM plasma concentration were validated according to the 2002/657/EC Directive (European Commission, 2002) . Linear regression showed that the coefficients of determination were greater than 0.99. The mean extraction recoveries of IVM from plasma were more than 90%. Intraday precision values were 5.1% [15%], interday precision values were 8.8% [15%], and the accuracy was between −12 and 8% [−20% to +10%]. The limit of quantification was established to be 5 ng/ml.

None of the bovines showed any clinical changes during the experimental period. Hematological and biochemical evaluations revealed that the values remained within the reference range for the species and were considered physiologically normal in all the evaluated animals (data not shown). In the same way, no animal showed a severe infection in the coproparasitological examination (data not shown).

The plasma concentrations of IVM in the bovines are presented in Figure 1, and the pharmacokinetic data are presented in Table 1. In trial one, the variation in IVM concentrations was significantly affected by the breed and time of evaluation; from the 1st day until the 18th day after IVM administration, the plasma concentrations of IVM were significantly increased in Tabapuã cattle compared to Angus cattle ($p < .05$). The plasma concentrations of IVM in crossbreed animals (Angus × Nelore) showed an intermediate profile between Tabapuã and Angus in all evaluations. The sustained higher IVM plasma concentration in Tabapuã cattle accounted for the greater AUC in this breed than in Angus cattle ($p < .05$). Similarly, the Tabapuã cattle C_{max} and $t_{1/2\beta}$ were higher than those of the Angus cattle ($p < .05$). In trial two, the plasma concentrations of IVM were very similar among the sexes; the plasma concentration was increased in females 7 and 11 days after IVM administration ($p < .05$). Sex was not responsible for any differences in pharmacokinetic patterns. In trial three, one and three days after IVM administration, the plasma concentrations in the IVM1% group were higher than those in the IVM 3.15% group ($p < .05$). From the 11th day until the 42nd day after IVM administration, the IVM plasma concentrations of the IVM3.15% group were significantly higher than those of the IVM1% group ($p < .05$). Concerning the pharmacokinetic data,





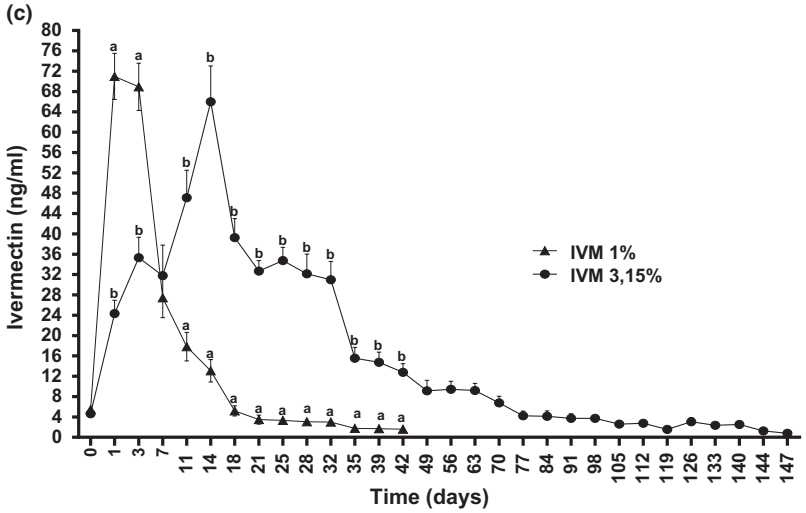


FIGURE 1 Mean ± *SEM* of the plasma concentration of ivermectin (ng/ml) in bovines. Breed (A) and sex (B) comparison after a single subcutaneous 1% ivermectin administration (0.2 mg/kg body weight). (C) Comparison of drug concentration after a single subcutaneous administration of 1% or 3.15% ivermectin (0.2 mg/kg or 0.63 mg/kg body weight, respectively). Different letters (a, b and c) at the same timepoint represent significant differences among the groups ($p < .05$)

the T_{max} in the IVM1% group was lower than that in the IVM3.15% group ($p < .05$). No significant difference was observed in the C_{max}. The maintenance of high IVM concentrations in the IVM 3.15% group for a longer period resulted in higher AUC and $t_{1/2\beta}$ in this group than in the IVM 1% group ($p < .05$).

