Population pharmacokinetics of a single intramuscular administration of tulathromycin in adult desert tortoises (Gopherus agassizii)

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INTRODUCTION

Habitat degradation, illegal collection, and upper respiratory tract disease (URTD) were identified as major factors in the decline of the Mojave desert tortoise population (Gopherus agassizii), which prompted listing them as a threatened species (Fish & Wildlife Service, 1990). Diagnosing, treating, and controlling URTD is a priority for many conservation programs due to the potential for severe morbidity and mortality in free-ranging desert tortoises (Fish & Wildlife Service, 1994). Clinical signs of URTD in desert tortoises include rhinitis, conjunctivitis, chemosis, nare and ocular discharge, periocular edema, and conjunctival hyperemia (Jacobson et al., 1991; Schumacher et al., 1993; Jacobson et al., 1995; Schumacher et al., 1997; Ledele et al., 1997; Homer et al., 1998; Christopher et al., 2003). The cause of URTD in desert tortoises is multifactorial with viral, Gram-negative bacterial, and mycoplasma etiologies identified (Jacobson et al., 1991; Schumacher et al., 1993; Brown et al., 1994; Jacobson et al., 1995; Snipes et al., 1995; Pettan-Brewer et al., 1996; Schumacher et al., 1997; Dickson et al., 2001; Brown et al., 2004; Johnson et al., 2005). Mycoplasma agassizii is of particular concern because this organism alone can cause morbidity and mortality in desert tortoises independent of co-infection with other viral or bacterial pathogens. Koch’s postulates have been fulfilled, further highlighting the importance of M. agassizii as a cause of morbidity in desert tortoises (Brown et al., 1994).

Enrofloxacin or clarithromycin are the mainstays of treatment in tortoises diagnosed with mycoplasmosis, and pharmacokinetic data have been published for both antibiotics (Prezant et al., 1994; Wimsatt et al., 1999; Wimsatt et al., 2008). The efficacy of enrofloxacin- or clarithromycin-based treatment protocols for mycoplasmosis have not been evaluated in chelonians in part because demonstration of mycoplasma clearance is difficult and subclinical infection is common. As such, the goal of most treatment protocols is alleviation of
clinical signs which has been anecdotally reported using both enrofloxacin and clarithromycin. Although potentially effective in alleviation of clinical URTD signs, intramuscular enrofloxacin administration may result in acute, moderate lameness 5–10 min following injection, injection site swelling at 24 h, muscle necrosis, skin depigmentation, and sloughing (Prezant et al., 1994). Pharmacokinetic data suggest administration every 24–48 h is needed to maintain therapeutic concentrations of enrofloxacin in chelonians (Prezant et al., 1994). The potential for lameness and swelling associated with injection and 24–48 h dosing frequency makes enrofloxacin use problematic for long-term management of URTD, especially when treating large numbers of tortoises. The use of clarithromycin is limited, as this antibiotic is formulated for oral administration, restricting its clinical use to animals that are continuing to eat or have an esophagostomy tube placed. Per rectum administration of clarithromycin did not result in therapeutic plasma levels and thus is not considered a useful administration route in anorexic chelonians (Wimsatt et al., 2008). As such, alternative treatment modalities should be explored.

Mycoplasma species are mollicute bacteria that lack a cell wall and may be commensal or opportunistic organisms that have the potential to cause clinical disease in a variety of animals including reptiles, birds, and mammals; (Brown et al., 2005; Kleven, 2008; Besser et al., 2008; Maunsell et al., 2011; Vranckx et al., 2012). Due to the lack of a rigid cell wall, antibiotics that inhibit muropeptide synthesis in the cell wall are ineffective against Mycoplasma spp. Tulathromycin is a macrolide antibiotic that inhibits protein synthesis by binding to the 50S ribosomal subunit. Activity does not depend on the presence of a cell wall to be effective. Efficacy is likely enhanced by its ability to accumulate and be released by host phagocytic cells. The tribasic structure of tulathromycin allows it to accumulate in susceptible bacteria. Tulathromycin is used in food animal medicine for treatment of respiratory disease caused by a variety of organisms including Mycoplasma spp. Swine experimentally inoculated with M. hyopneumoniae and treated with a single intramuscular dose of tulathromycin demonstrated reduced coughing compared to pretreatment assessments (McKelvie et al., 2005). Dairy calves experimentally infected with two pathogenic strains of M. bovis demonstrated improvement of clinical signs and less severe postmortem lung lesions when treated with tulathromycin administered intramuscularly (Godinho et al., 2005).

