OFFICE OF CLINICAL PHARMACOLOGY REVIEW

NDA:	22-134
Submission Date(s):	September 29, 2009; May 28, 2010
Brand Name	(b) (4) ™
Generic Name	Alcaftadine
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Applicant	Vistakon Pharmaceutical LLC
Relevant IND(s)	66,884
Submission Type; Code	1S (NME)
Formulation; Strength(s)	0.25% alcaftadine ophthalmic solution
Indication	For the prevention of itching associated with allergic conjunctivitis

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LIST OF ABBREVIATIONS:

AAG	α-1 acid glycoprotein
Ae	amount excreted
Ae, %	amount excreted as a percent of dose
ANOVA	analysis of variance
AUC	area under the plasma concentration-time curve
AUC0-∞	area under the concentration vs. time curve from time 0 extrapolated to infinity
AUClast	area under the concentration vs. time curve from time 0 to last measurable concentration
BQL	below quantifiable limit
C	Celsius
CAC	conjunctival allergen challenge
CL	clearance
CLr	renal clearance
CLcr	creatinine clearance
CLt	total systemic clearance
C_{max}	maximum plasma concentration
CYP450	cytochrome P450
CV%	percent coefficient of variation
ECG	electrocardiogram
EDTA	ethylenediaminetetra-acetic acid
GFR	glomerular filtration rate
h	hour(s)
HPLC	high performance liquid chromatography
HSA	human serum albumin
ITT	intent to treat (population)
i.v.	intravenous
kg	kilogram(s)
L	liter
LC-MS	liquid chromatographic-mass spectrometry
LC-MS/MS	liquid chromatographic-triple quadrupole mass spectrometry
LOQ	limit of quantification
LLOQ	lower limit of quantification
mg	milligram(s)
µg	microgram(s)
mL	milliliter(s)
µM	micromole
min	minute(s)
msec	millisecond
N	number
NA	not applicable
NC	not calculated
NR	not reported
P-gp	P-glycoprotein
PD	pharmacodynamic
PK	pharmacokinetic
q.d.	once daily
R	accumulation ratio
SD	standard deviation
t1/2	elimination half-life
t1/2α	elimination half-life during alpha phase
t1/2β	elimination half-life during beta phase
t_{max}	time to maximum plasma concentration
ULOQ	upper limit of quantitation
uv	ultraviolet

1. EXECUTIVE SUMMARY

(b) (4)™ (alcaftadine, R89674) is a new chemical entity developed in the research laboratories of the Janssen Research Foundation. (b) (4)™ ophthalmic solution is a product developed by Johnson & Johnson Consumer & Personal Products Worldwide (J&JCPPW), Skillman, New Jersey, and is intended to be marketed by J&JCPPW's affiliate company, Vistakon Pharmaceuticals, LLC.

The Applicant submitted this 505(b)(1) application for (b) (4)™ 0.25% ophthalmic solution with a once daily dosing regimen in the prevention of itching associated with allergic conjunctivitis.

In support of the NDA, the Applicant submitted 11 clinical studies, including one Phase 1 PK study (05-003-09) conducted in healthy adult subjects following single and multiple doses of 0.25% alcaftadine ophthalmic solution and three Phase 1 tolerability /comfort studies (04-003-09, 05-003-04, and 07-003-10) conducted to assess the relative safety and comfort of alcaftadine ophthalmic solution with various dose strengths in different formulations compared to placebo. In addition, the sponsor conducted one Phase 2 proof-of-concept (POC) study (04-003-10) to evaluate the efficacy of alcaftadine ophthalmic solutions compared to vehicle and Patanol® (olopatadine hydrochloride 0.1%) using the conjunctival allergen challenge (CAC) model, three pivotal Phase 3 efficacy studies (05-003-11, 05-003-13, and 09-003-05) to further establish efficacy of 0.25% alcaftadine ophthalmic solution compared to vehicle using the CAC model, and one pivotal Phase 3 safety study (05-003-10) to evaluate the safety of 0.25% alcaftadine ophthalmic solution over a six-week period.

1.1. Recommendation

The Clinical Pharmacology information provided by the Applicant in the NDA submission is acceptable.

The reviewer's proposed label changes in Appendix 4.1 should be forwarded to the sponsor.

1.2. Phase IV Commitments

None.

1.3. Summary of Important Clinical Pharmacology and Biopharmaceutics Findings

The protein binding of alcaftadine and the active metabolite are 39.2% and 62.7%, respectively. The metabolism of alcaftadine is mediated by non-CYP450 cytosolic enzymes to the active carboxylic acid metabolite. *In vitro* studies showed that neither alcaftadine nor the carboxylic acid metabolite substantially inhibited reactions catalyzed by major CYP450 enzymes. Thus, clinically relevant interactions based on inhibition of CYP450 enzymes are not to be expected for alcaftadine and concomitantly administered drugs.

Following bilateral topical ocular administration of 0.25% alcaftadine ophthalmic solution, the mean plasma C_{max} of alcaftadine was approximately 0.06 ng/mL and the median T_{max} occurred at 15 minutes. Plasma concentrations of alcaftadine were below the lower limit of quantification (0.01 ng/mL) by 3 hours after dosing. The mean C_{max} of the active metabolite was approximately 3 ng/m and occurred at 1 hour after dosing. Plasma concentrations of the active metabolite were below the lower limit of quantification (0.10 ng/mL) by 12 hours after dosing. There was no indication of systemic accumulation or changes in plasma exposure of alcaftadine or the active metabolite following daily topical ocular administration. The elimination half-life of the active metabolite is approximately 2 hr following topical ocular administration and the metabolite is primarily eliminated unchanged in the urine.

2. QUESTION BASED REVIEW

2.1. General Attributes of the Drug

2.1.1. What are the highlights of the chemistry and physical-chemical properties of the drug substance and the formulation of the drug product?

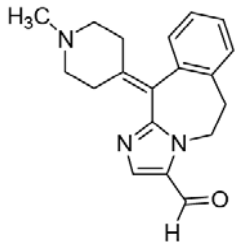
Alcaftadine is a white to yellow powder in which no polymorphism has been observed. The concentration of alcaftadine in the drug product solution is below the equilibrium solubility at 25°C in the formulation within the pH range of the formulation.

Structural Formula: $C_{19}H_{21}N_3O$

Molecular Weight: 307.39 Dalton

CAS Index Name: 6,11-dihydro-11-(1-methyl-4-piperidinylidene)-5*H*-imidazo[2,1-b] [3] benzazepine-3-carboxaldehyde

Chemical Structure:



Drug Product:
The drug product is a sterile ophthalmic solution containing 2.5 mg/mL alcaftadine. The primary packaging configuration consists of a low-density polyethylene (LDPE) bottle, dropper tip, and a polypropylene cap. The proposed commercial packaging configurations include a 5 mL bottle with a 3 mL fill volume (b) (4). The proposed commercial formulation (PD-F-5525) is shown in **Table 2.1.1-1**. To enhance ocular comfort, the buffer system in the formulation was modified several times and various formulations were studied in the development process (**Table 2.1.1-2**). The to-be-marketed formulation (i.e., PD-F-5525) was evaluated and supported by one pivotal Phase 3 efficacy study (09-003-05) and one Phase 1 tolerability/comfort study (07-003-10).

Table 2.1.1-1: Composition of Drug Product with the Formula PD-F-5525

Components	Reference to Quality Standard	Function	Concentration mg/mL
Alcaftadine	In-House	Active Ingredient	2.5
Sodium Phosphate Monobasic Monohydrate	USP	[redacted]	[redacted]
Edetate Disodium, Dihydrate	USP		
Benzalkonium Chloride (50% Solution) [1]	NF		
Sodium Chloride	USP		
Sodium Hydroxide (1N Solution) [2]	NF	pH Adjustment	pH Adjustment Target 7.0
Hydrochloric Acid (1N Solution) [2]			
Purified Water	USP	[redacted]	

[1] Equivalent to 0.05 mg/mL Benzalkonium Chloride.

[2] If needed, 1N NaOH solution and/or 1N HCl solution may be added to adjust the pH to 7.0.

Table 2.1.1-2: Clinical Studies and Formulations Evaluated with Alcaftadine Ophthalmic Solution (To-be-marketed formulation: PD-F-5525)

Component	PD-F-3089	PD-F-3090	PD-F-3091	PD-F-3092	NB: 10395-049	NB: 10395-050	NB: 10395-051	NB: 10395-052	PD-F-3549	PD-F-3552	PD-F-3730	PD-F-3785	PD-F-5525
Alcaftadine (mg/mL)	0.5	1.0	2.5	5.0	0.5	5.0	1.0	2.5	2.5	2.5	2.5	5.0	2.5
Buffer Conc. (mM)													
Sodium Chloride (mg/mL)													
BZK, 50% solution (mg/mL)													
EDTA (mg/mL)													
HPMC (mg/mL)													
Purified Water													
API Synthetic Vers.*													
Type of Study**													
14 day Rabbit Tox	04-1959-G1												
6 month Rabbit Tox											05-2456-G1		
Comfort 1					04-003-09								
Proof of Concept					04-003-10		04-003-10						
Comfort 2								05-003-04					
Phase 1 PK											05-003-09		
Phase 2 Pilot Relief											05-003-20		
Phase 3 & 3B Clinicals											05-003-10 05-003-11 05-003-13 06-003-09		
Comfort 3													07-003-10
Additional Phase 3 CAC & RSLs													09-003-05

* Chemical processing differences between the API synthetic versions can be seen in Cilag's DMF 20066.

** Study protocol numbers (xx-xxx-xx) are provided in the highlighted blocks.

2.1.2. What is the proposed mechanism of drug action and therapeutic indication?

Alcaftadine is a H_1, H_2, and H_4 histamine receptor antagonist. Alcaftadine inhibits allergen-induced changes in proteins [redacted] (b) (4) ™ 0.25% alcaftadine ophthalmic solution is indicated for the prevention of itching associated with allergic conjunctivitis.

2.1.3. What are the proposed dosage(s) and route(s) of administration?

0.25% alcaftadine ophthalmic solution with one drop in each eye once-daily.

2.2. General Clinical Pharmacology

2.2.1. What are the design features of the clinical pharmacology and clinical studies used to support dosing or claims?

The pharmacokinetics was studied in a Phase 1 study following single and multiple once-daily ocular instillation of 0.25% alcaftadine ophthalmic solution (05-003-09). Alcaftadine was evaluated in three Phase 1 single-dose tolerability/comfort studies (04-003-09, 05-003-04, and 07-003-10), one Phase 2 proof-of-concept (POC) study (04-003-10), three pivotal Phase 3 efficacy studies (05-003-11, 05-003-13, and 09-003-05), and one pivotal Phase 3 safety study (05-003-10) – **Table 2.2.1-1**.

