Administration of Doxycycline Hydrochloride via Drinking Water to Turkeys Under Laboratory and Field Conditions

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ABSTRACT A series of experiments were carried out in order to determine doxycycline hydrochloride (DoxHCl) plasma levels in 6-wk-old turkeys medicated via drinking water containing DoxHCl at a concentration of 250 mg/L under laboratory and field conditions. Maximal plasma concentration (Cmax) values of 5.7 (± 1.0) µg/mL and 4.9 (± 1.4) µg/mL obtained after DoxHCl administration during 2 and 7 d, respectively, were not significantly different. A significant difference was found between the area under the plasma concentration-time profile, calculated between 0 and 168 h (AUC(0–168)), Cmax, and the minimal plasma concentration (Cmin) values obtained after medication with a DoxHCl solution at a concentration of 250 mg/L (431.9 ± 96.6 µg·h/mL, 4.9 ± 1.4 µg/mL and 0.7 ± 0.3 µg/mL) and after medication with a DoxHCl solution at a concentration of 750 mg/L (1,176.5 ± 201.8 µg·h/mL, 12.5 ± 2.7 µg/mL and 2.9 ± 0.4 µg/mL), respectively. The increase in body weight was also significantly higher for turkeys medicated with a DoxHCl solution at a concentration of 750 mg/L (83.7 g/d) than for the lower concentration (35.6 g/d). The DoxHCl solution uptake significantly decreased with the increase of DoxHCl concentration. A Cmax value of 1.7 ± 0.6 µg/mL and a Cmin value of 0.5 ± 0.1 µg/mL were observed during the field experiment. Water consumption under laboratory conditions was followed for tap water (70 ± 50 mL/kg·d) and for a DoxHCl solution at a concentration of 250 mg/L supplemented with 1 g anhydrous citric acid/L (119 ± 6 mL/kg·d) and revealed to be not significantly different. The variability was significantly higher for tap water than for the DoxHCl solution. The stability of the DoxHCl solution containing 1 g citric acid/L over 24 h was 99% expressed as the percentage of the initial concentration.

(Key words: turkey, doxycycline, bioavailability, drinking water, field)

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INTRODUCTION

Drugs of the tetracycline group are widely used in poultry as prophylactic and therapeutic agents (Flammer, 1985; Gylsdorff, 1987; Satalowich et al., 1993). Chlamydiosis (ornithosis) in turkeys is caused by Chlamydia psittaci, a highly specialized, Gram-negative, obligate intracellular bacterium (Vanrompay et al., 1994) and tetracyclines are still the treatment of choice against this microorganism (Gherman et al., 1995). Due to the bacteriostatic action, their effect depends on establishing and maintaining therapeutic blood or tissue levels during treatment. It is a current practice to administer these antibiotics via drinking water, but no literature is available on the drinking behavior of turkeys and the plasma concentrations obtained after administration of doxycycline hydrochloride (DoxHCl) via drinking water. Küng and Wanner (1994) reported on Dox plasma levels in turkeys housed under laboratory conditions and receiving Dox via drinking water (100 mg/L). In the present paper, a simulation of DoxHCl plasma levels during drinking water medication was performed based on drinking water consumption data and the pharmacokinetics of DoxHCl in turkeys (Santos et al., 1996b). The DoxHCl plasma levels in turkeys were determined during 2 and 7 d administration of DoxHCl via drinking water under laboratory conditions. These plasma levels were compared with DoxHCl plasma levels obtained after administration of medicated drinking water for 4 d under farm conditions.

MATERIALS AND METHODS

Animals

For the experiments conducted under laboratory conditions, 5-wk-old turkeys were used and fed PLV
pellets\textsuperscript{2} from 1 wk before the start of the experiment until completion of the study. The pellets contained no antimicrobial or anti-parasitic agents. The field experiment was conducted in a turkey farm\textsuperscript{3} housing 2,100 animals at the age of 6 wk weighing 1.4 ± 0.2 kg at the start of the experiment. During the experiment a total number of 90 turkeys were killed and blood samples were collected for analysis.