The objective of this study was to determine the pharmacokinetics of tulathromycin in a population of captive held desert tortoises as a first step in evaluating the potential use of this antibiotic for treatment of URTD in chelonians.

MATERIALS AND METHODS

Adult desert tortoises (N = 110; 62 males and 48 females, mean weight ± SD, range = 3.2 ± 1.2, 1.4–6.8 kg) used in this study were housed at the Desert Tortoise Conservation Center in Las Vegas, NV, USA. Animals were maintained in outdoor enclosures (13.4–232 square meters) and provided supplemental water three times weekly and food (Zoo Med grassland tortoise food, Zoo Med Laboratories Inc, 3650 Sacramento Dr, San Luis Obispo, CA) twice weekly, except during the winter months (November–March). The study was conducted in April, and environmental temperature ranged from 14 to 29 °C. Animals were considered healthy based on physical examination immediately prior to commencement of the study. No study animals demonstrated clinical signs of active upper respiratory tract disease. During the study period, animals were housed individually in plastic containers lined with Timothy hay and allowed to bask in direct sunlight for a minimum of 4 h each day. Animals were soaked in water every other day throughout the duration of the study period. After physical examination and sex determination, tortoises were randomly assigned to one of eleven groups corresponding to the time point blood would be collected for drug concentration analysis, including one group of ten tortoises who served as controls and were not administered tulathromycin.

Tulathromycin (Draxxin®, 100 mg/mL injectable solution, Zoetis, New York, NY, 10017) was administered intramuscularly into the right forelimb musculature of 100 tortoises at a dose of 5 mg/kg body weight using 22-G needles and 3-mL syringes. A control group of 10 tortoises did not receive an injection of tulathromycin. The 5 mg/kg dose is twice the recommended weight-normalized dose of 2.5 mg/kg for cattle and swine. The 5 mg/kg dose was based on clinical observations at the Desert Tortoise Conservation Center that demonstrated minimal improvement in clinical signs in tortoises treated with 2.5 mg/kg, whereas tortoises administered 5 mg/kg demonstrated some improvement in clinical signs of URTD. After the drug was administered, digital pressure was placed on the right forelimb immediately after withdrawing the needle to prevent drug leakage from the injection site. The injected limb was visually evaluated daily for 5 days following injection for evidence of redness, swelling, or lameness. A single blood sample was collected from 10 tortoises at each of the following time points after tulathromycin administration: 0.25, 0.5, 1, 4, 8, 24, 48, 72, 120, and 240 h. Blood samples were collected from each tortoise only once during the study, including 10 tortoises that did not receive tulathromycin. During jugular blood collection, the tortoise was placed in right or left lateral recumbency, the head was extended to expose the lateral cervical region and the jugular vein was palpatated or visualized. Blood (1–3 mL) was collected from the jugular vein using a 22- or 23-gauge needle and 3-mL syringe. Pressure was applied to the blood collection site using either digital pressure or gauze attached to a tongue depressor. The sample was placed into a lithium heparin microtainer blood tube (3 mL, BD Franklin Lakes, NJ, USA), and immediately refrigerated. Within 6 h of blood collection, the blood samples were centrifuged for 15 min at 2594 g, the plasma was decanted, distributed into storage tubes, and frozen at ~80 °C until analyzed. If 1 mL of blood could not be collected within 5 min of the sampling time point, attempts at blood collection were not pursued further and the tortoise was removed from the study.
Samples were analyzed in singlet for tulathromycin using an ultra-performance liquid chromatography, tandem mass spectrometry (UPLC-MS) assay based on a previously described method (Galer et al., 2004). CP-66,458 was used as the internal standard. Samples were diluted with 0.05 M potassium phosphate and extracted by solid phase extraction (Isolute, 500 mg CBA, Biotage, Uppsala, Sweden). Most of the samples required extensive dilution to bring them within the standard curve range. The standard curve ranged from 2 to 500 ng/mL and consisted of eight concentrations. Analytical separation was on a UPLC system (Waters, Milford, MA) with an Ace C8, 2.1 x 50 mm, 3 μm column (MacMod, Chadds Ford, PA). Detection was carried out with a Thermo TSQ Quantum Discovery Max (Thermo Electron, West Palm Beach, FL) tandem quadrupole mass spectrometer with a heated electrospray ionization source operated in the positive ion mode. The limit of detection (LOD) and limit of quantification (LOQ) were based on manually defined peaks within the baseline at the expected retention time for time 0 samples. The standard curve was used to calculate the concentrations for the manually defined peaks. Calculated LOD and LOQ were based on 3 and 10 times the standard deviation (SD, respectively. The limit of detection was determined to be 1.2 ng/mL and the limit of quantification was 4.0 ng/mL. Recovery was 94.3% and interassay variability was 8.9% as measured by relative standard deviation of QC’s for 11 assay runs. Intra-assay variability was not determined due to limited volume of control matrix.

