

**CENTER FOR DRUG EVALUATION AND
RESEARCH**

APPLICATION NUMBER:
204251Orig1s000

PHARMACOLOGY REVIEW(S)

**DEPARTMENT OF HEALTH AND HUMAN SERVICES
PUBLIC HEALTH SERVICE
FOOD AND DRUG ADMINISTRATION
CENTER FOR DRUG EVALUATION AND RESEARCH**

PHARMACOLOGY/TOXICOLOGY NDA MEMORANDUM

TO:	The file
FROM:	Andrew J. McDougal, Ph.D., D.A.B.T. Pharmacologist. Division of Transplant and Ophthalmology Products (DTOP, Office of Antimicrobial Products (OAP), CDER, HFD-590
THROUGH:	Lori E. Kotch, Ph.D., D.A.B.T., DTOP
Review Division:	Division of Transplant and Ophthalmology Products (DTOP), Office of Antimicrobial Products (OAP), CDER, HFD-590
File #s:	NDA 204251 (Simbrinza®)
SPONSOR:	Alcon Research, Ltd. (Alcon)
PRODUCT:	Brinzolamide 1% / brimonidine tartrate 0.2 % ophthalmic suspension
INDICATION:	Treatment of elevated intraocular pressure in patients with open-angle glaucoma or ocular hypertension
SYNOPSIS	P/T labeling review

Summary

- This reviewer concludes that using the nonclinical exposure multiples from the Alphagan (brimonidine 0.2%) label (NDA 20613) to describe the nonclinical exposure multiples for the brimonidine exposure of Simbrinza (brimonidine 0.2%) is appropriate and consistent.
- For the brimonidine rat embryofetal study, the Applicant calculated an exposure margin of 24-fold (instead of 100-fold for Alphagan), but based on the data limitations, the Applicant proposes “to keep the label the same with respect to exposure multiples based on administered dose”

Background

- The P/T review (McDougal, 3/15/2013, NDA 2042510 proposed draft labeling revisions based on the Applicant’s 2/19/2013 draft label (SD #15).
- As documented in DARRTS (Milstein, 3/18/2013), two nonclinical comments were conveyed to the Applicant.
 - Question 1¹ was the subject of a teleconference (March 26, 2013); due to an

¹ “Regarding the two year rat and mouse oral carcinogenicity studies for brinzolamide (summarized in the label drafts and in NDA module 2.6.6), the study reports were not identified either in NDA 204251 or in the NDA for the listed drug. The CDER Carcinogenicity Assessment Committee (CAC)’s review of the study reports was not identified in the NDA or the referenced NDA. Please provide the location of these

- oversight, Alcon had not yet submitted the carcinogenicity studies (to any FDA file). DTOP concluded that the carcinogenicity studies are not pivotal to the safety review, and the study reports will not be reviewed prior to the Agency Action. Please see DARRTs for more information.
- Question 2¹ requested that the Applicant provide exposure multiples for the nonclinical doses mentioned in the label, if adequate data are available.
- The Applicant formally replied to the 3/18/2013 email on April 3, 20113 (SD #18). Regarding question 2:
 - For brinzolamide:
 - the Applicant owns the relevant nonclinical and clinical data (i.e. Alcon submitted NDA 20816)
 - Alcon proposes to keep expressing the nonclinical doses relative to the human ophthalmic dose (mg/kg/day). This is acceptable, and consistent with the previous brinzolamide label (NDA 20816).
 - In response to DTOP's request, Alcon attempted to calculate an exposure margin based on AUC: "using the data from the available PK/TK data provides a systemic exposure margin of 2.9 to 3.0 fold based on a human TID systemic exposure of 14.2 µM (TDOC-0014794), and the lowest mean rat (42.6 µM) and rabbit (40.8 µM) systemic exposures (described above)." However, Alcon does not consider this estimate reliable.
 - For brimonidine:
 - The Applicant reported that it does not have access to or right-of-reference to the nonclinical results described in the label.
 - In the 4/06/2013 response, the Applicant attempted to calculate an exposure estimate for the rat developmental study based on information gleaned from the Summary Basis of Approval (SBA) for Alphagan (i.e. the P/T review for NDA 20613). However, because Alcon does not have right-of-reference to the data used for their exposure estimate, their exposure estimate was not used to support safety or for labeling.

Revised P/T Labeling Recommendations:

Additional changes recommended to the internal April 8, 2013 draft label:

"8.1 Pregnancy ...

Developmental toxicity studies performed in rats with oral doses of 0.66 mg brimonidine base/kg revealed no evidence of harm to the fetus. Dosing at this level resulted in a plasma drug concentration **approximately 100** ^{(b) (4)} times higher than that seen in

study reports (e.g. date submitted, file numbers they were submitted to)."

¹ "It is preferable to provide exposure multiples based on systemic AUC data in nonclinical sections 8 and 13. If adequate pharmacokinetic/toxicokinetic data are available, please calculate exposure multiples based on systemic AUC data for label sections 8 and 13, and provide the datasets used to make these calculations."

humans at the recommended human ophthalmic dose. In animal studies, brimonidine crossed the placenta and entered into the fetal circulation to a limited extent.”

...

“13 NONCLINICAL TOXICOLOGY

13.1 Carcinogenesis, Mutagenesis, Impairment of Fertility

(b) (4) The following tests for mutagenic potential of brinzolamide were negative: (1) *in vivo* mouse micronucleus assay; (2) *in vivo* sister chromatid exchange assay; and (3) Ames *E. coli* test. The *in vitro* mouse lymphoma forward mutation assay was negative in the absence of activation, but positive in the presence of microsomal activation. In this assay, there was no consistent dose-response relationship to the increased mutation frequency and cytotoxicity likely contributed to the high mutation frequency. Carbonic anhydrase inhibitors, as a class, are not mutagenic and the weight of evidence supports that brinzolamide is consistent with the class. In reproduction studies of brinzolamide in rats, there were no adverse effects on the fertility or reproductive capacity of males or females at doses up to 18 mg/kg/day (180 times the recommended human ophthalmic dose).

Brimonidine tartrate was not carcinogenic in either a 21-month mouse or 24-month rat study. In these studies, dietary administration of brimonidine tartrate at doses up to 2.5 mg/kg/day in mice and 1 mg/kg/day in rats resulted in plasma drug concentrations (b) (4) 80 (b) (4) and 120 (b) (4) times higher than the human plasma drug level at the recommended clinical dose, respectively. Brimonidine tartrate was not mutagenic or cytogenic in a series of *in vitro* and *in vivo* studies including the Ames test, chromosomal aberration assay in Chinese Hamster Ovary (CHO) cells, a host-mediated assay and cytogenic studies in mice, and a dominant lethal assay. In reproductive studies performed in rats with oral doses of 0.66 mg brimonidine base/kg (b) (4) approximately 100 times the plasma drug concentration level seen in humans following multiple ophthalmic doses) (b) (4) fertility was not impaired.”

P/T review notes

- A theoretical concern regarding labeling has been resolved.
 - Alcon does not have right-of-reference to the brimonidine embryofetal study or carcinogenicity studies. This reviewer did not obtain the studies (for the PK results).
 - The Alphagan 0.2% label (NDA 20613) presents the exposure margins based on the systemic exposure in humans following topical ocular exposure to brimonidine 0.2%.
 - The concern was that if the clinical systemic exposure to brimonidine from Simbrinza was different than the exposure to brimonidine from Alphagan 0.2%, then P/T could not use the label information from Alphagan 0.2% for Simbrinza. The Applicant did not claim comparability in the 4/06/2013 response.

- This theoretical concern is resolved by the Clinical Pharmacology review of NDA 204251: Dr. Zhang's review (3/22/2013, NDA 20451) reports PK data for Alcon's clinical trial # C-10-010, which compared Alcon's Brinzolamide/Brimonidine 1%/0.2% solution against reference therapy including brimonidine tartrate ophthalmic solution 0.2% (1 drop in both eyes 3 times daily or 1 drop in both eyes 2 time daily). Dr. Zhang concluded that the "steady-state plasma PK of brimonidine following topical ocular administration of the fixed dose combination of Brinzolamide/Brimonidine dosed TID or BID, were comparable to those observed after administration of the individual active components alone"
 - For TID dosing of Brinz/Brim, the brimonidine plasma concentrations on D21 were $C_{max} = 0.0479$ ng/ml and $AUC = 0.183$ ng*hr/ml.
 - For TID dosing of Brinz/Brim, the brimonidine plasma concentrations on D107 were $C_{max} = 0.0419$ ng/ml and $AUC = 0.145$ ng*hr/ml.
 - The report for trial # C-10-010 (report # TDOC-0014794, submitted to the original NDA) notes (pdf page 79) that for the brimonidine 0.2%, "the test articles are identical to the marketed products of AZOPT and ALPHAGAN". For the brimonidine 0.2%, "lot # 10-501227-1, FID 101461" was used.
 - Therefore, this reviewer concludes that using the same brimonidine exposure margins from the NDA 20613 label for Simbrinza label is appropriate
- For NDA 20613 (Alphagan®, brimonidine 0.2%, discontinued), the most recent label¹ is dated 12/20/2001 (for both brimonidine 0.5% and 0.2%):
 - "Reproductive studies performed in rats with oral doses of 0.66 mg base/kg revealed no evidence of harm to the fetus due to ALPHAGAN®. Dosing at this level produced 100 times the plasma drug concentration level seen in humans following multiple ophthalmic doses."
 - "No compound-related carcinogenic effects were observed in either mice or rats following a 21 month and 24-month study, respectively. In these studies, dietary administration of brimonidine tartrate at doses up to 2.5 mg/kg/day in mice and 1.0 mg/kg/day in rats achieved ~77 and 118 times, respectively, the plasma drug concentration estimated in humans treated with one drop ALPHAGAN® into both eyes 3 times per day."
 - Note: The exposure multiples presented in the labels² for Alphagan P, brimonidine 0.15% (Allergan's NDA 21262) and brimonidine 0.1% (Allergan's NDA 21770) for the mouse and rat carcinogenicity studies do not quite scale linearly with the exposure margins for these studies as presented in the label for Alphagan (brimonidine 0.2%). The reviewer presumes that the difference in the exposure margins reflects observed differences in clinical exposure (i.e. exposure from the 0.1% and 0.15% doses do not quite scale linearly to the exposure from the 0.2% doses). However, the labeling lacks sufficient detail to check this presumption.

¹ Beginning on pdf page 8, via http://www.accessdata.fda.gov/drugsatfda_docs/label/2001/21262s6lbl.pdf

² http://www.accessdata.fda.gov/drugsatfda_docs/label/2010/021262s020_021770s004lbl.pdf

This is a representation of an electronic record that was signed electronically and this page is the manifestation of the electronic signature.

/s/

ANDREW J MCDOUGAL
04/09/2013

LORI E KOTCH
04/09/2013

**DEPARTMENT OF HEALTH AND HUMAN SERVICES
PUBLIC HEALTH SERVICE
FOOD AND DRUG ADMINISTRATION
CENTER FOR DRUG EVALUATION AND RESEARCH**

PHARMACOLOGY/TOXICOLOGY NDA REVIEW AND EVALUATION

Application number: 204251
Supporting documents (SD): SD0 (New NDA)
Applicant's letter date: June 19, 2012
CDER stamp date: June 19, 2012
Product: Brinzolamide 1%/ brimonidine tartrate 0.2%
suspension
Indication: Reduction of elevated intraocular pressure in
patients with open-angle glaucoma or ocular
hypertension
Applicant: Alcon Research, Ltd.
Review Division: Division of Transplant and Ophthalmology
Products (DTOP), Office of Antimicrobial
Products (OAP), CDER, HFD-590
Reviewer: Andrew J. McDougal, Ph.D., D.A.B.T., DTOP
Supervisor/Team Leader: Lori E. Kotch, Ph.D., D.A.B.T., DTOP
Division Director: Renata Albrecht, M.D., DTOP
Project Manager: Judit Milstein (temporary)

TABLE OF CONTENTS

1	EXECUTIVE SUMMARY	5
1.1	RECOMMENDATIONS	5
1.2	BRIEF DISCUSSION OF NONCLINICAL FINDINGS	8
2	DRUG INFORMATION	11
3	STUDIES SUBMITTED.....	17
4	PHARMACOLOGY	21
4.1	PRIMARY PHARMACOLOGY	21
4.2	SECONDARY PHARMACOLOGY	34
4.3	SAFETY PHARMACOLOGY	34
5	PHARMACOKINETICS/ADME/TOXICOKINETICS	35
5.1	PK/ADME.....	35
5.2	TOXICOKINETICS	43
6	GENERAL TOXICOLOGY	44
6.1	SINGLE-DOSE TOXICITY	44
6.2	REPEAT-DOSE TOXICITY	44
7	GENETIC TOXICOLOGY	65
8	CARCINOGENICITY	65
9	REPRODUCTIVE AND DEVELOPMENTAL TOXICOLOGY	66
10	SPECIAL TOXICOLOGY STUDIES.....	66
11	INTEGRATED SUMMARY AND SAFETY EVALUATION.....	66

Table of Tables

Table 1: Composition of the drug product	13
Table 2: CDER files referenced by IND 106293	15
Table 3: Manufacturing files (MFs) referenced by the applicant.....	15
Table 4: FDA-approved NDAs and ANDAs for brimonidine-containing products	16
Table 5: Brinzolamide inhibited HCA-I, HCA-II and HCA-IV activity <i>in vitro</i> (report # 001:39320:0696)	21
Table 6: Brinzolamide binds to HCA-I and HCA-2 <i>in vitro</i> (report # 051-39310-0796) ..	22
Table 7: Four treatments with 1% brinzolamide over 8 days reduced IOP and aqueous flow in pigmented rabbits (report # 002:41:0301)	24
Table 8: Topical ocular brinzolamide (1% and 2%) reduced IOP in normotensive and hypertensive eyes of monkeys (report # 311:39600:0692).....	26
Table 9: Topical brinzolamide reduced IOP in the eyes of Dutch belted rabbits (report # 288:39600:0592)	30
Table 10: 2% brinzolamide topical ocular administration decreased IOP and increased optic nerve head blood flow in rabbits (report # 018:39500:1097).....	34
Table 11: Formulations for the rabbit single-dose topical ocular distribution study (report # TDOC-000954).....	36
Table 12: Brimonidine PK for the rabbit single-dose distribution study (report # TDC-0009954)	39
Table 13: Brinzolamide PK for the rabbit ocular distribution study (report # TDC-0009954)	39
Table 14: Brinzolamide (AL-4862) PK after single bilateral dosing and 14 days bilateral dosing in male rabbits (report # TDOC-0014507)	42
Table 15: Brimonidine (AL-8923) PK after single bilateral dosing and 14 days bilateral dosing in male rabbits (report # TDOC-0014507)	43
Table 16: Brimonidine-induced penile erection observed in the 6-week rabbit toxicity study (report # TDOC-0010074).....	46
Table 17: The duration of rabbit penile erection showed a dose-response for brimonidine, and Alcon's brinzolamide 2%/brimonidine tartrate 0.2% appeared more active than Falcon's brimonidine tartrate 0.2% (report # TDOC-0010074).....	47
Table 18: Reported incidence of female urogenital swelling in the 6-week rabbit study (report # TDOC-0010074)	48
Table 19: Reported incidence of sedation in the 6-week rabbit study (report # TDOC-0010074)	48
Table 20: Treatment-related ophthalmic changes (increased corneal thickness, decreased IOP) in the 6-week rabbit study (report # TDOC-0010074)	49
Table 21: Selected serum chemistry (D38) for the 6-week rabbit study (report # TDOC-0010074)	50
Table 22: PK for the 6-week rabbit study (report # TDOC-0010074)	51
Table 23: Reported treatment-related clinical signs: penile erection, urogenital swelling and discoloration in the 9-month rabbit study (report # TDOC-0013267).....	56
Table 24: Selected data showing the slight treatment-related suppression of body weight gain in the 9-month rabbit study (report # TDOC-0013267).....	57

Table 25: Incidences of decreased food consumption in the 9-month rabbit study (report # TDOC-0013267).....	58
Table 26: Treatment-related ophthalmic changes (increased corneal thickness, decreased IOP) in the 9-month rabbit study (report # TDOC-0013267).....	58
Table 27: Treatment-related decrease in serum glucose in the 9-month rabbit study (report # TDOC-0013267)	60
Table 28: Results weakly suggest a treatment-related increase in liver weights in the 9-month rabbit study (report # TDOC-0013267).....	61
Table 29: Selected histopathology results for the 9-month rabbit study (report # TDOC-0013267)	63
Table 30: TK summary for the 9-month rabbit study (report # TDOC-0013267)	64
Table 31: Dose comparison: clinical label versus the 6-week & 9-month rabbit studies	67
Table 32: Clinical brimonidine summary PK data (TID).....	68
Table 33: Clinical brimonidine summary PK data (BID).....	68
Table 34: Clinical brimonidine summary PK data (TID).....	69
Table 35: Clinical brimonidine summary PK data (BID).....	69
Table 36: Clinical brimonidine summary PK data (TID or BID).....	70

Table of Figures

Figure 1: Structure of brinzolamide	11
Figure 2: Structure of brimonidine tartrate.....	13
Figure 3: Differences in brinzolamide uptake into the bulbar conjunctiva (C_{max} and AUC) for the rabbit single-dose distribution study (report # TDOC-0009954)	38
Figure 4: Differences in brimonidine uptake into the bulbar conjunctiva (C_{max} and AUC) for rabbit single-dose distribution study (report # TDOC-0009954)	38

1 Executive Summary

On June 19, 2012, Alcon Research, Ltd. submitted a new drug application (NDA for a prescription product (Rx) under the 505(b)(2) pathway via electronic media for the fixed combination ophthalmic suspension of Brinzolamide 1% and Brimonidine Tartrate 0.2% (Brin/Brim).

The applicant proposed an indication of “reduction of elevated intraocular pressure in patients with open-angle glaucoma or ocular hypertension.”

1.1 Recommendations

1.1.1 Approvability

P/T did not identify substantial new safety issues. Pending resolution of the question regarding the brinzolamide carcinogenicity studies and agreement on the label, P/T has no objection to approval.

1.1.2 Additional Non Clinical Recommendations

Two P/T issues are unresolved. This reviewer recommends that these draft comments be conveyed:

1. Regarding the two year rat and mouse oral carcinogenicity studies for brinzolamide (summarized in the label drafts and in NDA module 2.6.6), the study reports were not identified either in NDA 204251 or in the NDA for the listed drug. The CDER Carcinogenicity Assessment Committee (CAC)’s review of the study reports was not identified in the NDA or the referenced NDA. Please provide the location of these study reports (e.g. date submitted, file numbers they were submitted to).
2. It is preferable to provide exposure multiples based on systemic AUC data in nonclinical sections 8 and 13. If adequate pharmacokinetic/toxicokinetic data are available, please calculate exposure multiples based on systemic AUC data for label sections 8 and 13, and provide the datasets used to make these calculations.

1.1.3 Labeling

- For Azopt® (brinzolamide ophthalmic suspension 1%), the most recent label¹ (Alcon's NDA 20816) is dated March 8, 2011.
- Alcon's NDA 21764 for Qoliniana® (brimonidine tartrate ophthalmic solution 0.15%) has a recent label dated March 27, 2012.² This appears to be the most recently approved brimonidine label.