**Chemicals**

Doxycycline HCl and anhydrous citric acid were purchased from Alpha Pharma\textsuperscript{4} and Cerestar Bioproducts,\textsuperscript{5} respectively.

**Stability of DoxHCl Solution**

The stability of a DoxHCl solution in tap water containing 250 mg DoxHCl/L and 1 g/L anhydrous citric acid was determined at room temperature (22 °C). The pH and residual concentration were evaluated for 7 d. The stability was determined in drinking water containers made of plastic, glass, and metal, respectively. An additional stability experiment was performed in the presence of feces in the solution [1, 5, 10, and 20% (wt/vol)]. The stability of another DoxHCl solution in tap water containing 250 mg DoxHCl/L and 1,000 mg/L anhydrous citric acid kept at 30 °C was followed during 1 wk in glass containers.

**Water Consumption Behavior in 6-wk-old Turkeys**

Healthy turkeys (n = 9) weighing 1.6 ± 0.2 kg were housed together in a cage from the moment they were purchased at the age of 5 wk until completion of the study. Water and feed were available for ad libitum consumption throughout the experiment. A light cycle of 12 h was provided by means of a combination of natural and artificial light from 0700 to 1900 h. Complete darkness during the night was obtained by turning off the lights and by covering the cage with an opaque black plastic. All turkeys were assigned a number using black tape and their behavior was continuously followed by means of a video camera\textsuperscript{6} installed inside the cage and operating under infrared light. The video camera was connected to a video recorder placed in a separate room in order to enable changing of the video tapes without disturbing the animals. The drinking behavior was followed during 3 consecutive d for tap water (pH 7.65) and during 4 consecutive d for a DoxHCl solution (250 mg DoxHCl/L + 1 g anhydrous citric acid/L, pH 3.58) supplied in a plastic bucket. The solution was replaced daily and a sample was taken at Time 0 and after 24 h for the control of the DoxHCl stability. The turkeys were weighed at Time 0 and after 3 and 7 d. The average was calculated for tap water (1.7 ± 0.2 kg) and for the DoxHCl solution (1.9 ± 0.3 kg), respectively. The amount and frequency of water uptake were determined. For the determination of the amount consumed, results from previous experiments conducted under the same conditions were also considered. The temperature inside the cage was registered twice daily, at the start and at the end of each light period. The average temperature was 13.6 ± 2.6 °C (range 9 to 16 °C).

**Simulation Study**

The administration of drinking water medication to 6-wk-old turkeys was simulated over a 2-d period using a computer program.\textsuperscript{7} This simulation was done as a repeated administration of a DoxHCl solution for a total daily dose of 25 mg DoxHCl/kg BW, equally divided over 24 administrations (1.93 mg DoxHCl per administration) during the 12-h light period. The average weight for the 6-wk-old turkeys was assumed to be 1.85 kg and a normal daily water consumption of 100 mL/kg BW was adopted.

**Animal Experiments**

In a first experiment, 6-wk-old turkeys (n = 9) were housed together and received DoxHCl during 2 d via drinking water at a concentration of 250 mg DoxHCl/L supplemented with 1 g citric acid/L. The DoxHCl solution was replaced daily and a sample was taken at Time 0 and after 24 h for control of DoxHCl stability. The DoxHCl uptake was determined after each blood sampling time. The light cycle was kept at 12 h as described previously. Forty-eight hours after the start of the experiment, the DoxHCl solution was replaced by tap water. Blood samples were withdrawn from the wing vein before the start of the experiment (control) and at 3, 6, 9, 12, 20, 24, 27, 36, 44, 50, 60, and 72 h. All turkeys were weighed at Time 0 and at the end of the experiment and the overall average weight (1.7 ± 0.2 kg) was used to express water consumption. The average daily temperature recorded after each sampling time was 7.3 ± 1.4 °C (range 5 to 9 °C).