Table 2.2.1-1: Clinical Studies with Alcaftadine Ophthalmic Solution

Study Number /Study Phase	Study Design	Treatments (Number of Subjects Treated)	Duration of Treatment/Total Subjects/Age
Tolerability (Comfort) Studies			
04-003-09 Phase 1	Single-center, double-masked, randomized, placebo-controlled, contralateral study design in adult subjects with normal ocular health. One visit on Day 0.	(b) (4)™ at 4 concentrations: 0.05%, 0.1%, 0.25% and 0.5% in 1 eye (each N=32); Placebo (Tears Naturale® II) in contralateral eye (N=128)	1 day/128/18-50 yr
05-003-04 Phase 1	Single-center, double-masked, randomized, placebo-controlled, contralateral treatment study in adult subjects with normal ocular health. One visit on Day 0.	(b) (4)™ 0.25%, (N=93); Placebo (Tears Naturale® II) in contralateral eye (N=93)	1 day/93/18-77 yr
07-003-10 Phase 1	Single-center, double-masked, randomized, placebo-controlled, contralateral study design in adult subjects with normal ocular health. One visit on Day 0.	(b) (4)™ at 1 concentration: 0.25% in 1 eye (each N=30); Placebo (Tears Naturale® II) in contralateral eye (N=30)	1 day/30/24-69 yr
PK Study			
05-003-09 Phase 1	Single-center, open-label, pK and safety study in healthy adult subjects. Screening visit (between Days -14 to -3), and visits on Days 1, 2, 3-4, 5-6, 7, and 8.	(b) (4)™ 0.25% oph halmic solution instilled bilaterally (N=13)	Daily for 7 days/13/18-49 yr
POC Study			
04-003-10 Phase 2	Single-center, double-masked, randomized, active- & vehicle (placebo)-controlled CAC study in adult subjects with a history of ocular allergies and/or a posi ive skin allergies test. Visits on Days -21, -14, 0, and 14.	(b) (4)™ at 1 of 3 concentra ions ins illed bilaterally: 0.05% (N=34), 0.10% (N=34), or 0.25% (N=34); Vehicle (placebo) ophthalmic solution instilled bilaterally (N=34); Patanol® 0.1% instilled bilaterally (N=34)	2 days (14 days apart)/170 /18-71 yr
Efficacy CAC Studies			
05-003-11 Phase 3	Multi-center, double-masked, randomized, vehicle (placebo)-controlled, CAC study in subjects aged ≥10 years with a history of allergic conjunctivitis or allergic rhinoconjunctivitis. Visits on Days -21, -14, 0, and 14.	(b) (4)™ 0.25% oph halmic solution instilled bilaterally (N=40); (b) (4)™ 0.25% ophthalmic solution in 1 eye and placebo (vehicle) in contralateral eye (N=42); Vehicle (placebo) ophthalmic solution instilled bilaterally (N=44)	2 days (14 days apart)/126/12-78 yr
05-003-13 Phase 3	Single-center, double-masked, randomized, vehicle (placebo)- controlled, CAC study in subjects aged ≥10 years with a history of allergic conjunctivitis or allergic rhinoconjunctivitis. Visits on Days -21, -14, 0, and 14.	(b) (4)™ 0.25% oph halmic solution instilled bilaterally (N=30); (b) (4)™ 0.25% ophthalmic solution in 1 eye and vehicle in contralateral eye (N=29); Vehicle (placebo) ophthalmic solution instilled bilaterally (N=29)	2 days (14 days apart)/88/14-72 yr
09-003-05 Phase 3	Multi-center, double-masked, randomized, vehicle (placebo) controlled, CAC study in adult and pediatric subjects with a history of acute allergic conjunctivitis. Visits on Days -21, -14, 0 and 14	(b) (4)™ 0.25% instilled bilaterally (N=30); Vehicle (placebo) ophthalmic solution instilled bilaterally (N=30)	2 days (14 days apart)/60/11-72 yr
Safety studies			
05-003-10 Phase 3	Multi-center, randomized, double-masked, vehicle (placebo)-controlled, parallel-group study in healthy subjects aged ≥3 years. Visits on Days 0, 7, 21, and 42.	(b) (4)™ 0.25% instilled bilaterally (N=609); Vehicle (placebo) ophthalmic solution instilled bilaterally (N=300)	Daily for 42 days/909/3-81 yr
06-003-09 Phase 3b environmental	Multi-center, double-masked, randomized, parallel group, evaluating efficacy, safety and impact on QOL vehicle (placebo) or Patanol® 0.1% CAC study in adult and pediatric subjects wi h a history of seasonal allergic conjunctivi is. Visits on Days 0, 7, 21, and 42.	(b) (4)™ 0.25% ophthalmic solution instilled bilaterally (N=147); Patanol® 0.1% instilled bilaterally (N=146); Vehicle (placebo) ophthalmic solution instilled bilaterally (N=72)	Daily for 42 days/365/10-82 yr
Pilot Relief Study			
05-003-20 Phase 2	Single-center, double-masked, randomized, vehicle (placebo)- controlled, CAC relief study in adult subjects with a history of allergic conjunctivitis or allergic rhinoconjunctivitis. Visits on Days -7, 0, and 7.	(b) (4)™ 0.25% instilled bilaterally (N=16); Vehicle (placebo) ophthalmic solution instilled bilaterally (N=17)	2 days (7 days apart)/33/18-52 yr

CAC=Conjunctival Allergen Challenge, POC=Proof-of-Concept, QOL=Quality of Life

2.2.2. What is the basis for selecting the response endpoints (i.e., clinical or surrogate endpoints) or biomarkers (collectively called pharmacodynamics [PD]) and how are they measured in clinical pharmacology and clinical studies?

In the POC Phase 2 study (04-003-10) and pivotal Phase 3 efficacy studies (05-003-11, 05-003-13, and 09-003-05), the efficacy of 0.25% alcaftadine ophthalmic solution in the prevention of itching associated with allergic conjunctivitis in adults and pediatric subjects (≥10 years of age) was evaluated using the CAC model. The CAC model is a validated model that simulates an allergic response in an environmental setting. The two primary variables, ocular itching and conjunctival redness, were evaluated on a 0 to 4 scale, allowing for half increment scores, where 0 indicated absence of the symptom and 4 indicated the highest severity of the symptom. In this patient-reported 5-point scale (0-4), mean difference scores (active minus vehicle [placebo]-treated eye) of greater than 0.5 unit at all time points, with two of three time points demonstrating at least 1 unit difference for ocular itching and conjunctival redness assessment, would be necessary to establish the efficacy (i.e., prevention of ocular itching and redness) of 0.25% alcaftadine ophthalmic solution over vehicle (placebo).

2.2.3. Are the active moieties in the plasma (or other biological fluid) appropriately identified and measured to assess pharmacokinetic parameters and exposure response relationships?

Yes, the sponsor used liquid chromatography with tandem mass spectrometry (LC/MS/MS) to quantitate concentrations of alcaftadine and R90692 (the active metabolite) in human plasma (*Refer to Section 2.6*).

2.2.4. Exposure-response

2.2.4.1. What are the characteristics of the exposure-response relationships (dose-response, concentration-response) for efficacy? If relevant, indicate the time to the onset and offset of the desirable pharmacological response or clinical endpoint. (If necessary, indicate in your answer the degree of linearity or nonlinearity in the dose-concentration relationship and how PK parameters change with time on chronic dosing, however, do not provide data or details for those topics. Those topics are addressed in question 2.2.5.)

Dose-response of alcaftadine ophthalmic solution was evaluated in Study 04-003-10, the phase 2 POC study that evaluated the efficacy of three concentrations of alcaftadine ophthalmic solutions (0.05%, 0.10%, and 0.25%) in relation to each other and to vehicle (placebo) and Patanol® ophthalmic solution (olopatadine hydrochloride 0.1%) in the prevention of allergen-mediated conjunctivitis using the CAC model. The study involved two instillations of the assigned study medication (one drop in each eye) two weeks apart with the study duration of approximately 5 weeks. At 15 min post study medication instillation, of the three doses of alcaftadine tested, only the alcaftadine 0.25% group had a mean difference of approximately ≥1 unit which was statistically significant ($p<0.001$) compared to the vehicle (placebo) group for both primary endpoints (prevention of ocular itching and conjunctival redness). Clinically significant differences between 0.25% alcaftadine ophthalmic solution and the vehicle (placebo) groups were also observed in the prevention of ocular itching and conjunctival redness at 16 hours (duration of action) post-study medication instillation. Additionally, this dose demonstrated the most consistent efficacy for the secondary endpoints including ocular and nasal symptoms.

2.2.4.2. *What are the characteristics of the exposure-response relationships (dose-response, concentration-response) for safety? If relevant, indicate the time to the onset and offset of the undesirable pharmacological response or clinical endpoint.*

The dose-response relationship for safety was evaluated in Phase 1 study 04-003-09, the tolerability/comfort study 1 that studied the relative safety and comfort of four concentrations of alcaftadine ophthalmic solutions (0.05%, 0.1%, 0.25%, and 0.5%) following single administration compared to Tears Naturale® II (Placebo). A dose-dependent increase in eye discomfort following drop instillation was noted. Alcaftadine 0.5% seemed to lead to a higher mean comfort score (less desirable) than the other doses. In Phase 1 study 05-003-04 (the tolerability/comfort study 2) designed to evaluate the safety and comfort of alcaftadine 0.25% in three different formulations as compared to Tears Naturale® II (placebo), a reduced buffer concentration was selected for use in the Phase 3 clinical programs based on a clinically meaningful benefit in comfort score beyond that seen with the other two formulations.

2.2.4.3. Does this drug prolong the QT or QTc interval? (You must answer this question, unless this is addressed in the question above.)

Cardiovascular safety and tolerability was not evaluated following topical administration of 0.25% alcaftadine ophthalmic solution, but has been studied following oral administration of alcaftadine. There were no clinically relevant changes in QT-interval following up to a single 8 mg oral solution dose. Due to a lower dose (i.e., 0.18 mg daily with a 35-µL drop per eye) and lower bioavailability with topical administration compared with oral administration (Refer to *Appendix 4.2.4.* for more detail), 0.25% alcaftadine ophthalmic solution is not expected to prolong the QT or QTc interval following topical administration.

2.2.4.4. Is the dose and dosing regimen selected by the sponsor consistent with the known relationship between dose-concentration-response, and are there any unresolved dosing or administration issues?

The Phase 3 dose of 0.25% alcaftadine was selected based on the dose-response data from the POC study 04-003-10 (Refer to *Section 2.2.4.1*) and the acceptable comfort score of this dose in Phase 1 studies 04-003-09 and 05-003-04 (Refer to *Section 2.2.4.2*). There are no unresolved dosing or administration issues.

2.2.5. What are the PK characteristics of the drug and its major metabolite?

The plasma PK of alcaftadine and its active metabolite R90692 were studied in healthy adult volunteers following single and multiple once-daily topical administration of 0.25 % alcaftadine ophthalmic solution.

2.2.5.1. What are the single dose and multiple dose PK parameters?

Table 2.2.5.1-1: Summary of Pharmacokinetic Parameters (Mean ± SD) for Alcaftadine and R90692 following Single and Multiple Daily Doses of Alcaftadine 0.25% Ophthalmic Solution (n=13)

	Alcaftadine		R90692	
	Day 1	Day 7	Day 1	Day 7
Mean C_{max}, ng/mL(range)	0.051 ± 0.028 (0.01, 0.11)	0.060 ± 0.030 (0.02, 0.11)	3.228 ± 1.803 (1.40, 7.23)	2.715 ± 1.653 (1.03, 5.64)
Median T_{max}, hr (Min, Max)	0.25(0.15, 0.67)	0.25(0.25, 0.5)	1.05(0.8, 1.58)	1.00(1.00, 1.5)
Mean $AUC_{0\text{-last}}$, ng*hr/mL (range)	0.040 ± 0.030 (0.01, 0.11)	0.039 ± 0.028 (0.01, 0.11)	10.613 ± 4.655 (5.22, 20.3)	9.272 ± 5.709 (3.13, 22.57)
$T_{½}$. hr	1.1 ± 0.9	0.7 ± 0.5	2.0 ± 0.2	2.1 ± 0.4

2.2.5.2. How does the PK of the drug and its major active metabolites in healthy volunteers compare to that in patients?

The plasma PK of alcaftadine and R90692 were only studied in healthy adult volunteers following single and multiple once-daily topical administration of 0.25% alcaftadine ophthalmic solution.

2.2.5.3. What are the characteristics of drug absorption?

Following topical administration of 0.25% alcaftadine ophthalmic solution, T_{max} for alcaftadine and R90692 were approximately 0.25 hr and 1 hr post dose, respectively. C_{max} of alcaftadine was approximately $1/25^{th}$ of the C_{max} of R90692. The relative bioavailability of alcaftadine following topical administration has not been assessed. On a dose-adjusted basis, the bioavailability of R90692 following multiple daily ophthalmic administration of alcaftadine was approximately half of that obtained following multiple daily oral administration of alcaftadine (**Table 2.2.5.3-1**).