From the Applicant's proposed draft labeling (2/19/2013, SD #15):

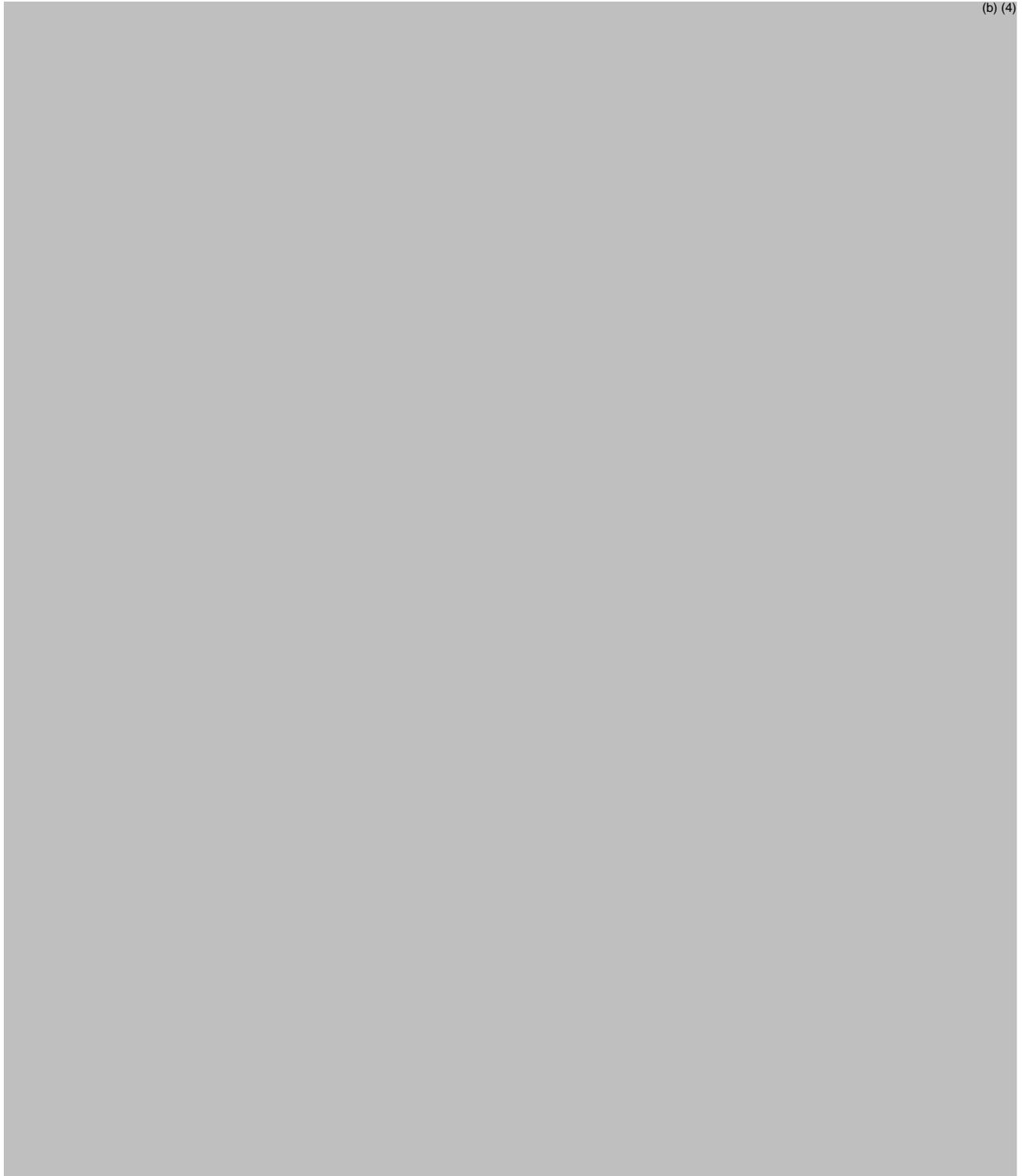


¹ Accessed via:

http://www.accessdata.fda.gov/drugsatfda_docs/label/2011/020816s012lbl.pdf

² Accessed via:

http://www.accessdata.fda.gov/drugsatfda_docs/label/2012/021764s005lbl.pdf



Basis of labeling revisions:

1. The animal-to-human dose conversion for brinzolamide used by the Applicant assumes one affected eye (i.e. not dosing both eyes), and a human body weight of 65 kg. This reviewer recommends rounding to the same number of significant digits (to avoid introducing the false appearance of precision).

2. For the brinzolamide rat development study mentioned in section 8.1, the Applicant appears to have used [REDACTED] (b) (4)
3. The term [REDACTED] (b) (4)
4. In section 13.1, Alcon's recent brimonidine label (Qolinia®) uses the language "In these studies, dietary administration of brimonidine tartrate at doses up to 2.5 mg/kg/day in mice and 1mg/kg/day in rats achieved ... times, respectively, the plasma drug concentration of ... estimated in humans treated with one drop of..."
5. In Section 13.1, the last sentence for brimonidine does not appear in Alcon's brimonidine label. The sentence should be [REDACTED] (b) (4)
6. For brinzolamide, the Ames test was described in the previous label as [REDACTED] (b) (4)

1.2 Brief Discussion of Nonclinical Findings

- Azopt® (brinzolamide ophthalmic suspension) is marketed in the U.S. solely by the Applicant as a 1% suspension. This NDA submission is for a fixed combination suspension that contains the same concentration of brinzolamide, 1% (brin 1%).
- Brimonidine tartrate is marketed under multiple NDAs and ANDAs, including by the Applicant at 0.2%. This NDA submission is for a fixed combination suspension that contains brimonidine tartrate at 0.2% (brim 0.2%)
- The Applicant submitted two topical ocular distribution studies for brin/brim in rabbits
- The Applicant submitted two topical ocular toxicology studies in rabbits, a 6-week study testing two doses (brin 1% / brim 0.15%; brin 2% / brim 0.2%) and a 9-month study testing two doses (brin 1% / brim 0.2%; brin 2% / brim 2%).
- Comparing pharmacokinetic (PK) parameters for the rabbit studies versus the clinical trials (e.g. NDA module 2.7.2 Summary of Clinical Pharmacology Studies), the Applicants concluded that systemic exposure to brimonidine is substantially higher in rabbits compared to patients. This reviewer concurs (see Section 11 of this review).

No new ocular toxicity identified

- The two topical ocular rabbit studies for the combination product did not identify no new concerns for the local/eye toxicity
 - Decreased intraocular pressure (IOP) is the intended pharmacological effect, and IOP was observed up to the D90 time point and earlier, but not at the D181 or D272 time points. The reason for the apparent lack of chronic activity in the rabbits is unclear, and the relevance to chronic patient dosing is unclear.

- Increased corneal thickness was also observed; the Applicant reports that this effect has been observed previously with brimonidine in rabbits and is a species-specific effect not observed in humans.
- The 9-month study detected a treatment-related increase in minimal conjunctival discharge. This effect has previously been noted clinically.

Severe hepatocellular cytoplasmic vacuolization and glycogen accumulation

- Brimonidine is an alpha-2 adrenergic agonist, and multiple nonclinical toxicology studies with brimonidine have reported increased serum glucose, weight loss/reduced weight gain, and liver weight changes in rats, mice, rabbits, and monkeys; however, changes in liver histopathology have not been noted in previous FDA P/T reviews (see section 3.3 of this review).
- Notably, hyperglycemia has not been reported clinically (for brimonidine alone or for brin/brim), suggesting these glucose and hepatic effects are species-specific.
- The 6-week rabbit study with Brin/Brim did not detect increased serum glucose, and no liver histopathology was observed. Urinary glucose was not measured in the 6-week study.
- In the 9-month rabbit study with Brin/Brim (Table 29 of this review):

A dramatic increase in the severity of hepatocellular cytoplasmic vacuolization was observed (standard hematoxylin and eosin staining), and PAS staining found cytoplasmic glycogen accumulation

- Minimal to slight/mild (grade 1-2) vacuolization was noted in the control rabbits
- Comparing the 3-month and 9-month groups treated with Brin/Brim, the severity increased
- Comparing the Brin 1%/ Brim 0.2% against the Brin 2% / Brim 0.2%, a dose-response is apparent. Because the dose of brimonidine is held constant, these data suggest that the increasing dose of brinzolamide contributes to the toxicity.
- Increased serum glucose was observed (+1.5-fold to +3-fold), beginning at the first time point measured (D90, which is later than the duration of the 6-week study).
- The male Brin/Brim groups exhibited diffuse hyperplasia of the islet cells of the pancreas (evaluated at 9 months but not 3 months), and the incidence increased with the increasing dose of brinzolamide (1% to 2%).
- Sedation was observed occasionally, but the study was not designed to assess clinical signs at T_{max} (therefore, the results likely underestimate the true incidence of sedation).
- Suppressed weight gain, decreased food consumption, and a slight increase in male liver weight (relative and absolute) were also observed. The extent to which these effects (increased serum glucose, sedation, reduced weight gain) are cause and effect is unclear.
- The authors and Applicant consider the liver histopathology secondary to the increased serum glucose, and therefore not relevant to patient safety.

- **Note:** Considering the dose-response apparent for the liver and pancreas effects, this reviewer concludes that brinzolamide is contributing to the liver toxicity of brimonidine. Therefore, the previous safety data (clinical and nonclinical) for brimonidine-alone may not be adequate to fully characterize the safety of the combination of brinzolamide plus brimonidine.
- **Note:** Following topical ocular exposure, rabbits did not exhibit liver histopathology at 6 weeks, and the severity of liver pathology increased from 3 to 9 months. Because the duration of exposure to Brin/Brim in the Phase 3 clinical trials was 3 months, these rabbit data raise a theoretical question regarding the potential liver toxicity of longer-duration clinical exposure to Brin/Brim.

Transient penile erection, urogenital swelling and discoloration

- Both the 6-week and 9-month rabbit studies observed increased incidence of penile erection in males, and urogenital swelling and discoloration in females.
 - This reviewer notes that the same urogenital changes were observed for the positive control in the 6-week study, 0.2% brimonidine, as for the Brin/Brim dose groups.
 - The Applicant reports that increased penile erection has been observed previously in rabbits treated with brimonidine; this reviewer was unable to identify P/T reviews or published literature documenting these observations for brimonidine.
- The data tables (Table 16 and Table 23) of this review likely under-represent the true incidence observed in the rabbit studies with Brin/Brim, because clinical signs were not evaluated at T_{max} . In the 6-week study, an additional observation on D42 detected a higher incidence.
- The Applicant and study authors attribute these effects to the alpha-2 adrenergic activity of brimonidine, and provided two papers regarding alpha-2 drugs as a class^{3,4}.
- The Applicant reports that similar effects were not observed clinically for brimonidine or Brin/Brim. This reviewer defers to the Clinical Reviewer regarding patient assessments.

Other systemic effects observed for Brin/Brim do not appear concerning

- The 6-week rabbit study detected a slight increase in serum cholesterol; this is consistent with previous data for brinzolamide. The 9-month study did not produce serum cholesterol data (inadequate assay results).
- The 9-month study detected an increased incidence of minimal spleen lymphoid depletion in the Brin 2%/Brim 0.2% male group.

³ Calabrese E. J. 2001. Adrenergic Receptors: Biphasic Dose Responses. Critical Review in Toxicology, 31 (4&5): 523-538.

⁴ Rampin O. 1999. Pharmacology of α -Adrenoceptors in Male Sexual Function. European Urology, 36(suppl 1):103-106.

2 Drug Information

The drug product is a fixed combination ophthalmic suspension of two drugs: Brinzolamide 1% and Brimonidine Tartrate 0.2%.

2.1 Drug – Brinzolamide 1%

2.1.1 CAS Registry Number

138890-62-7

2.1.2 Generic Name

Brinzolamide

2.1.3 Code Names

AL 4862, AL-4862, AL04862, ALO4862, AL-4862-95, KYCAI State 7

2.1.4 Chemical Name

(R)-4-(ethylamino)-3,4-dihydro-2-(3-methoxypropyl)-2H-thieno[3,2-e]-1,2-thiazine-6-sulfonamide-1,1-dioxide

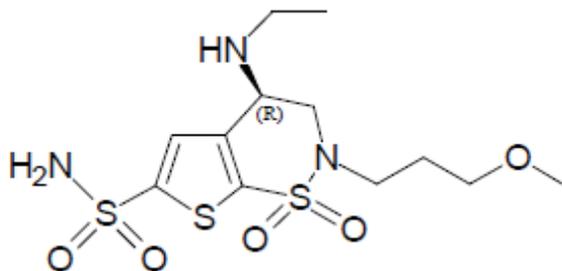
2.1.5 Molecular Formula/Molecular Weight

Molecular formula: $C_{12}H_{21}N_3O_5S_3$

Molecular weight: 383.51

2.1.6 Structure

Figure 1: Structure of brinzolamide



Note: The “R” in Figure 1 shows the one-chiral center in the structure of brinzolamide.

2.1.7 Pharmacologic class

Carbonic anhydrase inhibitor (CAI)

2.1.8 Relevant IND/s, NDA/s, and DMF/s

The applicant referenced Azopt®. Alcon Laboratories Inc. submitted NDA 020816 for brinzolamide, and received initial approval in 1998.

Please also see section 2.5 of this review (below).

2.2 Drug – Brimonidine tartrate 0.2%

2.2.1 CAS Registry Number

- 79570-19-7 (provided by the Applicant)
- Brimonidine tartrate is also registered under the CASRN 70359-46-5
- The CASRN for brimonidine is 59803-98-4

2.2.2 Generic Name

Brimonidine, brimonidine tartrate

2.2.3 Code Names

- brimonidine d-tartrate
- brimonidine purite
- AL-3823, AL-8923A (the "A" is used to denote the tartrate salt), AL-8923A-46
- AGN 109034-LF, AGN 109034LF

2.2.4 Chemical Name

The applicant notes three different common chemical names:

- 5-bromo-N-(4,5-dihydro-1H-imidazol-2-yl)-6-quinoxalinamine
- 5-bromo-6-(2-imidazolylamino)quinoxaline L-tartrate
- 5-bromo-6-(imidazol-2-ylamino)quinoxaline L-tartrate

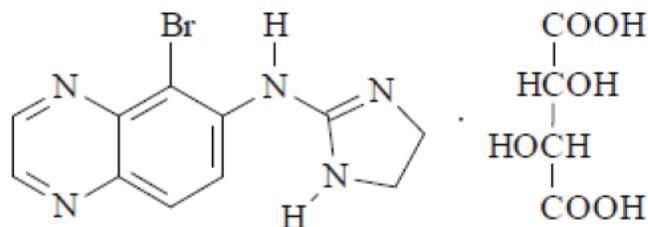
2.2.5 Molecular Formula/Molecular Weight

Molecular formula: $C_{11}H_{10}BrN_5 \cdot C_4H_6O_6$

Molecular weight:

2.2.6 Structure

Figure 2: Structure of brimonidine tartrate



2.2.7 Pharmacologic class

Alpha adrenergic receptor agonist

2.2.8 Relevant IND/s, NDA/s, and DMF/s

The Applicant referenced Bausch and Lomb's ANDA 76-260 for brimonidine tartrate. The cover letter of the submission sent August 2, 2012 notes that Allergan's Alphagan® (brimonidine tartrate), the subject of NDA 20613, has been discontinued.

Please also see section 2.5 of this review (below).

2.3 Clinical Formulation

2.3.1 Drug Formulation

The drug product is formulated as an ophthalmic suspension with 1% brinzolamide and 0.2% brimonidine tartrate, in a sterile multi-dose formulation containing preservatives. From the NDA (module 2.3.P.1 Description and Composition of Drug Product):

Table 1: Composition of the drug product

Component	% (weight/volume)	Applicant's statement of function
Brinzolamide	1	Active ingredient
Brimonidine tartrate	0.2	Active ingredient
Carbomer 974P		(b) (4)
Sodium chloride		
Mannitol		
Propylene glycol		
Tyloxapol		
Boric acid		

Benzalkonium chloride	0.003	Preservative
Sodium hydroxide and/or hydrochloric acid	<i>Quantum satis</i> (QS)	pH adjustment
Purified water	(b) (4)	

2.3.2 Comments on Novel Excipients

The applicant notes that the excipients are compendial, referring to the U.S. Pharmacopeial Convention (USP) and the National Formulary (NF).

2.4 Proposed Clinical Population and Dosing Regimen

The Applicant provided (most recently on 2/19/2013, SD 15) draft labeling, which states:

- Indications and usage: “SIMBRINZA™ is a fixed combination of a carbonic anhydrase II inhibitor and a selective alpha 2 adrenergic agonist indicated for the reduction of elevated intraocular pressure in patients with open-angle glaucoma or ocular hypertension.”
- Dosage and administration: “Instill one drop in the affected eye(s) three times daily. If more than one topical ophthalmic drug is being used, the drugs should be administered at least (b) (4) minutes apart.”
- Dosage forms and strengths: “Suspension containing 10 mg/ml brinzolamide and 2 mg/ml brimonidine tartrate” (brinzolamide/brimonidine tartrate ophthalmic solution 1%/0.2%)

The proposed label summarizes the clinical data submitted to support the NDA, stating (in section 14 Clinical Studies), (b) (4)

2.5 Regulatory Background

- Alcon Pharmaceuticals submitted NDA 204251 under the 505(b)(2) pathway and referenced Azopt® (NDA 20816) and brimonidine tartrate (ANDA 76260).
 - The NDA was initially submitted under the 505(b)(1) pathway with no listed drugs; this oversight was corrected in the August 2, 2012 submission (SD #4).
 - Because Alcon’s NDA 21764 for brimonidine tartrate was submitted under the 505(b)(2) pathway, this Applicant could not use their own NDA as the listed drug (discussed in SD #3, submitted July 27, 2012)
 - Because Allergan has discontinued the Alphagan 0.2% product (NDA 20613), Alcon referenced Bausch and Lomb’s ANDA for brimonidine tartrate ophthalmic solution 0.2%.

- IND 106293 is the predecessor IND of this NDA (i.e. the clinical trials supporting this NDA were performed under IND 106293). In DARRTS, IND 106293 makes reference to 4 files:

Table 2: CDER files referenced by IND 106293

File #	Company	Status	Product name	Review division
IND 106293	Alcon Research Ltd	Active	Brinzolamide / brimonidine tartrate ophthalmic suspension	DTOP
ANDA 76254	Alcon Pharmaceuticals	Approved	Brimonidine tartrate	OGD
NDA 20613	Allergan Inc.	Approved	Alphagan	DTOP
NDA 20816	Alcon Pharmaceuticals	Approved	Azopt	DAIP

- Section 1.4 (Reference Section) of the NDA provides letters of authorization to reference four manufacturing files (MFs):

Table 3: Manufacturing files (MFs) referenced by the applicant

File #	Company	Status	Product name	Review division
(b) (4)	(b) (4)	Active	(b) (4)	OBI/DRM
(b) (4)	(b) (4)	Active	(b) (4)	OBI/DRM
(b) (4)	(b) (4)	Active	(b) (4)	OBI/DRM
(b) (4)	(b) (4)	Active	(b) (4)	OBI/DRM

- Alcon Pharmaceuticals submitted NDA 020816 for brinzolamide (Azopt®) and received initial approval on April 1, 1998. No other NDAs or ANDAs have been approved for brinzolamide.
 - The predecessor IND for NDA 20816 is IND 040152 (Alcon Laboratories for ALO4862 ophthalmic suspension; not directly referenced by NDA 204251).
- Initial approval of brimonidine tartrate was granted to Allergan. Allergan submitted NDA 20613 for the 0.2% solution and received initial approval on September 6, 1996. Allergan has previously submitted NDA 20490 for the 0.5% solution, which was also approved, on March 13, 1997. Both NDAs are now discontinued. This reviewer did not identify the basis for discontinuation, but notes that Alphagan® is apparently still marketed outside the U.S.
- Alcon submitted NDA 21764 for brimonidine tartrate 0.15% solution, and received approval on May 22, 2006. Alcon also submitted ANDA 76254 for brimonidine tartrate 0.2%, and received approval on September 16, 2003.

