



Original Article

Disposition of posaconazole after single oral administration in large falcons (*Falco spp*): Effect of meal and dosage and a non-compartmental model to predict effective dosage

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Abstract

Posaconazole has been used anecdotally to treat aspergillosis in falcons resistant to voriconazole. In human medicine, it is used prophylactically in immunosuppressed human subjects with invasive pulmonary aspergillosis. So far, no studies have been performed in birds. The aim of this study was to evaluate the *in-vivo* pharmacokinetic behavior of oral posaconazole after a single administration in six large falcons (i.e gyrfalcons, saker falcons). Posaconazole oral suspension (Noxafil, 40 mg/ml, Schering-Plough) was administered *per os* without meal in a single dosage of 12.5 mg/kg in 3 falcons. A comparison was done in two more falcons, one with a natural fatty meal at the same single dose, and one with a natural fatty meal and a higher dosage (20 mg/kg). Finally, six falcons received posaconazole pre-dissolved in corn oil with a natural low-fat meal in the higher single dose (20 mg/kg). No side effects were observed in the falcons in any of the experiments. In starved state posaconazole was poorly absorbed, more so than in other species. As expected, absorption of posaconazole was higher with the administration of meal or in the presence of plant (corn) oil, with a fourfold increase in apparent bioavailability. Despite the preferential absorption in the presence of fat, for both dosing schemes the AUC₂₄:MIC ratio was lower than described in human medicine to achieve a therapeutic effect. The AUC_{inf}:MIC which is an indicator of efficacy after steady-state, while variable, did indicate that the drug is worth trying when susceptibility testing shows to be the only effective drug.

Lay Abstract

The focus of this work is to determine the pharmacokinetic parameters of oral posaconazole in large falcons for the first time after a single dose. Posaconazole has higher bioavailability when administered with meal and fatty components. No adverse reactions have been observed. The ratio of the area under the curve (AUC₂₄) to minimum inhibitory concentration was lower compared to the therapeutic level in human.

Key words: posaconazole, oral administration, falcon, meal influence, non-compartmental model, aspergillosis.

Introduction

Aspergillosis is a well-described, ubiquitous, invasive, fungal infection affecting human, mammals and birds.¹ Various

reviews have summarized the impact of the disease in birds, the predisposing factors, the diagnostic, and treatment options.^{1–7} Aspergillosis is one of the most prominent causes of morbidity

and mortality in falcons in the Middle East. Clinical signs in falcons with aspergillosis are often non-specific, early detection of the disease is still challenging and the development of novel diagnostic methods is an active field of research.^{5,8,9} Antifungal resistance has been described^{10,11} and clinically studied via culture and sensitivity testing,^{12,13} but recent research has focused in genotyping and molecular identification of *Aspergillus* species and strains and their resistance in common antifungals.^{14–19}

Posaconazole (PSC) is a triazole, belonging to the family of azoles, which are lanosterol 14 α -demethylase inhibitors. Azoles inhibit the enzyme cytochrome P450-dependent 14 α -sterol demethylase, which is required for the conversion of lanosterol to ergosterol. Exposed fungi become depleted of ergosterol and accumulate 14 α -methylated sterols. This action causes disruption of membrane structure and function, thereby inhibiting fungal growth.²⁰ Posaconazole is available in the market in the form of oral suspension, delayed release tablets and intravenous solution. In humans, posaconazole has been used prophylactically for invasive candidiasis and immunocompromised patients with aspergillosis, as well as for the treatment of oral candidiasis, invasive aspergillosis, zygomycosis and *Fusarium* infections in case other antifungals prove resistant.²⁰ In veterinary medicine, there are few pharmacokinetic studies^{21–23} but abundant data exists for experimental animals (mice, rats and rabbits) that have been used as experimental model for invasive aspergillosis and drug efficacy for human.^{24–29} Posaconazole has never been used in avian medicine broadly and so far, no detailed pharmacokinetic studies exist in avian species. Dubai Falcon Hospital (DFH) previously tried an empirical use in falcons for the treatment of resistant aspergillosis and candidiasis. The aim of this study was to determine the *in-vivo* pharmacokinetic behavior of the oral posaconazole suspension after a single oral administration and evaluate the effect of the dosage and the meal in large falcons. Further aim was to develop a pharmacokinetic model to predict the optimal AUC₂₄:MIC and define the dosage against *Aspergillus* spp.