Table 2.2.5.3-1: Summary of Pharmacokinetic Parameters for R90692 (Active Metabolite) Following Multiple Ophthalmic or Oral Daily Doses of Alcaftadine

Parameter	**Ophthalmic 0.25% Solution Day 7**	**0.5 mg Oral Day 8**	**1 mg Oral Day 8**	**2 mg Oral Day 8**
Cmax (ng/mL)	2.72 ± 1.72	13.6 ± 3.3	31.4 ± 5.7	55.8 ± 7.6
Median Tmax (h) Range	1.0 (1.0 -1.5)	1.0 (0.5 -1.0)	1.0 (0.5 – 2.0)	0.75 (0.5 – 2.0)
$AUC_{0\text{-last}}$ (ng·hr/mL)	9.27 ± 5.94	49.8 ± 10.3	110 ± 15	203 ± 20
Dose-adjusted AUC ($AUC_{0\text{-last}}$ /Dose)	54.5	99.6	110.0	101.5
t ½ (h)	2.10 ± 0.41	1.8 ± 0.2	1.7 ± 0.3	1.9 ± 0.2
Urinary excretion (% Dose)	NA	56.8 ± 2.8	60.1 ± 5.3	55.0 ± 6.6

Note: Total daily ophthalmic dose = 0.18 mg; The LLOQ in the oral pharmacokinetics study was 0.5 ng/mL for both alcaftadine and R90692. Alcaftadine concentrations were below the LLOQ following single dose (8 mg) or multiple dose (2 mg per day for 8 days) oral administration.

2.2.5.4. What are the characteristics of drug distribution?

Plasma protein binding of alcaftadine (5 ng/mL) and R90692 (20 ng/mL) in human plasma was 39.2% and 62.7%, respectively.

2.2.5.5. Does the mass balance study suggest renal or hepatic as the major route of elimination? (This may include table with results of mass balance study.)

A mass balance study was not performed.

2.2.5.6. What are the characteristics of drug metabolism? (This may include data on extraction ratio; metabolic scheme; enzymes responsible for metabolism; fractional clearance of drug.)

Based on the *in vitro* metabolism study using human liver microsomes, only one major metabolite, R90692, which accounted for approximately 4.3% of the total radioactivity, was present in the incubates. Therefore, CYP450 is not the major enzyme system involved in alcaftadine metabolism. Alcaftadine is metabolized *in vivo* to the acid metabolite R90692. The conversion is believed to be predominantly mediated by non-CYP450 cytosolic enzymes such as aldehyde dehydrogenase, alcohol dehydrogenase, aldehyde oxidase, and aldehyde reductase.

2.2.5.7. What are the characteristics of drug excretion?

The major metabolite R90692, following oral administration of alcaftadine, was primarily excreted unchanged in urine. Following ocular administration of 0.25% alcaftadine ophthalmic solution, the major metabolite R90692 is expected to be excreted unchanged mainly in urine as well.

2.2.5.8. Based on PK parameters, what is the degree of linearity or nonlinearity in the dose-concentration relationship?

Not evaluated because only a single dose PK study was conducted.

2.2.5.9. How do the PK parameters change with time following chronic dosing? (This may include time to steady-state; single dose prediction of multiple dose PK; accumulation ratio.)

As shown in **Table 2.2.5.1-1**, there was no accumulation of alcaftadine and R90692 following multiple daily administration of 0.25% alcaftadine ophthalmic solution. $AUC_{0\text{-last}}$, C_{max}, T_{max}, and $T_{1/2}$ for either alcaftadine or R90692 were similar between Days 1 and 7.

2.2.5.10. What is the inter- and intra-subject variability of PK parameters in volunteers and patients, and what are the major causes of variability?

The CV% for $AUC_{0\text{-last}}$ and C_{max} values for both alcaftadine and R90692 were > 50% among healthy subjects (**Table 2.2.5.1-1**). This level of inter-subject variability in systemic exposure is not considered unexpected following topical ocular administration. PK was not studied in patients.

2.3. Intrinsic Factors

2.3.1. What intrinsic factors (age, gender, race, weight, height, disease, genetic polymorphism, pregnancy, and organ dysfunction) influence exposure (PK usually) and/or response, and what is the impact of any differences in exposure on efficacy or safety responses?

In the Phase 1 PK study (05-003-09), a limited number of healthy subjects (n=13) were enrolled. The sponsor has not evaluated the effect of the commonly known intrinsic factors including race, age, and renal or hepatic impairment on the PK of alcaftadine and R90692 following topical administration of 0.25% alcaftadine. Given the low systemic exposure following topical administration, however, dose adjustment is likely not warranted in patients based on the commonly known intrinsic factors.

2.3.1.1. Gender

Based on the data from the Phase 1 PK study (05-003-09) which enrolled 13 healthy subjects (n=13 total) including 6 male and 7 female, no gender difference (p >0.05) was observed in alcaftadine and R90692 exposure in terms of plasma C_{max} and AUC.

2.4. Extrinsic Factors

2.4.1. What extrinsic factors (drugs, herbal products, diet, smoking, and alcohol use) influence dose-exposure and/or -response and what is the impact of any differences in exposure on response?
Based upon what is known about exposure-response relationships and their variability, what dosage regimen adjustments, if any, do you recommend for each of these factors? If dosage regimen adjustments across factors are not based on the exposure-response relationships, describe the basis for the recommendation.

Not applicable.

2.4.2. Drug-drug interactions

2.4.2.1. Is there an in vitro basis to suspect in vivo drug-drug interactions?

No. The metabolism of alcaftadine occurs primarily via cytosolic enzymes, presumably by aldehyde dehydrogenase, alcohol dehydrogenase, aldehyde oxidase or aldehyde reductase. *In vitro* studies suggested that no clinically relevant interactions based on inhibition of CYP450 enzymes would be expected for the metabolism of alcaftadine. In addition, both alcaftadine and R90692 are not expected to inhibit the metabolism of other drugs via CYP450 enzymes following topical administration of 0.25% alcaftadine ophthalmic solution.

2.4.2.2. Is the drug a substrate of CYP enzymes? Is metabolism influenced by genetics?

Based on the *in vitro* metabolism study using human liver microsomes, only one major metabolite, R90692, which accounted for approximately 4.3% of the total radioactivity, was present in the incubates. Therefore, CYP450 enzyme is not likely the major enzyme system involved in alcaftadine metabolism. Based on studies *in vitro*, CYP 2A6, 3A4, and 2C19 may be involved in the metabolism of alcaftadine to the acid R90692. However, the metabolism of R897674 is not expected to be influenced by CYP450 polymorphisms because the *in vivo* metabolism of alcaftadine is mainly mediated by non CYP450 enzymes.

In a clinical drug interaction study (Study No: N122433) following 2 mg oral dose of alcaftadine, co-administered ketoconazole (a CYP3A4 inhibitor) was found not to inhibit the *in vivo* metabolism of alcaftadine as there was no changes in the plasma concentrations of alcaftadine and the AUC of the metabolite R90692 increased 10% following co-administration of ketoconazole. The 10% increase in R90692 AUC is not considered to be clinically relevant.

2.4.2.3. Is the drug an inhibitor and/or an inducer of CYP enzymes?

Neither alcaftadine (up to 100 ng/mL) nor R90692 (up to 1000 ng/mL) substantially inhibited reactions catalyzed by major CYP450 enzymes. Whether alcaftadine or R90692 is an inducer of CYP450 enzymes has not been assessed.

2.4.2.4. Is the drug a substrate and/or an inhibitor of P-glycoprotein transport processes?

It is unknown whether alcaftadine is a substrate and/or an inhibitor of P-glycoprotein transport.

2.4.2.7. What other co-medications are likely to be administered to the target patient population?

Numerous medications are likely to be co-administered with alcaftadine including other topical ophthalmics and and systemic drugs.

2.4.2.8. Are there any in vivo drug-drug interaction studies that indicate the exposure alone and/or exposure-response relationships are different when drugs are co-administered?

Not studied.

2.4.2.9. Is there a known mechanistic basis for pharmacodynamic drug-drug interactions, if any?

Yes, antihistamines such as alcaftadine has the potential to antagonize topical cholinergic agonists (e.g., pilocarpine).

2.4.2.10. Are there any unresolved questions related to metabolism, active metabolites, metabolic drug interactions, or protein binding?

There are no unsolved questions related to metabolism, active metabolites, metabolic drug interaction, or protein binding.

2.4.3. What issues related to dose, dosing regimens, or administration are unresolved and represent significant omissions?

No issues related to dose, dosing regimens, or administration remain unresolved.

2.5. General Biopharmaceutics

Not applicable. Alcaftadine is formulated as an ophthalmic solution for topical administration.

2.6. Analytical Section

2.6.1. How are the active moieties identified and measured in the plasma in the clinical pharmacology and biopharmaceutics studies?

The sponsor used liquid chromatography with tandem mass spectrometry (LC/MS/MS) to quantitate concentrations of alcaftadine and R90692 (active metabolite) in K3 EDTA human plasma. Samples were prepared using solid phase extraction and subsequently analyzed by LC/MS/MS using positive turbo-ionspray ionization mode and operating the instrument in the multiple-reaction-monitoring (MRM) mode.

2.6.2. Which metabolites have been selected for analysis and why?

One carboxylic acid metabolite R90692 was selected for analysis because it is the primary and pharmacologically active metabolite of alcaftadine.

2.6.3. For all moieties measured, is free, bound, or total measured? What is the basis for that decision, if any, and is it appropriate?

The reported concentrations of alcaftadine and R90692 represent total concentrations. The mean plasma protein binding of alcaftadine is 39.7% at 5 ng/mL. The mean plasma protein binding of R90692 is 62.7% at 20 ng/mL. Free concentrations in the plasma are not considered clinically relevant to the indicated efficacy following ocular topical administration.

2.6.4. What bioanalytical methods are used to assess concentrations?

Refer to *Section 2.6.1.* for further information.

2.6.4.1. What is the range of the standard curve? How does it relate to the requirements for clinical studies? What curve fitting techniques are used?

The standard curve in plasma ranged from 0.010 ng/mL to 2.50 ng/mL for alcaftadine and from 0.100 ng/mL to 25.0 ng/mL for R90692. The observed plasma concentrations of either alcaftadine or R90692 in clinical studies did not exceed the upper limit of quantitation for the respective standard curve. The linear regression of the curves for peak area ratios *versus* concentration was weighted $1/x^2$ for the standard curves for both alcaftadine and R90692.

2.6.4.2. What are the lower and upper limits of quantification (LLOQ/ULOQ)?

The lower and upper limits of quantitation were 0.010 ng/mL and 2.50 ng/mL for alcaftadine and 0.100 ng/mL and 25.0 ng/mL for R90692, respectively.

2.6.4.3. What are the accuracy, precision, and selectivity at these limits?

The accuracy (%RE) and precision (%CV) ranges for alcaftadine were -5.3% to 4.7% and 1.1% to 11.9%, respectively.

The accuracy (%RE) and precision (%CV) ranges for R90692 were -6.1% to 2.5% and 1.7% to 13.2%, respectively.

Selectivity was demonstrated by the lack of interference by potential endogenous interfering substances in six distinct lots of K3 EDTA human plasma.

2.6.4.4. What is the sample stability under the conditions used in the study (long-term, freeze-thaw, sample-handling, sample transport, autosampler)?

Both alcaftadine and R90692 was shown to be stable in plasma at room temperature up to 24 hours, in extracted samples for 2 days when stored refrigerated, following three freeze/thaw cycles, in stock solution stored at -20°C for more than 120 days, and in plasma when stored at -70°C for a period of 29 days.

2.6.4.5. What is the QC sample plan?

The concentrations of the QC samples consisted of 0.030, 0.400, and 2.00 ng/mL for alcaftadine, and 0.300, 4.00, and 20.0 ng/mL for R90692, respectively. Between-run and within-run accuracy and precision were evaluated using replicates (n=6) from each of the three concentrations for either alcaftadine or R90692.

3. LABELING RECOMMENDATIONS

See Appendix 4.1. for detailed labeling recommendations.

4. APPENDICES

4.1. Proposed Package Insert (Original and Annotated)

3 pp of draft labeling withheld in full immediately after this page as (b)(4) CCI/TS.

4.2. Individual Study Review

4.2.1. Studies on Plasma Protein Binding and Distribution in Blood

Study Number: N125358
Dates: March 20, 1997 to June 24, 1997
Title: The plasma protein binding and the distribution of alcaftadine and its carboxylic acid metabolite (R90692) in blood

Objectives:
To assess the *in vitro* plasma protein binding and distribution between plasma and blood cells of alcaftadine and R90692 in human and various animal species.

