

**CENTER FOR DRUG EVALUATION AND
RESEARCH**

APPLICATION NUMBER:

21-730

PHARMACOLOGY REVIEW(S)



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

PHARMACOLOGY/TOXICOLOGY REVIEW AND EVALUATION

NDA NUMBER: 21-730
SERIAL NUMBER: 000
DATE RECEIVED BY CENTER: May/11/04
PRODUCT: Xopenex HFA (levalbuterol tartrate HFA)
Inhalation Aerosol
INTENDED CLINICAL POPULATION: Adults and children 4 years and older with reversible
obstructive airway disease
SPONSOR: Sepracor
DOCUMENTS REVIEWED: Submission 000/May 11, 2004
Submission 000/February 4, 2005
REVIEW DIVISION: Division of Pulmonary and Allergy Drug Products
(HFD-570)
PHARM/TOX REVIEWER: VWhitehurst, Ph.D.
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Date of review submission to Division File System (DFS): February 23, 2005

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EXECUTIVE SUMMARY

I. Recommendations

A. Recommendation on approvability

The NDA application is approvable from a nonclinical perspective

B. Recommendation for nonclinical studies

No further nonclinical studies are needed

C. Recommendations on labeling

The labeling is approvable pending recommended revisions. The recommended changes include adjustment of animal to human exposure ratios and the modification of proposed text referring to _____

II. Summary of nonclinical findings

A. Brief overview of nonclinical findings

Preclinical studies in rats and dogs reveal that the target organ for levalbuterol is the cardiovascular system and spleen. Cardiovascular toxicities include decreases in blood pressure with a concurrent tachycardia, changes in EKG, prolongation of QTc, flattening of the T-wave and ST segment depression, increases in mean heart weight and myocardial necrosis. Spleen toxicity consists of multi-focal capsulitis and capsular thickening.

B. Pharmacologic activity

In vitro and *in vivo* studies show levalbuterol (composed of the (R)-enantiomer) stimulates beta₂-adrenergic receptors on airway smooth muscle leading to activation of adenylyl cyclase and to an increase in cyclic AMP. Increases in cyclic AMP induce the relaxation of airway smooth muscle. Also, increases in cyclic AMP are associated with inhibition of release of mediators from the mast cells in the airway. Levalbuterol induced dose-related decreases in blood pressure and concurrent increases in heart rate in rats and dogs. Additionally, the drug caused decreases in plasma potassium as well as increases in plasma glucose concentrations in muscle tissues. Levalbuterol decreased TNF α -induced hyperreactivity, while (S)-albuterol was not as potent, indicating that the ability to offset airway hyperreactivity resides primarily with the (R)-enantiomer.

C. Nonclinical safety issues relevant to clinical use:

The primary nonclinical safety issues associated with the use of levalbuterol are related to cardiovascular effects, including decreases in blood pressure, increases in heart rate, EKG changes, arrhythmogenic potential (due mainly to loss of

serum potassium) and myocardial necrosis. Also, albuterol is teratogenic in animal studies and the drug will be listed as a Category C for Pregnancy.

**APPEARS THIS WAY
ON ORIGINAL**

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ON ORIGINAL**

2.6 PHARMACOLOGY/TOXICOLOGY REVIEW

2.6.1 INTRODUCTION AND DRUG HISTORY

NDA number: NDA 21-730

Review number: 001

Sequence number/date/type of submission: 000/May 11, 2004/original submission

000/February 4, 2005/BZ

Information to sponsor: Yes (X) No ()

Sponsor and/or agent: Sepracor Inc., Marlborough, MA

Manufacturer for drug substance: 3M Pharmaceuticals, Northridge, CA 91324-3298

Reviewer name: Virgil Whitehurst Ph.D.