Methods

Six large, healthy, captive-bred falcons (two male Gyrfalcons, three female saker falcons, 1 male saker falcon) were included in the study after anamnesis, physical examination, radiology, hematology, biochemistry, electrophoresis, bacteriology, parasitology, and endoscopy. Falcons with an indication of disease and especially aspergillosis were excluded. The trained falconry falcons were housed indoors during the experiment (48 h) in DFH. Their age was 2–3 years old and their average weight 1000 \pm 200 g. During the experiments, the falcons were kept on a block with artificial grass, in a 2 m \times 3 m air-conditioned room, which was cleaned daily. Birds received daily adequate food (quail, pigeon) and free access to water. Stressors were minimized (quiet rooms, hood, dimmed light, same caretaker). The falcons were inspected daily by experienced personnel and an avian

specialist for signs of illness and discomfort. If there was any indication of health compromise, individual falcons were excluded from the experiments. The study was approved by the DFH direction and the ethical committee of the American University of Sharjah, United Arab Emirates (UAE).

For the experiment, posaconazole (PSC) in the form of oral suspension (Noxafil, 40 mg/ml, Schering-Plough®) was used. For the first pilot pharmacokinetic data, three falcons (1 Gyrfalcon, 2 female Saker falcons) were gavaged, using a metal crop tube, with oral PSC (12.5 mg/kg) without meal and fasted. The selected dose was 12.5 mg/kg as this has been used empirically in UAE and extrapolated from the oral dose of voriconazole in falcons.³⁰ To allow for the further evaluation of meal and dosage, with the minimal sampling in a second pilot, one falcon previously gavaged, using a metal crop tube, with PSC (12.5 mg/kg) alone was forced fed natural fatty meal (80 g beef meat/20 g of beef fat). A second falcon was gavaged, using a metal crop tube, with a high dose of PSC (20 mg/kg) and forced fed natural fatty meal (80 g beef meat/20 g beef fat). For the pivotal pharmacokinetic study, six falcons were gavaged, using a metal crop tube, with prediluted PSC (20 mg/kg) with plant (corn) oil (1 : 1) and then fed low fat meal (eviscerated quail).

In addition to obtaining basic pharmacokinetic information, the other rationale for the early pharmacokinetic studies was to evaluate the effect of a high fat meal in comparison to the fasted state at the same dose of 12.5 mg/kg (Saker falcon 1), and the impact of PSC at a higher dose (20 mg/kg) with a high-fat meal. The main pharmacokinetic study finally simulated common practices of feeding falcons with quail (low fat meal) and enhancing posaconazole activity by having it pre-diluted in plant (corn) oil. The rationale is to prevent the recovering animals from accumulating fat as these birds are speed athletes. Based on the results of the pilot study, the main study used the higher dose of 20 mg/kg.

For the blood sampling, the falcons were manually restraint in a special raptor jacket, while the leg was extended. Blood was taken by the medial metatarsal vein of the left or the right leg. For each falcon, 9 bleeding points were selected namely before injection and, at 0.5, 1, 2, 4, 6, 8, 12 and 24 h after injection. For each time point, 0.5 ml plasma was drawn from the medial metatarsal veins after manual restraint and directly poured in a pediatric lithium-heparin container (MiniCollect CE®). Plasma was immediately centrifuged after collection and frozen at -40° Celsius until laboratory analysis. The sample collected before oral administration served as control that no drug was already present in the falcons. The posaconazole in the plasma was detected and quantified by high performance liquid chromatography (HPLC), Nexera-i, LC-2040C 3D with LC-2030/2040 PDA Detector, Shimadzu Scientific Instruments, Japan. A volume of 500 μ l of methanol was added to 200–300 μ l of plasma and the sample was mixed on a vortex mixer for 1 min. Then 500 μ l of acetonitrile was added, the sample was mixed on a vortex mixer for 1 min and centrifuged at 6000 rpm for 5 min. The

Table 1. Relevant pharmacokinetic parameters presented as geometric means from the pilot study in which three falcons (one Gyrfalcon and two Saker falcons) received a single oral administration of posaconazole at a dose 12.50 mg/kg in the absence of a fatty meal and in the fasted state.