Methods:
Human and animal blood was collected. The protein binding of alcaftadine and R90692 was determined in individual human plasma samples, and in individual or pooled plasma samples from animals (male Beagle dogs, male and female Wistar rats, female Cunistar-MDL rabbits, and male Swiss mice). Human plasma from five male subjects was fortified with 5 ng ^{3}H-alcaftadine/mL or with 20 ng ^{3}H-R90692/mL. Dog and rabbit plasma, and pooled rat plasma and pooled mouse plasma were fortified with 100 ng ^{3}H-alcaftadine/mL or with 1,000 ng ^{3}H-R90692/mL. Dog plasma was also fortified with 5 ng ^{3}H-alcaftadine/mL or with 20 ng ^{3}H-R90692/mL. Duplicate samples were subjected to equilibrium dialysis. (b) (4)

The binding of ^{3}H-alcaftadine (10 ng/mL) and of ^{3}H-R90692 (50 ng/mL) to purified human serum albumin (HSA) and human α_1-acid glycoprotein were also studied. Duplicate samples were subjected to equilibrium dialysis against 0.067 M Sorensen phosphate buffer pH 7.40.

To study the distribution of alcaftadine and R90692 in blood, blank whole blood obtained from the same subjects and animals (rats, rabbits, mice, and dogs) as in the plasma protein binding experiments was fortified with ^{3}H-alcaftadine and ^{3}H-R90692 at the concentrations used to study the plasma protein binding. Duplicate fortified blood samples were incubated at 37 °C for 30 min in test tubes in a (b) (4) incubation bath. The tubes were gently twisted around from time to time during the incubation. (b) (4)

Analytical Methods:
Radioactivity levels were measured in duplicate samples of fortified plasma before equilibrium dialysis, and of the contents of plasma, protein and buffer compartments of the dialysis cells. Radioactivity levels were measured in triplicate blood and plasma samples obtained from the experiments on the distribution in blood. Radioactivity was measured in a liquid scintillation

spectrometer with automatic calculation from counts per minute (cpm) into disintegrations per minute (dpm).

Calculation:
The fraction of the unbound drug (f_u) was calculated as the ratio of the unbound concentration, (C_u) to the total concentration (C) as determined by radioactivity measurements in the buffer (C_u) and plasma or protein (C) compartments of the dialysis cells: $f_u = C_u/C$.

The ratio of blood to plasma concentration (C_b/C) was calculated from the experiments on the distribution in blood. The fractions of the drug distributed in blood to plasma water, $f_{ub}(1–H)$, to plasma proteins, $f_{pp}(1 – H)$, and to blood cells ($f_{BC}.H$) were calculated according to the following formulae:

$f_{ub}(1 – H) = C \times f_u (1–H)/C_b$ H = haematocrit value

$f_{pp}(1 – H) = C \times f_b (1–H)/C_b$ with $f_b = 1 - f_u$

$f_{BC}.H = 1 – C \times (1–H)/C_b$

Results:

The stability of ^{3}H-alcaftadine and ^{3}H-R90692 in plasma under the experimental conditions of the equilibrium dialysis and in blood during the experiments on the distribution was measured by radio-HPLC analysis.

^{3}H-R90692 was stable in both blood and plasma of all species studied. In contrast, ^{3}H-alcaftadine stability in blood and plasma varied among species. In human, dog, and mouse plasma and in human and dog blood, alcaftadine was considered relatively stable as a less than (b) (4)% degradation was observed for alcaftadine after 30-min incubation in blood or 4-hour incubation in plasma at 37°C. Alcaftadine was considered unstable in rat, mouse and rabbit blood, and rat and rabbit plasma because substantial ((b) (4)%) degradation was observed. Therefore, alcaftadine plasma protein binding was only reported on human, dog and mouse, and alcaftadine red blood cell to plasma concentration ratios were only reported on human and dog.

Plasma protein binding
The results are shown in **Table 1**. Plasma protein binding of alcaftadine in human, dog and mouse was 39.2%, 41.4%, and 52.1%, respectively. Plasma protein binding of R90692 in human, dog , rat, mouse and rabbit was 62.7%, 19.9%, 11.5-13.8%, 12.8%, and 28.7%, respectively.

Table 1: Plasma Protein Binding of Alcaftadine and R90692

Species	R89674 % bound (mean ± SD)	R90692 % bound (mean ± SD)
Human (m)	39.17 ± 1.4	62.7 ± 1.4
Dog (m)	41.4 ± 1.2	19.9 ± 1.1
Rat (m)	NA	11.5 ± 0.9
Rat (f)	NA	13.8 ± 1.4
Mouse (m)	52.1 ± 2.3	12.8 ± 1.4
Rabbit (f)	NA	28.7 ± 1.9

m=male; f=female

Binding to purified human plasma proteins
The binding of alcaftadine to HSA and α1-acid glycoprotein at their physiological concentrations was 27 % and 10%, respectively. The binding R90692 to HSA and α_1-acid glycoprotein at their physiological concentrations was 72% and < 5%, respectively.

Blood to plasma ratio
The results are shown in Table 2. Blood to plasma ratio of alcaftadine in human and dog was 1.40 and 1.45, respectively. Blood to plasma ratio of R90692 in human, dog, rat, mouse and rabbit was 0.63, 0.81, 0.89, 0.85 and 0.75, respectively.

Table 2: Red Blood Cell to Plasma Concentration Ratios for Alcaftadine and R90692

Species	**R89674 C_{blood}/C_{plasma} (mean ± SD)**	**R90692 C_{blood}/C_{plasma} (mean ± SD)**
Human (m)	1.40 ± 0.05	0.63 ± 0.01
Dog (m)	1.45± 0.03	0.81 ± 0.02
Rat (m/f)	NA	0.89 ± 0.01
Mouse (m)	NA	0.85 ± 0.02
Rabbit (f)	NA	0.75 ± 0.01

m=male; f=female

Conclusions:
Plasma protein binding of alcaftadine and R90692 in human and various preclinical species can be classified as low to moderate. Both alcaftadine and R90692 bind to HSA at a higher degree than that to α1-acid glycoprotein. In addition, the higher binding of R90692 to HSA versus alcaftadine is consistent with a higher plasma protein binding of R90692 versus alcaftadine.

Alcaftadine has an affinity for red blood cells in human and dog blood. In contrast, R90692 is largely concentrated in the plasma.

These studies are acceptable from a clinical pharmacology perspective.

4.2.2. In Vitro Metabolism Study on Alcaftadine

Study Number: N125328
Dates: February 18, 1997 to June 1997
Title: In Vitro Metabolism of ^{14}C-alcaftadine in Human Microsomal Fractions

Objectives:
To characterize the involvement of human CYP450 enzymes in the microsomal metabolism of alcaftadine; to evaluate the potential inhibition of alcaftadine metabolism by diagnostic CYP450 inhibitors; and to evaluate the potential inhibition by alcaftadine and its major metabolite R90692 on the metabolism of probe CYP450 substrates.

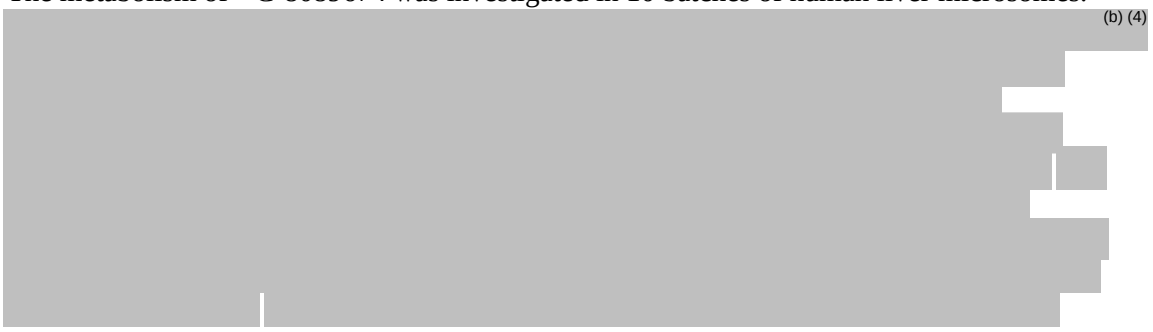
Methods:
^{14}C-alcaftadine (labeled in the benxylic bridgehead carbon) was incubated with 10 different batches of human liver microsomes in the presence of an NADPH-generating system. The correlation between alcaftadine metabolism and the metabolism of isoenzyme-specific CYP450 substrates in 10 different batches of human liver microsomes was calculated. In addition, inhibition experiments with diagnostic CYP450 inhibitor or substrates were performed.

The diagnostic CYP450 inhibitors were dissolved in methanol, including 7, 8-benzoflavone (2 mM, CYP1A2), furafylline (10 mM, CYP1A2), mephenytoin (100 mM, CYP2C19), gestodene (8 mM, CYP3A4), phenacetin (10 mM, CYP1A2), tolbutamide (10 mM, CYP2C8/9), phenytoin (10 mM, CYP2C), sulphaphenazole (2 mM, CYP2C10), coumarin (5 mM, CYP2A6), aniline (10 mM, CYP2E1), *p*-nitrophenol (10 mM, CYP2E1), quinidine (2 mM, CYP2D6), and troleandornycin (4 mM, CYP3A4).

The probe CYP450 substrates were prepared as stock solutions including tolbutamide (240 mM in DMSO; CYP2C8/9), chlorzoxazone (100 mM in DMSO; CYP2E1), cyclosporin-A (1 mM in DMSO, 5.78 MBq/rnl; CYP3A4), caffeine (50 nM in 50 mM Na, K-phosphate buffer pH 7.4, 103 .6 kBq/ml; CYP1A2), coumarin (2 mM in 50:50 /methanol: water; CYP2A6), lauric acid (10 mM in methanol, 370 kBq/ml; CYP4A), testosterone (10 mM in methanol, 745 kBq/ml), and debrisoquine (48.8 pM dissolved immediately in the microsomal suspension, 46.7 kBq/ml; CYP2D6).

Reviewer's comments: *Some diagnostic CYP450 inhibitors and probe CYP450 substrates the applicant used in this in vitro metabolism study are not in agreement with those suggested by the recent Guidance for Industry "Drug Interaction Studies-Study Design, Data Analysis, and Implications for Dosing and Labeling (2006).*

Metabolism of 089674 in 10 batches of human liver microsomes
The metabolism of ^{14}C-8089674 was investigated in 10 batches of human liver microsomes.



Inhibition of ^{14}C-alcaftadine metabolism by diagnostic CYP450 inhibitors

(b) (4)

(b) (4)

Inhibition of specific CYP450 activities by ^{14}C-alcaftadine and R90692

The metabolism of specific CYP450 substrates was studied in a batch of human liver microsomes (the same as above) in the absence and presence of different concentrations of alcaftadine (0, 1, 10 and 100 ng/ml) and R90692 (0, 50, 200 and 1000 ng/ml). (b) (4)

Results*:*

The metabolism of alcaftadine was comparable in the 10 different batches of human liver microsomes. Only one major metabolite, R90692, which accounted for 4.25 ± 2.24% of the injected radioactivity, was present in the incubates. The main *in vitro* metabolic pathway was aldehyde oxidation of alcaftadine to the acid metabolite R90692.

The correlations between the rate of overall metabolism of alcaftadine (or rate of formation of R90692) and the rate of metabolism of various specific CYP450 substrates in 10 different batches of human liver microsomes were calculated by linear regression and the Spearman rank correlation analysis. Although linear regression analysis indicated a correlation between the CYP2A6 activity and the overall metabolism of alcaftadine, the correlation coefficients obtained after Spearman rank correlation analysis showed no significant correlation between the cytochrome P-450 probe substrates and the metabolism of alcaftadine or the formation of the acid metabolite R90692.

The data on the percent inhibition of the metabolism of alcaftadine by each of the diagnostic CYP450 inhibitors (**Table 1**) indicated that alcaftadine metabolism and R90692 formation were most sensitive to inhibition by mephenytoin (CYP2C19), gestodene (CYP3A4) and troleandomycin (CYP3A4), suggesting the involvement of CYP3A4 and CYP2C19 in the microsomal metabolism of alcaftadine.

The involvement of CYP450 in the metabolism of alcaftadine is, however, most likely to be of minor importance. R90692 was the only metabolite observed after incubation with human liver

microsomes and represented only 4.25% of the injected radioactivity, thus, CYP450 is probably not the major enzyme system involved in the metabolism of alcaftadine. The enzymes involved in catalyzing aldehyde oxidation are aldehyde dehydrogenase and the aldehyde oxidase, which are cytosolic enzymes. The lack of significant involvement of CYP3A4 in the metabolism of alcaftadine was confirmed by a clinical study (No: N122433) following 2 mg oral dose of alcaftadine indicating that ketoconazole was not an inhibitor of the *in vivo* metabolism of alcaftadine.