A noncompartmental analysis of the time–concentration profile was performed using the sparse sampling option in Phoenix® (Certara, Cary, NC). This option calculates the pharmacokinetic parameters from the mean curve of the pooled data. Parameters for this analysis are the maximum observed plasma concentration (Cmax), time to that maximum observed plasma concentration (Tmax), and slope of the terminal portion of the time–concentration profile [elimination rate constant (kel)]. The area under the time–concentration [area under the curve (AUC)] profile was calculated using the trapezoidal rule and extrapolated to infinity by adding the ratio of the last measured concentration and slope of the terminal portion [plasma concentration at the last observed time point (Clast)/Kel]. The clearance and volume of distribution, both corrected for unknown bioavailability, are then calculated as the ratios Dose/Area under the curve extrapolated to infinity (AUClast) and Dose/(AUClast × Kel), respectively.

A disadvantage of the naïve-pooled noncompartmental analysis is that there is no way to estimate variability in pharmacokinetic parameter values in the population. Therefore, the data were also fitted to one- and two-compartment models with first-order absorption using the Pmetrics nonparametric population modeling and simulation package for R (Neely et al., 2012). Population approaches are ideal for study designs that must incorporate sparse sampling, such as the one animal – one sample design in the current study. The primary parameters for this model are the absorption rate constant (Ka), the elimination rate constant (Kel) and the volume of the central compartment, corrected for unknown bioavailability (Vc/F).

The two-compartment model additionally has transfer rate constants from the central to a peripheral compartment (KCP) and from the peripheral back to the central compartment (KPC). The program creates a nonparametric population model consisting of discrete support points, each with a set of estimates for all parameters in the model plus an associated probability of that set of estimates. There can be at most one point for each individual in the population. As doses were weight adjusted, we did not include this covariate in any model. No other covariates were available. Models were discriminated using the Akaike Information Criterion (AIC), by inspection of the observed versus predicted plots, and minimizing bias and imprecision of predicted concentrations relative to observations. Bias was calculated as the mean weighted error of prediction minus observation, and imprecision was calculated as the mean weighted squared error, adjusted for bias. All predicted concentrations were calculated using either the median of the population values or the median of each tortoise’s Bayesian posterior parameter value distribution. Predicted time–concentration profiles were then simulated for each animal in the population and the noncompartmental parameters AUC0–∞, Cmax, Tmax and T½ calculated to compare the average with the noncompartmental analysis of the naïve-pooled data.

Because the measurements in all assays have some imprecision, in Pmetrics, observations during the fitting process are weighted according to the reciprocal of the variance (standard deviation squared) of the tulathromycin assay at that concentration. To calculate the standard deviation (SD) for any measured tulathromycin concentration, we initially used a function in Pmetrics to fit replicate standard measurements to a first-order polynomial. However, because of the sampling strategy, lower concentrations from animals at later time points were overly de-weighted; hence, we used Pmetrics to estimate the coefficients for the polynomial directly from the observed tulathromycin concentrations in the study population, such that SD = 0.001 – 2*10^-7*C, where C is the concentration of tulathromycin. This is essentially a constant SD of 0.001.