4 | DISCUSSION

It is well known that the kinetic behavior of IVM is largely influenced by many factors (Flajs & Grabnar, 2002; Canga et al., 2009). Considering that knowledge of the pharmacokinetic parameters of a

TABLE 1 Pharmacokinetic data of ivermectin in the bovine, breed, and sex comparisons after a single subcutaneous administration of 1% ivermectin (0.2 mg/kg body weight) and comparison of the drug concentration after a single subcutaneous administration of 1% or 3.15% ivermectin (0.2 mg/kg or 0.63 mg/kg body weight, respectively)

	Breed comparison		
Kinetic parameters	**Angus (7)^n**	**Tabapuã (9)**	**Nelore × Angus(5)**
C_{max} (ng/ml)	25.84 ± 3.18^a	44.49 ± 4.03^b	30.99 ± 3.99^{ab}
T_{max} (days)	4.71 ± 0.81	6.25 ± 1.31	3.80 ± 0.80
$t_{1/2\beta}$ (days)	4.61 ± 0.45^a	6.17 ± 0.36^b	5.9 ± 0.54^{ab}
K_{el} (h^{-1})	0.16 ± 0.02	0.12 ± 0.01	0.12 ± 0.01
Cl (L/h)	360.70 ± 44.52	266,08 ± 21.13	362.23 ± 56.76
AUC (ng d/ml)	276.1 ± 14.7^a	590,3 ± 45.4^b	426.6 ± 63.7^{ab}
	Sex comparison		
	Female (10)	**Male (7)**	**Castrated male (5)**
C_{max} (ng/ml)	36.6 ± 5.03	31.00 ± 3.99	38.50 ± 1.88
T_{max} (days)	6.20 ± 0.80	3.80 ± 0.80	4.14 ± 0.74
$t_{1/2\beta}$ (days)	5.01 ± 0.23	6.28 ± 0.81	4.88 ± 0.28
K_{el} (h^{-1})	0.14 ± 0.01	0.12 ± 0.01	0.14 ± 0.01
Cl (L/h)	282.19 ± 42.85	362.23 ± 56.76	280.20 ± 17.96
AUC (ng d/ml)	444.1 ± 39.0	426.4 ± 63.7	374.0 ± 20.3
	Drug concentration		
	1% Ivermectin (10)		**3.15% Ivermectin (9)**
C_{max} (ng/ml)	75.57 ± 4.31		66.22 ± 6.96
T_{max} (days)	1.80 ± 0.33^{ass}		15.22 ± 1.22^b
$t_{1/2\beta}$ (days)	7.19 ± 0.42^a		22.84 ± 1.67^b
K_{el} (h^{-1})	0.10 ± 0.01^a		0.03 ± 0.00^b
Cl (L/h)	89.97 ± 8.00		105.60 ± 12.98
AUC (ng d/ml)	661.4 ± 42.5^a		1904.0 ± 128.3^b

Abbreviations: AUC, area under the concentration–time curve from time zero to the last time with a measurable concentration; Cl, clearance; C_{max}, peak plasma concentration; K_{el}, elimination constant; $t_{1/2\beta}$, elimination half-life; T_{max}, time of peak plasma concentration.
Within a row, mean kinetic parameters lacking a common superscript letter are significantly different at $p < .05$.

drug is a basis not only for predicting and optimizing drug efficacy but also for determining the residual concentration in edible tissues and withdrawal times, it is quite desirable to understand all factors involved in IVM disposition. Thus, in addition to the pharmacokinetic evaluation of IVM considering "traditional" factors such as animal species, sex, age, routes of administration and formulations (Fink & Porras, 1989; Canga et al., 2009), other conditions such breed could also be relevant in the disposition of macrocyclic lactones (Vercruysse, Deprez, Everaert, Bassissi, & Alvinerie, 2008). To the best of our knowledge, no other research has been performed that compares the pharmacokinetic profile of IVM or any other macrocyclic lactones among *B. taurus*, *B. indicus*, and crossbreed animals under the same experimental conditions (i.e., in the same: geographic location, time of the year, semiconfined condition, and feed).