Parameter	Units	Gyrfalcon ID 18390	Saker ID 18487	Saker ID 18465	GMean 18447.29	SEM 29.36
C _{max}	ug/ml	0.23	0.21	0.25	0.23	0.01
T _{max}	H	6.00	4.00	6.00	5.24	0.67
AUC _{last}	ug/ml * h	2.06	1.36	3.18	2.07	0.53
AUC _{extra}	ug/ml * h	0.89	0.78	0.89	0.85	0.04
AUC _{inf}	ug/ml * h	2.95	2.13	4.07	2.95	0.56
%AUC _{extra}	%	30.12	36.52	21.91	28.89	4.23
L _z	h-1	0.13	0.09	0.07	0.09	0.02
AUM _{last}	ug/ml * h ²	12.28	7.68	32.51	14.53	7.63
thalf	h	5.47	7.72	9.47	7.37	1.16
MRT	h	10.16	12.05	16.24	12.57	1.80
Cl/F	l/h * Kg	4.24	5.86	3.07	4.24	0.81
V _z /F	l/Kg	33.46	65.26	41.97	45.09	9.50
V _{ss} /F	l/Kg	43.07	70.57	49.90	53.33	8.27

C_{max} - maximum plasma concentration; T_{max} - Time to maximum concentration; AUC_{last} - Area under the curve to the last time point; AUC_{extra} - Area under the curve for the extrapolated curve from the last time point to infinity; AUC_{inf} - Area under the infinity curve; %AUC_{extra} - % of the area under infinity curve made up of the extrapolated portion of the curve; L_z - Elimination constant; AUM_{last} - Area under the moment curve; thalf - Half-life of elimination; MRT - Mean residence time; Cl/F - plasma clearance; V_z/F - Apparent volume of distribution; V_{ss}/F - Volume of distribution at steady state.

supernatant was filtered through at 0.2 μm PTFE syringe filter (MAS scientific, China). The sample was diluted 1 : 1 with the mobile phase and 50 μl of sample was injected into the HPLC system. The mobile phase was 55 : 45 (v/v) acetonitrile-water at a flow rate of 0.2 ml/min on a RESTEK, Ultra C18, 5 μm, 2.1 × 250 mm (Agilent Technologies, USA) and detection was by UV absorption at 260 nm. Calibration standards were 0.1, 0.25, 0.5, 1.0, 5.0, and 10.0 μg/ml (R² = 0.9968) and control samples were 1.0, 2.0, 5.0, and 10.0 μg/ml. The assay limit of detection and quantification were 0.14 and 0.43 μg/ml, respectively. Average recoveries of PSC from control samples were 94.0%.

Statistical and pharmacokinetic analysis

All pharmacokinetic parameters were calculated with Kinetica 5.1 (Thermo) using a non-compartmental model. The maximum plasma concentration (C_{max}) and the time to maximum concentration (T_{max}) were read directly of the concentration versus time plasma profile. The area under curve to the last quantifiable time point (AUC_{last}) was determined using the linear trapezoidal rule. The total area under curve extrapolated to infinity (AUC_{inf}) was calculated as AUC_{inf} = AUC_{last} + AUC_{ext} = AUC_{last} + C_{Last}/λ with C_{Last} being the computed last measured concentration and λ being the terminal elimination rate constant. The area under the moment curve from the time point zero to the last measured time point (AUM_{last}) was also calculated using the linear trapezoidal rule. The half-life (t_{1/2}), clearance (Cl/F) and volume of distribution during terminal phase (V_z/F) and volume of distribution at steady state (V_{ss}/F) and the mean residence time (MRT) was determined as t_{1/2} = ln(2)/λ; V_z = Cl/λ =

Dose/(AUC_{tot} * λ); V_{ss} = (Dose * MRT)/AUC, Cl = dose/AUC_{tot} and MRT = AUM_{tot}/AUC_{tot}. For efficacy, the model suggested for human was used.³¹ In the model, efficacy is based on the ratio of AUC₂₄ to MIC of 163.