The percent inhibition of CYP450 catalyzed reactions (**Table 2**), assayed after incubation of 100 ng/mL alcaftadine and 1000 ng/mL R90692 with the diagnostic CYP450 substrates, indicated that no substantial inhibition (≤ 25%) of any of the metabolism of any of the diagnostic substrates occurred, even at the highest concentrations of alcaftadine (100ng/mL) and R90692 (1000 ng/mL) tested.

Table 1: Percent Inhibition of the Metabolism of alcaftadine by Diagnostic CYP450 Inhibitors

Inhibitor	Cytochrome P-450 form inhibited	% Inhibition of metabolism	
		Overall	R90692 formation
7,8-Benzoflavone (10 μM)	CYP1A2	20.8	8.9
Phenacetin (50 μM)	CYP1A2	9.4	-11.2
Furafylline (100 μM)	CYP1A2	26.9	22.3
Coumarin (25 μM)	CYP2A6	3.3	-1.8
Tolbutamide (50 μM)	CYP2C8/9/10	-3.6	-24.6
Phenytoin (50 μM)	CYP2C8/9/10	22.3	17.0
Sulphaphenazole (10 μM)	CYP2C8	12.4	-9.8
Mephenytoin (500 μM)	CYP2C19	45.2	53.1
Quinidine (10 μM)	CYP2D6	6.4	3.6
P-Nitrophenol (50 μM)	CYP2E1	-8.9	-11.2
Aniline (50 μM)	CYP2E1	-20.3	6.3
Gestodene (40 μM)	CYP3A4	49.8	57.1
Troleandomycin (20 μM)	CYP3A4	50.5	61.2

Table 2: Percent Inhibition of CYP450 Catalyzed Reactions by alcaftadine or R90692

Enzyme activity	CYP P-450 Form	% Inhibition at 100 ng/mL R89674	% Inhibition at 1000 ng/mL R90692
Caffeine N3-demethylation	CYP1A2	-25.0	25.0
Coumarin 7-hydroxylation	CYP2A6	-14.1	-31.3
Tolbutamide hydroxylation	CYP2C9/10	-22.6	-24.2
Debrisoquine 4-hydroxylation	CYP2D6	-13.8	3.1
Chlorzoxazone 6-hydroxylation	CYP2E1	-3.1	0
Testosterone 6-β hydroxylation	CYP3A4	-5.8	6.5
Cyclosporin A-oxidation	CYP3A4	-21.7	-6.3
Lauric acid hydroxylation	CYP4A	-5.0	-19.9

Conclusion:

The metabolism of alcaftadine to the acid metabolite R90692 was likely predominately mediated by non- CYP450 enzymes (presumably by aldehyde dehydrogenase, alcohol dehydrogenase, aldehyde oxidase, and aldehyde reductase). Microsomal metabolism by CYP2A6, 3A4, and 2C19 appeared to account for only a small percentage of alcaftadine metbaolism. Neither alcaftadine nor R90692 could substantially inhibit reactions catalyzed by major CYP450 isoenzymes.

Clinically relevant drug interactions based on inhibition of the CYP450 isoenzymes is not expected for the metabolism of alcaftadine or by alcaftadine or R90692 on the metabolism of other drugs following topical administration of alcaftadine.

Some diagnostic CYP450 inhibitors and probe CYP450 substrates the Applicant used in this *in vitro* metabolism study are not in agreement with those suggested by the recent Guidance for Industry "Drug Interaction Studies-Study Design, Data Analysis, and Implications for Dosing and Labeling (2006). However, the study is considered acceptable from a clinical pharmacology perspective and no additional studies are warranted, given the low systemic exposure to both alcaftadine and R90692 following topical instillation, the notion that only marginal role CYP450 enzymes play in alcaftadine metabolism, and the fact that the study was conducted before the publication of the Guidance.

COMMENTS:
The major limitation of this study is that the positive controls were not included in the metabolism and inhibition studies, rendering the experimental conditions questionable. Despite that the Applicant stated in the submission dated on May 28, 2010 that the human liver microsomes used were tested for metabolic activity prior to use, the reviewer recommends that the metabolism data from study N125328 should be used for internal purposes only and not used to support regulatory decisions.

Study Number: N126519
Dates: January 28, 1997 to September 1997
Title: The in vitro metabolism of alcaftadine and R90692 in hepatocytes and liver subcellular fractions of male and female, adult and neonatal mice, male and female rats, female rabbits, male dog and human

Objectives:
To investigate the *in vitro* biotransformation of alcaftadine and R90692 in liver subcellular fractions and hepatocytes (suspensions and primary cell cultures) of males and female rats, female rabbits, male dog and human.

Reviewer's comments: *This review is focused on the data pertaining to in vitro metabolism of alcaftadine and R90692 in <u>human</u> hepatocyte suspension and subcellular fractions.*

Methods:

The hepatocytes suspension was characterized for protein and CYP450 contents and for the metabolism of two substrates: 7-ethoxycoumarin and scoparone. Both substrates undergo Phase I metabolism (**Figure 1**) and their Phase I metabolites are further metabliszed via glucuronidation. The subcellular fractions were also characterized for protein and CYP450 contents, but not for the metabolism of two substrates.

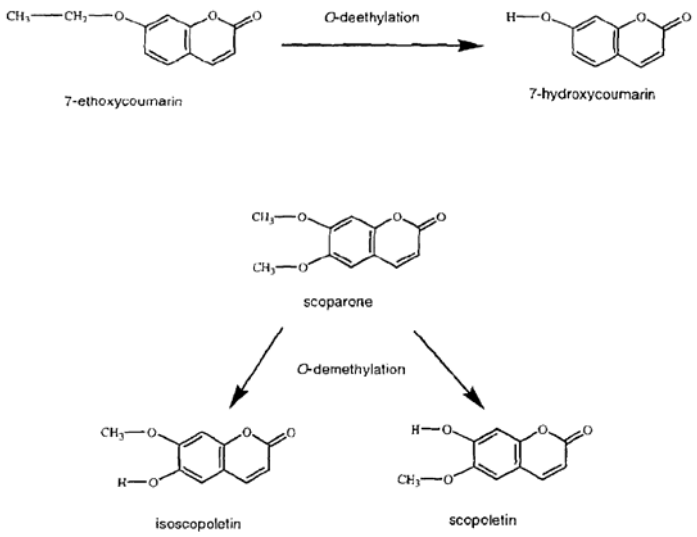


Figure 1: Phase I metabolic pathways of 7-ethoxycoumarin and scoparone

^{14}C-alcaftadine and ^{14}C-R90692 was incubated at 10 μM (final radioactive concentration of 5.9 kBq/mL) with human hepatocyte suspension containing 4 ×10^6 cells/mL. The incubates were mixed and incubated at 37 °C for 120 minutes. The incubations of ^{14}C-alcaftadine and ^{14}C-R90692 (final concentraions at 10 μM) with human liver subcellular fraction (both microsomes and 12000 ×g supernatant fractions) were performed in 10-mL tubes at 37 °C for 120 minutes.

Results:

Phase I and Phase II metabolic activities of the human hepatocyte preparation were demonstrated by two probe substrates 7-ethoxycoumarin and scoparone, suggesting that the preparation was suitable to investigate the metabolism of alcaftadine and R90692.

Alcaftadine was metablized into R90692 (47% converted) and two other metabolites (**Table 1**, **Figure 2**) in human hepatocyte suspension. Alcaftadine was metablized into R90692 in a greater extent (95.2% converted) in human liver 12000×g supernatant. The majority of alcaftadine (81.4%) remained unchanged in human liver microsomes.

In contrast, R90692 appeared to be stable and did not undergo further metablism in all the preparations.

Table 1: Mass balance of alcaftadine (R89674) and R90692 and of their major metablites in incubates of ^{14}C-R89674 (10 μM) and ^{14}C-R90692 (10 μM) with human hepatocyte suspension and human liver subcellular samples (12000×g supernatants and microsomes) after 120-minute incubation.

	^{14}C-R89674		
	Hepatocyte	12000×g supernatants	Microsomes
R89674	38	-	81.4
R90692	47	95.3	12.1
Metabolite 3	10.3	2.2	-
Metabolite 5	2.2	-	-
Sum %	95.2	97.5	93.5
	^{14}C-R90692		
	Hepatocyte	12000×g supernatants	Microsomes
R90692	102.5	99.5	101
Metabolites	-	-	-
Sum %	102.5	99.5	101

Reviewer comments: It appears that the reported values were from the n=1 experiments.

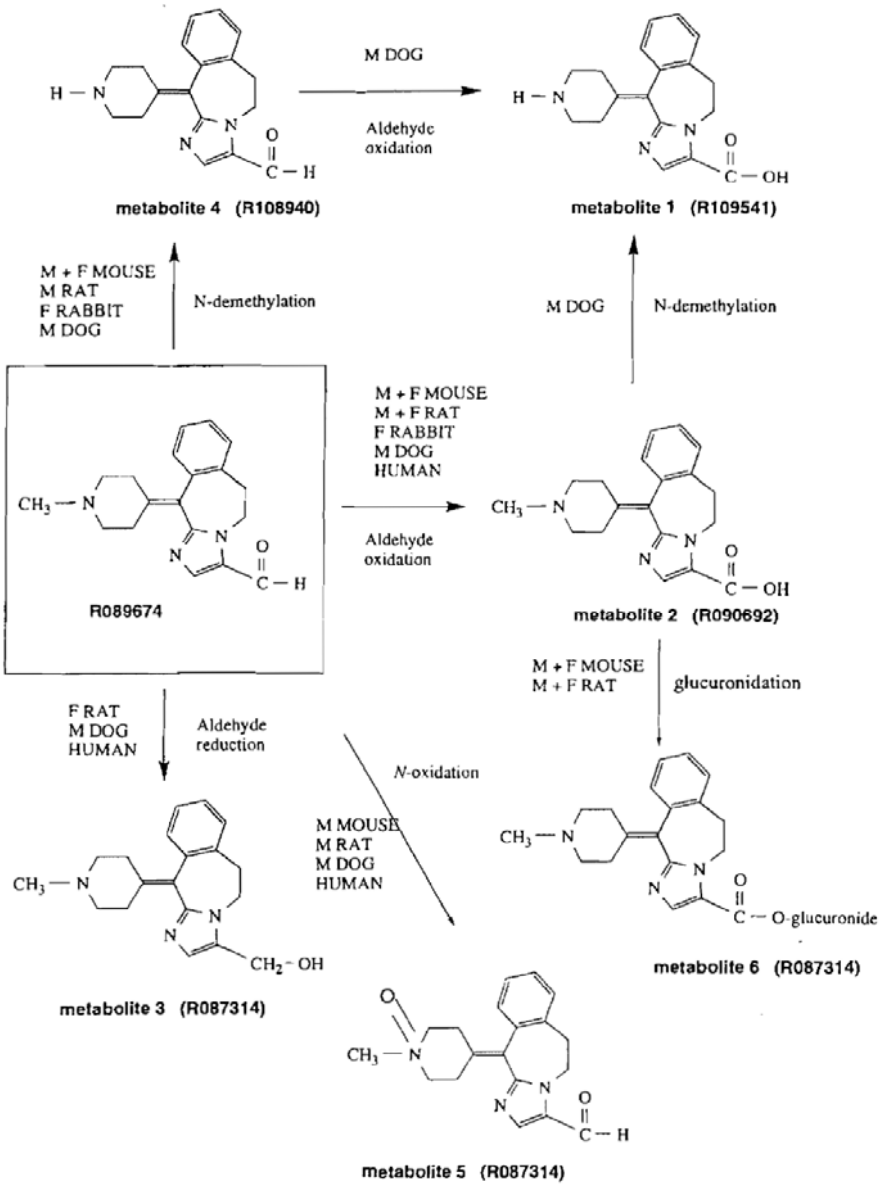


Figure 2: *In vitro* metabolic pathways proposed for alcaftadine (R89674) in the liver of preclinical speices and human. The identification of metablites was via LC/MS.