Division name: Division of Pulmonary and Allergy Drug Products

HFD #: HFD 570

Review completion date: February 23, 2005

Drug:

Trade name: Xopenex HFA (levalbuterol tartrate HFA) Inhalation Aerosol

Generic name: Levalbuterol

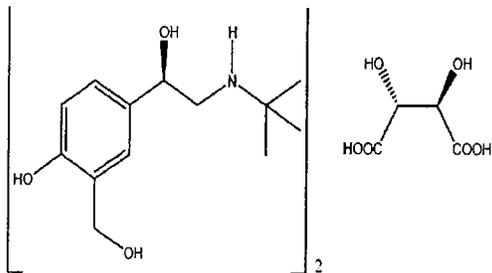
Code name: None

Chemical name: (R)- α -1-[(1,1-dimethylethyl)methyl]-4-hydroxy-1,3-benzenedimethanol hemi-tartrate salt

CAS registry number: 661464-94-4

Molecular formula/molecular weight: $(C_{13}H_{21}NO_3)_2 \cdot C_4H_6O_6 / 628.71$

Structure:



Relevant INDs/NDAs/DMFs: IND 47, 363, Levalbuterol HCL Inhalation Solution; NDA 20-837, Xopenex (levalbuterol HCL) Inhalation Solution, NDA 20-503, Proventil Inhalation Aerosol and NDA 19-243, Proventil Inhalation Solution, DMF _____; DMF _____; DMF _____; DMF _____

Drug class: Beta 2 adrenergic agonist

Intended clinical population: Treatment or prevention of bronchospasm in patients (adults and children, 4 years and older) with reversible obstructive airway disease and attacks of bronchospasm. Maximum daily recommended dose is 2 inhalations (90 mcg) up to 6 times a day or a maximum of 540 mcg/day.

Clinical formulation:

Components	Function	Amount/actuation	Amount/Canister
Levalbuterol tartrate	Active ingredient		
Oleic acid			
Dehydrated alcohol			
HFA	Propellant		
Total	---	60.mg	15.00 g

* Equivalent to .mcg of levalbuterol free base (ex-valve) to deliver 45 mcg levalbuterol free base/59 mcg of levalbuterol tartrate (ex-actuator).

Route of administration: Oral inhalation

Disclaimer: Tabular and graphical information are constructed by the reviewer unless cited otherwise.

Data reliance : Except as specifically identified below, all data and information discussed below and necessary for approval of NDA 21-730 are owned by Sepracor or are data for which Sepracor has obtained a written right of reference. Any information or data necessary for approval of NDA 21-730 that Sepracor does not own or have a written right to reference constitutes one of the following: (1) published literature, or (2) a prior FDA finding of safety or effectiveness for a listed drug, as described in the drug's approved labeling. Any data or information described or referenced below from a previously approved application that Sepracor does not own (or from FDA reviews or summaries of a previously approved application) is for descriptive purposes only and is not relied upon for approval of NDA 21-730.

Studies reviewed within this submission:**Pharmacology Studies:****Pharmacodynamics and Primary Pharmacology**