Results

Following treatment, none of the six falcons showed any adverse effects due to handling or the gavage of posaconazole or fed meal. Minor hematomas around the medial metatarsal veins were treated with local ointment (Hirudoid®) for 7 days. All falcons had normal appetite, feces, and casts after the experiment. All plasma samples were within normal range (color, viscosity). In case the time point was not accurate, the actual time that the sample was taken was recorded in an accuracy of 1 min, to be taken into consideration for the statistical analysis.

Following the pilot study, the three falcons (1 Gyrfalcon, 2 female Saker falcons) treated with oral PSC (12.5 mg/kg) in the fasted state achieved very low maximum concentrations of 0.23 ± 0.01 ug/m in 5.24 ± 0.67 h (Table 1). The area under curve was also low at 2.07 ± 0.53 ug/ml * h and the half-life of elimination was rapid at 7.37 ± 1.16 h. Despite monitoring the pharmacokinetic profile over a 24-h period, the evaluation was only able to capture 72% of the drugs' concentration versus time profile. In both species of falcon, the profile was characterized by a biphasic absorptive profile (Figure 1). PSC plasma concentrations also failed to reach even the low MIC₂₅ reported for *Aspergillus* spp.

Following treatment for the fat portion of the pilot study, the bioavailability of 12.5 mg/kg dose increased fourfold, with

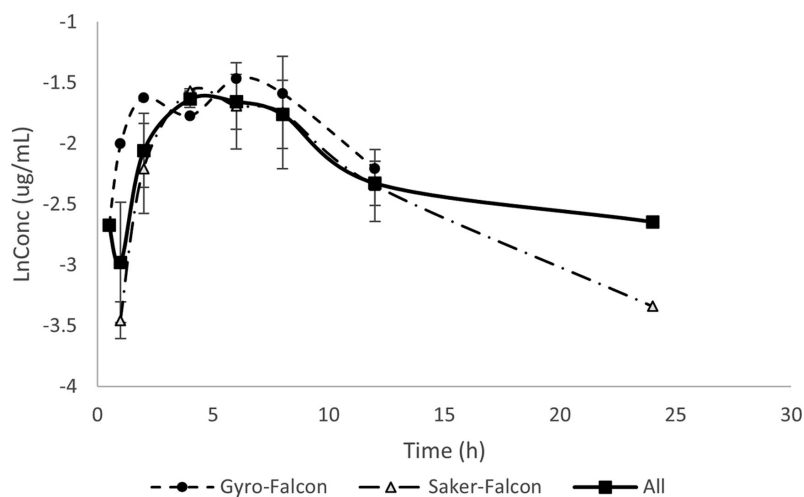


Figure 1. Plasma concentration versus time profile for the main pharmacokinetic study following the treatment of 3 falcons (1 Gyr and 2 Saker falcons) with posaconazole at 12.5 mg/kg in the fasted state on the natural logarithmic scale monitored over 24 h. The posaconazole was administered in the absence of a fatty meal or with oil.

Table 2. Relevant pharmacokinetic parameters from the pilot study in which two Saker falcons were exposed to a high fat meal in the presence of a dose of posaconazole administered *per os* either at 12.50 or 20 mg/kg. Also presented in the relevant parameters exploring the extent of exposure for the 20 mg/kg dose scaled to 12.5 mg/kg.

Parameter	Unit	ID 18465		ID 18487		Scaled
		Fasted	Fatty	Fasted	Fatty	
Dose	mg/kg	12.5	12.5	12.5	20	12.5
C_{max}	$\mu\text{g/ml}$	0.25	0.73	0.21	0.99	0.62
T_{max}	h	6.00	8.00	4.00	12.00	
AUClast	$\mu\text{g/ml} \cdot \text{h}$	3.18	13.87	1.36	15.10	9.44
AUCextra	$\mu\text{g/ml} \cdot \text{h}$	0.89	13.62	0.78	8.42	5.26
AUCinf	$\mu\text{g/ml} \cdot \text{h}$	4.07	27.49	2.13	23.53	14.70
%AUCextra		21.91	49.54	36.52	35.80	22.38
Lz	1/h	0.07	0.03	0.09	0.06	
AUMClast	$\mu\text{g/ml} \cdot (\text{h})^2$	32.51	165.73	7.68	194.58	121.61
thalf	h	9.47	20.91	7.72	11.86	
MRT	h	16.24	32.86	12.05	22.99	
Cl/F	L/h * kg	3.07	0.45	5.86	0.85	
Vz/F	L/kg	41.97	13.71	65.26	14.55	
Vss/F	L/kg	49.90	14.94	70.57	19.54	