Conclusion:

Alcaftadine metabolism to the active metabolite R90692, presumably mediated by non-CYP450 enzymes, appears to be more extensive (qualitatively) in human hepatocytes and human liver 12000×g supernatant than in human liver microsomes. One limitation of the study is that the study was not conducted in replicates; therefore data reproducibility could not be evaluated.

The study is acceptable from a clinical pharmacology perspective.

4.2.3. In Vitro Drug-drug Interaction Studies

Study Number: N125331
Dates: March 3, 1997 to June 13, 1997
Title: *In vitro* determination of the drug-drug interaction potential of alcaftadine in human liver microsomes: effects of possible co-medication on the metabolism of alcaftadine

Objectives:
To study the effect of possible co-medications (adrenaline, beclomethasone, budesonide, codeine, diphenhydramine, erythromycin, ketotifen, loratadine, salbutamol, terfenadine, carboxyterfenadine, and theophylline) on the metabolism of ^{14}C-alcaftadine in human liver microsomes and human liver 12,000 x g supernatant fractions.

Methods:
The inhibition potential was studied in the presence of increasing inhibitor concentrations, based on typical clinical plasma concentrations of the inhibitors. Following incubation with the microsomal preparations or 12,000 x g supernatant fractions, samples were analyzed by radio-HPLC. The major metabolites in the incubation samples were characterized by comparison of HPLC chromatogram of the samples to that of a co-chromatographed mixture of reference samples.

Results and Discussion:
The metabolism of ^{14}C-alcaftadine was slow in human liver microsomes and led to the formation of the carboxylic acid metabolite R90692. R90692 was the only significant metabolite observed and represented 6.33 ± 0.35% of the injected radioactivity following the 30 min incubation. Following incubation with the 12,000 x g human liver supernatant fraction, the formation of R90692 was rapid and complete by 30 min. These results indicate that the metabolism of alcaftadine occurs primarily via non-CYP450 enzymes, presumably by aldehyde dehydrogenase, alcohol dehydrogenase, aldehyde oxidase or aldehyde reductase.

The overall metabolism of alcaftadine and the formation of the major metabolite R90692 in human liver microsomes or in human liver 12,000 x g supernatant were not inhibited after incubation with adrenaline, beclomethasone, budesonide, codeine, diphenhydramine, salbutamol, terfenadine, carboxyterfenadine, and theophylline. Ketotifen, loratidine, and erythromycin slightly affected the overall metabolism of alcaftadine and the formation of R90692 after incubation with human liver 12,000 × g. R90692 formation was significantly inhibited when alcaftadine was incubated with human liver microsome in the presence of ketotifen, loratidine and erythromycin. Erythromycin is an inhibitor of CYP3A4, however the other CYP3A4 inhibitors tested, budesonide, terfenadine and loratidine, did not result in competitive inhibition of alcaftadine metabolism at the concentrations tested. The potential for inhibition by CYP3A4 inhibitors would be of minor importance, however, since the cytosolic enzymes present in the 12,000 x g supernatant are involved in the biotransformation of alcaftadine, resulting in rapid oxidation to R90692. The lack of CYP3A4 in the metabolism of alcaftadine was further substantiated by a clinical drug interaction study following 2 mg oral dose of alcaftadine, in which co-administered ketoconazole was found not to inhibit the *in vivo* metabolism of alcaftadine as there was no changes in the plasma concentrations of alcaftadine and the AUC of the metabolite R90692 increased 10% following co-administration of ketoconazole.

Conclusion:
No clinically relevant interaction from the potential co-medications investigated in the present study on the metabolism of alcaftadine was to be expected.

These studies are acceptable from a clinical pharmacology perspective.

4.2.4. PK Study Following Ocular Administration

Study Number: 05-003-09
A Prospective, Single-Center, Open-Label Study of the Plasma Pharmacokinetics and Safety following Topical Administration of alcaftadine 0.25% Ophthalmic Solution as a Single and Repeated Dose in Healthy, Adult Subjects (Phase 1)

Dates: 17 September, 2005 to 2 October, 2005
Clinical investigator: (b) (4)
Analytical site: (b) (4)

OBJECTIVES:

The objectives of this study were to characterize the plasma pharmacokinetics and safety profile of alcaftadine 0.25% ophthalmic solution following a single bilateral dose and following multiple once-a-day doses in healthy adult subjects.

FORMULATION & ADMINISTRATION

Alcaftadine (Lot number: PD-0-5090) was formulated (Formulation #: PD-F-3730) as a sterile, light yellow, (b) (4) buffered ophthalmic solution containing benzalkonium chloride as a preservative and alcaftadine 0.25% as the active ingredient.

The inactive ingredients of the ophthalmic solution included the following:
- Dibasic sodium phosphate dihydrate USP (United States Pharmacopoeia, USP)
- Monobasic potassium phosphate NF (National Formulary, NF)
- Sodium chloride USP
- Edetate disodium dihydrate USP
- Benzalkonium chloride NF
- Purified water USP

A single drop (~34 μL/eye) of alcaftadine 0.25% ophthalmic solution was administered once daily in each eye for seven days (instilled between 7 am and 9 am).

STUDY DESIGN:

This was a prospective, single-center, open-label, Phase 1 study to characterize the plasma pharmacokinetics and overall safety of alcaftadine 0.25% ophthalmic solution, administered as a single drop in each eye for 7 days, in 13 healthy male and female adult volunteers (**Table 1**). Plasma pharmacokinetics was also assessed for R90692, the primary metabolite of alcaftadine.

Serial plasma samples were taken from each subject at the following time-points:
- Day 1 at pre-dose, and 0.25, 0.5, 1, 1.5, 2, 3, 4, 6, 8, 12 and 18 hours post-dose;
- Day 2 at 24-hour post-dose;
- Days 5 and 6 at pre-dose;
- Day 7 at pre-dose, and 0.25, 0.5, 1, 1.5, 2, 3, 4, 6, 8, 12, 18 and 24 hours post-dose.

Table 1: Demographic Characteristics

	Total (N=13)
Age (Years)	
n	13
Mean (SD)	31.6 (9.67)
Median	28
(Min,Max)	(18, 49)
Sex, n (%)	
Male	6 (46.2)
Female	7 (53.8)
Race, n (%)	
American Indian	0 (0.0)
Asian	0 (0.0)
African American	5 (38.5)
Pacific Islander	1 (7.7)
White	6 (46.2)
Other[a]	1 (7.7)
Ethnicity, n (%)	
Hispanic/Latino	2 (15.4)
Not Hispanic/Latino	11 (84.6)

ASSAY METHODOLOGY:

Plasma samples were assayed for the parent drug, alcaftadine, and its primary active metabolite, R90692. Plasma Samples were prepared using solid phase extraction and subsequently analyzed by LC/MS/MS using positive turbo-ionspray ionization mode and operating the instrument in the multiple-reaction-monitoring (MRM) mode. The validated concentrations ranged from 0.0100 ng/mL to 2.50 ng/mL for alcaftadine, and from 0.100 ng/mL to 25.0 ng/mL for R90692, using a 200 µL sample size.

Criterion	Alcaftadine	R90692	Comments
Conc. range, ng/mL	0.0100 – 2.50	0.100 – 25.0	satisfactory
LLOQ, ng/mL	0.0100	0.100	satisfactory
Linearity, r^2	> 0.99	> 0.99	satisfactory
Accuracy, % RE	-5.0 – 4.0 [a] -5.3 – 4.7 [b]	-4.7 – -0.2 [a] -6.1 – 2.5 [b]	satisfactory
Precision, % CV	1.1 – 11.9 [a] 2.3 – 6.4 [b]	1.7 – 12.2 [a] 2.1 – 6.2 [b]	satisfactory
Recovery	Evaluated for each analyte and internal standards at three QC concentrations (low, medium and high)		satisfactory
Specificity	Interference by endogenous compounds evaluated		satisfactory
Stability	Stable in plasma at room temperature up to 24 hours, extracted samples for 2 days when stored refrigerated, , stock solution stored at -20°C for more than 120 days, plasma when stored at -70°C for a period of 29 days, and following three freeze/thaw cycles		satisfactory

[a], QC samples; [b], calibration standards.

DATA ANALYSIS

Non-compartmental analysis was used to estimate key pharmacokinetic parameters including C_{max}, T_{max}, $AUC_{0\text{-}last}$, and the elimination $T_{1/2}$ for those subjects with plasma concentration values above the lower limits of quantification (0.0100 ng/mL for alcaftadine; 0.100 ng/mL for R90692).

RESULTS:

Alcaftadine
Plasma concentrations of alcaftadine on both Days 1 and 7 reached C_{max} around 0.25 hour post dose and were near or below the LLOQ (0.01 ng/mL) after approximately 2 hours post dose (**Figure 1 & 2)**. The mean C_{max} value (0.060 ng/mL) at Day 7 was higher than that at Day 1 (0.051 ng/mL). When C_{max} and AUC values were compared between Day 1 and Day 7 for each subject, the C_{max} ratio (Day7/Day1) ranged from 0.3 to 3.5, and the AUC ratio (Day7/Day1) for each subject ranged from 0.3 to 2.8 (**Table 2**).

No accumulation of alcaftadine in plasma was observed following repeated daily dosing, consistent with a short elimination half-life (< 1.5 hour) of alcaftadine in plasma.

When the paired t-test was used to assess if there is any statistically significant difference in C_{max} or AUC for alcaftadine between Day 1 and Day 7 for these 13 subjects, no significant difference (*p >0.05*) was observed for both C_{max} and AUC (**Table 3**).

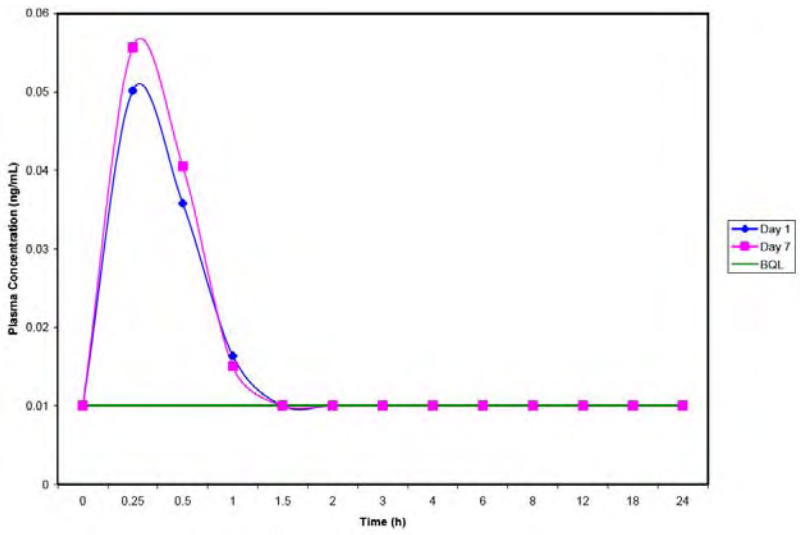


Figure 1: Mean plasma concentration-time profiles of alcaftadine on Day 1 and Day 7.
Concentrations at 2 hour post dose or later were below the lower limit of quantitation (0.0100 ng/mL, marked by the solid green line). *Source: Study report pk-0500309*

Table 2: Individual C_{max}, AUC, and within-subject ratios (Day7/Day1) for alcaftadine