Although the t_{max} values of the three breeds evaluated were equivalent, the AUC of the Tabapuã cattle was more than twofold greater than that of the Angus cattle. The AUC of the crossbreed cattle was an intermediate value between *B. taurus* and *B. indicus*; in fact, no significant differences were detected between the AUC of Angus × Nelore and those of both Tabapuã and Angus. In fact, the C_{max} obtained in Tabapuã was much higher than that verified in Angus. Moreover, it could be clearly observed that the elimination half-life ($t_{1/2\beta}$) in Tabapuã cattle was significantly higher than that of Angus, and in the same manner, no significant differences were detected between the $t_{1/2\beta}$ of the crossbred animals and those obtained from both Tabapuã and Angus cattle.

Ndong et al. (2005), after studying the plasma kinetics of IVM in Zebu Gobra cattle (*B. indicus*) and comparing the values of the pharmacokinetic parameters obtained in their trial with those from *B. taurus* available in the literature (Lanusse et al., 1997; Lifschitz et al., 1999; Toutain et al., 1988), proposed that drug concentration has lower in the Gobra breed than in *B. taurus* breeds. However, our

pharmacokinetic data clearly revealed the opposite of Ndong´s et al. conclusion, as we verified here that the AUC of IVM in Zebu (Tabapuã) was much higher than that in Angus. A possible explanation for this discrepancy could be related to the different experimental conditions when comparing pharmacokinetic data of Gobra and *B. taurus* breeds. In fact, although Ndong´s comparison took into account that in all the studies selected from the literature, European cattle also received the same commercial formulation of IVM (Ivomec®) by the same route (SC) and dose (0.2 mg/kg) as the Gobra cattle in their trial, other factors that strongly interfere in IVM pharmacokinetics, such as age, environmental conditions, nutritional status, parasitic infection, production system, and sampling times (Flajs & Grabnar, 2002), could not be controlled.

Due to its high lipophilicity, IVM is characterized by a large distribution and long excretion time (Steel, 1993). It is widely known that cattle of European origin have, characteristically, greater "marbling" (i.e., an accumulation of intramuscular fat) than in to Zebu cattle (Huffman, Williams, Hargrove, Johnson, & Marshall, 1990); thus, we expected that the value of the half-life of elimination in Angus to be higher than that in Tabapuã cattle. Nevertheless, our results undoubtedly showed the contrary; the highest value was verified in Tabapuã cattle, followed by the crossbred animals and then Angus cattle. We have few hypotheses to explain this unexpected result at this moment, since we did not perform studies to detect IVM residue in tissues. However, it should be considered that Zebu cattle possess a much higher incidence of intramuscular fat in the hump (*Rhomboideus* muscle) than do any other cattle breed (Pedrão et al., 2009). Hence, knowing that the fat-containing tissues in cattle permit the prolonged residence of IVM (Lifschitz et al., 2000), it is feasible to hypothesize that the hump could sequester the drug, acting as a drug reservoir and in this manner, promoting the half-life of elimination higher in Zebu cattle.

P-glycoprotein (P-gp) is a plasma membrane protein belonging to the ATP-binding cassette superfamily and has been identified as the main factor that controls the concentration of many drugs by affecting their in vivo absorption, distribution, and excretion in the host (Kwei et al., 1999; Schinkel, Wagenaar, Mol, & Deemter, 1996). P-gp is expressed in many tissues, including the brush border epithelial cells of the intestinal mucosa, and actively effluxes many drugs, including IVM, out of cells into the biliary and intestinal lumen, thus reducing plasma concentrations (Bodó, Bakos, Szeri, Váradi, & Sarkadi, 2003).

Considering that intestinal secretion is a major route for the elimination of IVM, which undergoes very little metabolism, as most of the excreted drug is unchanged (Canga et al., 2009), another possibility to explain the higher persistence of IVM in Tabapuã cattle than in Angus cattle is that Zebu could have relatively lower expression of P-gp at the intestinal level, causing a decrease in the intestinal clearance of IVM. Corroborating this assumption, a study by Laffont, Toutain, Alvinerie, and Bousquet-Melou (2002) conducted in rats that coadministered the P-gp blocker verapamil with IVM, verified a significant 50% reduction in the elimination capacity of the jejunum, which resulted in a 30% decrease in the overall elimination of IVM by the small intestine. In the same manner, two additional studies, in vitro (Ballent, Lifschitz, Virkel, Sallovitz, & Lanusse, 2006) and in vivo in rats (Ballent et al., 2006; Lifschitz, Ballent, Virkel, Sallovitz, & Lanusse, 2006), using another P-gp blocker, itraconazole, verified that the IVM concentrations measured in the intestinal wall tissue of different sections of the gastrointestinal tract was significantly higher (between 60% and 100%) in the itraconazole-treated group compared with the group treated with IVM alone. However, to better clarify this hypothesis, further immunohistochemistry evaluation should be conducted to assess the P-gp protein expression in the small intestine of the three breeds of cattle studied in this research.