C_{max} - maximum plasma concentration; T_{max} - Time to maximum concentration; AUClast - Area under the curve to the last time point; AUCextra - Area under the curve for the extrapolated curve from the last time point to infinity; AUCinf - Area under the infinity curve; %AUCextra - % of the area under infinity curve made up of the extrapolated portion of the curve; Lz - Elimination constant; AUMClast - Area under the moment curve; thalf - Half-life of elimination; MRT - Mean residence time; Cl/F - plasma clearance; Vz/F - Apparent volume of distribution; Vss/F - Volume of distribution at steady state. The relevant parameters for 20 mg/kg dose scaled to 12.5 mg/kg.

C_{max} reaching the MIC_{50} for *Aspergillus spp* (Table 2). In an unexpected manner, the administration of the increased dose of 20 mg/kg dose did not substantially increase the extent of exposure above that for the 12.5 mg/kg dose. When the 20 mg/kg dose was scaled to 12.5 mg/kg, it becomes evident that lower concentration achieved for the higher 20 mg/kg dose was due to limitation in the absorptive process (Figure 2). For both doses, the presence of the high fat meal resulted in a longer half-life of elimination and a slightly longer time to maximum concentration akin to a depot-like effect.

In the pivotal pharmacokinetic study, six falcons were gavage, using a metal crop tube, with prediluted PSC (20 mg/kg) with plant (corn) oil (1 : 1) and then fed low fat meal (eviscerated quail). The study was able to confirm the results from the pilot study. The applicable pharmacokinetic parameters are presented in Table 3 and illustrated in Figure 3. As expected, the oil enhanced the bioavailability of posaconazole with the C_{max} being reached 4–5 h after administration at a concentration above the MIC_{50} for 24 h. The oil formulation once again resulted in the prolonged rate of absorption and a depot type of effect,

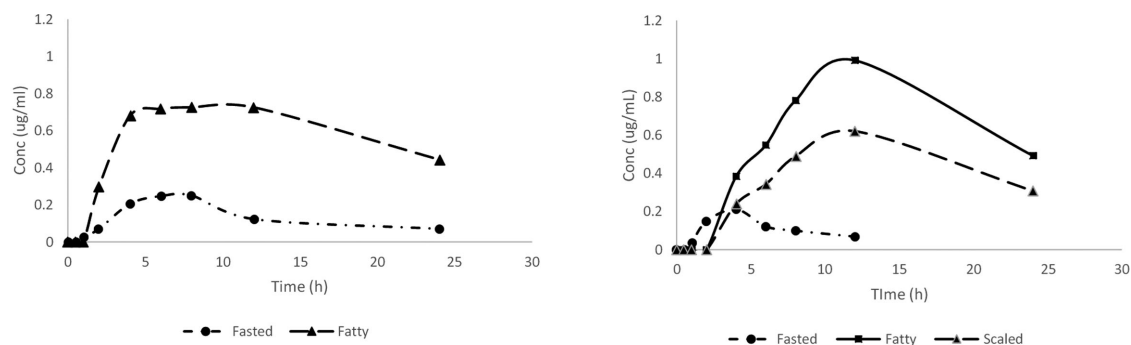


Figure 2. Plasma concentration versus time profile for two Saker falcons with Bird 1 (left) treated with posaconazole at 12.5 mg/kg in the fasted state or the equivalent dose administered with a fatty meal and the bird 2 (right) treated with posaconazole at 12.5 mg/kg in the fasted state or 20 mg/kg in the presence of a fatty meal, also illustrated is the administered 20mg/kg dose corrected to 12.5 mg/kg to allow for comparison between the birds and doses. Results are presented on the natural logarithmic scale.