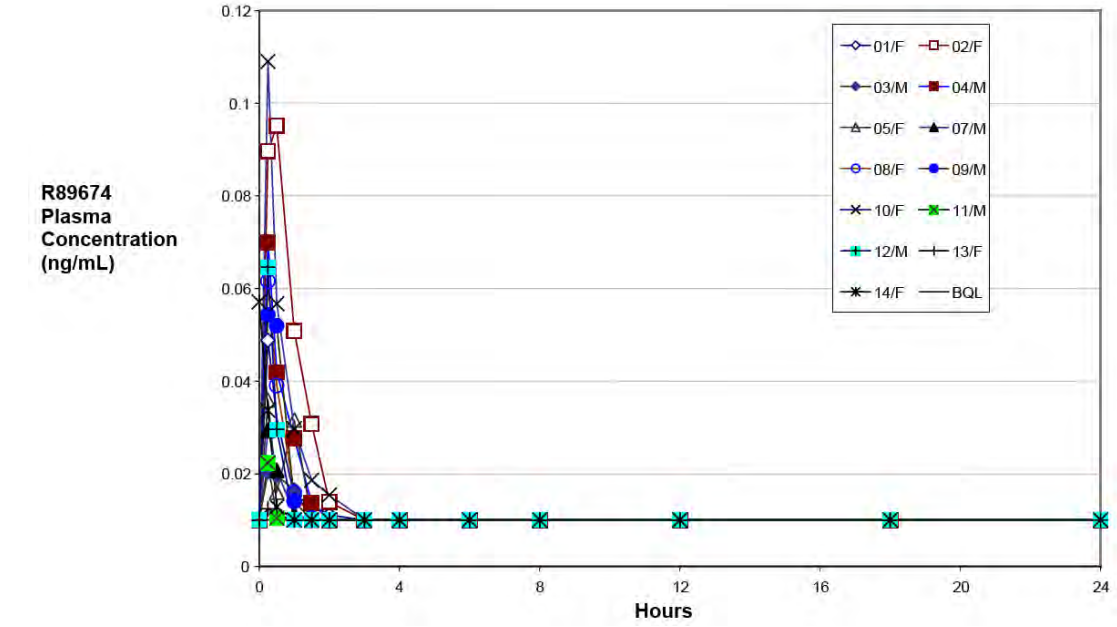
Subject No.	Day 1 Cmax, ng/mL	Day 7 Cmax, ng/mL	Cmax ratio: Day7/Day1	Day 1 AUC, ng*hr/mL	Day 7 AUC, ng*hr/mL	AUC ratio: Day7/Day1
1 F	0.049	0.04	0.8	0.045	0.024	0.5
2 F	0.095	0.09	0.9	0.108	0.106	1.0
3 M	0.02	0.022	1.1	0.017	0.007	0.4
4 M	0.07	0.107	1.5	0.052	0.057	1.1
5 F	0.036	0.028	0.8	0.023	0.02	0.9
7 M	0.029	0.074	2.6	0.017	0.041	2.4
8 F	0.062	0.111	1.8	0.042	0.068	1.6
9 M	0.054	0.057	1.1	0.038	0.042	1.1
10 F	0.109	0.062	0.6	0.093	0.066	0.7
11 M	0.022	0.076	3.5	0.012	0.021	1.8
12 M	0.065	0.019	0.3	0.02	0.006	0.3
13 F	0.012	0.03	2.5	NC	0.01	NA
14 F	0.034	0.064	1.9	0.013	0.036	2.8
Mean	0.051	0.060	1.5	0.040	0.039	1.2
SD	0.028	0.030	0.9	0.030	0.028	0.7

NC, not caculated because this subject had only one measurable concentration in samples from Day 1
NA, not applicable

Table 3: Paired t-test on the C_{max} and AUC values between Day 1 and Day 7 for alcaftadine and R90692 (Confidence interval = 0.95; two-sided)

	p-value for Cmax, Day 1 vs Day 7	p-value for AUC, Day 1 vs Day 7
R89674	0.325	0.821
R90692	0.038	0.104

(a)



(b)

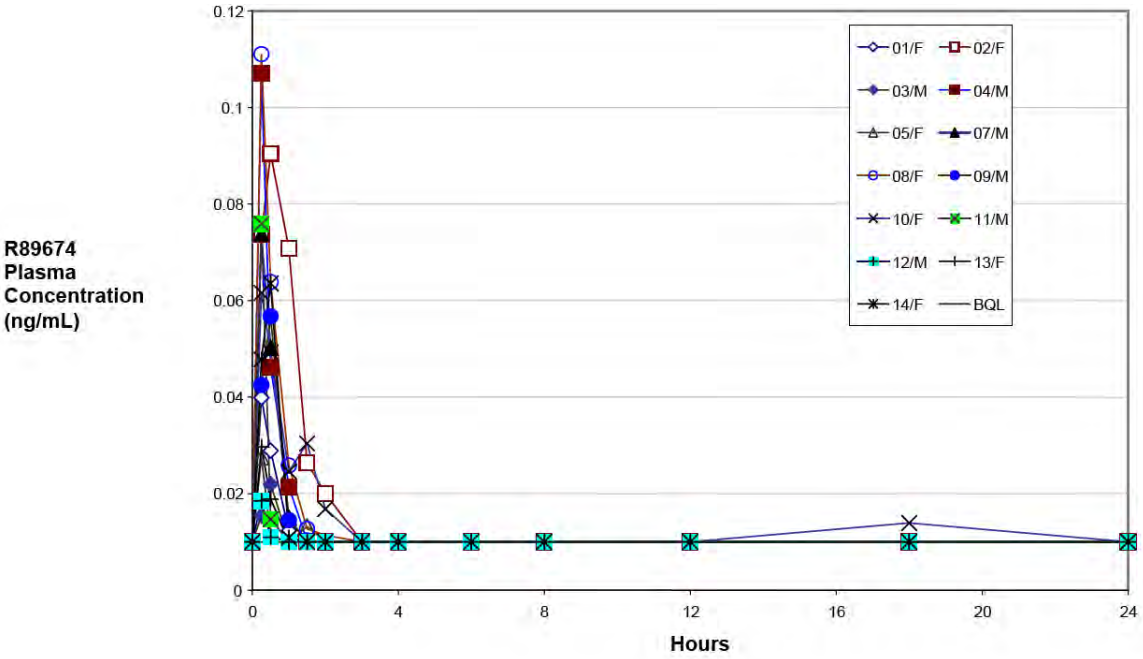


Figure 2: Individual plasma concentration-time profile of alcaftadine on Day 1(a) and Day 7(b). The lower limit of quantitation (0.0100 ng/mL) was marked by the solid line.

Source: Study report pk-0500309

R90692

Plasma concentrations of R90692 on both Days 1 and 7 reached C_{max} around 1 hour post dose and were near or below the LLOQ (0.1 ng/mL) after approximately 12 hours (**Figure 3 & 4)**. The mean C_{max} value (2.715 ng/mL) at Day 7 was lower than that at Day 1 (3.228 ng/mL). When C_{max} and AUC values were compared between Day 1 and Day 7 for each subject, the C_{max} ratio (Day7/Day1) ranged from 0.4 to 1.3 with a mean ratio at 0.8, and AUC ratio (Day7/Day1) ranged from 0.4 to 1.2 with a mean ratio at 0.9 (**Table 4**).

No accumulation of R90692 in plasma was observed following repeated daily dosing, consistent with a short elimination half-life (< 2.5 hour) of R90692 in plasma. Following topical administration, plasma exposure of the active metabolite R90692 is approximately 55-fold higher (C_{max}) or 25-fold higher (AUC) than that of alcaftadine.

When the paired t-test was used to assess if there is any statistically significant difference in C_{max} and AUC for R90692 between Day 1 and Day 7 for these 13 subjects, there was a significant difference in C_{max} between Day 1 and Day 7 (p=0.038), but not in AUC values (**Table 3**). The significantly higher C_{max} in Day 7 compared to that in Day 1 is not expected to have clinical significance.

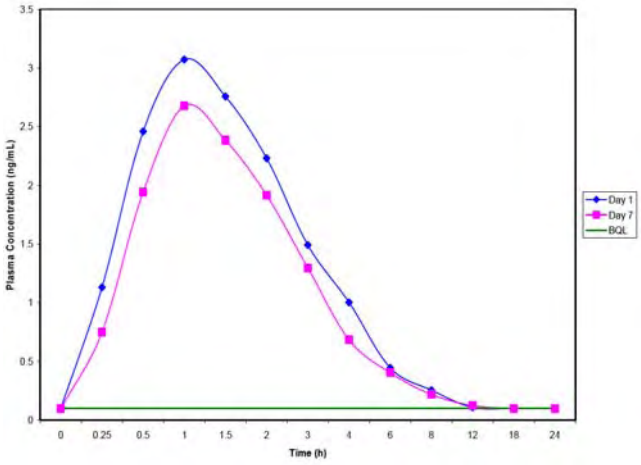
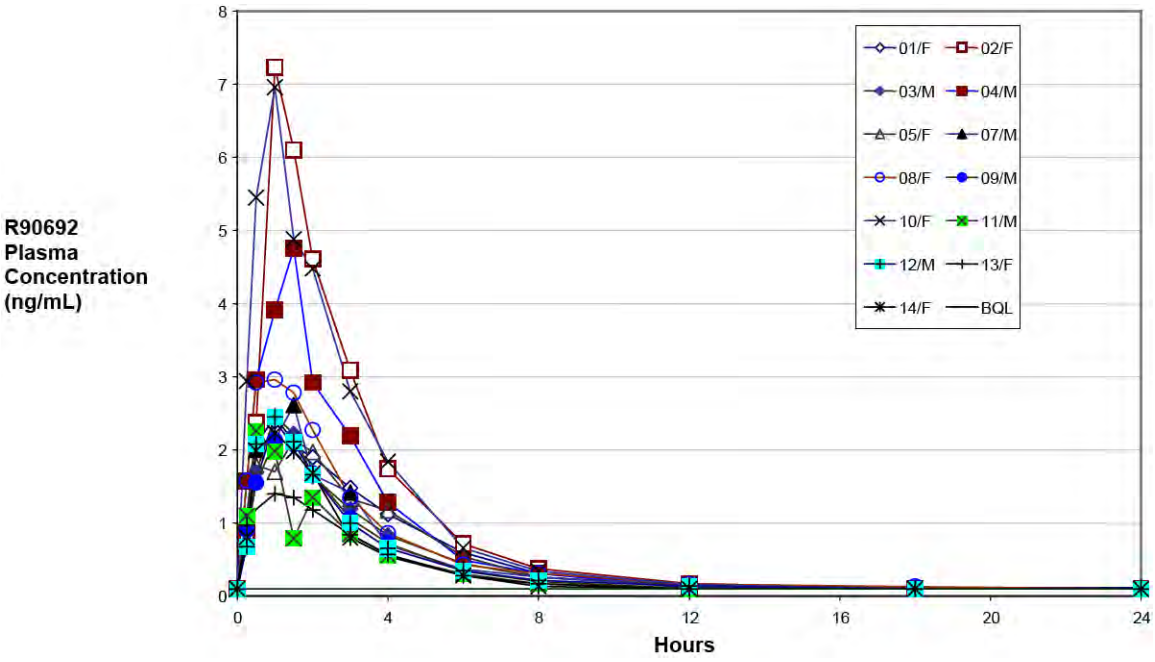


Figure 3: Mean plasma concentration-time profiles of R90692 on Day 1 and Day 7
Concentrations at 12-24 hour post dose were near or below the lower limit of quantitation (0.10 ng/mL, green solid line). *Source: Study report pk-0500309*

Table 4: Individual C_{max}, AUC, and within-subject ratios (Day7/Day1) for R90692

Subject No.	Day 1 Cmax, ng/mL	Day 7 Cmax, ng/mL	Cmax ratio: Day7/Day1	Day 1 AUC, ng*hr/mL	Day 7 AUC, ng*hr/mL	AUC ratio: Day7/Day1
1 F	2.39	2.35	1.0	11.42	7.7	0.7
2 F	7.23	5.64	0.8	19.87	22.57	1.1
3 M	2.46	1.31	0.5	8.78	5.61	0.6
4 M	4.75	5.31	1.1	13.96	15.83	1.1
5 F	2.14	1.34	0.6	9.7	4.01	0.4
7 M	2.61	1.58	0.6	8.74	5.62	0.6
8 F	2.96	3.73	1.3	11.54	12.4	1.1
9 M	2.13	2.2	1.0	7.87	7.37	0.9
10 F	6.96	5.43	0.8	20.3	17.35	0.9
11 M	2.26	2	0.9	5.8	6.96	1.2
12 M	2.45	1.04	0.4	8.14	3.13	0.4
13 F	1.4	1.03	0.7	5.22	4.62	0.9
14 F	2.23	2.34	1.0	6.63	7.37	1.1
Mean	3.228	2.715	0.8	10.613	9.272	0.9
SD	1.803	1.653	0.2	4.655	5.709	0.3

(a)



(b)