Table 4: FDA-approved NDAs and ANDAs for brimonidine-containing products

NDA/ANDA #	Company	Drug name	Date of initial approval	Most recent label on Drugs@FDA
NDA 20490	Allergan	Alphagan (0.5%)	March 13, 1997 ^a	December 20, 2001 ⁵
NDA 20613	Allergan	Alphagan (0.2%)	September 6, 1996 ^a	
NDA 21262	Allergan	Alphagan P (0.5%)	March 16, 2001	August 13, 2010 ⁶
NDA 21770	Allergan	Alphagan P (0.1%)	August 19, 2005	
NDA 21764	Alcon Pharms. Ltd.	Brimonidine tartrate, 0.15%	May 22, 2006	March 27, 2012 ⁷
ANDA 76254	Alcon Pharms. Ltd.	Brimonidine tartrate, 0.2%	September 16, 2003	Not available
ANDA 76260	Bausch and Lomb	Brimonidine tartrate, 0.2%	May 28, 2003	
ANDA 76372	Teva Parenteral	Brimonidine tartrate, 0.2%	September 10, 2004 ^a	

⁵ Accessed via:

http://www.accessdata.fda.gov/drugsatfda_docs/label/2001/21262s6lbl.pdf

⁶ Accessed via:

http://www.accessdata.fda.gov/drugsatfda_docs/label/2010/021262s020,021770s004lbl.pdf

⁷ Accessed via:

http://www.accessdata.fda.gov/drugsatfda_docs/label/2012/021764s005lbl.pdf

ANDA 76439	Akorn	Brimonidine tartrate, 0.2%	March 14, 2006	
ANDA 78075	Sandoz	Brimonidine tartrate, 0.2%	January 30, 2008	
ANDA 78480	Apotex Inc.	Brimonidine tartrate, 0.1%	January 27, 2012 ^b	
NDA 21398	Allergan	Combigan (brimonidine tartrate 0.2%, timolol maleate 0.5%)	October 30, 2007	October 11, 2012 ^b
ANDA 91086	Hitech Pharma	brimonidine tartrate 0.2%, timolol maleate 0.5%)	April 6, 2011 ^b	Not available
ANDA 01087	Sandoz		May 11, 2011 ^b	
ANDA 91574	Alcon		August 3, 2010 ^b	

^a Discontinued

^b no marketing status, tentative approval

3 Studies Submitted

3.1 Studies Reviewed

Report #	Title	GLP status	# of pages
Primary pharmacodynamics (module 4.2.1.1)			
001:39320:0696	The IC ₅₀ s of key carbonic anhydrase inhibitors	No	3
002:3930:1196	The IC ₅₀ results of carbonic anhydrase inhibitors, AL-125353	No	3
051-39310-0796	<i>In vitro</i> binding (K1) to human carbonic anhydrase isozymes I and II for AL04822A, AL07118A, and standards acetazolamide (AL04408) and dorzolamide (AL04217A)	No	5
022:41:0301	Effects of brinzolamide on aqueous humor dynamics in pigmented rabbits	No	10
311:3900:0692	Comparison of effect of 600 µg vs 300 µg AL04862 in carbopol 934 suspension on intraocular pressure in monkeys during two days of BID dosing	No	31

⁸ Accessed via:

http://www.accessdata.fda.gov/drugsatfda_docs/label/2012/021398s006lbl.pdf

193:3900:0694	Effect of 300 µg AL06218 vs 300 µg AL04862 on intraocular pressure in monkeys during one day of BID ocular instillation (crossover)	No	16
288:39600:0592	Effect of AL04862 on intraocular pressure in rabbits after a single topical ocular instillation (dose response and viscosity effect)	No	64
093:39600:0294	The effect of AL04862 on ocular hemodynamics systemic blood pressure, heart rate and acid-base balance in anesthetized cats and New Zealand albino rabbits	No	98
018:39500:1097	The effects of one-week topical BID dosing of 2% AL-4862, 2% Trusopt, or AL-4862 vehicle on optic nerve head blood flow, systemic blood pressure, heart rate, and acid-base balance in the Dutch-Belted rabbit	No	53
Safety pharmacology (module 4.2.1.3)			
0010263	Effects of AL-4862 and a combination of AL-4862 and AL-8923a on hERG tail current recorded from stably transfected HEK293 cells	Yes	47
Pharmacokinetics – absorption (module 4.2.2.2)			
0009895	Toxicokinetics of AL-4862 and AL-8923 in study N-09-085	Yes	120
0011778	Toxicokinetics of LA-4862, AL-8520 and AL-8923 in toxicology study N-10-083	Yes	266
Pharmacokinetics – distribution (module 4.2.2.3)			
0009954	Uptake of brimonidine (AL-8923) and brinzolamide (AL-4862) in ocular tissues following topical ocular administration to New Zealand White/Red F1 cross rabbits	No	210
0014507	Ocular tissue distribution of brinzolamide (AL-4862) and brimonidine (AL-8923) following topical ocular administration of brinzolamide 1% / brimonidine 0.2%, AZOPT or brimonidine (Falcon) to New Zealand White/Red F1 cross rabbits	No	375

Toxicology – repeat-dose toxicity (module 4.2.3.2)			
N-10-083	Final: Nine-month topical ocular safety evaluation of AL-4862 (brinzolamide) and AL-8923A (brimonidine tartrate) with a 3-month interim in F1 pigmented rabbits	Yes	1051
N-08-085	6-week topical ocular safety evaluation of AL-4862 (brinzolamide) and AL-8923A (brimonidine tartrate) in F1 pigmented rabbits	Yes	476
N-10-083	Final: Nine-month topical ocular safety evaluation of AL-4862 (brinzolamide) and AL-8923A (brimonidine tartrate) with a 3-month interim in F1 pigmented rabbits	Yes	380

3.2 Studies Not Reviewed

Report #	Title	GLP status	# of pages
Pharmacokinetics – analytic methods and validation reports (module 4.2.2.1)			
0009948	UPLC/UV analysis of brinzolamide and n-desethylbrinzolamide in rabbit blood	No	45
0010467	Brimonidine rabbit K2EDTA plasma validation at (b) (4)	Yes	190

3.3 Previous Reviews Referenced

This reviewer references the prior P/T reviews documented:

- For IND 106293
 - P/T review of the original IND submission (Wild 1/15/2010)
 - Minutes of the End-of-Phase 2 meeting held between DTOP and Alcon on November 15, 2010 (Chambers, 12/15/2010)
- For IND 40152 (the predecessor IND for Alcon’s NDA 204251 for brinzolamide) , no P/T reviewers were found (in DARRTS or elsewhere)
- For NDA 20816 for Azopt® (brinzolamide ophthalmic suspension), this reviewer references the Pharmacology review (Coulter, 1997)⁹. No other P/T reviews were located for this NDA in DARRTS.

⁹ Accessed in two parts, via:

http://www.accessdata.fda.gov/drugsatfda_docs/nda/98/20816_AZOPT_PHARMR_P1.PDF and

- For Allergan's NDA 20490 and NDA 20613, no P/T reviews were located in DARRTS or on Drugs@FDA, but redacted P/T reviews were identified via Pharmapendium:
 - The original P/T review for NDA 20490 (Minigi, 1995)¹⁰
 - An earlier P/T review for NDA 20490 that includes evaluation of the bioassays (Adeyemo, 12/28/1998)¹¹
 - The original P/T review for NDA 020613 (Mukherjee, 2/26/1996)¹²
- For brimonidine tartrate, the P/T reviews of the original NDAs publicly available for
 - Allergan's NDA 21262 (Tandon, 2000)¹³ and NDA 21770 (Mukherjee, 1/24/2005)¹⁴ for Alphagan P
 - NDA 21764 (Mukherjee, 5/26/2004)¹⁵ for Alcon's brimonidine tartrate 0.15%
 - NDA 21398 (Chen, 1/17/2002)¹⁶ for Allergan's Combigan®

http://www.accessdata.fda.gov/drugsatfda_docs/nda/98/20816_AZOPT_PHARMR_P2.PDF . Review is not available via DARRTS. The review lists a completion date of May 31, 1997 and has a stamp date of Jun 25, 1997.

¹⁰ Accessed via:

<https://www.pharmapendium.com/fda.do?query=brimonidine&includeSynonyms=true&drugName=Brimonidine+Tartrate&document=f78e91a316266bff88467dedad8f855f>

¹¹ Accessed via:

<https://www.pharmapendium.com/fda.do?query=brimonidine&includeSynonyms=true&drugName=Brimonidine+Tartrate&document=f78e91a316266bff88467dedad8f855f>

¹² Accessed in two parts via:

<https://www.pharmapendium.com/fda.do?query=brimonidine&includeSynonyms=true&drugName=Brimonidine+Tartrate&document=f78e91a316266bff88467dedad8f855f> and <https://www.pharmapendium.com/fda.do?query=brimonidine&includeSynonyms=true&drugName=Brimonidine+Tartrate&document=f78e91a316266bff88467dedad8f855f>

¹³ Accessed via: http://www.accessdata.fda.gov/drugsatfda_docs/nda/2001/21-262_Alphagan%20P%20Ophthalmic_pharmr.pdf Review completion date October 27, 2000; signed November 22, 2000.

¹⁴ Accessed via:

http://www.accessdata.fda.gov/drugsatfda_docs/nda/2005/021770s000_PharmR.pdf

¹⁵ http://www.accessdata.fda.gov/drugsatfda_docs/nda/2006/021764s000_PharmR.pdf

¹⁶ Accessed via:

http://www.accessdata.fda.gov/drugsatfda_docs/nda/2007/021398s000_PharmR.pdf

4 Pharmacology

4.1 Primary Pharmacology

Alcon submitted primary pharmacology data for brinzolamide in support of approved NDA 20816, which was reviewed previously.⁹ The applicant submitted nine primary pharmacology study reports for brinzolamide (as a single agent) to this NDA; they are reviewed below. No new safety concerns were identified.

- Under the *in vitro* conditions tested, brinzolamide (compound AL04862) inhibited of human carbonic anhydrase (HCA) I, II and IV
 - The results were provided to the NDA in two short study reports that summarized assays to determine the concentrations of brinzolamide that inhibited the activity of three carbonic anhydrase enzymes *in vitro* (IC₅₀).
 - Both assays were conducted by Alcon Laboratories, Forth Worth, Texas, using the same assay method.
 - The first report is titled: “**The IC₅₀s of key carbonic anhydrase inhibitors – number 96-001**” Report # 001:39320:0696. Signed by the authors on December 12, 1996
 - The second report is titled: “**The IC₅₀ results of carbonic anhydrase inhibitor, AL-12353 – Number 96-002**”. Report # 002:39320:1196. Signed by the authors on December 11 or December 12, 1996.
 - The IC₅₀ values for HCA I, HCA II and HCA IV are presented in Table 5:

Table 5: Brinzolamide inhibited HCA-I, HCA-II and HCA-IV activity *in vitro* (report # 001:39320:0696)

Brinzolamide activity	HCA I IC ₅₀ (nM)	HCA II IC ₅₀ (nM)	HCA IV IC ₅₀ (nM)
Report # 001:39320:069	1367 ± 82	3.19 ± 0.30	45.3 ± 0.3
Report # 002:39320:1196	234 ± 0	2.91 ± 0.05	101 ± 6

Data presented as means ± standard deviation (average of two measurements)

- Methods notes for reports # 001:39320:069 and # 002:39320:1196
 - The inhibition of human carbonic anhydrase (HCA) I, II and IV was measured *in vitro*. The source and buffer composition was not reported.
 - The assay calculated the rate of carbon dioxide hydration based on the amount of base needed to maintain the original pH over 4 minutes.
 - As a positive control for enzyme inhibition, a 50:50 mixture of ethanol and water was used.

- Under the *in vitro* conditions tested, brinzolamide (compound AL04862A) bound to HCA I and II.
 - The results were provided to the NDA in a short report titled "*In vitro* binding (K_i) to HCA isozymes I and II for AL04862A, AL07118A, and standards acetazolamide (AL04408) and dorzolamide (AL04217A). Report # 051-38310-0796. Signed by the authors July 19, 1996. The study report was written at Alcon Laboratories, Forth Worth Texas, but the actual study was performed by (b) (4) (location not reported).

Table 6: Brinzolamide binds to HCA-I and HCA-2 *in vitro* (report # 051-39310-0796)

Compound	K_i (nM)	
	HCA-I	HCA-II
Brinzolamide	32.1 ± 0.99	0.13 ± 0.03
Acetazolamide	673 ± 81.8	33.8 ± 4.90
Dorzolamide	1240 ± 417	0.51 ± 0.09

- Method notes:
 - Commercial HCA I and HCA II, purified from human erythrocytes, were used
 - The assay measured the fluorescence of 5-dimethylaminonaphthalene -I-sulfonamide (DNSA) as an indirect endpoint for competitive binding / displacement of DNSA from HCA, at 37°C
 - As negative controls, a 50:50 mixture of ethanol and water, and 100% ethanol, were used.
 - The positive controls, acetazolamide (Diamox®) and dorzolamide (Trusopt®), are marketed carbonic anhydrase inhibitors.
 - The test concentrations were 0.5, 1, 5, 10, 50 and 100 nM. Each agent was tested in triplicate
- Administrative note: The brinzolamide data for binding to HCA-I was conducted under this report. However, the brinzolamide data for binding to HCA-II are reported in this report, but the HCA-II experiment was apparently conducted under another report (# 017:39310:0496). This reviewer concludes that requiring submission of the original report (# 017:39310:0496) is not necessary, because reviewing the original report is not critical to understanding the results.

Study title: Effects of brinzolamide on aqueous humor dynamics in pigmented rabbits (contract study)	
Study no.:	002:41:0301
Study report location:	NDA module 4.2.1.1 Primary pharmacodynamics
Conducting laboratory and location:	(b) (4)
Report date:	Signed April 17, 2001
GLP compliance:	No
Drug, lot #:	1% brinzolamide (purity and formulation not reported)

Key Study Findings

- The authors conclude, and this reviewer concurs, that a two doses of 1% brinzolamide (topical ocular administration, two doses 16 to 20 hours apart) had no apparent effect in the eyes of rabbits; but that a second series of doses administered one week later did reduce IOP (-4 to -11%) and aqueous flow (-19%)
- The applicant reports (Module 2.6.2 Pharmacology Written Summary) that this report showed “Following topical ocular administration, brinzolamide inhibits aqueous humor formation and reduces elevated IOP.” Because these rabbits did not have elevated IOP, this study was not designed to detect the activity of brinzolamide to reduce “elevated IOP”. The applicant’s conclusion for this study is not entirely accurate.

Method notes:

- Twelve female pigmented rabbits were used (approximately 2.2 kg body weight, approximately 9 months of age)
- At baseline,
 - Fluorescein was applied to the cornea (over 10 to 20 seconds, using iontophoresis).
 - The next morning, the following endpoints were measured: IOP, pulsatile ocular blood flow (POBF), cornea thickness and anterior chamber depth, and aqueous flow
 - After the baseline measurements, rabbits received either timolol (0.5 topically to the cornea of each eye) or intravenous acetazolamide (20 mg/kg), presumably to verify normal response to positive control. IOP, aqueous flow, and uveoscleral outflow was measured/calculated (results for timolol and acetazolamide not provided).
 - The report does not indicate which rabbits received which doses, or whether the rabbits were randomized to these treatments
 - Reportedly, these treatments did not affect IOP
- First series of doses: a week after the baseline treatments, eyes were randomized to treatment or control (no treatment).

- For the first dose, rabbits received two doses of 25 µl of 1% brinzolamide: first dose at 4:00 pm, second dose between 8:00 am and 12 noon the following day.
- This reviewer presumes that each rabbit had one control eye and one brinzolamide-treated eye, but this is not explicitly stated. The reporting appears to allow for some rabbits to have two treated eyes and other rabbits to have two control eyes.
- Endpoints measured after the first brinzolamide doses include IOP and aqueous flow
- Second series of doses: conducted a week after the first series of doses, each eye was re-dosed
 - 6 rabbits were evaluated for IOP (time points not reported) and aqueous flow.
 - 6 rabbits were evaluated for IOP (time points not reported); the rabbits were then anesthetized and uveoscleral flow was measured

Results notes:

- The first doses of 1% Brinzolamide (two doses approximately 16 to 20 hours apart) did not affect IOP or aqueous flow.
- The second doses of 1% Brinzolamide (two doses approximately 16 to 20 hours apart) were associated with statistically significant decreases in IOP (-4 to -11% compared to control) and aqueous flow (-19% compared to control)
- It is not clear to this reviewer how baseline treated eye versus control eye was reported by the authors. The report indicates rabbits received either timolol in both eyes, or intravenous acetazolamide (which presumably distributes to both eyes equally). This reviewer infers that the authors considered either timolol or acetazolamide to be “treated” and the other to be “control”, but the reporting is not explicit.

Table 7: Four treatments with 1% brinzolamide over 8 days reduced IOP and aqueous flow in pigmented rabbits (report # 002:41:0301)

Endpoint	Treated eye	Control eye	P-value (treated versus control)
Baseline (12 rabbits)			
IOP at ~ 10:00 am	23.7 ± 2.3	23.1 ± 1.4	0.35
IOP at ~ 12 noon	23.2 ± 2.3	23.1 ± 1.7	0.82
Aqueous flow (µl/min)	2.33 ± 0.46	2.63 ± 0.72	0.25
After first treatments with 1% brinzolamide (two 25 µl topical ocular doses) (12 rabbits)			
IOP at ~ 10:00 am	22.6 ± 2.2	23.7 ± 2.2	0.15
IOP at ~ 12 noon	21.6 ± 2.6	22.7 ± 1.6	0.07
Aqueous flow (µl/min)	2.14 ± 0.53	2.40 ± 0.40	0.12

Outflow facility ($\mu\text{l}/\text{min}/\text{mm Hg}$)	0.27 ± 0.07	0.29 ± 0.10	0.83
After second treatments with 1% brinzolamide (two 25 μl topical ocular doses) (6 rabbits)			
IOP1 ^a	23.1 ± 2.4	24.7 ± 2.3	0.03
IOP2 ^a	21.6 ± 2.5	22.5 ± 1.9	0.05
IOP3 ^a	21.3 ± 2.1	23.8 ± 1.9	0.001
IOP4 ^a	22.2 ± 2.6	23.5 ± 1.5	0.08
Aqueous flow ($\mu\text{l}/\text{min}$)	2.2 ± 0.5	2.7 ± 0.6	0.02
After second treatments with 1% brinzolamide (two 25 μl topical ocular doses) (6 rabbits)			
IOP ^a	20.33 ± 3.20	24.00 ± 4.47	0.03
Outflow facility ($\mu\text{l}/\text{mg}/\text{mm Hg}$)	0.34 ± 0.09	0.29 ± 0.13	0.42

^a The report does not specify when these IOP measurements were taken.