RESULTS

Following tulathromycin injection, no lameness, erythema, or swelling was noted in any of the tortoises. Blood was collected from the jugular vein of 101/110 tortoises (92%). Inability to extend the neck, lack of visualization or palpation of the jugular vein, and movement of the patient during restraint prevented blood collection in nine animals at the 4 (n = 3), 24 (n = 1), 48 (n = 3), 72 (n = 1), and 120 (n = 1) hr sampling time points. In spite of applying digital pressure to the phlebotomy site, lateral cervical hematomas occurred in two animals. After these hematomas were noted, the postblood collection procedure was altered and instead of applying digital pressure to the cervical region, gauze attached to a tongue depressor was used to allow for continued pressure application when the neck was retracted. No further cervical hematomas were observed.
Liquid chromatography mass spectrometry detected the parent form of tulathromycin in all of the plasma samples from tortoises administered tulathromycin. A mean (±SD) Cmax of 36.2 ± 29.7 µg/mL was observed at 0.25 ± 0 h (Tmax). Tulathromycin was detected in all plasma samples 240 h following administration (mean Clast = 0.6 ± 0.2 µg/mL). Coefficient of variance (CV%) for measured drug concentrations at sampling times ranged from 26 to 80% (data not shown). Results of the noncompartmental analysis of the naive pooled data were T½ = 77.1 h, CL/F = 4.7 mL/h/kg and V/F = 0.53 L/kg.

The AUCs for the one-compartment model versus the two-compartment model were 271 and −188, respectively, with the lower value for the two-compartment suggesting greater likelihood. The choice of the two-compartment model is further supported by comparing plots of the individual predicted time–concentration profiles for the one- and two-compartment models versus the observed concentrations (Fig. 1a,b). The population parameter estimates for the two-compartment model are summarized in Table 1. The values for KCP and KPC were fixed after being varied manually to achieve a good fit. This was necessary due to the one sample per animal study design, because the early phase animals forced the fit to a one-compartment model, although this did not describe the overall trend in concentrations well (Fig. 1a). There is a broad distribution of points in the population predictions vs. observations (Fig. 2a). However, when the median Bayesian posterior parameter values for the individuals were used to calculate predictions, agreement with observed concentrations was excellent, with very low bias (−0.0396) and imprecision (0.307). The regression of posterior predictions and observations had an R² of 0.99 (Fig. 2b). Table 2 compares the noncompartmental parameters calculated from the predicted profiles with those from the naïve-pooled data. The average values for the elimination half-life (T½el), maximum plasma concentration (Cmax), and AUC∞ were essentially the same as those calculated from the naïve pooled data, although the range of latter parameter was very wide (278.6–8270.4 h*µg/mL) and not normally distributed.

**DISCUSSION**

Tulathromycin was rapidly absorbed following intramuscular injection of a single 5 mg/kg dose and did not result in local adverse reactions in healthy desert tortoises. Using liquid chromatography mass spectrometry, plasma tulathromycin was detectable at all sampling time points starting at 0.25 h and extending to 240 h posttulathromycin injection. Noncompartmental analysis of pooled time–concentration data suggested tulathromycin was rapidly absorbed, achieved high concentrations, distributed into a volume that is suggestive of total body fluid without sequestration, and was slowly eliminated from the body.

Compartamental analysis of the pharmacokinetic data using a nonparametric approach revealed tulathromycin was absorbed relatively rapidly and distributed initially to a small central compartment, resulting in high initial plasma concentrations (Kₐ = 0.2 1/h, Vc/F = 5.4 mL/kg). The drug was then rapidly distributed to a peripheral compartment (KCP = 4.0), from which it slowly distributed back into the central compartment (KPC = 0.04). This resulted in a rapid initial decline in the high plasma concentrations, followed by lower concentrations that were sustained for a prolonged period of time.

The consequence is that the mean AUC∞ calculated from the predicted curves from the population model is lower than the AUC∞ calculated from the naïve-pooled data. From a clinical perspective, this offers the advantage that when considering a pharmacodynamic parameter such as AUC/mean inhibitory concentration (MIC), the population modeling would not overestimate the AUC∞. However, given the single-sample per animal study design, it is not possible to identify a
Definitive explanation for the second peak, and both curves must be considered equally valid; however, for the purposes of dose calculation, the more conservative approach would be to use the smaller AUC0-∞.