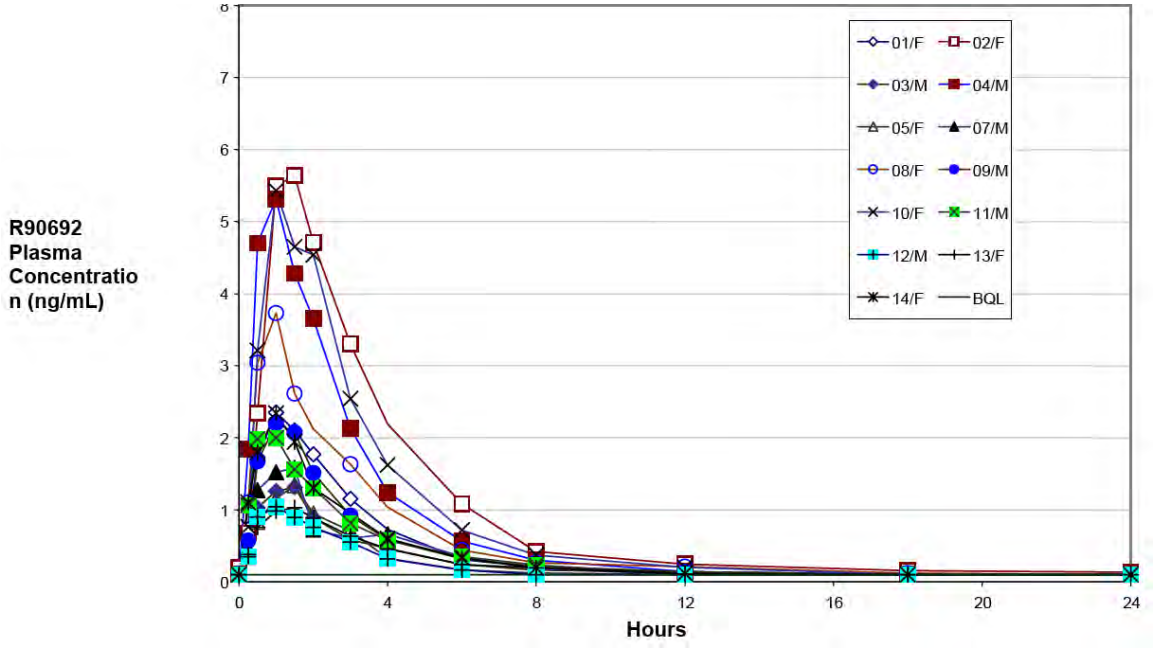


Figure 4: Individual plasma concentration-time profile of R90692 on Day 1(a) and Day 7(b).
The lower limit of quantitation (0.10 ng/mL) was marked by the solid line.
Source: Study report pk-0500309

Pharmacokinetic parameters for R89774 and R90692 are summarized in **Table 5**.

Table 5: Summary of Pharmacokinetic Parameters (Mean ± SD) for Alcaftadine (i.e. R89674) and R90692 Following Single and Multiple Ocular Doses of Alcaftadine 0.25% Ophthalmic Solution (n=13).

	R89674		R90692	
	Day 1	Day 7	Day 1	Day 7
Mean C_{max}, ng/mL(range)	0.051 ± 0.028 (0.01, 0.11)	0.060 ± 0.030 (0.02, 0.11)	3.228 ± 1.803 (1.40, 7.23)	2.715 ± 1.653 (1.03, 5.64)
Median T_{max}, hr (Min, Max)	0.25 (0.15, 0.67)	0.25(0.25, 0.5)	1.05(0.8, 1.58)	1.00 (1.00, 1.5)
Mean AUC, ng*hr/mL (range)	0.040 ± 0.030 (0.01, 0.11)	0.039 ± 0.028 (0.01, 0.11)	10.613 ± 4.655 (5.22, 20.3)	9.272 ± 5.709 (3.13, 22.57)
$T_{½}$. hr	1.1 ± 0.9	0.7 ± 0.5	2.0 ± 0.2	2.1 ± 0.4

Gender difference
Potential gender difference was evaluated using the student t-test to compare C_{max} and AUC values (**Table 6**) for alcaftadine and R90692, no statistically significant differences (*p>0.05*) were observed between male and females subjects (**Table 7**).

Table 6: The mean (SD) C_{max} and AUC values (pooled data from Days 1 and 7) of alcaftadine and R90692 in male and female healthy subjects

	Alcaftadine		R90692	
	C_{max} (SD) ng/mL	$AUC_{0\text{-last}}$ (SD) ng*hr/mL	C_{max} (SD) ng/mL	$AUC_{0\text{-last}}$ (SD) ng*hr/mL
Male	0.051(0.029)	0.028(0.018)	2.508(1.275)	8.151(3.557)
Female	0.059(0.031)	0.050(0.035)	3.369(2.091)	11.478(6.241)

Table 7: The p-values from the Student t-test to assess the potential gender differences for C_{max} and AUC values of alcaftadine and R90692. (Confidence interval = 0.95; two-sided)

	male vs female, Cmax	male vs female, AUC
R89674	0.538	0.053
R90692	0.227	0.116

SAFETY RESULTS:

All 13 subjects completed the study. There were no ocular adverse events, serious adverse events, deaths reported in this study.

COMPARISON WITH ORAL PHARMACOKINETICS

The pharmacokinetics of alcaftadine and the metabolite R90692 following oral administration were investigated in healthy subjects following single (0.25, 0.5, 1, 2, 4, and 8 mg*; Report N122431-not reviewed*) and multiple daily dosing (0.5, 1, and 2 mg daily for 8 days; *Report N122432-not reviewed*). The plasma samples were assayed by HPLC and GC/MS methods with a

lower quantification limit (0.5 ng/mL). At the highest single dose of 8 mg, the plasma concentrations of alcaftadine at all timepoints were below 0.5 ng/mL. No accumulation for either alcaftadine or R90692 was observed following oral multiple dialy dosing. On a dose-adjusted basis, the bioavailability of R90692 following multiple daily ophthalmic administration of alcaftadine was approximately half of that obtained following multiple daily oral administration of alcaftadine (**Table 8**). R90692 exhibited similar T_{max} and $t_{1/2}$ following ophthalmic dose as those following oral dose of alcaftadine.

The urinary excretion of R90692 following oral administration of alcaftadine was determined in the oral dose studies. Following multiple oral doses of 0.5 to 2 mg of alcaftadine, the renal excretion of R90692 was from 55% to 60% of the alcaftadine dose. Following ophthalmic dose of alcaftadine, R90692 is expected to be excreted primarily unchanged in the urine as well.

Table 8: Summary of Pharmacokinetic Parameters for R90692 Following Multiple Ophthalmic or Oral Daily Doses of alcaftadine

Parameter	Ophthalmic 0.25% Solution Day 7	0.5 mg Oral Day 8	1 mg Oral Day 8	2 mg Oral Day 8
Cmax (ng/mL)	2.72 ± 1.72	13.6 ± 3.3	31.4 ± 5.7	55.8 ± 7.6
Median Tmax (h) Range	1.0 (1.0 -1.5)	1.0 (0.5 -1.0)	1.0 (0.5 – 2.0)	0.75 (0.5 – 2.0)
AUC $_{0\text{-last}}$ (ng·hr/mL)	9.27 ± 5.94	49.8 ± 10.3	110 ± 15	203 ± 20
Dose-adjusted AUC ($AUC_{0\text{-last}}$ /Dose)	54.5	99.6	110.0	101.5
t ½ (h)	2.10 ± 0.41	1.8 ± 0.2	1.7 ± 0.3	1.9 ± 0.2
Urinary excretion (% Dose)	NA	56.8 ± 2.8	60.1 ± 5.3	55.0 ± 6.6

Note: Total daily ophthalmic dose = 0.18 mg. The LLOQ in the oral pharmacokinetics study was 0.5 ng/mL for both alcaftadine and R90692. Alcaftadine concentrations were below the LLOQ following single dose (8 mg) or multiple dose (2 mg per day for 8 days) oral administration.

SPONSORS CONCLUSIONS:

Systemic exposure (C_{max} and AUC) of the parent drug (alcaftadine) after a single bilateral dose (Day 1) of alcaftadine 0.25% ophthalmic solution were comparable to those observed after multiple daily doses (Day 7). Plasma concentrations were low and declined rapidly, falling near or below the lower limit of quantification (0.010 ng/mL) by approximately 3 hours after dosing. No accumulation was observed.

Systemic exposure (C_{max} and AUC) of the primary metabolite, R90692, after a single bilateral dose (Day 1) of alcaftadine 0.25% ophthalmic solution were comparable to those observed after multiple daily doses (Day 7). Plasma concentrations were low and declined rapidly, falling near or below the lower limit of quantification (0.100 ng/mL) by 12 hours after dosing. No accumulation was observed.

Alcaftadine was found to be safe when administered once daily as a single drop of 0.25% ophthalmic solution in both eyes to healthy adult volunteers for a period of 7 days.

REVIEWER'S ASSESSMENT & RECOMMENDATION:

Results from Study 05-003-09 adequately described the pharmacokinetics of alcaftadine and its active metabolite R90692 in plasma following repeated daily ocular topical administration of

alcaftadine 0.25% ophthalmic solution in healthy, adult subjects. The sponsor's conclusions are valid. The reviewer has additional comments as follows:

- A significantly higher plasma C_{max} value for R90692 on Day 1 was observed compared to that on Day 7 (p =0.038). However, this difference is not expected to be clinically significant because the systemic exposure is considered low and the pharmacological target is not in the blood.
- No gender difference was observed in alcaftadine and R90692 exposure in terms of plasma C_{max} and AUC (p>0.05).
- Following topical administration, plasma exposure of the active metabolite R90692 was approximately 55-fold higher (C_{max}) or 25-fold higher (AUC) than that of alcaftadine.
- Following ophthalmic administration of alcaftadine, R90692 is expected to be excreted primarily unchanged in the urine.

Application Type/Number	Submission Type/Number	Submitter Name	Product Name
NDA-22134	ORIG-1	VISTAKON PHARMACEUTICALS LLC	(b) (4) OPHTHALMIC SOLUTION

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/s/

YONGHENG ZHANG
05/28/2010

CHARLES R BONAPACE
05/28/2010

CLINICAL PHARMACOLOGY NDA FILEABILITY CHECKLIST

NDA:	22-134
Drug Name:	Alcaftadine (b) (4) ™)
Applicant:	Vistakon Pharmaceutical LLC
Submission Date:	September 29, 2009
Filing Date:	November 28, 2009
PDUFA Date:	July 28, 2010
OCP Primary Reviewer:	Yongheng Zhang, Ph D
OCP Team Leader:	Charles Bonapace, Pharm D

QUESTION	***YES***	***NO***	***NA***	***COMMENTS***
Fileability: ***Is the Clinical Pharmacology section of the application fileable?*** ***(if 'NO', please comment as to why it is not fileable)***	Yes			
Fileability Review Components				
1. Is the clinical pharmacology section of the NDA organized in a manner to allow substantive review to begin (including a table of contents, proper pagination, reference links, etc.)?	☒	☐	☐	This is an eCTD NDA. TOC, hyperlinks, reference et al OK
2. Are the clinical pharmacology studies of appropriate design and breadth of investigation to meet the basic requirements for approvability of this product?	☒	☐	☐	
3. If multiple formulations were used in the clinical development of the product, does the NDA contain appropriate biopharmaceutics information to allow comparison between the clinical development and to-be-marketed product(s) (i.e. pivotal BE)?	☒	☐	☐	• Formulations changed in the development • The proposed commercial formulation supported by one Phase 1 safety study & one Phase 3 pivotal efficacy study
4. If unapproved products or altered approved products were used as active controls, was bioequivalence to the approved product demonstrated?	☐	☐	☒	
5. Are complete and relevant bioanalytical reports included in the NDA submission?	☒	☐	☐	
6. If applicable, was the sponsor's request for a waiver of the requirement for submission of in vivo bioavailability data included in the NDA submission?	☐	☐	☒	
7. Are complete datasets supporting the clinical pharmacology studies included in the NDA submission?	☒	☐	☐	

OCP Primary Reviewer _____ Date _____

OCP Team Leader _____ Date _____

Application Type/Number	Submission Type/Number	Submitter Name	Product Name
NDA-22134	ORIG-1	VISTAKON PHARMACEUTICALS LLC	(b) (4) OPHTHALMIC SOLUTION

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/s/

YONGHENG ZHANG
11/16/2009

CHARLES R BONAPACE
11/17/2009