Study title: Comparison of effect of 600 μg vs 300 μg AL04862 in carbopol 934 suspension on intraocular pressure in monkeys during two days of BID dosing	
Study no.:	311:39600:0692
Study report location:	NDA module 4.2.1.1 Primary pharmacodynamics
Conducting laboratory and location:	Alcon Laboratories Forth Worth, Texas 76134
Report date:	Signed by authors on July 2 and July 7, 1992
GLP compliance:	No
Drug:	1% and 2% brinzolamide (AL04862). Purity and lot # not reported

Key Study Findings

- 1% and 2% Brinzolamide decreased IOP in both the normal and hypertensive eyes of conscious cynomolgus monkeys. No difference apparent between the two doses.
- Note: The applicant reported (NDA module 2.6.2.2 Primary Pharmacodynamics) that this study showed “Brinzolamide lowered IOP by 20%-30% in dose-dependent manner in lasered ocular hypertensive cynomolgus monkeys”. This statement is not completely accurate – although brinzolamide lowered IOP by 20 to 32%, no dose-dependence was observed in this study.

Method notes:

- Disease model:
 - Cynomolgus monkeys (mixed sex adults) from a permanent colony at Alcon were used (i.e. presumably non-naïve).
 - Each animal has undergone laser trabeculoplasty of the right eye, resulting in ocular hypertension. The left eyes were normal and normotensive.
 - The monkeys had been trained to sit in restraint chairs designed for ocular studies, and to undergo IOP measurement without sedation.
- Brinzolamide was formulated (b) (4)
- Groups of 7 monkeys received one drop (30 µl) of either 1% brinzolamide (300 µg) or 2% brinzolamide (600 µg) into the right eye at 9:00 am and 9:00 pm of day 1 and day 2 (4 total doses). Left eyes were untreated. IOP was measured under local proparacaine anesthetic at baseline, at 1, 3, 6 and 12 hours after the first and third doses, and 12 hours after the second and fourth dose.
- IOP statistical significance was evaluated using Student's t-test for each time point from baseline, and ANOVA for differences between groups.

Results notes:**Table 8: Topical ocular brinzolamide (1% and 2%) reduced IOP in normotensive and hypertensive eyes of monkeys (report # 311:39600:0692)**

Dose	IOP time point	IOP OD (treated hypertensive)	OD % change in IOP	IOP OS (untreated normotensive)	OS % change in IOP
1% Brinzolamide (300 µg)	Pre-dose	33.0	0	21.0	0
	After 1st dose – 1 hr	28.7	-12.8**	21.4	+2.5
	After 1st dose – 3 hr	23.7	-26.9**	20.3	-3.2
	After 1st dose – 6 hr	22.4	-30.8**	20.3	-3.3
	After 1st dose – 12 hr	22.4	-30.3**	19.3	-7.4
	After 2 nd dose – 12 hr	25.0	-23.8**	19.3	-8.0*
	After 3 rd dose – 1 hr	23.4	-28.2**	19.7	-5.8
	After 3 rd dose – 3 hr	21.7	-32.8**	19.6	-6.5
	After 3 rd dose – 6 hr	22.4	-30.8**	20.3	-3.1

	After 3 rd dose – 12 hr	21.1	-34.8**	18.6	-10.9**
	After 4th dose – 12 hr	25.4	-22.2*	18.6	-10.7*
2% Brinzolamide (600 µg)	Pre-dose	31.7	0	22.0	0
	After 1st dose – 1 hr	26.4	-15.4*	23.0	+5.6
	After 1st dose – 3 hr	22.9	-26.9**	21.7	+0.1
	After 1st dose – 6 hr	23.3	-26.0**	22.4	+3.6
	After 1st dose – 12 hr	22.6	-28.6**	21.0	+2.7
	After 2 nd dose – 12 hr	24.9	-21.9**	21.1	+2.5
	After 3 rd dose – 1 hr	23.3	-29.3**	22.0	+1.1
	After 3 rd dose – 3 hr	21.1	-32.9**	21.3	-1.8
	After 3 rd dose – 6 hr	22.4	-28.9**	21.3	-1.5
	After 3 rd dose – 12 hr	21.3	-32.4**	18.6	-13.5*
	After 4th dose – 12 hr	23.1	-26.4**	19.6	-9.9**

* statistically significant, $p < 0.05$

** statistically significant, $p < 0.01$

Study title: Effect of 300 µg AL06218 vs 300 µg AL04862 on intraocular pressure in monkeys during one day of BID topical ocular instillation (crossover)

Study no.: 193:39600:0694
 Study report location: NDA module 4.2.1.1 Primary pharmacodynamics
 Conducting laboratory and location: Alcon Laboratories
 Fort Worth, Texas
 Date of study initiation: April 28, 1994
 Report date: October 4, 1994
 GLP compliance: No
 Drug, lot #: • 1% Brinzolamide (AL04862) ophthalmic suspension, lot # 92-2864-1, pH 7.5 (purity not reported)
 • 1% "AL06218"

Key Study Findings

- As expected, 1% brinzolamide lowered IOP in the lasered (hypertensive) eyes of non-naïve cynomolgus monkeys for at least 12 hours
- This reviewer infers that this study was conducted to investigate the IOP pharmacological activity of AL06218, with brinzolamide included as a positive control.

Method notes:

- The identity of “AL06218” was not determined by this reviewer (either by searching the NDA or searching the published literature). The code does not refer to brimonidine. This study report identifies AL06218 as a CAI (like brinzolamide).
- Twelve cynomolgus monkeys (6/sex) were used, from a glaucoma research colony
 - The monkeys had undergone argon laser trabeculoplasty of the right eye to induce hypertension
 - The monkeys had undergone trabeculoplasty 6 to 12 years prior to the initiation of this study.
- In a cross-over design, 3/sex received either two doses of 1% brinzolamide (twelve hours apart) or two doses of 1% AL06218 (also twelve hours apart). IOP was measured by pneumatic tonometry pre-dose, after the first dose (1, 3, 6 and 12 hours) and after the second dose (at 12 hours). Light corneal anesthesia (proparacaine) was used for each IOP measurement, and then eyes were washed with saline
- Four weeks later, the groups were reversed using the same treatment protocol. Monkeys were returned to colony after completion of the experiments.

Results notes:

- Slight blinking was observed following dosing, for both compounds (report page 2; no further details about the blinking were reported, such as the time to onset of blinking, duration, or recoverability).
 - The formulation was not unusual (data provided report pages 10-11), and therefore not a likely cause of the blinking.
 - Based on the lack of blinking noted in the other brinzolamide studies, this reviewer does not consider these observations of blinking to be clearly adverse.
- Brinzolamide lowered IOP, statistically significant compared to baseline. Mean decreases:
 - By 24.7% (9.2 mm g) at 1 hour after the first dose
 - By 35.8% (13.5 mm g) at 3 hour after the first dose
 - By 26.5% (10.2 mm g) at 6 hour after the first dose
 - By 23.5% (9 mm g) at 12 hour after the first dose
 - By 17.6% (6.9 mm g) at 12 hour after the second dose
- No other toxicology endpoints reported
- Authors conclude that no drug-drug interaction was apparent

Study title: Effect of AL04862 on intraocular pressure in rabbits after a single topical ocular instillation (dose response and viscosity effect)	
Study no.:	288:39600:0592
Study report location:	NDA module 4.2.1.1 Primary pharmacodynamics
Conducting laboratory and location:	Alcon Laboratories Fort Worth, Texas
Date of study initiation:	April 16, 1992
Report date:	July 7, 1992
GLP compliance:	No
Drug, lot #:	Brinzolamide (AL04862), batch # 92-2493, purity not reported

Key Study Findings

- Brinzolamide decreased intraocular pressure in the eyes of rabbits, with a clear dose-response. Under the conditions tested, higher viscosity formulations were associated with earlier onset of activity, greater magnitude of the IOP decrease, and a longer duration of response.

Method notes:

- In four separate experiments, groups of 7 female Dutch belted rabbits received two topical ocular instillations of 25 µl of test article (the duration between the two doses is not specified) into the one eye; the contralateral eye was instilled with saline. The doses were:
 - 250 µg of brinzolamide (0.5% solution)
 - 500 µg of brinzolamide (1% solution)
 - 1 mg of brinzolamide (2% solution)
 - Formulation with 200 cps viscosity, 500 µg of brinzolamide (1% solution)
 - Formulation with 5000 cps viscosity, 500 µg of brinzolamide (1% solution)
 - Formulation with 1 cps viscosity, 500 µg of brinzolamide (1% solution)
- IOP was measured by pneumatic tonometry at baseline, and after dosing at 30 minutes; at one hour, and then hourly (to 4 or 6 hours post-dose). Light corneal anesthesia (proparacaine) was used for each IOP measurement, and then eyes were washed with saline
- The report used centipoise (CPS) to describe the viscosity. For context, reportedly:
 - The viscosity 1 cps is approximately the same as water
 - Viscosity of 200 cps is approximately the same viscosity as motor oil
 - Viscosity of 5000 cps is approximately the same viscosity of honey
- No necropsy; no reporting of other toxicology endpoints

Results notes:

- Brinzolamide decreased IOP. Over four separate experiments, the 1 mg dose was tested twice. Almost all of the data points in Table 9 were statistically significantly different from baseline.

Table 9: Topical brinzolamide reduced IOP in the eyes of Dutch belted rabbits (report # 288:39600:0592)

Brinzolamide dose & formulation	Time (hour)	Mean % decrease in IOP from baseline
Dose response experiments		
250 µg	0.5	2.1
	1	10.6
	2	11.8
	3	9.7
	4	9.1
	5	7.0
	6	5.2
500 µg	0.5	11.8
	1	19.8
	2	21.6
	3	21.0
	4	13.8
	5	15.9
	6	7.9
1 mg (first experiment)	0.5	1.7
	1	12.9
	2	22.2
	3	13.7
	4	12.9
	5	7.1
	6	7.3
1 mg (second experiment)	0.5	15.4
	1	27.5
	2	25.1
	3	20.6
	4	16.3
	5	14.6
	6	11.1
Viscosity experiments		
500 µg, 1 cps	0.5	8.8
	1	13.3
	2	15.4
	3	16.8

	4	14.5
500 µg, 200 cps (third experiment)	0.5	13.5
	1	24.3
	2	18.2
	3	17.7
	4	13.5
	5	16.1
	6	15.3
500 µg, 200 cps (fourth experiment)	0.5	8.8
	1	13.3
	2	15.4
	3	16.8
	4	14.4
500 µg, 5000 cps	0.5	22.2
	1	20.6
	2	23.4
	3	18.3
	4	19.9
	5	19.6
	6	20.5

Study title: The effect of AL04862 on ocular hemodynamics [sic] systemic blood pressure, heart rate and acid-base balance in anesthetized cats and New Zealand albino rabbits

Study no.: 093:39600:0294
Study report location: NDA module 4.2.1.1 Primary pharmacodynamics
Conducting laboratory and location: Alcon Laboratories
Fort Worth, Texas
Report date: June 19, 1996
GLP compliance: No
Drug, lot #: Brinzolamide (AL04862), lot # 86513, 70241, and 86532 (purity not reported)

Key Study Findings

- The results presented in this study report do not raise safety concerns.
- Based on the expectation that brinzolamide-induced decrease in IOP would decrease vascular resistance in the eye, several exploratory experiments were conducted in cats and rabbits to characterize ocular blood flow, optic nerve head microvascular blood flow (ONHBF), and secondary parameters (acid-base balance, as an indicator of metabolic activity).

- Under the conditions tested, daily topical administration of 1% brinzolamide did not clearly affect optic nerve head microvascular blood flow (ONHBF) in anesthetized cats
- Intravenous brinzolamide administered to anesthetized cats (5 mg/kg) and anesthetized rabbits (0.5, 2.5, 5 mg/kg) increased ONHBF, blood flow to the eye (only measured in rabbits; anterior and posterior uveal tissues), and arterial acidosis (increased arterial carbon dioxide and decreased pH)
- No brinzolamide-induced decrease in IOP was apparent in the anesthetized animals; the authors speculate that the anesthesia-induced decrease in IOP obscured the effect of brinzolamide.

Method notes

- Prior to dosing, animals underwent surgery to implant arterial, venous and tracheal cannulations.
- A group of seven adult domestic short haired cats were dosed with brinzolamide, a group of three cats were used as concurrent controls.
 - The cats were dosed (topical ocular administration) with one drop (reported by the authors as approximately 30 μ l) of 0 or 1% brinzolamide twice daily for seven days, anesthetized on the eighth day, and ONHBF was measured.
 - After a week of recovery, the procedure was repeated.
 - After a month of additional recovery, the cats were dosed with 0 or 1% of topical ocular brinzolamide (right eye only), and measurements were taken (ONHBF, blood pressure, heart rate, IOP, hematocrit, arterial pH, pO₂ and pCO₂)
 - After an unreported additional recovery period, the cats were dosed with 0 or 5 mg/kg of brinzolamide intravenously, and measurements were taken 10 and 30 minutes post-dose (ONHBF, blood pressure, heart rate, IOP, hematocrit, arterial pH, pO₂ and pCO₂)
- The number of New Zealand white rabbits was not reported. Under anesthesia, each of the rabbits received a series of bolus iv doses: 0.5 mg/kg, then 2.5 mg/kg, then 5 mg/kg, with 10 minutes between doses.

Results notes

- Topical ocular administration of brinzolamide did not affect ONHBF under the conditions tested in either cats or rabbits.

Study title: The effects of one-week topical BID dosing of 2% AL-4862, 2% Trusopt, or AL-4862 vehicle on optic nerve head blood flow, systemic blood pressure, heart rate, and acid-base balance in the Dutch-Belted rabbit

Study no.:	018:39500:1097
Study report location:	NDA module 4.2.1.1 Primary pharmacodynamics
Conducting laboratory and location:	Alcon Laboratories Fort Worth, Texas
Report date:	May 2, 2001
GLP compliance:	No
Drug, lot #:	Brinzolamide: <ul style="list-style-type: none">• Alcon's AL-4862, lot # AQE-2870 and 88984• Merck's Trusopt, lot # 0210E

Key Study Findings

- The authors' justification for this study is that other CAIs are used systemically to determine cerebral blood flow reserve, and therefore the potential activity of topical brinzolamide on ocular blood flow was of interest
- Twice daily topical ocular dosing with 2% brinzolamide increased ONHBF by 8.4 to 11.2% and decreased IOP, compared to baseline in rabbits. Slight changes in blood gas and acid-base were noted, and of unclear relevance to treatment and unclear biological significance. No changes in heart rate or arterial blood pressure were apparent.

Method notes

- A preliminary study (only summary results provide) was conducted, comparing three male New Zealand albino rabbits and three Dutch-Belted rabbits for baseline ONHBF
- A three-way cross-over design was used with three groups of 9 male Dutch-Belted rabbits. One drop of vehicle control, Alcon's 2% brinzolamide, or Trusopt (2% brinzolamide) were administered, twice daily for 7 days.
 - Endpoints (measured baseline and 90 minutes after the last dose on D8): IOP, ONHBF, blood pressure, heart rate, arterial pH, arterial pO₂ and arterial CO₂
 - After a 7-14 day wash-out period, the groups were switched to another treatment

Results notes:

- Both formulations of 2% brinzolamide reduced IOP and increased ONHBF. No difference was apparent between the two formulations

Table 10: 2% brinzolamide topical ocular administration decreased IOP and increased optic nerve head blood flow in rabbits (report # 018:39500:1097)

Treatment	IOP (% change)	ONHBF (% change)	pH (% change)	PO ₂ (% change)	PCO ₂ (% change)
Vehicle	3.6 ± 3.6	-6.6 ± 3.2	0.5 ± 0.2	-2.9 ± 1.9	2.4 ± 2.9
2% AL-4862 (Alcon's)	-16.8 ± 2.2 *	11.2 ± 1.8 *	-0.5 ± 0.2 *	8.5 ± 2.4 *	-6.7 ± 3.8 *
2% Trusopt (Merck's)	-20.7 ± 3.9 *	8.4 ± 4.3 *	-0.7 ± 0.2 *	6.9 ± .8 *	-10.2 ± 2.7 *

Values are means + standard deviation. Endpoints were measured 90 minutes after the last dose

* Statistically significantly different from baseline, $p \leq 0.05$

4.2 Secondary Pharmacology

No nonclinical secondary pharmacology studies were submitted to the NDA.

4.3 Safety Pharmacology

The applicant submitted one safety pharmacology study: no hERG inhibition was apparent for brinzolamide or a mixture of brinzolamide/brimonidine *in vitro*, under the conditions tested.

Study title: Effects of AL-4862 and a combination of AL-4862 and AL-8923a on hERG tail current recorded from stably transfected HEK293 cells	
Study no.:	TDOC-0010263
Study report location:	NDA module 4.2.1.3 Safety Pharmacology
Conducting laboratory and location:	(b) (4)
Date of study initiation:	July 6, 2009
Report date:	August 2009
GLP compliance:	Yes, signed
Quality assurance:	Yes, signed
Drug, lot #, and % purity:	<ul style="list-style-type: none"> • Brinzolamide (AL-4862), lot # 00222105, purity 99.70% • Brimonidine (AL-8923A), lot # 00216234, purity 99.40%

Key Study Findings

- Vehicle control, a positive control, and four test articles were tested:
 - 3 µg/ml of AL-4863
 - 0.3 µg/ml of AL-4862 + 0.1 µg/ml of AL-3823A
 - 1 µg/ml of AL-4862 + 0.3 µg/ml of AL-3823A
 - 3 µg/ml of AL-4862 + 1 µg/ml of AL-3823A
- No inhibition of human ether-a-go-go-related gene (hERG) tail current was apparent in HEK293 cells stably transfected with hERG cDNA and exposed to the brinzolamide and brinzolamide/brimonidine test articles, compared to vehicle.
- The positive control, 100 nM of E-4031, inhibited hERG tail current as expected.
- The methods and reporting appear adequate¹⁷.
- The authors (report page 16) justify the selection of test concentrations with the conclusion that the concentrations tested are above the maximum anticipated human plasma levels. This reviewer concurs.
 - As noted in the proposed label for this NDA, following topical ocular brinzolamide administration in patients, plasma levels of brinzolamide (and N-desethyl brinzolamide) were reportedly below assay quantitation limits (< 10 ng/ml).
 - Following topical ocular administration of brimonidine tartrate in patients, peak plasma concentrations were detected in the range of ~ 100 pg/ml (varying by dose level)

5 Pharmacokinetics/ADME/Toxicokinetics

5.1 PK/ADME

The sponsor submitted two rabbit ocular distribution studies. In both, topical ocular administration of Alcon's combination product resulted in higher local exposure than the commercial products tested for comparison.

¹⁷ The authors cite, and this reviewer consulted, ICH S7B Nonclinical Evaluation of the Potential for Delayed Ventricular Repolarization (QT Interval Prolongation) by Human Pharmaceuticals. Accessed online via: <http://www.fda.gov/downloads/Drugs/GuidanceComplianceRegulatoryInformation/Guidances/UCM074963.pdf>

Study title: Uptake of brimonidine (AL-8923) and brinzolamide (AL-4862) in ocular tissues following topical ocular administration to New Zealand white/red F1 cross rabbits

Study no.: TDOC-0009954
 Study report location: NDA module 4.2.2.3 Pharmacokinetics – Distribution
 Conducting laboratory and location: Alcon Research, Ltd.
 Forth Worth, Texas 76134
 Date of study initiation: April 20, 2009
 Report date: August 24, 2009
 GLP compliance: No
 Drugs: Brimonidine / Brinzolamide ophthalmic suspension

Key Study Findings

- Three brimonidine/brinzolamide ophthalmic suspensions prepared by the applicant were compared against commercial brimonidine (Alphagan P) and commercial brinzolamide (Azopt®).
 - However, Alcon's formulation did not precisely match either commercial formulation
- Rabbits were dosed once in each eye, and concentrations of brimonidine and brinzolamide were measured in the aqueous humor, bulbar conjunctiva, and iris-ciliary body
- Distribution into the bulbar conjunctiva was higher for Alcon's brimonidine and brinzolamide compared to the commercial brimonidine and brinzolamide.
- A clear dose-response was observed for the three Alcon formulations tested.
- No pharmacodynamic or toxicology endpoints were assessed.