Among other factors, sex has been reported to have a significant influence on different pharmacokinetic parameters (Mugford & Kedderis, 1998). In this sense, studies have shown kinetic discrepancies between males and females treated with macrocyclic lactone compounds in different animal species (Dupuy et al., 2004; Dupuy, Eeckhoutte, Sutra, Mage, & Alvinerie, 1999). A study conducted by Toutain et al. (1997) showed higher IVM plasma concentrations and consequently 10% higher AUC in heifers compared to steers. In an additional study to elucidate the difference between the IVM pharmacokinetics of sexes, Lifschitz et al. (2006) verified that although both sexes of rats showed an increase in AUC ratio after the coadministration of IVM with itraconazole, the values of this parameter were approximately six times higher in males than in females. The authors attributed this response to the higher P-gp expression/activity in the male intestine, which causes greater intestinal clearance of IVM and consequently lower bioavailability. In fact, in the present study, we also detected an increase in the IVM plasma concentrations of females compared to those of males and castrated males; however, the pharmacokinetic profiles of the three studied groups were similar.

The effectiveness of parasitic control in livestock is directly related to the duration of the antiparasitic effect of the drug on the animal (Bridi, Carvalho, Cramer, & Barrick, 2001; Rehbein, Visser, Winter, & Maciel, 2002). To extend the antiparasitic effect of a single treatment, long-acting IVM drugs were developed by pharmaceutical industries. Thus, we decided to evaluate the main pharmacokinetic differences between the IVM1% and IVM-LA3.15% treatments in Zebu cattle.

Thus, considering the efficacy of IVM against *Boophilus microplus*, the main parasite target in tropical countries (Rodriguez-Vivas, Jonsson, & Bhushan, 2018), it was consistently shown that the efficacy of a single administration of the same commercial product, IVM, at different concentrations, 1% and 3.15%, in European cattle lasts less than 15 days (Toutain, Upson, Terhune, & McKenzie, 1997) and 42 days (Davey, Pound, Miller, & Klavons, 2010), respectively, which corresponds to IVM plasma concentrations higher than 8 ng/ml (Davey et al., 2010). In our study, we verified that this plasma concentration (8 ng/ml) of the antiparasitic drug in Nelore cattle persisted for 17 days after IVM1% administration and for 65 days after IVM-LA3.15% administration. In addition, in previous studies conducted by Toutain et al. (1997) and Davey et al. (2010), European cattle presented C_{max} values of 31.7 and 26.2 ppb after IVM1% and

IVM-LA3.15% administration, respectively. However, in the present study, we observed a C_{max} twice as high in Nelore cattle, 75.5 and 66.2 ng/ml after the administration of IVM1% and IVM-LA3.15%, respectively. Hence, although the present study and the studies with European cattle were not conducted under the same experimental conditions, and as we noted before, experimental conditions could directly interfere with pharmacokinetic parameters, it is feasible to consider that IVM could present more therapeutic and persistent efficacy in combating ticks in Zebu cattle than in European cattle; thus, to better clarify this conjecture, future studies will be performed to evaluate the plasma concentrations of IVM after the administration of the two concentrations employed in this study in both breeds, *B. taurus* and *B. indicus*, under the same experimental conditions.

This study emphasizes the differences between Indian and European cattle with regard to the pharmacokinetic profile of IVM. The results clearly indicated that the IVM plasma concentrations in Tabapuã (Indian cattle) were much higher than that in Angus (European cattle). The practical food safety implications of these pharmacokinetic differences between breeds, and the possible repercussion on altered withdrawal periods of IVM in Zebu cattle may require further evaluation.

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CONFLICT OF INTEREST

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AUTHORS' CONTRIBUTION

A.T.G., R.L.F., and S.L.G. conceived and designed the experiments; A.T.G. and R.L.F. performed the experiments; A.T.G. and R.L.F. analyzed the data; F.B., C.A.T., L.S.B., T.C., and R.L.F. performed the analytical method and analyzed the samples; and A.T.G. and S.L.G. wrote the paper.

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