In a second study, 6-wk-old turkeys (n = 9) were housed together and received DoxHCl solution for 1 wk under the same conditions as described above. One hundred sixty 8 h after the start of the experiment, the DoxHCl solution was replaced by tap water. Blood samples were withdrawn from the wing vein before the start of the experiment and at 12, 24, 36, 48, 60, 72, 84, 96, 108, 120, 132, 144, 156, and 168 h. The turkeys were weighed at the start (1.6 ± 0.7 kg) and at the end of the experiment (1.8 ± 0.7 kg) and the overall mean weight was 1.7 ± 0.1 kg. The average daily temperature was 6.6 ± 0.9 °C (range 5 to 8 °C).

In a third study, 6-wk-old turkeys (n = 9) were housed together and had free access to a DoxHCl solution with a

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\textsuperscript{6}Avicom N.V., Zevergem, Belgium.
\textsuperscript{7}MaxSim version 3.01, Uppsala, Sweden.
concentration of 750 mg DoxHCl/L and 10 g citric acid/L during 1 wk. The DoxHCl solution was replaced daily and a sample was taken at time zero and after 24 h for control of DoxHCl stability. Blood sampling was performed as described for the second study. Turkeys were weighed at the start (1.3 ± 0.1 kg) and after completion of the study (1.9 ± 0.2 kg). The consumption of DoxHCl solution was determined daily and expressed as a function of the overall average BW (1.6 ± 0.3 kg). The average daily temperature was 9.0 ± 1.3 C (range 7 to 10 C).

A fourth study was conducted on a turkey farm with 2,100 6-wk-old healthy turkeys (1.4 ± 0.2 kg), housed in one garner. Medicated drinking water was given for a period of 4 d (36 g DoxHCl + 120 g anhydrous citric acid in 120 L of tap water, every 12 h) for a total daily dose of 25 mg DoxHCl/kg BW. The drinking water system consisted of individual drinking bowls connected via plastic piping to a plastic storage tank of 120 L. The level in the tank was kept constant by means of a floating-gauge. At 0700 and 1900 h the mixture of DoxHCl and anhydrous citric acid was added to the tank as a concentrated solution and homogenized. A light cycle of 24 h was maintained in the farm. Samples of the DoxHCl solution were taken at 0700 and 1900 h in the tank, and every 3 h between 0700 and 1900 h in the drinking bowls. Blood sampling was performed at the start of the experiment and at time 0, 6, and 12 h each day. On the 5th d, a blood sample was collected at 0700 h. For each sampling point, six turkeys were taken in a random way, weighed, and euthanatized. The ambient temperature was kept constant within a range of 20 to 23 C.

Sample Treatment and Analysis

For the stability determinations, samples of the DoxHCl solution were injected directly into the chromatographic system. For plasma level determination, 1 mL of blood was collected in sodium citrate tubes. The blood was centrifuged for 10 min at 2,000 rpm immediately after collection and the plasma was frozen at −20 C pending analysis. The DoxHCl concentration in plasma was determined after extraction using a validated HPLC method (Santos et al., 1996a) with demeclocyclineHCl as the internal standard. The chromatographic system consisted of an isocratic HPLC pump, a reversed phase column equipped with a precolumn and a variable wavelength ultraviolet detector. The mobile phase consisted of water:acetonitrile: 70% HClO4 [699:298.5:2.5 (vol/vol)], Na2EDTA (0.6 mmol/L), and oxalic acid (5 mmol/L). The pH was adjusted to 2.5 using 1 N NaOH.

The mobile phase was degassed before use. The flow rate was 1 mL/min and the analysis was performed at ambient temperature (22 C). The detection took place at a wavelength of 350 nm. Linear calibration curves (r2 > 0.99) were obtained in plasma between 0 and 600 µg/mL. Good recoveries for DoxHCl (> 66%) and demeclocycline (> 72%) were obtained in both water and plasma. The limit of detection was 0.1 µg/mL in plasma samples.

Statistical and Pharmacokinetical Analysis of Data

The pharmacokinetic parameters [AUC (area under the plasma concentration-time profile, calculated up to the last sampling point), Cmax (maximal plasma concentration), Cmin (minimal plasma concentration), and tmax (time at which the maximal plasma concentration is achieved)] were determined using a computer program. Integration and interpolation were performed using a mixed algorithm. Doses were expressed as a function of body weight.