Table 3. Relevant pharmacokinetic parameters from the pivotal pharmacokinetic study and the geometric means following a single oral administration of posaconazole at a dose of 20 mg/kg in four Saker and two Gyrfalcons in equal ratios with plant oil by oral gavage (*per os* with crop tube).

Parameter	Unit	Saker				Gyrfalcon		Gmean	SEM
		18487	18486	18465	18288	16228	18389		
C_{max}	µg/ml	0.54	0.98	0.75	0.92	0.88	0.61	0.76	0.07
T_{max}	h	8.00	8.00	6.00	12.00	12.00	8.00	8.73	1.00
T_{lag}	h	1.00		0.50	2.00	2.00	2.00	1.32	0.29
AUC _{last}	µg/ml * h	8.83	15.65	10.63	15.12	14.52	10.68	12.29	1.17
AUC _{extra}	µg/ml * h	5.57	7.80	4.66	35.16	331.71	40.41	21.37	52.56
AUC _{inf}	µg/ml * h	14.40	23.45	15.29	50.28	346.23	51.09	40.77	52.98
%AUC _{extra}		38.66	33.26	30.45	69.94	95.81	79.10	52.42	11.20
L_z	1/h	0.05	0.05	0.06	0.02	0.00	0.01	0.02	0.01
AUM _{Clast}	µg/ml * (h) ²	109.11	181.71	118.49	219.11	225.32	149.40	160.86	20.29
thalf	h	14.65	13.02	12.49	34.00	269.42	56.58	32.75	41.15
MRT	h	25.02	21.98	20.54	55.45	396.04	86.47	52.71	59.95
Cl/F	L/h * kg	1.39	0.85	1.31	0.40	0.06	0.39	0.49	0.22
V_z	L/kg	29.35	16.02	23.57	19.51	22.45	31.95	23.18	2.44
V_{ss}	L/kg	34.76	18.75	26.87	22.06	22.88	33.85	25.86	2.68
AUC ₂₄ : MIC		17.67	31.30	21.27	30.23	29.04	21.36	24.58	2.34

C_{max} - maximum plasma concentration; T_{max} - Time to maximum concentration; AUC_{last} - Area under the curve to the last time point; AUC_{extra} - Area under the curve for the extrapolated curve from the last time point to infinity; AUC_{inf} - Area under the infinity curve; %AUC_{extra} - % of the area under infinity curve made up of the extrapolated portion of the curve; L_z - Elimination constant; AUM_{Clast} - Area under the moment curve; thalf - Half-life of elimination; MRT - Mean residence time; Cl/F - plasma clearance; V_z /F - Apparent volume of distribution; V_{ss} /F - Volume of distribution at steady state.

as evident by the plateau like profile after C_{max} was reached. The latter was also more pronounced than for the pilot study, where the dose when administered with a naturally fatty meal, to such an extent that in some cases the half-life was substantially increased and not properly quantifiable. While the mean half-life of elimination in the main study was prolonged compared to the earlier studies, for the bird included in both phases (Bird 18465) the half-life at 20 mg/kg was shorter than when the bird was dosed at 12.5 mg/kg with a fatty meal. Unfortunately, while we were not able to follow the full profile for the curve, the results over the 24-h monitoring period provide some insight into the efficacy of the product, which was the main aim

of the study i.e., to determine if posaconazole could be effective in treatment of *Aspergillus* through PK/PD modeling. Using the MIC reported for the various strains of *aspergillus* the average ratio of AUC₂₄ : MIC varied from 50 to 100 for the listed strains, which was below the 167 required for efficacy. However the ratio of the AUC_{inf} : MIC for the same strains varied from 163 to 339.