Table 11: Formulations for the rabbit single-dose topical ocular distribution study (report # TDOC-000954)

Group #	Formulation	Batch #
1	Brimonidine 0.1% / Brinzolamide 1%	09-55042-1
2	Brimonidine 0.15% / Brinzolamide 1%	09-55043-1
3	Brimonidine 0.2% / Brinzolamide 1%	09-54989-2
4	Alphagan P (Brimonidine) 0.15%	Allergan, # 58108
5	Azopt® (brinzolamide) 1%	Alcon # 157574F

Methods

Doses: Single 30 µl topical ocular administration into the right eye followed by the left eye (total dose 60 µl/rabbit)
 Frequency of dosing: Single dose to each eye

Route of administration:	Topical ocular administration
Formulation/Vehicle:	<ul style="list-style-type: none"> For Alcon's formulations, the suspension consisted of (b) (4) Carbopol 974P, (b) (4) tyloxapol, (b) (4) boric acid, (b) (4) mannitol, (b) (4) propylene glycol, (b) (4) sodium chloride, 0.003% benzalkonium chloride, pH 6.5 (same as clinical formulation) For Alphagan P, the suspension contained 0.005% Purite®, and unspecified concentrations of sodium carboxymethylcellulose, sodium borate and boric acid, sodium chloride, potassium chloride, calcium chloride, and magnesium chloride, pH 6.6 to 7.4 For Azopt®, the suspension contained 0.01% benzalkonium chloride and unspecified concentrations of mannitol, Carbopol 974P, tyloxapol, edentate disodium, pH 7.5
Species/Strain:	Male New Zealand white/red F1 cross rabbits
Number/Sex/Group:	<ul style="list-style-type: none"> 30 males for the three Alcon suspensions (sub-groups of 6 per time point) 15 males for the Alphagan P and Azopt® suspensions (sub-groups of 3 per time point)
Weight:	3 kg (mean weight)

Method notes:

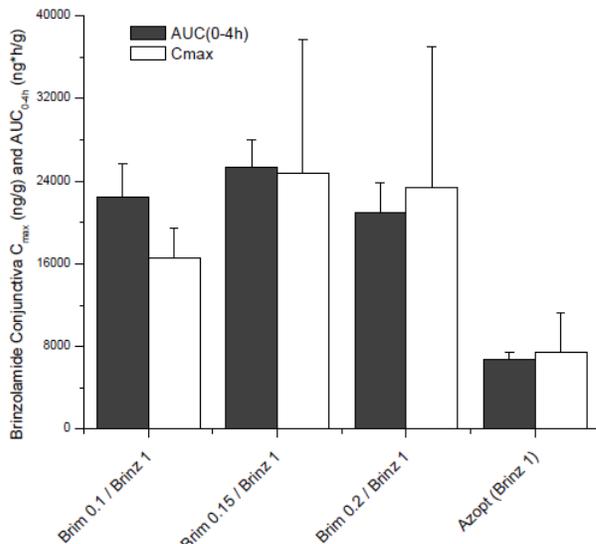
- Rabbits were not fasted
- Five sampling time points: 0.25, 0.5, 1, 2 and 4 hours post-dose.
- For the Alcon suspensions, the right eye was analyzed from brinzolamide and N-desethyl brinzolamide (a metabolite of brinzolamide) and the left eye was analyzed for brimonidine (i.e. 6 eyes/dose/time point)
 - For the commercial suspensions, both eyes were analyzed (i.e. 6 eyes/dose/time point), because the suspensions were not mixtures (i.e. Alphagan P is brimonidine only, and Azopt® is brinzolamide only)
- Rabbits were euthanized by intravenous injection of an "appropriate euthanizing agent" (report page 32); the aqueous humor, bulbar conjunctiva and iris ciliary body were immediately collected into eppendorf tubes, and cooled to 5°C. The conjunctiva and iris ciliary body were weighed, and then the tissues were frozen (-70°C).

Results notes:

- The authors concluded, and this reviewer agrees, that the brinzolamide uptake in to the bulbar conjunctiva for the three Alcon formulations was greater than the

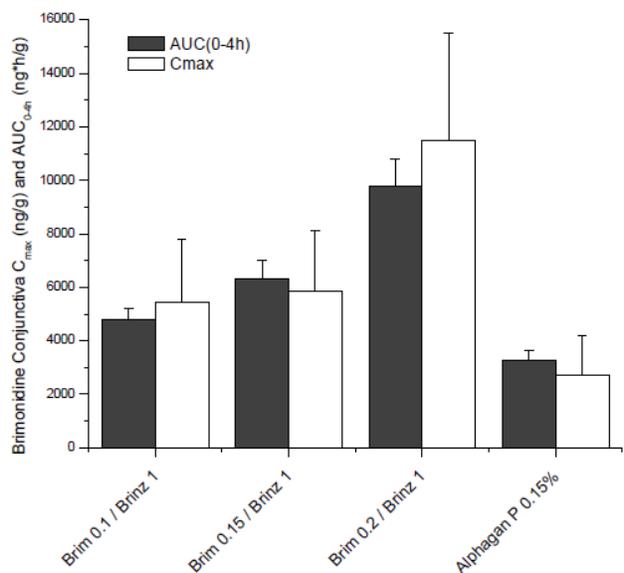
brinzolamide uptake for Azopt. The authors report that the difference was statistically significant ($p \leq 0.05$). From the report (page 22):

Figure 3: Differences in brinzolamide uptake into the bulbar conjunctiva (C_{max} and AUC) for the rabbit single-dose distribution study (report # TDOC-0009954)



- Although not noted by the authors, a similar difference in brimonidine distribution into the bulbar conjunctiva is apparent (not statistically significant at the $p \leq 0.05$ level). From report page 19:

Figure 4: Differences in brimonidine uptake into the bulbar conjunctiva (C_{max} and AUC) for rabbit single-dose distribution study (report # TDOC-0009954)



- No other remarkable differences between the commercial products and Alcon's products were noted. A clear dose-response is apparent for the three Alcon

formulations tested. N-desethyl brinzolamide (AL-8520) was not detected in any of the samples at any time point.

Table 12: Brimonidine PK for the rabbit single-dose distribution study (report # TDC-0009954)

Treatment	Aqueous humor		Bulbar conjunctiva		Iris-ciliary body	
	AUC _{0-4h} (ng*h/ml)	C _{max} (ng/ml)	AUC _{0-4h} (ng*h/ml)	C _{max} (ng/ml)	AUC _{0-4h} (ng*h/ml)	C _{max} (ng/ml)
Brimonidine 0.1% + Brinzolamide 1%	400 ± 20	290 ± 50	4800 ± 400	5500 ± 2300	15200 ± 900	4700 ± 1500
Brimonidine 0.15% + Brinzolamide 1%	540 ± 30	390 ± 130	6300 ± 700	5900 ± 2200	23800 ± 1800	8000 ± 1800
Brimonidine 0.2% + Brinzolamide 1%	680 ± 60	540 ± 320	9800 ± 970	11500 ± 4000	31300 ± 2000	10000 ± 2600
Alphagan P (brimonidine 0.15%)	440 ± 30	290 ± 60	3300 ± 340	2700 ± 1500	18000 ± 1000	6700 ± 1800

Data presented as mean ± standard deviation. Values rounded by this reviewer for readability.

Table 13: Brinzolamide PK for the rabbit ocular distribution study (report # TDC-0009954)

Treatment	Aqueous humor		Bulbar conjunctiva		Iris-ciliary body	
	AUC _{0-4h} (ng*h/ml)	C _{max} (ng/ml)	AUC _{0-4h} (ng*h/ml)	C _{max} (ng/ml)	AUC _{0-4h} (ng*h/ml)	C _{max} (ng/ml)
Brimonidine 0.1% + Brinzolamide 1%	890 ± 60	340 ± 100	22500 ± 3200	16500 ± 2900	5900 ± 500	1930 ± 660
Brimonidine 0.15% + Brinzolamide 1%	800 ± 80	320 ± 110	25300 ± 2700	24800 ± 12900	5600 ± 400	2210 ± 530
Brimonidine 0.2% + Brinzolamide 1%	680 ± 70	220 ± 80	21000 ± 2800	23400 ± 13600	5700 ± 500	2040 ± 660
Alphagan P (brimonidine 0.15%)	770 ± 70	270 ± 90	6700 ± 800	7450 ± 3840	5500 ± 400	1840 ± 580

Data presented as mean ± standard deviation. Values rounded by this reviewer for readability.

Study title: Ocular tissue distribution study of brinzolamide (AL-4862) and brimonidine (AL-8923) following topical ocular administration of brinzolamide 1% / brimonidine 0.2%, AZOPT, or brimonidine (Falcon) to New Zealand White/Red F1 cross rabbits

Study no.: TDOC-0014507
Study report location: NDA module 4.2.2.3 Pharmacokinetics – Distribution
Conducting laboratory and location: Alcon Research, Ltd.
Forth Worth, Texas 76134
Report date: February 20, 2012
GLP compliance: No
Drug, lot #: • Brinzolamide 1% / Brimonidine 0.2%, lot # 18543-01 (purity not reported)
• Falcon's brimonidine 0.2%, lot # 18300F
• Azopt, lot # 171289F

Key Study Findings

- One Alcon formulation (brinzolamide 1% / brimonidine tartrate 0.2%) was evaluated for ocular tissue distribution in male rabbits following D1 and D14 of BID bilateral topical dosing. Both Azopt 1% and Falcon's brimonidine tested for comparison.
- Brinzolamide accumulation was observed in all tissues measured (comparing D14 versus D1). Brimonidine accumulation was observed in all tissues except bulbar conjunctiva (comparing D14 versus D1).
- For brimonidine: the highest concentrations were found in the iris/ciliary body; iris ciliary body > choroid > conjunctiva > retina, cornea, aqueous humor > lens > plasma.
- For brinzolamide: distribution on D1 was cornea > choroid > conjunctiva > iris ciliary body > blood > retina > aqueous humor > lens. However, brinzolamide distribution on D14 was retina > choroid > blood > cornea > conjunctiva > aqueous humor > lens.
- No toxicology endpoints were measured in these experiments.

Methods	
Doses:	<p>First experiment:</p> <ul style="list-style-type: none"> Alcon's brinzolamide 1% / brimonidine 0.2% Azopt 1% (brinzolamide) Falcon's brimonidine 0.2% <p>Second experiment:</p> <ul style="list-style-type: none"> Alcon's brinzolamide 1% / brimonidine 0.2% Falcon's brimonidine 0.2%
Frequency of dosing:	<p>First experiment: single bilateral dose or BID bilateral dosing for 14 days. Second experiment: BID bilateral dosing only for 14 days bilateral (30 µl to the right eye and 30 µl) dosing twice daily x 14 (total dose = 60 µl/eye/day = 120 µl/rabbit/day)</p>
Route of administration:	Topical ocular dosing to both eyes
Formulation/Vehicle:	<ul style="list-style-type: none"> For the Alcon product: (b) (4) mannitol, (b) (4) sodium chloride, (b) (4) propylene glycol, (b) (4) boric acid, (b) (4) carbopol 974P, (b) (4) tyloxapol, 0.003% benzalkonium chloride, water (b) (4) pH 6.5 For Azopt: (b) (4) mannitol, (b) (4) sodium chloride, (b) (4) carbopol 974P, (b) (4) tyloxapol, 0.1% benzalkonium chloride, (b) (4) edentate disodium, water (b) (4) pH 7.5
Species/Strain:	Male New Zealand White/Red F1 cross rabbits
Number/Sex/Group:	<p>First experiment:</p> <ul style="list-style-type: none"> 72 rabbits for Alcon's brinzolamide 1% / brimonidine 0.2% (6 rabbits/time point) 36 rabbits for Azopt (3 rabbits/time point) 36 rabbits for Falcon's brimonidine (3 rabbits/time point) <p>Second experiment:</p> <ul style="list-style-type: none"> 18/group (3 rabbits/time point)
Weight:	Mean 2.78 kg

- For the first experiment:
 - Rabbits were harvested on D1, following the first dose (at 0.5, 1, 2, 4, 8 and 12 hours) and on D14, following the last dose (0.5, 1, 2, 4, 8 and 12 hours).
 - For the Alcon brinzolamide 1% / brimonidine 0.2% group only: right eye analyzed for brinzolamide and N-desethyl brinzolamide; left eye analyzed for brimonidine (3 eyes/time point)
 - Tissues collected: whole blood, plasma, aqueous humor, iris capillary body, bulbar conjunctiva, cornea, lens cortex/nucleus and lens capsule combined, retina, choroid
 - Bioanalysis for brimonidine in the iris ciliary body were above the upper limit of quantitation at D14. The authors attributed this to an inadequate

standard curve, and conducted a second experiment to obtain iris ciliary body data

- For the second experiment:
 - Rabbits were dosed for 14 days prior to tissue harvest, after the last dose: 0.5, 1, 2, 4, 8 and 12 hours.
 - Only the iris ciliary body and aqueous humor were collected.

Results notes:

- N-desethyl brinzolamide (AL-8520) was detected in the choroid of 3 rabbits at 12 hours post-dose on D1, and in the choroid of four rabbits, beginning at 1 hour post-dose on D14. N-desethyl brinzolamide was not detected in any other tissue at any time point.
- Brinzolamide = AL-4862; brimonidine = AL-8923

Table 14: Brinzolamide (AL-4862) PK after single bilateral dosing and 14 days bilateral dosing in male rabbits (report # TDOC-0014507)

Treatment Group	Matrix	DAY 1			DAY 14		
		AUC _{0-12h} (ng*h/g or ng*h/ml)	C _{max} (ng/g or ng/ml)	T _{max} (h)	AUC _{0-12h} (ng*h/g or ng*h/ml)	C _{max} (ng/g or ng/ml)	T _{max} (h)
1% brinzolamide / 0.2% brimonidine	Aqueous humor	1350	277	2	1800	354	1
	Blood	16,400	1760	12	62,500	602	4
	Choroid	36,700	5030	4	89,900	9260	0.5
	Conjunctiva bulbar	15,800	11,000	0.5	26,600	8980	0.5
	Cornea	16,200	4950	0.5	24,000	5500	1
	Iris ciliary body	19,400	2540	4	26,600	3300	4
	Lens	BLQ	BLQ	NC	4880	460	1
	Retina	3770	442	4	90,600	8220	0.5
Azopt 1%	Aqueous humor	1740	389	1	2300	681	1
	Blood	16,500	1640	12	64,100	6140	0.5
	Choroid	25,400	3000	2	55,500	7630	1
	Conjunctiva bulbar	16,500	3410	0.5	28,500	3850	0.5
	Cornea	18,700	4890	0.5	24,300	5950	1
	Iris ciliary body	20,900	2390	2	40,400	5580	2
	Lens	374	46	8	5490	493	12
	Retina	3210	333	1	87,500	7760	8

Data presented as means. BLQ = below the limit of quantitation. NC = not calculated.

Table 15: Brimonidine (AL-8923) PK after single bilateral dosing and 14 days bilateral dosing in male rabbits (report # TDOC-0014507)

Treatment Group	Matrix	DAY 1			DAY 14		
		AUC _{0-12h} (ng*h/g or ng*h/ml)	C _{max} (ng/g or ng/ml)	T _{max} (h)	AUC _{0-12h} (ng*h/g or ng*h/ml)	C _{max} (ng/g or ng/ml)	T _{max} (h)
Experiment 1							
1% brinzolamide / 0.2% brimonidine	Aqueous humor	572	396	0.5	1170	575	0.5
	Plasma	2.03	1.71	0.5	3.82	2.77	0.5
	Choroid	30,000	2930	8	752,000	75,200	1
	Conjunctiva bulbar	7470	2900	0.5	11,400	3900	0.5
	Cornea	3680	2840	0.5	6470	3480	0.5
	Iris ciliary body	98,800	9890	12	ALQ	ALQ	NC
	Lens	77.6	10	1	685	74.8	2
	Retina	742	91.7	0.5	13,300	2130	12
Falcon's 0.2% brimonidine	Aqueous humor	339	237	0.5	557	264	0.5
	Blood	2.47	1.43	0.5	3.35	2.67	0.5
	Choroid	10,000	1010	4	280,000	28,200	1
	Conjunctiva bulbar	1570	802	0.5	4720	1010	0.5
	Cornea	2220	1480	0.5	3420	163	0.5
	Iris ciliary body	56,500	5940	12	ALQ	ALQ	NC
	Lens	48.8	6.11	1	350	45.3	1
	Retina	851	104	1	14,900	1980	4
Experiment 2							
1% brinzolamide / 0.2% brimonidine	Iris ciliary body	Not measured			1,260,000	136,000	0.5
Falcon's 0.2% brimonidine	Iris ciliary body	Not measured			1,110,000	105,000	2

Data presented as means. ALQ = above the limit of quantitation. NC = not calculated.

5.2 Toxicokinetics

The applicant submitted separate TK study reports for the 6-week and 9-month studies in NDA module 4.2.2.2 Pharmacokinetics -> Absorption. Review of the data from these reports is documented below.

6 General Toxicology

6.1 Single-Dose Toxicity

The applicant did not conduct or identify any single-dose toxicity studies with the brinzolamide/brimonidine combination. Single-dose toxicity studies with each (as a single agent) are referenced.

6.2 Repeat-Dose Toxicity

The applicant submitted two repeat-dose toxicity studies, using topical ocular dosing to administer Alcon's brinzolamide and brimonidine formulations to rabbits for 6-weeks and 9-months.

For the 9-month study, the Applicant submitted both the 3-month interim report, and the final 9-month report (which included the 3-month data). For readability, review of both reports is combined below.

Study title: 6-Week topical ocular safety evaluation of AL-4862 (brinzolamide) and AL-8923A (brimonidine tartrate) in F1 pigmented rabbits	
Study no.:	TDOC-0010074
Study report location:	NDA module 4.2.3.2 Repeat dose toxicity – rabbit – topical - medium
Conducting laboratory and location:	Alcon Research, Ltd. Fort Worth, Texas 76134
Date of study initiation:	July 1, 2009
GLP compliance:	Yes
QA statement:	Yes
Drug, lot #, and % purity:	<ul style="list-style-type: none"> • Brinzolamide 1% / brimonidine tartrate 0.15% ophthalmic suspension, lot # 09-55565-1 • Brinzolamide 2% / brimonidine tartrate 0.2% ophthalmic suspension, lot # 09-55375-1 <p>The brinzolamide was $\geq 99.7\%$ pure The brimonidine tartrate was $\geq 99.4\%$ pure</p>

Key Study Findings

- Five groups of rabbits, 4/sex/dose, were treated daily for 43 days and necropsied on D44. Two dose-levels of Alcon's combination product were tested: brinzolamide 1% / brimonidine 0.15%, and brinzolamide 2% / brimonidine 0.2%.
- No new or unexpected findings were apparent. The authors consider the NOAEL to be the high-dose (Brin 2%/ Brim 0.2%).