The statistical analysis of data was performed using nonparametric statistics (Siegel and Castellan, 1988). Parametric statistics (F test) (Sokal and Rohlf, 1981) were used to compare the variance for the water consumption in turkeys receiving tap water with the water consumption in turkeys given medicated drinking water containing DoxHCl at a concentration of 250 mg DoxHCl/L. The influence of the DoxHCl dose on the weight gain and water consumption was evaluated using the Wilcoxon-Mann-Whitney test. The level of significance was set at 0.05.

RESULTS

Stability of the DoxHCl Solution

The presence of feces in the solution kept at room temperature (22 C) and the use of a metal receptacle might dramatically affect the DoxHCl stability (Table 1). Plastic and glass containers were revealed to have no influence on stability. Together with a decrease of the pharmaceutical availability of DoxHCl, an increase of the pH was observed.

The residual DoxHCl content of a DoxHCl solution (250 mg DoxHCl/L) containing 1 g citric acid/L and kept at 30 C was 98% after 48 h and 91% after 1 wk.

Tap Water and DoxHCl Solution Consumption by 6-wk-old Healthy Turkeys

The addition of 250 mg DoxHCl/L and 1 g anhydrous citric acid/L to tap water had no influence on drinking water consumption. The mean daily volume consumed and expressed per kilogram of BW was not significantly different for tap water (70 ± 50 mL/kg·d, range 7.5 to 138.6
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FIGURE 1. Simulation of the plasma concentration-time profile for a doxycycline hydrochloride (DoxHCl) administration via drinking water to 6-wk-old turkeys at a dose of 25 mg DoxHCl/kg body weight, over a 48-h period.

FIGURE 2. Mean plasma concentration-time profile (±SD) for a doxycycline hydrochloride (DoxHCl) administration via drinking water to 6-wk-old turkeys (n = 9). The light period was kept at 12 h and the average room temperature was 6.6 ± 0.9°C. The cumulative daily dose (milligrams per kilogram of BW) of DoxHCl consumed by the turkeys throughout the experiment is indicated (◊).

FIGURE 3. Mean plasma concentration time profiles (±SD) after 1 wk doxycycline hydrochloride (DoxHCl) administration via drinking water at a concentration of 250 mg DoxHCl/L (◊) and 750 mg DoxHCl/L (⁄) to 6-wk-old turkeys (n = 9). The light period was kept at 12 h. Mean daily amount of DoxHCl (milligrams per kilogram of BW) consumed by the turkeys receiving DoxHCl at a concentration of 250 mg DoxHCl/L (o) and 750 mg DoxHCl/L (∫) throughout the experiment.

DoxHCl Medication via Drinking Water

The simulated plasma concentration-time profile of DoxHCl for a repeated administration of a DoxHCl solution at a total daily dose of 25 mg DoxHCl/kg BW to 6-wk-old turkeys is shown in Figure 1. The maximal plasma concentration (Cmax) reached between 3 and 4 mg/mL both at 12 and 36 h, coinciding with the end of the light period each day. During the night, plasma levels dropped below 1 mg/mL.

The mean plasma concentration-time profile and the cumulative daily uptake of DoxHCl (milligrams per kilogram of BW) administered during 2 d via drinking water (250 mg DoxHCl/L) in laboratory conditions is shown in Figure 2. An overall Cmax value of 5.7 (±1.0) mg/mL was observed after 9 h in six turkeys and after 27 h in three turkeys, whereas a Cmin of 0.7 ± 0.4 mg/mL was observed after 24 h. The Cmin value corresponds with the end of the night period. During the light period of the 1st d (0 to 12 h), turkeys consumed a total of 111 mL of DoxHCl solution expressed per kilogram of BW, corresponding to a dose of 27.75 mg DoxHCl/kg BW. During the light period of the 2nd d (24 to 36 h), the value was similar, being 112 mL of DoxHCl solution/kg BW, which corresponds to a dose of 28 mg DoxHCl/kg BW.
TABLE 1. Effect of the presence of turkey feces in the medicated drinking water and of the bucket material on doxycycline hydrochloride (DoxHCl) stability after 1, 2, and 7 d at room temperature (22°C). The initial DoxHCl concentration was 250 μg/mL and the pH was 3.64.