Discussion

Posaconazole is one of the newer antifungal agents introduced on the market.³⁰⁻³² The drug is registered for use in people for

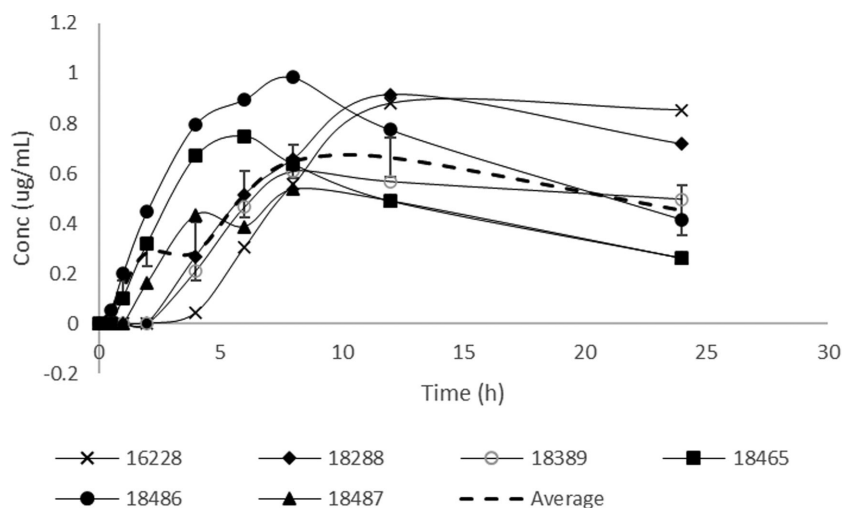


Figure 3. Plasma concentration versus time profile for the main pharmacokinetic study following treatment of 6 falcons with posaconazole at 20 mg/kg in the fasted state on the natural logarithmic scale monitored over 24 h. The posaconazole was predissolved in corn oil at a ratio of 1 : 1 and administered *per os*.

the management of systemic fungal infections especially in the presence of immune suppression. The drug is characterized by a high lipid solubility which as a result needs to be administered either in smaller dose fairly frequently or with a high fat meal to achieve optimal bioavailability, of which the latter is recommended means of administration in people. Also, as a result of its lipid solubility, posaconazole has a large volume of distribution and good penetration into tissue fluid. The drug enjoys excellent penetration into the lungs, and it has been reported to achieve alveolar concentration approximately twice to four-fold higher than that of the plasma.²⁹ Based on the dose-dependent decline in galactomannan concentration, it was proposed that the best surrogate for efficacy was $AUC_{24} : MIC$, with a ratio of 167 being associated with the half-maximal antifungal effect.^{24,27}

For this study, we were interested in determining if the drug could be effective in the treatment of falcons, which as sporting animals are routinely hospitalized with respiratory-related infections. From the pilot study, it was evident that posaconazole showed extremely poor absorption when administered in the absence of food or a fatty meal as seen for other species. However, in comparison, the C_{max} achieved in this study at 0.23 ug/ml was substantially lower than that for the dog also following the administration of an oral suspension. In dogs treated at 6 mg/kg (half the dose for this study), the C_{max} was 0.42 ug/ml.²¹ The same was evident from humans, where a dose of 200 mg (average weight of 77 kg = 2.5 mg/kg) achieved a C_{max} of 0.094.6 $\mu\text{g/ml}$ in the fasted stated (scaled to 0.47 $\mu\text{g/ml}$ for 12.5 mg/kg dose).³² This leads to the conclusion that firstly the absorption of posaconazole is highly variable and species dependent. This is not a surprising finding as lipid soluble drugs are known to have variable absorption as a result of unpredictable nature of the drug dissolution, which is needed to facilitate trans-membrane transport by the enterocytes. The particularly low absorption in comparison to

mammals was however unexpected. While it is not clear why this transpired, it is possible that the drug suspension administered in the absence of food was trapped in the crop for a longer period. As a structure the crop is present in raptors and allows for temporary storage of a consumed meal, for subsequent slow release into proventriculus. The other contributing factor is likely gastrointestinal transit time, which is significantly faster in raptors than in mammals. As a result, the administered dose may not have had sufficient time for dissolution and subsequent absorption to occur. The latter is best illustrated in people, where dividing a single large dose administered once every 24 h, to four smaller doses every 6 h allowed the drug sufficient time for absorption with a 200% increase in the fraction of absorption despite the same dose being administered.³⁰