- Decreased IOP, sedation, penile erection and swelling of the urogenital area were observed in both combination-treated groups. The Applicant considers these effects pharmacological rather than toxicological.
- This reviewer considers sedation to be unintended and undesirable, and therefore adverse.
- This Applicant attributes the penile erection and urogenital swelling to the known effects of brimonidine in rabbits, considering it a species-specific effect.
- Slight treatment-related changes in serum cholesterol were detected. The Applicant concludes, and this reviewer concurs, that the effect was not adverse (due to the small magnitude of the change)
- Following topical ocular administration of the combination products, systemic exposure to brinzolamide, N-desethyl brinzolamide, and brimonidine were detected. No accumulation was apparent.

Methods																					
Doses:	<p>Single drop (approximately 30 µl)</p> <ul style="list-style-type: none"> ● Untreated control ● Vehicle control ● Brinzolamide 1% / brimonidine tartrate 0.15% <ul style="list-style-type: none"> ○ 1.8 mg brinzolamide / 0.27 mg brimonidine ● Brinzolamide 2% / brimonidine tartrate 0.2% <ul style="list-style-type: none"> ○ 3.6 mg brinzolamide / 0.36 mg brimonidine ● Falcon’s brimonidine tartrate 0.2% (marketed comparator) <ul style="list-style-type: none"> ○ 0.36 mg brimonidine <p>(doses calculated by the authors, report p 14)</p>																				
Frequency of dosing:	Three times per day (3 hours apart; at approximately 7:30 am, 11:30 am and 3:30 pm) bilaterally for 43 days (necropsy on D44)																				
Route of administration:	Topical ocular in both eyes																				
Dose volume:	30 µl/eye x 3/day																				
Formulation/Vehicle:	<table border="1"> <thead> <tr> <th>Component</th> <th>%</th> </tr> </thead> <tbody> <tr> <td>Mannitol</td> <td>(b) (4)</td> </tr> <tr> <td>Sodium chloride</td> <td>(b) (4)</td> </tr> <tr> <td>Propylene glycol</td> <td>(b) (4)</td> </tr> <tr> <td>Boric acid</td> <td>(b) (4)</td> </tr> <tr> <td>Carbopol 974p</td> <td>(b) (4)</td> </tr> <tr> <td>Tyloxapol</td> <td>(b) (4)</td> </tr> <tr> <td>Benzalkonium chloride</td> <td>0.003%</td> </tr> <tr> <td>Sodium hydroxide & hydrochloric acid</td> <td>To pH 6.5 ± 0.2</td> </tr> <tr> <td>Purified water</td> <td>(b) (4)</td> </tr> </tbody> </table>	Component	%	Mannitol	(b) (4)	Sodium chloride	(b) (4)	Propylene glycol	(b) (4)	Boric acid	(b) (4)	Carbopol 974p	(b) (4)	Tyloxapol	(b) (4)	Benzalkonium chloride	0.003%	Sodium hydroxide & hydrochloric acid	To pH 6.5 ± 0.2	Purified water	(b) (4)
Component	%																				
Mannitol	(b) (4)																				
Sodium chloride	(b) (4)																				
Propylene glycol	(b) (4)																				
Boric acid	(b) (4)																				
Carbopol 974p	(b) (4)																				
Tyloxapol	(b) (4)																				
Benzalkonium chloride	0.003%																				
Sodium hydroxide & hydrochloric acid	To pH 6.5 ± 0.2																				
Purified water	(b) (4)																				

	<i>This formulation is identical to the clinical formulation</i>
Species/Strain:	Rabbit – New Zealand White x Red, F1 <i>Oryctolagus cuniculus</i>
Number/Sex/Group:	4/sex/group
Age:	4 to 5 months at initiation of dosing
Weight:	3.0 to 3.3 kg at initiation of dosing

Observations and Results

Mortality

- No mortalities occurred prior to scheduled necropsy
- Checked twice daily (morning and afternoon)
- Food consumption and feces production were noted

Clinical Signs

- Once weekly cage side observation
- Clinical signs were also noted during removal of the rabbit from the cage (i.e. body weight measurement)
- The timing of clinical sign evaluations was not reported.
- Penile erection was observed in all male brimonidine-treated rabbits
 - First observation on D23
 - The authors consider this effect clearly treatment-related, and attribute it to the known pharmacology of brimonidine (α_2 -activation) in rabbits.

Table 16: Brimonidine-induced penile erection observed in the 6-week rabbit toxicity study (report # TDOC-0010074)

Groups (N=4 males)	Penile erection incidence on study days							
	1,8,15	23	27	30	34	37	41	42
Untreated control	0	0	0	0	0	0	0	0
Vehicle control	0	0	0	0	0	0	0	0
Brinzolamide 1% / Brimonidine Tartrate 0.15%	0	4	4	4	4	4	4	4
Brinzolamide 2% / Brimonidine Tartrate 0.2%	0	3	4	4	4	4	4	4
Falcon's Brimonidine Tartrate 0.2%	0	3	3	3	4	4	4	4

- On D42, the duration of penile erection was assessed after the first dose (at 7:30 am) and third dose (at 3:30 pm).
 - A dose-response was apparent for duration of response, comparing the males exposed to 0.2% brimonidine versus those exposed to 0.15% brimonidine.
 - After the third dose on D42, Alcon's brinzolamide 2% / brimonidine tartrate 0.2% appeared to induce erection longer than Falcon's brimonidine tartrate. The authors did not comment, and this reviewer is uncertain whether this is a true difference, or incidental.

Table 17: The duration of rabbit penile erection showed a dose-response for brimonidine, and Alcon's brinzolamide 2%/brimonidine tartrate 0.2% appeared more active than Falcon's brimonidine tartrate 0.2% (report # TDOC-0010074)

Treatment	Male ID #	Duration (hr) after the first dose (7:30 am)	Duration (hr) after the third dose (3:30 pm)
Brinzolamide 1% / Brimonidine Tartrate 0.15%	3001	2	1.5
	3002	2	1.5
	3003	2	2
	3004	2	1
	Mean	2	1.5
Brinzolamide 2% / Brimonidine Tartrate 0.2%	4001	2	1.5
	4002	2	2
	4003	2	2
	4004	2.5	1.5
	Mean	2.1	1.8
Falcon's Brimonidine Tartrate 0.2% positive control	5001	2	2
	5002	2	1
	5003	2	1
	5004	2	1.5
	Mean	2	1.4

- Female rabbits exhibited slight-to-moderate urogenital swelling from D23 to D42. The authors considered these observations treatment-related, and attributed them to brimonidine exposure; this reviewer concurs. The incidence was notably higher in the brimonidine positive control group compared to either the low- or high-dose groups.
 - Note: the data in Table 18 likely under-represent the true incidence; rabbits were evaluated more carefully on D42.

Table 18: Reported incidence of female urogenital swelling in the 6-week rabbit study (report # TDOC-0010074)

Day	Untreated	Vehicle	Brin 1%/Brim 0.15%	Brin 2%/Brim 0.2%	Brim 0.2% positive control
23	0/4	0/4	0/4	2/4	2/4
27	0/4	1/4	2/4	1/4	0/4
30	0/4	0/4	2/4	0/4	3/4
34	0/4	0/4	1/4	0/4	4/4
37	0/4	0/4	1/4	0/4	4/4
41	0/4	0/4	0/4	1/4	3/4
42	0/4	0/4	0/4	0/4	3/4
Total #	0	1	6	4	22

- Sedation was observed in brimonidine-exposed rabbits; the authors considered the effect treatment-related, and consistent with the known pharmacology of brimonidine (α_2 -activation). Two brimonidine-treated rabbits exhibited head tilt, and one exhibited unsteady gait; this reviewer considers these signs secondary to sedation.
 - Note: the data in Table 19 likely under-represent the true incidence; rabbits were evaluated more carefully on D42.

Table 19: Reported incidence of sedation in the 6-week rabbit study (report # TDOC-0010074)

Day	Untreated	Vehicle	Brin 1%/Brim 0.15%	Brin 2%/Brim 0.2%	Brim 0.2% positive control
20	0/8	0/8	0/8	1/8	3/8
23	0/8	0/8	0/8	0/8	5/8
27	0/8	0/8	3/8	2/8	6/8
30	0/8	0/8	1/8	2/8	6/8
34	0/8	0/8	1/8	1/8	6/8
37	0/8	0/8	1/8	0/8	6/8
41	0/8	0/8	1/8	0/8	6/8
42	0/8	0/8	7/8	5/8	7/8
Total #	0	0	14	11	45

Data from report page 27 (both sexes combined per treatment group)

Body Weights

- No treatment-related effects apparent
- Measured pre-dose, and on D6, 13, 27, 41 and 44

Feed Consumption

- No treatment-related effects apparent. Food consumption was assessed as part of the daily mortality check.
- Rabbits were fasted prior to necropsy.

Ophthalmoscopy

- Evaluations were conducted for both eyes of each rabbit:
 - Biomicroscopy (slit lamp) pre-dose, D7, D14, D28 and D42 for: conjunctiva, cornea, anterior chamber, light reflex, lens, and iris
 - Dilated lens evaluations, indirect ophthalmoscopic examination, and specular microscopy at pre-dose and on D42
 - Pachymetry and IOP pre-dose, D6, D27 and D41
- Treatment decreased IOP; statistical significance was achieved occasionally.
- The authors considered the small apparent increase in corneal thickness observed in female rabbits to be incidental (report page 30).

Table 20: Treatment-related ophthalmic changes (increased corneal thickness, decreased IOP) in the 6-week rabbit study (report # TDOC-0010074)

Endpoint	Day	Males					Females				
		Un-treated	Vehicle	Brin 1/ Brim 0.15	Brin 2 / Brim 0.2	Brim 0.2	Un-treated	Vehicle	Brin 1/ Brim 0.15	Brin 2 / Brim 0.2	Brim 0.2
Pachymetry OD (µm)	-2	350	362	347	367	342	359	354	369	370	342
	6	365	365	358	389	348	372	368	385	383	340
	27	365	371	369	377	346	365	367	374	376	353
	41	358	374	375	391	361	372	359	397 *	389	349
Pachymetry OS (µm)	-2	357	365	348	372	346	370	356	362	367	344
	6	360	367	358	380	345	370	358	384	375	333
	27	367	359	362	383	348	359	369	376	382	347
	41	361	357	374	390	355	373	347	396 **	381	338
IOP OD (mm Hg)	-2	24	25	22	25	25	22	21	25	23	20
	6	23	22	19	20	14 **	21	19	21	16	15
	27	25	24	26	22	16 **	24	23	24	20	16 **
	41	21	21	16	19	17	22	19	21	17	14
IOP OS (mm Hg)	-2	23	23	21	21	24	22	19	25	21	20
	6	21	21	18	18	13 **	19	17	18	15	13 **
	27	25	23	24	22	17 **	23	21	25	20	16 **
	41	21	19	15	17	15	21	19	17	15	12 **

Values rounded by this reviewer for readability. Statistically significant values bolded for readability.

* statistically significant by 1 way analysis of variance, $p < 0.05$.

** statistically significant by 1 way analysis of variance, $p < 0.01$

Hematology

- No treatment-related effects were apparent

- Blood was collected on D38 only for hematology, coagulation, and clinical chemistry (standard panel of endpoints), with fasting overnight prior to the blood draw

Clinical Chemistry

- Blood was collected on D38 only.
- Both Brin/Brim treated groups exhibited decreased cholesterol. The authors considered this effect treatment-related but not adverse.
- No serum glucose differences were apparent among groups for either sex.
- No other treatment-related effects were apparent.

Table 21: Selected serum chemistry (D38) for the 6-week rabbit study (report # TDOC-0010074)

Endpoint	Males					Females				
	un-treated	Vehicle	Brin 1/ Brim 0.15	Brin 2/ Brim 0.2	Brim 0.2	Vehicle	un-treated	Brin 1/ Brim 0.15	Brin 2/ Brim 0.2	Brim 0.2
Cholesterol (mg/dl)	33	31	29	27	26	57	70	51	40 *	35 *
Glucose (mg/dl)	87	80	76	82	84	72	74	80	75	72

Values rounded by this reviewer for readability. * statistically significant by 1 way analysis of variance, $p < 0.05$.

Gross Pathology

- No treatment-related effects were apparent
 - Notably, no gross lesions were reported for any animal (report pages 134-139)
- All rabbits were necropsied on D44

Organ Weights

- No treatment-related effects were apparent
- Weights were collected for the adrenal, brain, heart, kidney, liver, ovary, spleen and testes only.

Histopathology

Adequate Battery: yes. Histopathology was performed at [REDACTED] (b) (4) in accordance with GLP.

- Tissues/organs collected were: eyes, adnexa, adrenal, aorta, bone, bone marrow, brain, cervix, epididymis, esophagus, gall bladder, salivary gland, heart, colon, duodenum, cecum, ileum jejunum, rectum, kidney, larynx, liver, lungs, mesenteric and submandibular lymph nodes, mammary gland, nasal lacrimal region, oviduct, ovary, pancreas, pituitary, prostate, parathyroid, ribs, spinal cord, skin, skeletal muscle, sciatic nerve, spleen, sacculus rotundus, stomach, sternum, seminal vesicle, testes, thyroid, thymus, tongue, trachea, urinary bladder, urethra, ureter, uterus, vagina, penis and vulva.
- Eyes and adnexa were evaluated for all rabbits. Other tissues were only evaluated for the vehicle-control group and the Brin 2% / Brim 0.2% group.

Peer Review: no

Histological Findings

- No treatment-related effects were apparent
- The authors concluded, and this reviewer agrees, that the observed histopathological lesions appear incidental (e.g. mononuclear cell infiltrate, epidermal hyperplasia,

Endpoints Not Assessed

No ECG or urinalysis endpoints

Toxicokinetics

- Blood was collected on D1, D15 and D43
 - For the controls at 0 and 1 hour post-dose
 - For the treated groups at 0, 0.5, 1, and 3 hours post-dose
- Blood was analyzed for brinzolamide, N-desethyl brinzolamide and brimonidine.
- No accumulation was apparent.
- Comparing doses on D43, the increase in brinzolamide was less than dose-proportional, and the increase in brimonidine was dose-proportional.

Table 22: PK for the 6-week rabbit study (report # TDOC-0010074)

Group	Study day	Brimonidine		Brinzolamide		N-desethyl brinzolamide	
		AUC _{0-3h} (ng*h/ml)	C _{max} (ng/ml)	AUC _{0-3h} (ng*h/ml)	C _{max} (ng/ml)	AUC _{0-3h} (ng*h/ml)	C _{max} (ng/ml)
Brin 1%/ Brim 0.15%	1	2.8	2.4	16200	5790	NQ	NQ
	15	2.7	2.2	19500	6920	280	120
	43	1.4	1.4	20500	7190	NQ	120
Brin 2%/	1	3.9	2.8	19100	6770	NQ	NQ

Brim	15	2.4	2.0	19800	6990	370	150
0.2%	43	1.0	0.81	22300	7860	380	130
Brim	1	0.94	0.87	-			
0.2%	15	3.0	2.7				
	43	2.0	1.9				

Values presented as means, rounded by this reviewer for readability.

NQ: not quantified; authors considered the number of data points above the LLOQ insufficient to calculate a reliable value

Stability and Homogeneity

The Brin/Brim test articles were “shaken well prior to dosing, as per label instructions” (report page 15). As with the 9-month study (reviewed below) no mention is made in this report regarding shaking the test articles for the dose analysis, and this is a study limitation.

Pre- and post-study analyses of the test suspensions were conducted for brinzolamide, brimonidine, and benzoalkonium concentration; all were within 5.5% of nominal (report page 79).

Study title: Final report: nine-month topical ocular safety evaluation of AL-4862 (brinzolamide) and AL-8923A (brimonidine tartrate) with a 3-month interim in F1 pigmented rabbits

Study no.:	<ul style="list-style-type: none"> • TDOC-0013267 (for the completed study) • TDOC_0011992 (for the 3-month interim report)
Study report location:	NDA module 4.2.3.2 Repeat dose toxicity – rabbit – topical - long
Conducting laboratory and location:	Alcon Research, Ltd. Fort Worth, Texas 76134
Date of study initiation:	June 16, 2010
Report date:	February 22, 2012
GLP compliance:	Yes
QA statement:	Yes
Drug, lot #, and % purity:	<ul style="list-style-type: none"> • Brinzolamide 1% / brimonidine tartrate 0.2% ophthalmic suspension, lot # 10-59208-1 • Brinzolamide 2% / brimonidine tartrate 0.2% ophthalmic suspension, lot # 10-59193-1 • Purity of brinzolamide \geq 98.5% • Purity of brimonidine \geq 98%

Key Study Findings

- The study authors and the Applicant consider the high-dose, Brin 2%/Brim 0.2%, to be the NOAEL, because they consider the observed treatment-related effects to be pharmacological and not adverse. This reviewer disagrees.
- For ocular toxicity, this reviewer concurs that the high-dose was a NOAEL.
- No NOAEL for systemic toxicity was observed. Several systemic adverse effects appear to be species-specific and not expected to be relevant to human patients:
 - Corneal thickness – the Applicant reports that this effect has been observed in previous rabbit studies with brinzolamide, but not in humans or other animal models
 - Penile erection, female urogenital swelling and discoloration - consistent with the 6-week study
- Hyperglycemia and liver toxicity (slight increases in weight, hepatocellular cytoplasmic vacuolation)
 - The observed increase in serum glucose is attributed to brimonidine, and has been observed in previous studies in rabbits and other animal models. However, increased serum glucose has not been detected as a clinical adverse effect.

- Notably, a dose-response was observed for liver toxicity, suggesting that the effects attributed to brimonidine (0.2% in both the low- and high-dose groups) were affected by the dose level of brinzolamide (1% versus 2%).

Methods																					
Doses:	<ul style="list-style-type: none"> • Untreated control • Vehicle control • Brinzolamide 1% + brimonidine tartrate 0.2% ophthalmic suspension (Brin 1%/Brim 0.2%) • Brinzolamide 2% + brimonidine tartrate 0.2% ophthalmic suspension (Brin 2%/Brim 0.2%) 																				
Dosing:	<p>One drop (via droptainer) to both eyes (OU) three times daily (TID) at approximately 4 hours apart. Drop sizes were measured:</p> <ul style="list-style-type: none"> • 32.8 µl for vehicle control • 36.3 µl for brin 1%/ brim 0.2% • 32.8 µl for brin 2% / brim 0.2% 																				
Route of administration:	<p>Topical ocular</p> <ul style="list-style-type: none"> • Upper eyelid was lightly retracted • The suspensions were “shaken well prior to dosing” 																				
Formulation/Vehicle:	<table border="1"> <thead> <tr> <th>Component</th> <th>%</th> </tr> </thead> <tbody> <tr> <td>Mannitol</td> <td>(b) (4)</td> </tr> <tr> <td>Sodium chloride</td> <td>(b) (4)</td> </tr> <tr> <td>Propylene glycol</td> <td>(b) (4)</td> </tr> <tr> <td>Boric acid</td> <td>(b) (4)</td> </tr> <tr> <td>Carbopol 974p</td> <td>(b) (4)</td> </tr> <tr> <td>Tyloxapol</td> <td>(b) (4)</td> </tr> <tr> <td>Benzalkonium chloride</td> <td>0.003%</td> </tr> <tr> <td>Sodium hydroxide & hydrochloric acid</td> <td>To pH 6.5 ± 0.2</td> </tr> <tr> <td>Purified water</td> <td>(b) (4)</td> </tr> </tbody> </table> <p><i>This formulation is identical to the clinical formulation</i></p>	Component	%	Mannitol	(b) (4)	Sodium chloride	(b) (4)	Propylene glycol	(b) (4)	Boric acid	(b) (4)	Carbopol 974p	(b) (4)	Tyloxapol	(b) (4)	Benzalkonium chloride	0.003%	Sodium hydroxide & hydrochloric acid	To pH 6.5 ± 0.2	Purified water	(b) (4)
Component	%																				
Mannitol	(b) (4)																				
Sodium chloride	(b) (4)																				
Propylene glycol	(b) (4)																				
Boric acid	(b) (4)																				
Carbopol 974p	(b) (4)																				
Tyloxapol	(b) (4)																				
Benzalkonium chloride	0.003%																				
Sodium hydroxide & hydrochloric acid	To pH 6.5 ± 0.2																				
Purified water	(b) (4)																				
Species/Strain:	Rabbit – New Zealand White x Red, F1 <i>Oryctolagus cuniculus</i> (pigmented strain)																				
Number/Sex/Group:	<ul style="list-style-type: none"> • Interim group: 4/sex/dose (necropsied on D92) • Terminal group: 5/sex/dose (necropsied on D274) 																				
Age:	4.5 to 5.2 months at initiation of dosing																				
Weight:	3.0 to 3.4 kg at initiation of dosing																				

The authors calculate the doses (report pages 1-2):

- Brin 1%/Brim 0.2% as 2.18 mg/animal/day of brinzolamide and 0.44 mg/animal/day for brimonidine

- Brin 2%/Brim 0.2% as 3.9 mg/animal/day of brinzolamide and 0.39 mg/animal/day for brimonidine

Observations and Results

Mortality

- No unscheduled mortalities occurred.
- Mortality and morbidity were assessed twice daily (morning and afternoon).