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Feces concentration in solution (wt/vol)</th>
<th>Drining water container</th>
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<tbody>
<tr>
<td>pH</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Day 1</td>
<td>4.0</td>
<td>3.5</td>
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<tr>
<td>Day 2</td>
<td>4.0</td>
<td>3.5</td>
</tr>
<tr>
<td>Day 7</td>
<td>6.2</td>
<td>3.5</td>
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<tr>
<td>Residual DoxHCl, %</td>
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<td></td>
</tr>
<tr>
<td>Day 1</td>
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<tr>
<td>Day 7</td>
<td>54.7</td>
<td>100.0</td>
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</tbody>
</table>

obtained after DoxHCl administration at a concentration of 250 mg DoxHCl/L during 2 d (5.7 ± 1.0 μg/mL) was not significantly different from the C_max value of 4.9 ± 1.4 μg/mL obtained after administration of a similar DoxHCl solution during 1 wk. The administration of DoxHCl at a dose of 75 mg/kg showed values of 1,176.5 ± 201.8 μg·h/mL, 12.3 ± 2.7 μg/mL, 2.9 ± 0.4 μg/mL, and 81.0 ± 42.0 h, for AUC(0–168), C_max, C_min, and t_max, respectively (Table 2). All parameters were determined as the average of the individual values throughout the experiment. The mean daily amount of DoxHCl solution consumed per turkey was significantly higher (P < 0.001) for the 250 mg DoxHCl/L solution with 1 g anhydrous citric acid/L (117.2 ± 15.1 mL/kg) than for the 750 mg DoxHCl/L solution (86.0 ± 6.9 mL/kg). This results in a DoxHCl dose of 29.3 ± 3.8 mg/kg and 64.5 ± 5.2 mg/kg, respectively.

In the field experiment, DoxHCl plasma levels and the DoxHCl concentration in the drinking bowls were measured as a function of time (Figure 4). The initial DoxHCl concentration in the reservoir ranged from 92 to 347 μg/mL. This variation could be due to a poor homogenization of the solution or to a different water volume in the reservoir each time the DoxHCl was added. The maximal concentration measured in the drinking bowls ranged from 130 to 386 μg/mL (Figure 4). The DoxHCl concentration in the drinking bowls dropped to zero within 12 h.

The AUC(0–168) value for the mean Dox plasma concentration-time profile was 107.8 μg·h/mL. This value might be underestimated due to the lack of sampling points during the second period of the cycle (1900 to 0700 h). An average maximal plasma concentration (C_max) of 1.7 ± 0.6 μg/mL (range 0.9 to 2.6 μg/mL) was obtained 54 h after the start of the administration. During the administration period a C_min value of 0.5 ± 0.1 μg/mL (range 0.3 to 0.6 μg/mL) was observed after 36 h (Figure 4).

DISCUSSION

The success of a medication depends among others on factors such as sensitivity of the pathogen to antimicrobials used, the pharmacokinetic behavior of the drug, i.e., the rate and extent of absorption, the drug concentration obtained at the infection site, and the residence time of the drug in tissues. The route of drug administration and the formulation can play an important role in the effectiveness of the treatment. Because sick birds eat less but generally still drink, it is advisable, for oral medication, to administer drugs via drinking water. We analyzed the influence of formulation parameters and drinking water uptake on the plasma levels of DoxHCl in turkeys under laboratory and field conditions.

The addition of 0.1 and 1% of anhydrous citric acid to the formulation was necessary for the stability and solubility of DoxHCl in tap water at a concentration of 250 mg DoxHCl/L and 750 mg DoxHCl/L, respectively. The DoxHCl added to tap water without citric acid precipitated. This effect is in agreement with the findings reported by Dorrestein et al. (1981). It was also observed that the palatability of a DoxHCl solution containing citric acid was good, as water consumption did not change significantly for medicated water. The
material of the drinking system and the conditions of hygiene in the pen can play an important role on the stability of the DoxHCl solution.