Posaconazole in contrast to other antifungals like voriconazole is highly dependent on food for absorption. This was evident in a study in hawks, where it was demonstrated that food had a negative impact on voriconazole absorption and resulted in a marginally lower extent and a lag in absorption.³³ As expected, the administration of both the fatty meal and plant oil enhanced the absorption of posaconazole. However, the depot like effect seen in this study was unexpected as no such effect was reported in either the mouse, man, or dog. From the information for the in-oil or the high fat administrations, it is clear that the presence of lipids enhances absorption by stimulating the release of bile acid which allows for emulsification and the drug subsequently having access to cell membrane surface for transcellular movement. The long-lasting effect is however more difficult to explain and likely results from the normal action of GIT enzyme action. For lipid soluble (hydrophobic) drugs administered in the presence of an oily vehicle, it is known that the drug dissolves within its oily vehicle. Subsequently for the drug to be released from this lipid solution, the fat portion has to undergo enzyme induced lipolysis in the GIT so that it can enter into the aqueous phase through

Table 4. MIC values used for estimations in this study and the corresponding AUC₂₄ and AUC_{inf} ratio determined from the results obtained in the pivotal study were the birds were treated at a dose of 20 mg/kg in equal ratios with plant oil. (Silvanose et al. 2006; Silvanose et al. 2011).

Strain	Minimum inhibitory concentration (MIC)	Area under curve after single dose AUC ₂₄ : MIC		Area under curve infinity AUC _{inf} : MIC	
		Mean	Range	Mean	Range
<i>A. fumigatus</i>	0.12	102.41	73.61–130.41	339.74	120–2885
<i>A. flavus</i>	0.25	49.16	35.33–62.59	163.07	57.6–1384
<i>A. niger</i>	0.185	66.43	47.75–84.59	220.37	77.84–1871
<i>A. terreus</i>	0.25	49.16	35.33–62.59	163.07	57.60–1384

Area under curve to s4 h (AUC₂₄) and area under the area curve to infinity (AUC_{inf}) of 12.29 and 40.77 ug/ml * h was used respectively for calculations. The MIC values were obtained from Silvanose et al. (2006).

the action of bile salt micelles.³⁴ The depot like effect evident in this study is thus likely the result of a slow lipolysis process. This effect would also explain the lower absorption of the 20 mg/kg dose if the volume of oil in the administered mixture was insufficient, i.e., the oil would be completely metabolized prior to the contained dose having a chance to enter into its micellar solution. Once the oil has been metabolized the hydrophobic drug would subsequently precipitate and follow the same pharmacokinetic process as seen in the absence of a fatty meat. As a result, one possible way of overcoming this would be to double the volume of the administered dose of oil.

The main aim of the study was to determine if posaconazole would be efficacious in falcons. In human the AUC₂₄ : MIC ratio of 167 has been suggested as a surrogate indicator of efficacy,³¹ as this would allow for sufficient concentration of drug to be reached in the lungs where the infection is present as a result of the drug's ability to accumulate in the lungs. For this study the AUC₂₄ and AUC_{inf} ratio to the MIC was ascertained based on the results from previous studies for aspergillosis isolated from falcons (Table 4).^{12,13} In this model, the AUC₂₄ represents efficacy after a single dose while the AUC_{inf} represented the efficacy after steady state is reached. While it may be argued that the MICs are dated, these are the only results that we are aware of evaluating *Aspergillus* strains isolated from falcons. We are however confident that the MIC values are representative as they are very similar to a more recent study looking at avian strains from Argentina with the exception that the *A. fumigatus* strain, they identified was more susceptible.³⁵ Based on the results from the pivotal pharmacokinetic studies it is evident that the ability to reach this ratio is unlikely to happen due to the poorer absorption of the drug in the falcon especially when compared to that seen in humans and other mammalian species. However, from the calculated AUC_{inf}, which is a good indicator of AUC achievable after steady state, showed a strong likelihood of achieving the required effect after multiple dose administrations. With posaconazole looking promising for the treatment of falcons, further investigation is necessary. For this we recommend a repeat-dose study with daily dosing at a 24-h interval to allow for the

determination of the pharmacokinetic profile at steady state. Use of peak and trough data as previous evaluated following the 14 days of dosing in the red-tailed hawk, would likely be a suitable model.³⁶ We suggest that further studies focus on a target dose of 20 mg/kg in oil ratio of 1 : 2 to avoid the lower absorption seen in this study.

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Declaration of interest

The authors report no conflicts of interest. The authors are responsible for the content and the writing of the paper.

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