Clinical Signs

Treatment-related sedation, penile erection, female urogenital swelling and discoloration were noted. Because these effects are expected to be transient, they may have resolved prior to the evaluation of clinical signs. Therefore, this reviewer presumes that the reported incidences dramatically under-represent the true incidences

- Note that dosing was performed 3x daily, 4 hours apart, at approximately 7:30 am, 11:30 am, and 3:30 pm (report page 18)
- As part of the daily morning mortality/morbidity check, food consumption and fecal/urine output were evaluated.
- Cage-side observations were made once weekly (time of day not reported; e.g. page 20, pages 92-95, pages 453-493)
- Once weekly when the animal was removed from the cage (i.e. for body weight measurement; time of day not reported).
- Sedation was observed in two low-dose males and one high-dose female on D1.
 - The authors conclude, and this reviewer concurs, that the effect was expected for brimonidine.
 - Notably, the duration of sedation on D1 was not monitored
- On D1, all Brin/Brim treated males exhibited penile erection. Brim/Brin females exhibited urogenital swelling (5/5 in the low-dose group, 1/5 in the high-dose group) and urogenital discoloration (4/5 females in the low-dose group, 3/5 females in the high-dose group). These changes were noted rarely after D1.
 - The authors note (page 4) that these effects “were expected, have been characterized in a previous study with brimonidine-containing formulations, and may be a species specific effect.”
 - The authors consider these effects pharmacological and not adverse. This reviewer disagrees, and considers them adverse.
- Although the authors did not note the increased incidence of eye discharge, this reviewer considers the effect treatment-related and adverse. This effect has been reported clinically.

Table 23: Reported treatment-related clinical signs: penile erection, urogenital swelling and discoloration in the 9-month rabbit study (report # TDOC-0013267)

Observation	Days	Untreated	Vehicle	Brin 1%/Brim 0.2%	Brin 2%/Brim 0.2%
Males					
Penile erection	# of observations	0	2	9	15
	# of affected animals	0	2	9	9
	Days from - to	-	188-223	D1 only	1-118
Sedation	# of observations	0	0	2	0
	# of affected animals	0	0	2	0
	Days from - to	-	-	D1 only	-
Eye discharge	# of observations	2	7	3	13
	# of affected animals	1	5	3	5
	Days from - to	251-274	84-240	167-226	55-268
Females					
Urogenital discoloration	# of observations	0	1	5	3
	# of affected animals	0	1	4	3
	Days from - to	-	D6 only	1-181	D1 only
Urogenital swelling	# of observations	10	0	5	1
	# of affected animals	1	0	5	1
	Days from - to	62-92	-	D1 only	D1 only
Sedation	# of observations	0	0	0	1
	# of affected animals	0	0	0	1
	Days from - to	-	-	-	D1 only
Eye discharge	# of observations	0	6	6	15
	# of affected animals	0	3	3	2
	Days from - to	-	97-209	69-265	62-272

Body Weights

- Body weight was measured weekly (the first measurement was pre-dose, and the second body weight measurement was on D6)
- For males in both Brin/Brim groups, slight decreases in weight gain were intermittently apparent (0 to 4% less than controls per week, statistically significant on D6, 13, 62, 69, 90, 104, 111, 139 and 274), consistent with the observed decrease in food consumption.
- The authors acknowledged the decrease in the results section (e.g. report page 28) but not in the conclusions (report page 3)
- These small changes in weight gain are not adverse *per se*, but provide context for understanding the biological relevance of the treatment-related sedation and hyperglycemia.

Table 24: Selected data showing the slight treatment-related suppression of body weight gain in the 9-month rabbit study (report # TDOC-0013267)

Treatment → Day ↓	Males				Females			
	Untreated	Vehicle	Brin 1 /Brim 0.2	Brin 2/ Brim 0.2	Untreated	Vehicle	Brin 1 /Brim 0.2	Brin 2/ Brim 0.2
-2	3150	3140	3140	3150	3200	3220	3220	3210
6	3280	3280	3210 (-2%)	3180 * (-3%)	3290	3330	3280 (-2%)	3270 (-2%)
92	3410	3480	3430 (-1%)	3270 * (-6%)	3630	3620	3530 (-3%)	3550 (-2%)
274	3820	3660	3560 * (-3%)	3530 * (-4%)	3980	3890	3760 (-3%)	3650 (-6%)

Data presented as means, rounded by this reviewer for readability. Values in parenthesis are the percent change from vehicle control. N = 9/sex/group for D-2 to D92, and N = 5 for D274.

* Statistically significant $p \leq 0.05$ (1 way analysis of variance)

** Statistically significant $p \leq 0.01$ (1 way analysis of variance)

Feed Consumption

- Rabbits were individually housed, and received a measured amount of feed (i.e. not *ad libitum*) with supplementation (kale, carrots, apples). No data were provided regarding the amount of supplement provided/consumed.
- Food consumption was decreased in the Brin/Brim treated groups, with a higher incidence apparent in females compared to males.
 - The authors attributed the decreased food consumption to brimonidine-induced sedation. This reviewer disagrees – the data suggest a dose-response for increasing brinzolamide, suggesting that brinzolamide may have contributed to the activity of brimonidine.
 - The authors consider the effect pharmacological, secondary to sedation, pharmacology, and not adverse. This reviewer disagrees - sedation and resulting toxicities are adverse effects.

- From report page 29:

Table 25: Incidences of decreased food consumption in the 9-month rabbit study (report # TDOC-0013267)

Incidences→ Treatment↓	Male				Female			
	D1-90	D91-180	D181-274	Total	D1-90	D91-180	D181-274	Total
Untreated control	0	0	1	1	6	18	23	47
Vehicle	0	0	1	1	5	50	50	105
Brin 1%/Brim 0.2%	3	4	9	16	59	43	83	191
Brin 2%/Brim 0.2%	16	3	37	56	67	66	81	221

Note: these data were collected mainly as clinical observations, but the sums also reflect data collected during the daily morbidity/mortality checks.

Ophthalmoscopy

- Endpoints (both eyes):
 - Slit lamp biomicroscopy (of the conjunctiva, cornea, anterior chamber, light reflex, lens, and iris) was performed pre-dose, on D3, D7 and weekly thereafter
 - Dilated lens evaluation, indirect ophthalmoscopic examination (of the fundus), pachymetry (of central corneal thickness), and IOP were measured pre-dose, D91, 182 and 273
 - Specular microscopy (of the central corneal endothelium) was performed pre-dose, D87 and D269
- Brin/Brim treatment decreased IOP on D6 and D90. Notably, IOP was not decreased on either D181 or D272.
- Brin/Brim treatment increased corneal thickness compared to the controls.
 - The authors note that this effect has been observed previously in brinzolamide-treated rabbits, and appears to be a species-specific effect.
 - In the absence of slit-lamp or pathology changes, the authors considered the increase in corneal thickness to be non-adverse.

Table 26: Treatment-related ophthalmic changes (increased corneal thickness, decreased IOP) in the 9-month rabbit study (report # TDOC-0013267)

Treatment → /endpoint ↓	Day	Males				Females			
		Untreated	Vehicle	Brin 1/ Brim 0.2	Brin 2/ Brim 0.2	Untreated	Vehicle	Brin 1/ Brim 0.2	Brin 2/ Brim 0.2
Pachymetry	-2	358	357	356	365	360	363	358	358

OD (μm)	6	363	361	382 *	388 **	360	359	376	375
	90	367	379	400 **	398 **	375	382	389	393
	181	382	380	411 *	421 **	373	388	400	411 **
	272	395	389	426 *	421 *	392	393	411 *	411 *
Pachymetry OS (μm)	-2	347	351	348	356	359	353	358	356
	6	364	363	383 *	384*	357	360	367	377
	90	373	377	402 **	398 **	373	378	388	390
	181	375	376	414 **	422 **	369	386	400 *	413 **
IOP OD (mm Hg)	272	395	388	423 *	422 *	384	389	408 *	414 **
	-2	23.6	23.7	22.9	23.8	22.7	24.8	24.3	23.3
	6	21.8	21.6	17.7 **	16.7 **	22.7	22.4	17.8 **	17.4 **
	90	25.2	21.8 *	18.6 **	17.1 **	22.2	21.8	18.1 **	17.8 **
	181	19.5	18.6	22.1	19.1	18.4	17.5	20.9	16.1
IOP OS (mm Hg)	272	17.6	22.7	19.8	22.0	20.7	17.7	19.6	17.7
	-2	23.7	23.2	22.1	23.9	21.9	23.8	22.7	22.2
	6	22.1	19.6	15.9 **	16.2 **	21.4	21.1	15.1 **	15.6 **
	90	22.0	22.1	17.0 **	15.3 **	21.0	22.5	17.8	17.7
	181	19.4	22.0	19.2	24.2 *	20.1	19.3	21.0	19.3
272	18.2	18.6	19.7	19.0	18.8	18.1	15.3	18.0	

Data presented as means, rounded by this reviewer for readability. N = 9/sex/group for D-2 to D90, and N = 5 for D181 and 272.

* Statistically significant $p \leq 0.05$ (1 way analysis of variance)

** Statistically significant $p \leq 0.01$ (1 way analysis of variance)

Hematology

- No treatment-related changes were apparent.
- Blood was collected for hematology, coagulation and clinical chemistry on D91, 213 and 273. Rabbits were fasted overnight prior to each blood draw.

Clinical Chemistry

- Increased serum glucose levels were observed
 - The authors attribute this effect to the known activity of brimonidine, and did not consider the effect clinically relevant
- Although the 6-week study observed treatment-related changes in serum cholesterol, this study was unable to reliably measure cholesterol (LLOQ = 2 mg/dl), gamma glutamyl transferase (LLOQ = 10 U/L), or total bilirubin (LLQC = 0.1 mg/dl). The lack of cholesterol data is a minor study limitation.
- No other treatment-related clinical chemistry changes were apparent.

Table 27: Treatment-related decrease in serum glucose in the 9-month rabbit study (report # TDOC-0013267)

Treatment → Endpoint ↓	Day	Males				Females			
		Untreat ed	Vehicle	Brin 1 /Brim 0.2	Brin 2/ Brim 0.2	Untreat ed	Vehicle	Brin 1 /Brim 0.2	Brin 2/ Brim 0.2
Glucose (mg/dl)	91	109	114	229 * (+2.1x)	222 * (+2.0x)	109	109	282 * (+2.6x)	235 * (+2.2x)
	213	103	99.4	311 * (+3.0x)	212 * (+2.1x)	106	98	280 * (+2.6x)	161 (+1.5x)
	273	110	110	285 * (+2.6x)	239 * (+2.2x)	106	108	232 * (+2.2x)	183 * (+1.7x)

Values presented as means, with the fold-increase from untreated in parenthesis.

Values rounded by this reviewer for readability.

N = 4/sex/group for D91. N = 5/sex/group for D213 and D 273.

* statistically significant, one way analysis of variance, $p < 0.05$

Urinalysis

- Urine was collected on D274 only (i.e. not for the 3-month interim rabbits) from the bladder of each rabbit (following euthanasia). Endpoints were urine electrolytes (sodium, potassium), pH, color and appearance, specific gravity, leukocytes, blood, protein, ketones, glucose, bilirubin, urobilinogen, and nitrite (report page 23).
- The authors concluded that no treatment-related effects were apparent.
 - The summary tables (report pages 156-158) only list sodium, potassium, and pH.
 - The individual data (report pages 682-688) show that one mid-dose female (#3508) and high-dose male (#4007) had detectable glucose in the blood – reported as 100 mg/dl for both animals. One high-dose urine sample was not analyzed (# 4505)
- This reviewer concludes that the detection of urinary glucose on D274 for two Brin/Brim treated rabbits is consistent with the observed hyperglycemia.

Gross Pathology

- No treatment-related effects apparent.
- Tissues examined for all groups were: eyes, ocular adnexa (evaluated as one organ/tissue), adrenal gland, ureter, ovary, pituitary gland, mammary gland, skin, ribs, sternum, stomach, duodenum, pancreas, mesenteric lymph node, jejunum, ileum, cecum, colon, sacculus rotundus (a rabbit-specific organ), spleen, liver, gall bladder, kidney, urinary bladder, urethra, rectum, uterus, cervix, vagina, oviduct, testes, epididymis, seminal vesicle, prostate, salivary gland, submandibular lymph node, esophagus, larynx, tongue, trachea, lungs, thymus, thyroid, parathyroid, heart, aorta, brain, nasal lacrimal region, skeletal muscle, sciatic nerve, bone, bone marrow, spinal cord, penis and vulva.

Organ Weights

- Treatment affected liver weight; the effect appears consistent with the observed increase in serum glucose and the observed liver histopathology
 - The authors report (in the conclusions section, page 5) “no adverse changes in organ weight”, without explicitly mentioning liver weight
 - The authors report in the results section (pages 3839) that treatment increased liver weight at D92 and D274, but consider the effect unrelated to the liver histopathology.
- Interim-group (D92) and terminal (D274) organ weights were collected for all groups for the: adrenals, brain, heart, liver, kidneys, spleen, testes, and ovaries.

Table 28: Results weakly suggest a treatment-related increase in liver weights in the 9-month rabbit study (report # TDOC-0013267)

Treatment→ /endpoint ↓		Males				Females			
		Untreat ed	Vehicle	Brin 1 /Brim 0.2	Brin 2/ Brim 0.2	Untreat ed	Vehicle	Brin 1 /Brim 0.2	Brin 2/ Brim 0.2
D92 liver wt	Absolute (g)	73.3	96.8 *	94.4 *	81.3	78.3	74.0	73.7	81.6
	Liver: body wt	2.15	2.79 *	2.75 *	2.49	2.16	2.04	2.08	2.29
D274 liver wt	Absolute (g)	86.3	86.7	86.0	104.2 *	82.4	71.0	74.8	76.7
	Liver: body wt	2.25	2.37	2.42	2.95 *	2.07	1.81	2.00	2.10

Values presented as means, with the fold-increase from untreated in parenthesis.

Values rounded by this reviewer for readability.

N = 4/sex/group for D91. N = 5/sex/group for D 274.

* Statistically significant, one way analysis of variance, $p < 0.05$

Histopathology

Adequate Battery - yes

- For the interim group animals (D92 necropsy), eyes, ocular adnexa (defined as eyelids, nictitating membrane, harderian gland, and lacrimal gland) and livers were collected for histopathology.
- For the terminal group animals (D274 necropsy):
 - Full histopathology (tissues listed above under gross pathology) was performed for the untreated control group and the high-dose group (Brin 2%/ Brim 0.2%).
 - For the vehicle control and the low-dose group (Brin 1%/ Brim 0.2%), only the eyes, ocular adnexa, spleen pancreas and livers were collected for histopathology.

Peer Review: Partial. Peer review was conducted for the 9-month histopathology samples (i.e. untreated controls and high-dose group), all liver samples (i.e. 3 and 9 months), and all spleen and pancreas samples at 9 months

Histological Findings

- Treatment with Brin/Brim increased the severity of liver hepatocellular cytoplasmic vacuolization and glycogen accumulation (see Table 29 below).
 - The authors attributed this effect to the known hyperglycemic activity of brimonidine in animal models, and did not consider it adverse.
 - The pathology report further detailed the observations of ‘hepatocytes, vacuolization, cytoplasmic’ (report page 987) as “clear lace-like cytoplasmic vacuolization of hepatocytes (centrilobular to panlobular) which ranged from minimal to severe; the more extensive cases of cytoplasmic vacuolization (moderate to severe) resulted in variable hepatocellular swelling”.
 - To characterize the hepatocyte vacuolization, liver samples taken at 9 months were stained with Periodic acid-Schiff stain (PAS) with and without diastase (alpha-amylase) to visualize the presence of glycogen. The pathologists conclude that the vacuolization is “consistent with hepatocellular cytoplasmic glycogen accumulation”, and notes a treatment-related increase in severity.
 - The study pathologist (page 987) noted the increase in absolute and liver weight for the high-dose males, and considered the effect consistent with the observed histopathology
- The study pathology report (report page 987) considered the pancreas islet cell hyperplasia and the lymphoid depletion in the spleen in the high-dose males to be treatment-related (report pages 988-989).
- Note: No treatment-related histopathology was apparent for the eye or adnexa.
 - The following tissues were evaluated for all rabbits: anterior chamber, choroid, ciliary body, cornea, eyelid, harderian gland, iris, lacrimal gland, lens, nictitating membrane, optic nerve, retina, clear, and vitreous.
 - Additionally, the nasal turbinate and nasolacrimal duct were evaluated for untreated control and the high-dose group only (at 3 and 9 months)

Table 29: Selected histopathology results for the 9-month rabbit study (report # TDOC-0013267)

Treatment→ / Lesion↓	Seve rity ^a	Males				Females			
		Untreat ed	Vehicle	Brin 1 /Brim 0.2	Brin 2/ Brim 0.2	Untreat ed	Vehicle	Brin 1 /Brim 0.2	Brin 2/ Brim 0.2
3 month data									
Liver (N=4/sex/dose)									
Hepatocytes, glycogen, cytoplasmic	Total	4	4	4	4	4	4	4	4
	1	0	0	0	1	4	3	1	1
	2	4	3	0	0	0	1	2	1
	3	0	1	3	3	0	0	1	2
	4	0	0	1	0	0	0	0	0
Hepatocytes, vacuolization, cytoplasmic	Total	3	4	4	3	0	1	1	3
	1	3	1	0	0	0	1	0	0
	2	0	2	0	0	0	0	0	0
	3	0	1	3	3	0	0	1	3
	4	0	0	1	0	0	0	0	0
Inflammation, chronic	1	2	1	0	0	0	0	0	0
9 month data									
Liver (N=5/sex/dose)									
Hepatocytes, glycogen, cytoplasmic (PAS)	Total	5	5	5	5	5	5	5	5
	1	1	2	0	0	1	2	0	0
	2	2	1	0	1	3	2	2	3
	3	3	2	4	0	1	1	2	2
	4	0	0	1	2	0	0	1	0
	5	0	0	0	2	0	0	0	0
Hepatocytes, vacuolization, cytoplasmic	Total	5	5	5	5	4	1	4	3
	1	3	3	1	0	4	1	3	2
	2	0	2	2	1	0	0	1	1
	3	2	0	2	3	0	0	0	0
	4	0	0	0	0	0	0	0	0
	5	0	0	0	1	0	0	0	0
Inflammation, chronic	1	0	2	0	0	2	0	0	0
Pancreas (N=5/sex/dose)									
Islet cells, hyperplasia, diffuse	Total	0	0	2	5	0	0	0	0
	1	0	0	1	5	0	0	0	0
	2	0	0	1	0	0	0	0	0
Spleen (N=5/sex/dose)									
Lymphoid depletion	1	0	0	0	3	0	0	0	0

^aSeverity scores: 1 = minimal, 2 = slight/mild, 3 = moderate, 4 = moderately severe, 5 = severe/high.