The maximum DoxHCl plasma concentrations found after drinking water medication at a concentration of 250 mg DoxHCl/L, during 2 (C_max 5.7 ± 1.0 µg/mL) and 7 d (C_max 4.9 ± 1.4 µg/mL) were not significantly different. The C_max values were in both cases higher than the values found for the simulation calculation. This result could be explained by a possible positive influence of citric acid on the intestinal absorption of Dox in poultry as was reported for other tetracyclines in chickens (Pollet et al., 1993) and in turkeys (Pollet and Glatz, 1984). This aspect was not taken into account for the determination of the pharmacokinetic parameters used for the simulation (Santos et al., 1996b). The average minimum plasma concentration reached during a 4-d DoxHCl administration via drinking water at a dose of 25 mg/kg BW in 6-wk-old turkeys kept in a pen was 1.7 ± 0.6 µg/mL. Plasma levels observed in the field experiment were lower than those obtained in the laboratory experiment. This result could be explained by the drinking water system with a continuous adjustment to the water level. As a consequence, the DoxHCl concentration in the reservoir and in the drinking bowls continuously decreased. It has to be emphasized that the field experiment was conducted with a 24-h light cycle. This lighting method means that the turkeys could drink over a 24-h period, whereas in the laboratory experiments the drinking period was limited to 12 h. Taking into account the dilution and the light cycle, the total daily dose of Dox (25 mg/kg BW) was provided in twice 12.5 mg/kg BW to obtain sufficient high plasma levels over 24 h. In our experiments, the C_max values were at least four times higher than the value 0.4 µg/mL reported for 12-wk-old turkeys medicated with Dox via drinking water at a concentration of 100 mg/L during 4 d (Kung and Wanner, 1994). These authors administered a 2.5 lower Dox concentration than that tried in our studies and they used 12-wk-old turkeys. The absolute bioavailability of DoxHCl in 12-wk-old turkeys (25 ± 9%) was reported to be significantly lower than in 6-wk-old turkeys (37 ± 7.5%) (Santos et al., 1996b). It is likely that the use of citric acid in our formulation could also contribute to the higher plasma levels found in our studies in comparison with those reported by Kung and Wanner (1994). The addition of citric acid to improve the oral absorption of other tetracyclines, such as chlorotetracycline (CTC), has been reported in turkeys (Pollet and Glatz, 1984) and chickens (Pollet et al., 1983), Dorrestein et al. (1984) reported the effect of calcium ions present in the small intestine on the pharmacokinetics of Dox in pigeons. These authors suggested that citric acid might improve Dox absorption from the gastrointestinal tract but no further reports have been published on this aspect. Citric acid could also improve the palatability of the DoxHCl solution with an increase in the water uptake. The minimum plasma levels found for a dose of 25 mg/kg BW, 0.7 µg/mL for the experiments conducted under laboratory conditions and 0.5 mg/mL for the field experiment, are clearly above the value of 0.03 and 0.06 µg/mL reported as the minimum inhibitory concentration (MIC) of Dox on 12 strains of Chlamydia psittaci (Henning and Krauss, 1986) and Strain X-73 of Pasteurella multocida (George et al., 1977), respectively. Recent studies on 12 Belgian isolates of C. psittaci showed a MIC value ranging from 0.05 to 0.2 µg/mL (P. Butaye, 1996, Department of Avian Medicine and Pathology and Laboratory for Veterinary Bacteriology and Mycology, Salisburylaan, 133, 9820 Merelbeke, Belgium, personal communication). With respect to the comparison of MIC values and plasma levels, one has to take into account that only the free drug is available for antimicrobial activity. In the case of Dox, 90% protein binding has been observed in turkey plasma (Santos et al., 1996b). Considering that only 10% of the determined concentration of DoxHCl is free, our C_min values expressed as free DoxHCl are 0.07 µg/mL (0.06 µg/mL expressed as free Dox base) and 0.05 µg/mL (0.04 µg/mL, expressed as free Dox base) for the DoxHCl administration at a dose of 25 mg/kg BW during 1 wk in the laboratory and during 4 d in the field, respectively. Both values are still above the MIC for C. psittaci reported in literature (Henning and Krauss, 1986), but under the recent MIC values reported for this antibiotic on C. psittaci. Doxycycline is known to have a high distribution volume but also high tissue binding. This high volume could result in a smaller fraction of free