Despite adjusting the tulathromycin dose by animal weight, there was considerable interanimal variability in drug exposure, as evidenced by the broad distribution of points in the population predictions vs. observations (CV% for measured drug concentrations at sampling times ranged from 26 to 80%). This suggests that there are unmeasured covariates affecting the disposition of the drug in these animals, such as age, sex, body condition score, ambient temperature, or underlying disease conditions affecting renal and hepatic function. In addition, the AUC0-∞ of the predicted time-concentration profiles was highly variable and not normally distributed. As 75% of the population were predicted to have an AUC0-∞ lower than the value calculated from the naïve pooled data, dosage regimens based on the latter could potentially be ineffective in the majority of animals.

The extremely close fit of the posterior predictions strongly suggests that our two-compartment model and parameter value distributions are representative of tulathromycin pharmacokinetic disposition in this tortoise species. This model will be useful to optimize sampling times for future studies. In addition, future collection of covariate data such as body condition, age, and sex may help to identify factors leading to the moderate interindividual pharmacokinetic variability in the population.

The rapid absorption of tulathromycin in desert tortoises (Tmax = 0.25) is similar to that observed in rabbits (1.0 h), lactating goats (0.5 h), pigs (0.25 h), and calves (1.12 h) administered intramuscular tulathromycin (2.5 mg/kg) (Tohamy et al., 2011; Zhao et al., 2011; Abo-El-Sooud et al., 2012; Amer et al., 2012). While absorption times are similar, higher tulathromycin plasma concentration is observed in tortoises (Cmax = 32.2 μg/mL) compared to other species. Tortoises had peak plasma tulathromycin concentrations that were nearly 50–100 times higher than rabbits (Cmax = 0.71 μg/mL), lactating goats (Cmax = 0.73 μg/mL), foals (Cmax = 0.76 μg/mL), calves (Cmax = 0.33 μg/mL), and pigs (Cmax = 0.75–0.84 μg/mL) (Scheuch et al., 2007). Similarly, the area under the curve (extrapolated to infinity) in desert tortoises (AUC0-∞ = 1055.7 μg/h/mL) was larger than those reported in rabbits (8.44μg/mL), goats (27.4μg/mL), pigs (15.77–18.53μg/mL), and calves (33.90μg/mL); it must be noted that the tulathromycin dose administered to tortoises in this study was 5 mg/kg compared to 2.5 mg/kg in all other studies. Anatomical and physiologic differences, such as poikilothermy and the presence of a shell, between mammals and chelonians may partially explain the observed pharmacokinetic differences. This is the first report of tulathromycin pharmacokinetics in a reptile, which limits the ability to make generalizations about the behavior of tulathromycin between these diverse phylogenetic groups. From a clinical perspective, the high Cmax and large AUC following tulathromycin administration are interesting in regards to the potential efficacy of tulathromycin. Any estimates of efficacy are speculative due to the lack of MIC of tulathromycin for pathogens commonly associated with URTD in desert tortoises.

The present study has a number of limitations. The interindividual variability is likely due to a combination of inherent properties of tulathromycin and study design. Despite all efforts to collect blood at exactly the nominal sampling times postdrug administration, blood collection directly from the jugular vein proved challenging. There were times where it was not possible to extend the head to facilitate jugular vein visualization, and for this reason, sample collection was allowed to occur up to 5 min before or after the designated collection time point. Therefore, blood was collected from tortoises at 0.16–0.33 h following tulathromycin administration which likely contributed to data variability at the 0.25 h time point. For population modeling of the data, the actual sampling times were used; however, this sampling scheme poses more of a problem for naïve pooled data analysis. When designing the study, even though we anticipated the jugular venipuncture to be a challenge, we still chose to collect blood directly from the jugular vein to avoid lymphatic contamination. Jugular vein catheterization was considered; however, heavy sedation or light anesthesia would have been required, and we sought to limit confounding variables. Furthermore, maintaining a jugular catheter for 10 days was determined to be impractical in this species. In future studies, phlebotomy from the brachial or caudal dorsal vein may provide a more reliable venous access point that is not as dependent on cooperation of the tortoise. Phlebotomy from the subcarapacial sinus could be performed; however, the possibility of lymphatic contamination would have to be considered.