- Because brinzolamide has been associated with renal toxicity, this reviewer carefully evaluated the kidney endpoints (organ weight, gross pathology,

histopathology of the kidney, urethra and urinary bladder), and verified that no renal treatment-related effects are apparent.

Toxicokinetics

- Blood samples on D1, 86 and 268 for the last 4 rabbits/sex/dose (i.e. the same 4 at each time point)
 - Blood collected at 0 and 1 hour for the control groups
 - Blood collected at 0, 0.5, 1 and 3 hours post-dose for the Brin/Brim groups.
- Accumulation of brinzolamide was apparent: statistically significant ($p < 0.05$) increases in C_{max} and AUC_{0-3h} were apparent on D86 and D268 compared to D1
- No apparent brimonidine accumulation was apparent
- No differences were apparent for either drug's PK between D86 and D268; the authors conclude that steady state had been achieved.

Table 30: TK summary for the 9-month rabbit study (report # TDOC-0013267)

Treatment	Study Day	Brinzolamide		N-desethyl brinzolamide		Brimonidine	
		C_{max} (ng/ml)	AUC_{0-3h} (ng*h/ml)	C_{max} (ng/ml)	AUC_{0-3h} (ng*h/ml)	C_{max} (ng/ml)	AUC_{0-3h} (ng*h/ml)
Brin 1%/Brim 0.2%	1	5200 ± 300	14300 ± 1000	BLQ	BLQ	3.4 ± 1.0	4.1 ± 1.3
	86	6200 ± 300	17700 ± 700	BLQ	BLQ	2.2 ± 1.3	2.8 ± 1.4
	268	6500 ± 400	18500 ± 1200	131 ± 18	310 ± 130	2.2 ± 0.8	3.0 ± 1.3
Brin 2%/Brim 0.2%	1	5900 ± 600	17000 ± 1800	BLQ	BLQ	3.1 ± 1.1	4.0 ± 1.4
	86	6200 ± 600	17700 ± 1600	BLQ	BLQ	1.8 ± 0.6	2.0 ± 0.6
	268	6400 ± 700	18300 ± 1600	179 ± 27	503 ± 72	1.6 ± 0.6	2.0 ± 0.7

BLQ = below the lower limit of quantitation (LLOQ)

Values rounded by this reviewer for readability. Brinzolamide and N-desethyl brinzolamide were measured in whole blood (LLOQ = 100 ng/ml). Brimonidine was measured in plasma (LLOQ = 10 pg/ml)

Stability and Homogeneity

- The suspensions were “shaken well” prior to dosing, and the proposed label states (section 16 Storage and Handling) “Shake well before use”. The report lacks details regarding the test article analysis (i.e. does not state whether the suspension was shaken, or how homogenous the suspension is with and without shaking). This is a minor study limitation.
- Test articles were analyzed pre-study and post-study for the presence/absence of brinzolamide, brimonidine, and benzalkonium chloride, and for the concentrations of brinzolamide and brimonidine. The test articles were within 98 to 104% of nominal (report page 84)

7 Genetic Toxicology

The applicant did not submit genetic toxicology studies. No genetic toxicology testing for the brinzolamide / brimonidine combination was identified by the applicant or this reviewer. NDA 204251 was submitted under 505(b)(2), and Alcon referenced Azopt, Alphagan, and Brimonidine Tartrate. The genetic toxicology data incorporated by reference for brinzolamide (as a single agent) and brimonidine (as a single agent) are deemed adequate. The results were summarized in NDA module 2.6.6 Toxicology Written Summary.

8 Carcinogenicity

The applicant did not submit carcinogenicity studies in this NDA.

Brinzolamide

- The Applicant provided a statement (NDA Module 2.6.6. Toxicology Written Summary, “Brinzolamide was characterized as noncarcinogenic based on 2-year oral dosing studies in rats and mice. CD-1 mice and Fischer rats were dosed daily by oral gavage up to 10 mg/kg/day and 8 mg/kg/day, respectively. There was no indication of a carcinogenic effect in any organ after at least 104 weeks of administration.”
- The Applicant proposed language for the label (b) (4)

This reviewer has found **no prior mention** of these carcinogenicity studies for brinzolamide. Notably:

- The label approved March 8, 2001 for Alcon's NDA 20816 for brinzolamide (Azopt®) states “Carcinogenicity data on brinzolamide are not available “
- IND 40152 was submitted by Alcon on July 22, 1992 for ALO4862 ophthalmic suspension for glaucoma therapy. The file is paper.
 - No P/T reviews in DARRTS
 - This reviewer consulted the annual reports (in the CDER Document Room) for October 2012 (no new P/T information), October 2011 (Dr. William Taylor NAI'd this AR on 11/04/2011 without comment) and October 2009;
 - The 2010 AR was not on the shelf (NAI'd by Dr. Wendelyn Schmidt, 5/11/2011 without comment)
- Alcon has three other INDs for brinzolamide (IND 78215, IND (b) (4) and IND 106293); this reviewer found no P/T reviews relevant to these carcinogenicity studies.
- An electronic search of the internal CDER Carcinogenicity Assessment Committee (CAC) database (via [\\Cdsnas\ptcc\CAC](#)) found no relevant files (e.g. protocol agreement or reviews)

- For NDA 20816, the Applicant submitted SD #251 on February 1, 2013, which includes a proposed package insert (internally available via [\\cdsesub4\NONECTD\NDA020816\5227234\DISC1](#)) that states, “Carcinogenicity data on brinzolamide are not available.”

This reviewer tentatively concludes that the Applicant’s claim of completed carcinogenicity studies for brinzolamide is a typographic error of some kind.

Brimonidine

The carcinogenicity toxicology data incorporated by reference for brimonidine (as a single agent) are deemed adequate. The results are also summarized in NDA module 2.6.6.

Combination

No carcinogenicity testing for the brinzolamide / brimonidine combination was identified by the applicant or this reviewer.

9 Reproductive and Developmental Toxicology

The applicant did not submit studies for developmental and reproductive toxicity (DART). No DART testing for the brinzolamide / brimonidine combination was identified by the applicant or this reviewer. The DART data incorporated by reference for brinzolamide (as a single agent) and brimonidine (as a single agent) are deemed adequate. The results were summarized in NDA module 2.6.6.

10 Special Toxicology Studies

The applicant did not submit special toxicology studies.

11 Integrated Summary and Safety Evaluation

- Both brinzolamide and brimonidine induce lowering of IOP in animal models and patients by targeting the ciliary epithelium to reduce aqueous humor production. The mechanisms of action are different:
 - Brinzolamide is a carbonic anhydrase inhibitor (CAI). CAIs as a class inhibit carbonic anhydrase in the ciliary epithelium, reducing production of bicarbonate ions, which diminishes sodium and fluid transport across the ciliary epithelium, which results in decreased aqueous humor production.
 - Brimonidine is a selective alpha-2 adrenergic agonist. Brimonidine activates alpha-2 adrenergic receptors expressed by the ciliary epithelium, which activates GTP-binding protein, which inhibits the activity of intracellular adenylyl cyclase, which decreases aqueous humor production.

- Repeat dose brimonidine reduces IOP by another, complementary mechanism. Prolonged alpha adrenergic stimulation increases local prostaglandin release, which relaxes the ciliary muscle and allows increased uveoscleral outflow.
- In addition to the P/T reviews identified (see section 3.3 of this review) authors affiliated with Allergan have published reviews of the nonclinical safety of brimonidine, mentioning the one-year systemic and ocular toxicity studies in monkeys, and the six-month ocular/systemic studies in rabbits.^{18,19}
- The clinical formulation, brin 1% / brim 0.2% is a suspension of 10 mg/ml brin and 2 mg/ml brim. The rabbit studies provide a margin of safety of exposure above the clinical dose.

Table 31: Dose comparison: clinical label versus the 6-week & 9-month rabbit studies

Species & study	Dose level	Dose	Dosing notes	Total µg/day of brinzolamide	Total µg/day of brimonidine
Human	Label dose	Brin 1% / brim 0.2%	one drop TID in one or both eyes	2100 ^a	420 ^a
Rabbit – 6 week study	Low-dose	Brin 1% / Brim 0.15%	One drop TID in both eyes	1800 (0.9x)	270 (0.6x)
	High-dose	Brin 2% / Brim 0.2%		3600 (1.7x)	360 (0.9x)
Rabbit – 9 month study	Low-dose	Brin 1% / Brim 0.2%		2180 (1x)	440 (1x)
	High-dose	Brin 2% / Brim 0.2%		3900 (1.9x)	390 (0.9x)

^a presuming both eyes dosed daily with a drop size of 35 µl

The daily doses presented for the rabbit studies are those calculated by the study authors. The margin of exposure is presented in parentheses.

- Comparing pharmacokinetic (PK) parameters for the rabbit studies versus the clinical trials (e.g. NDA module 2.7.2 Summary of Clinical Pharmacology Studies), the Applicants concluded that systemic exposure to brimonidine is substantially higher in rabbits compared to patients. Comparing the rabbit PK data (e.g. Table 12, Table 13, Table 15, Table 30 of this review) to the clinical PK data (copied below), this reviewer concurs. Therefore, the results of the rabbit

¹⁸ Angelov OV, Wiese AG, Tang-Liu DD, Acheampong AA, Ismail IM, Brar BS. 1996. Preclinical safety profile of brimonidine. *Eur J Ophthalmol*. 6(1):21-25.

¹⁹ Burke J, Schwartz M. 1996. Preclinical evaluation of brimonidine. *Surv Ophthalmol*. 41 Suppl 1:S9-18.

studies may overpredict the systemic effects of brimonidine compared to clinical use.

Table 32: Clinical brimonidine summary PK data (TID)

From NDA module 2.7.2 page 9:

Table 2.7.2.2-5
Least Squares Mean Ratio of Brimonidine (AL-8923) Plasma for Brinzolamide
1%/Brimonidine Tartrate 0.2% TID and Brimonidine TID on Day 21
(Pharmacokinetic Data)

Parameter	Least Squares Means		Least Squares Mean Ratio	Lower 90% CI	Upper 90% CI
	Brinz/Brim TID	Brimonidine TID			
C_{max} (ng/mL)	0.0479	0.0508	0.943	0.724	1.23
AUC_{0-t} (ng*hr/mL)	0.183	0.205	0.893	0.668	1.19

Brinz/Brim TID = Brinzolamide 1%/Brimonidine Tartrate 0.2% Ophthalmic Suspension, 3 times a day dosing
 Brimonidine TID = Brimonidine Tartrate Ophthalmic Solution, 0.2%, three times a day dosing
 ANOVA performed on ln-transformed data to calculate least squares means

Table 33: Clinical brimonidine summary PK data (BID)

From NDA module 2.7.2 page 9:

Table 2.7.2.2-6
Least Squares Mean Ratio of Brimonidine (AL-8923) Plasma for Brinzolamide
1%/Brimonidine Tartrate 0.2% BID and Brimonidine BID on Day 21
(Pharmacokinetic Data)

Parameter	Least Squares Means		Least Squares Mean Ratio	Lower 90% CI	Upper 90% CI
	Brinz/Brim BID	Brimonidine BID			
C_{max} (ng/mL)	0.0643	0.0602	1.07	0.863	1.32
AUC_{0-t} (ng*hr/mL)	0.174	0.227	0.765	0.614	0.954

Brinz/Brim BID = Brinzolamide 1%/Brimonidine Tartrate 0.2% Ophthalmic Suspension, 2 times a day dosing
 Brimonidine BID = Brimonidine Tartrate Ophthalmic Solution, 0.2%, two times a day dosing
 ANOVA performed on ln-transformed data to calculate least squares means

Table 34: Clinical brimonidine summary PK data (TID)

From NDA module 2.7.2 page 10:

Table 2.7.2.2-7
Least Squares Mean Ratio of Brimonidine (AL-8923) Plasma of Visit Days 107 and 21
for Brinzolamide 1%/Brimonidine Tartrate 0.2% TID
(Pharmacokinetic Data)

Parameter	Least Squares Means		Least Squares Mean Ratio	Lower 90% CI	Upper 90% CI
	107	21			
C_{max} (ng/mL)	0.0419	0.0479	0.875	0.640	1.20
AUC_{0-t} (ng*hr/mL)	0.145	0.183	0.791	0.562	1.11

Brinz/Brim TID = Brinzolamide 1%/Brimonidine Tartrate 0.2% Ophthalmic Suspension, three times a day dosing

107 = Visit Day 107

21 = Visit Day 21

ANOVA performed on ln-transformed data to calculate least squares means

Table 35: Clinical brimonidine summary PK data (BID)

From NDA module 2.7.2 page 10:

Table 2.7.2.2-8
Least Squares Mean Ratio of Brimonidine (AL-8923) Plasma of Visit Days 107 and 21
for Brinzolamide 1%/Brimonidine Tartrate 0.2% BID
(Pharmacokinetic Data)

Parameter	Least Squares Means		Least Squares Mean Ratio	Lower 90% CI	Upper 90% CI
	107	21			
C_{max} (ng/mL)	0.0592	0.0643	0.921	0.694	1.22
AUC_{0-t} (ng*hr/mL)	0.172	0.174	0.991	0.749	1.31

Brinz/Brim BID = Brinzolamide 1%/Brimonidine Tartrate 0.2% Ophthalmic Suspension, two times a day dosing

107 = Visit Day 107

21 = Visit Day 21

ANOVA performed on ln-transformed data to calculate least squares means

Table 36: Clinical brimonidine summary PK data (TID or BID)

From NDA module 2.7.2 page 14:

Table 2.7.2.3-4

Comparison of Brimonidine Mean (Minimum to Maximum) PK Parameters on Day 21 after Administration of Brinzolamide 1%/Brimonidine Tartrate 0.2% (TID or BID) or Brimonidine 0.2% (TID or BID) in Healthy Subjects

Day	PK Parameters	Brinzolamide 1%/ Brimonidine Tartrate 0.2% Ophthalmic Suspension BID	Brimonidine 0.2% BID	Brinzolamide 1%/ Brimonidine Tartrate 0.2% Ophthalmic Suspension TID	Brimonidine 0.2% TID
21	N	24	24	23	24
	C _{max} (ng/mL)	0.0724 (0.0234-0.179)	0.0639 (0.0279-0.114)	0.0545 (0.0186-0.122)	0.0574 (0.0085-0.137)
	T _{max} (hr) ^a	0.50 (0.25-1.00)	0.75 (0.25-2.00)	0.67 (0.25-1.50)	1.00 (0.25-1.52)
	AUC _{0-last} (ng*hr/mL)	0.196 (0.0580-0.408)	0.243 (0.0985-0.457)	0.215 (0.0530-0.538)	0.233 (0.0249-0.496)
	t _{1/2} (hr)	2.57 (1.37-4.69)	2.38 (1.75-3.99)	2.43 (1.69-3.35)	2.48 (1.48- 6.86)

^aT_{max} is expressed as median with range (minimum to maximum)

This is a representation of an electronic record that was signed electronically and this page is the manifestation of the electronic signature.

/s/

ANDREW J MCDOUGAL
03/15/2013

LORI E KOTCH
03/15/2013

PHARMACOLOGY/TOXICOLOGY FILING CHECKLIST FOR NDA

NDA Number: 204251

Applicant: Alcon Research, Ltd.

Stamp Date: 6-19-2012

Drug Name: Brinzolamide

NDA Type: 505(b)(2)

1%/Brimonidine Tartrate 0.2%
suspension

On **initial** overview of the NDA application for filing:

	Content Parameter	Yes	No	Comment
1	Is the pharmacology/toxicology section organized in accord with current regulations and guidelines for format and content in a manner to allow substantive review to begin?	x		
2	Is the pharmacology/toxicology section indexed and paginated in a manner allowing substantive review to begin?	x		This NDA was submitted electronically, and the indexing appears adequate. This reviewer verified the location of each nonclinical report, and that each report opened in the viewing software.
3	Is the pharmacology/toxicology section legible so that substantive review can begin?	x		
4	Are all required (*) and requested IND studies (in accord with 505 b1 and b2 including referenced literature) completed and submitted (carcinogenicity, mutagenicity, teratogenicity, effects on fertility, juvenile studies, acute and repeat dose adult animal studies, animal ADME studies, safety pharmacology, etc)?		x	See comment 8 below
5	If the formulation to be marketed is different from the formulation used in the toxicology studies, have studies by the appropriate route been conducted with appropriate formulations? (For other than the oral route, some studies may be by routes different from the clinical route intentionally and by desire of the FDA).	x		Two combination studies (chronic rabbit ocular toxicity) were conducted with the clinical formulation.
6	Does the route of administration used in the animal studies appear to be the same as the intended human exposure route? If not, has the applicant <u>submitted</u> a rationale to justify the alternative route?	x		
7	Has the applicant <u>submitted</u> a statement(s) that all of the pivotal pharm/tox studies have been performed in accordance with the GLP regulations (21 CFR 58) <u>or</u> an explanation for any significant deviations?	x		

**PHARMACOLOGY/TOXICOLOGY FILING CHECKLIST FOR
NDA**

	Content Parameter	Yes	No	Comment
8	Has the applicant submitted all special studies/data requested by the Division during pre-submission discussions?		x	The sponsor elected not to conduct a nonclinical embryofetal study using the to-be-marketed combination, as recommended by the nonclinical reviewer for the predecessor IND 106293. The applicant's justification claims a lack of concern regarding teratogenicity of individual components, and claims a sufficient exposure margin exists for individual components for the proposed route of administration (ocular administration). The adequacy of the applicant's scientific justification will be assessed during the NDA review cycle.
9	Are the proposed labeling sections relative to pharmacology/toxicology appropriate (including human dose multiples expressed in either mg/m ² or comparative serum/plasma levels) and in accordance with 21 CFR 201.57?		x	Section 13 of the proposed label (b) (4) Potential changes to the proposed label will be discussed with review team, and presented to sponsor during the review cycle.
10	Have any impurity – etc. issues been addressed? (New toxicity studies may not be needed.)			PENDING - No issues have been previously identified. The applicant reports (module 2.6.6) that the brinzolamide impurities are "biologically qualified." Upon preliminary review, the specifications and reported lot results appear to meet ICH Q3 guidelines. In conjunction with ONDQA, the final impurity profile and adequacy of qualification of APIs/excipients will be assessed during the NDA review cycle.
11	Has the applicant addressed any abuse potential issues in the submission?			Not applicable – Abuse potential is not a regulatory concern for this drug.
12	If this NDA/BLA is to support a Rx to OTC switch, have all relevant studies been submitted?			Not applicable

IS THE PHARMACOLOGY/TOXICOLOGY SECTION OF THE APPLICATION FILEABLE? yes

No potential review issues for the 74-day letter have been identified at this time.

This is a representation of an electronic record that was signed electronically and this page is the manifestation of the electronic signature.

/s/

ANDREW J MCDOUGAL
08/17/2012

LORI E KOTCH
08/17/2012