### Table 2. Pharmacokinetic parameters (mean ± SD, range) for a doxycycline hydrochloride (DoxHCl) administration via drinking water during 1 wk at a concentration of 25 mg DoxHCl/L supplemented with 1 g citric acid/L and 750 mg DoxHCl/L supplemented with 10 g citric acid/L to 6-wk-old turkeys

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Mean ± SD</th>
<th>Range</th>
<th>Mean ± SD</th>
<th>Range</th>
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<tbody>
<tr>
<td>AUC(0–168) µg·h/mL</td>
<td>431.9 ± 96.6</td>
<td>275.7 to 526.6</td>
<td>1,176.5 ± 201.8</td>
<td>986.5 to 1,612.1</td>
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<tr>
<td>C_max µg/mL</td>
<td>4.9 ± 1.4</td>
<td>2.7 to 6.9</td>
<td>12.3 ± 2.7</td>
<td>9.7 to 17.6</td>
</tr>
<tr>
<td>C_min µg/mL</td>
<td>0.7 ± 0.3</td>
<td>0.3 to 1.2</td>
<td>2.9 ± 0.4</td>
<td>2.1 to 3.3</td>
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<tr>
<td>t_max h</td>
<td>72.0 ± 42.0</td>
<td>12.0 to 156.0</td>
<td>81.0 ± 42.0</td>
<td>12.0 to 156.0</td>
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<td>Dox solution consumption, (mL/d·kg BW)</td>
<td>117.2 ± 15.1</td>
<td>92 to 130</td>
<td>86.0 ± 6.9</td>
<td>75.0 to 96.1</td>
</tr>
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1 AUC(0–168) = area under the plasma concentration-time profile, calculated between 0 and 168 h; C_max = maximal plasma concentration; C_min = minimal plasma concentration; t_max = time at which maximal plasma concentration is achieved.
Dox at the site of action. Nevertheless, the studies reported thus far do not include tissue concentrations of Dox, which may be a more important indicator of potential therapeutic effect than the plasma concentration.

A threefold increase of the DoxHCl concentration induced a significant decrease ($P < 0.001$) of the water uptake and this resulted in a 2.2 times higher dose. Proportional increases of $AUC_{(0-168)}$ and $C_{\text{max}}$ values were seen. The $C_{\text{min}}$ value ($2.9 \pm 0.4 \mu g/mL$) was four times higher than that observed with DoxHCl solution at a concentration of 250 mg DoxHCl/L ($0.7 \pm 0.3 \mu g/mL$). The mean weight gain after 1 wk treatment with medicated drinking water at a concentration of 750 mg DoxHCl/L was significantly higher ($P < 0.0001$) than the mean weight gain after the treatment with medicated drinking water at a concentration of 250 mg DoxHCl/L during the same period of time. Similar findings were reported by George et al. (1977) on young chickens medicated with Dox 50 ppm (base). This result is not in agreement with the findings reported by Kung and Wanner (1994); however, those experiments were conducted in adult turkeys (12-wk-old) and for a shorter period of time (4 d).

It can be concluded that the administration of DoxHCl via medicated drinking water to turkeys kept under laboratory conditions yielded higher plasma levels than the same treatment carried out under farming conditions. This result suggests the strong need for a validation of the drinking water distribution system, the water uptake, the light cycle, the temperature, etc., in order to assure an effective therapeutic treatment in the field. Considering the MIC values described in the literature for Dox on $C.\ psittaci$, the medication of turkeys kept under the field conditions reported in this work, with a doxycycline solution at a concentration of 250 mg DoxHCl/L might be insufficient to reach the plasma levels required for a successful therapy against this microorganism.

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