To further understand the utility of tulathromycin in desert tortoises for treatment of upper respiratory tract disease, we have identified a number of future research directions. First, it is essential to establish tulathromycin MICs for M. agassizii. The MIC50 of tulathromycin has been established for Mycoplasma bovis (>64 μg/mL) and Mycoplasma hyopneumoniae.

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Mean (predicted curves)</th>
<th>Median (predicted curves)</th>
<th>Range (predicted curves)</th>
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<td>AUC0-∞ (h*μg/mL)</td>
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<td>636.3</td>
<td>278.6–8270.4</td>
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<td>0.6</td>
<td>0.4–27.6</td>
<td>0.25</td>
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</table>
(0.05–0.06 μg/mL) for bovine and porcine species, respectively (Godinho, 2008). The large difference in MIC90 for *Mycoplasma bovis* and *Mycoplasma hyopneumoniae* demonstrates caution that must be taken when attempting to extrapolate MICs between mycoplasma organisms. Second, as peripheral tissue concentrations of macrolide antibiotics are consistently higher than blood concentrations and plasma drug concentrations poorly represent peripheral tissue concentrations, evaluation of tulathromycin concentration in target tissues such as respiratory epithelium may prove to be useful (Williams & Sefton, 1993). In pigs, tulathromycin concentrations in the lungs were 24.9–181 times higher than those measured in plasma following intramuscular injection, which suggests plasma concentrations likely underestimate *in vivo* antibiosis (Benchaoui et al., 2004). In goats, high tulathromycin concentrations have been detected in the lung tissue 5 days after subcutaneous injection (Clothier et al., 2011; Clothier et al., 2012). The harvesting and homogenization of respiratory epithelium of desert tortoises antemortem was not possible in this population of animals, and euthanasia of healthy tortoises was deemed unethical. Efficacy trials to determine whether administration of tulathromycin results in clinical improvement should be performed in desert tortoise and are currently underway. Resolution of clinical signs has been demonstrated in some animals following tulathromycin dose at 5 mg/kg intramuscularly at the Desert Tortoise Conservation Center. Because URTD in desert tortoises is often a chronic condition and can involve more than one etiologic agent requiring prolonged treatment, the safety and efficacy of repeated dosing should also be explored. Anecdotally, 5 mg/kg intramuscular tulathromycin has been administered to an Indo-gangetic flap-shelled turtle (*Lissemys punctata andersonii*) every 7 days for four treatments with no clinical adverse effects or changes in complete blood count or plasma biochemical values, which suggests multidose treatment protocols may be clinically useful in a variety of chelonian species. Our results demonstrate intramuscular tulathromycin is detectable in plasma 10 days following injection, which suggests less frequent dosing may be appropriate, decreasing the amount of handling necessary and limiting the amount of stress to the patient and labor required of caregivers. A multidose pharmacokinetic study would need to be performed to determine a more specific dosing protocol and to determine if there are any adverse effects following repeated injections.

Although the time–concentration data were sparse due to the constraints of conducting a pharmacokinetic study in this reptilian species, the amount of information gleaned was maximized by a population pharmacokinetic approach to the data analysis. We were able to minimize the noise in the data by using actual rather than nominal sampling times, and multiple models (one- vs. two-compartment) were tested to find the best fit for the data, and we were able to estimate interindividual
variability. As a result, the model could accurately predict individual plasma drug concentration–time profiles, which is important for predicting the efficacy of different dosage regimens. Future work needed to build on this model includes collecting co-variate data (e.g. age, sex, body condition score) to explain some of the interindividual variability and establishing tulathromycin MICs that are specific for pathogens of this chelonian species.

This study represents the first step in evaluating the clinical utility of tulathromycin in desert tortoises for treatment of URTD and emphasized that population-based pharmacokinetic data modeling might offer advantages for studies with sparse sampling and where there is substantial variability in the data. The demonstrated efficacy against mycoplasma organisms in other species, lack of documented adverse effects in desert tortoises, and prolonged detectable plasma concentrations suggest tulathromycin should be considered as an alternative treatment modality for URTD in tortoises. The demonstrated efficacy against non-mycoplasma species such as Gram-negative bacteria and anecdotal use in multiple chelonian species suggests the utility of this antibiotic may not be limited to treatment of clinical mycoplasmosis and may be useful when morbidity due to a bacterial etiology is suspected in chelonians.

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REFERENCES