- Report 090-480: Effect of RS-, R-, and S-Albuterol and RR, SS-, RR-, SS-Formoterol on Tumor Necrosis Factor-alpha (TNF-alpha) Induced Hyperreactivity in Tracheal Smooth Muscle of the Guinea-Pig In Vitro
- Report 051-560: Effects of (R)-, (S)-, and (RS)-Albuterol on Basal Airway Reactivity and Allergen-Induced Early and Late Asthmatic Reactions, Airway Hyperreactivity and Inflammation
- Report 051-496: Effects of R-, S-, and RS-Albuterol on Allergic and Non-Allergic Airway Responses In Vivo, Ex Vivo, and In Vitro
- Report 051-545: Effect of a Single Isomer and Racemic β_2 Receptor Agonist on Neurogenic Inflammation in a Rat Model of Respiratory Syncytial Virus Bronchiolitis
- Report 051-498: Evaluation of the Effects of Albuterol Enantiomers on Mitogen-stimulated Proliferation and Cytokine Production in Airway Smooth Muscle
- Report 051-551: Effect of Formoterol and Albuterol Stereoisomers on the Expression and Activity of G-Proteins, Intracellular Free Ca^{2+} and Nuclear Transcription Factor NFkB in Human Airway Smooth Muscle Cells
- Report 051-567: The Effects of Enantiomers of Albuterol on Indices of Remodeling in Human Lung Fibroblasts
- Report 051-568: The Effects of Enantiomers of Albuterol Non-Contractive Function of Human Airway Smooth Muscle Cells
- Report 051-561: Comparison of Albuterol, Salmeterol, and Formoterol Activation of Human β_2 -Adrenergic Receptors
- Report 051-577: Effects of Racemic Albuterol and Its Enantiomers on Cholinergic Twitch Contractions of Guinea Pig and Bovine Trachea
- Report 051-555: (S)-Albuterol, but Not Other β_2 -Agonists, Has Stimulatory Effects on Mucin Secretion and Changes in Gene Expression on Human Airway Epithelium

Secondary Pharmacology

- Report 051-534: Effect of S-Albuterol on R-Albuterol Enhanced Mucociliary Transport in Calf Trachea
- Report 051-563: The Effects of Enantiomers of Albuterol on Monocyte and Macrophage Function
- Report 051-535: Study of Four Compounds on Various Cytokine Secretions Using Basal Models
- Report 051-538: Effect of (RS)-, (R)- and (S)-Albuterol on Sperm Motility
- Report 051-546: Attenuation of Inflammation-Induced Morphologic Changes and Apoptosis in Human Neutrophils by Albuterol Is Isomeric Specific
- Report 051-518: Differential Effects on Albuterol Enantiomers on the Asthma-Like Syndrome in Rats
- Report 051-531: Study of Four Compounds in Various Inflammation Assays
- Report 051-574: Differential Effects of Racemic Albuterol, S-Albuterol, and R-Albuterol on Early and Late Phase Reactions, Airway Hyperresponsiveness and BAL Eosinophilia in the Guinea Pig Model of Asthma

Report 051-516: Effect of Methacholine, (RS)-, (R)- and (S)-Albuterol, and (RR, SS)-, (RR)- and (SS)-Formoterol on Mucociliary Transport in Calf Trachea
 Report 051-486: Effects of Four Compounds on Basal IL-5 Secretion by Human PBMC

Pharmacokinetic/ADME Studies:

Report 051-515: Toxicokinetics of R- and S-Albuterol during a Subchronic (90 days) Repeat Dose Inhalation Toxicity Study in Rats
 Report 051-578: Toxicokinetics of Levalbuterol During a 13 Week Inhalation Study in Dogs
 Report 051-504: ³H-(R)-Albuterol, ³H-(S)-Albuterol and ³H-(RS)-Albuterol: Brain Transfer of Radioactivity in Rats
 Report 051-554: Report for Chiral Analysis of Non-Radioactive R- and S- Albuterol in Rat Brain Tissue
 Report 051-553: Report for Chiral Analysis of Radioactive R-, S-, and RS-Albuterol in Rat Brain Tissue and Rat Plasma
 Report 051-503: [3H]-(R)-Albuterol: In Vitro Blood to Plasma Partitioning and Protein Binding in the Dog and Human
 Report 052-402: [3H]-(S)-Albuterol: In Vitro Blood to Plasma Protein Binding in the Dog and Human
 Report 051-526: [3H]-(R)-Albuterol: In Vitro Blood-to-Plasma Partitioning and the Plasma Protein Binding of [3H]-(R)-Albuterol in Human and the Effect of (S)-Albuterol, (R,R/S, S)-Formoterol and (R,R)-Formoterol on the Protein Binding of (R)-Albuterol

Toxicology Studies:

Report 051-824: Xopenex HFA Pressurized Metered Dose Inhaler: Dose Range-Finding Study in the Dog
 Report 051-825A2: Xopenex HFA Pressurized metered Dose Inhaler: 13 Week Inhalation Qualification study in the Dog. Amendment 2, Amendment 1, and Final Report

Mutagenicity Studies:

Report 051-822: Chromosomal Aberration in Chinese hamster Ovary (CHO) Cells With R-Albuterol, S-Albuterol and RS-Albuterol
 Report 051-823: In Vivo Mouse Micronucleus Assay

Special Studies:

Report 051-820: Acute Dermal Irritation Study of Levalbuterol and Albino Rabbits — in
 Report 051-821: Acute Eye Irritation Study of Levalbuterol and Albino Rabbits — in

Studies not reviewed within this submission:

Report 051-537: Validation Report: HPLC Analysis of Albuterol Enantiomers in Mouse Plasma

Report 051-511: Method Validation Report: Determination of (R)-Albuterol and (S)-Albuterol in Rat Plasma by High Performance Liquid Chromatography
Report 051-474A3: Method Validation Report: Determination of Chiral Albuterol in Rabbit Plasma. With Addenda I-III
Report 051-575: Method Validation Report: Determination of Albuterol Enantiomers in Dog Plasma by LC/MS/MS
Report 051-510: Determination of Chiral Albuterol in Rat Plasma by High Performance Liquid Chromatography
Report 051-576: LC/MS/MS Analysis of (R)-Albuterol and (S)-Albuterol in Dog Plasma (in Support of — Protocol 1487/002) [Located in Report 051-825A2, Annex to Final Report]

These studies were not reviewed as they were validation assays for various kinetic techniques.

Levalbuterol is the (R)-isomer of racemic albuterol. This moiety was approved as Xopenex Inhalation Solution (NDA 20-837) was approved in the US in March 1999. All preclinical information conducted for the approved NDA is incorporated by reference into this application. Additionally, carcinogenicity and reproduction toxicology data for racemic albuterol by reference to NDA 19-243 (Proventil) are relied upon. Initial development of Xopenex HFA was conducted under IND 62,906.

2.6.2 PHARMACOLOGY

2.6.2.1 Brief summary

In vitro and *in vivo* studies reviewed under this NDA and NDA 20-837 show levalbuterol (composed of the (R)-enantiomer) stimulates beta₂-adrenergic receptors on airway smooth muscle leading to activation of adenylyl cyclase and to an increase in cyclic AMP. Increases in cyclic AMP result in the relaxation of airway smooth muscle. Also, increases in cyclic AMP are associated with inhibition of release of mediators from the mast cells in the airway.

Studies conducted previously under NDA 20-837 demonstrate that the binding potential of R- and racemic albuterol for beta-adrenergic receptors were similar in membrane assays using rat heart and guinea pig lung tissue. R-albuterol was 2 times more potent than racemic albuterol in binding to beta₂-adrenergic receptors and in stimulating maximum cAMP elevations. Both R-albuterol and racemic albuterol desensitize beta adrenoceptors with greater potency demonstrated by the racemic form. R-albuterol and racemic albuterol attenuated the increase in pulmonary resistance in guinea pigs induced by spasmogens.

Newly conducted pharmacology studies submitted to this application demonstrated the efficacy of RS-, R-, and S- albuterol. *In vitro* studies using isolated guinea pig, bovine and human tissues reveal that R-albuterol induced reduced airway responses to various

challenges such as Tumor Necrosis Factor- α (TNA- α), histamine, and methacholine, inhibited GM-CSF production and DNA synthesis in human airway smooth muscle cells, and increased COL3A1 mRNA. It also inhibits in vitro mucociliary transport in bovine trachea and enhances the release of the IL 10 from human monocytes but inhibits stimulated production of various cytokines in cell lines. In vivo studies demonstrated that R-albuterol induced decreases in vascular permeability in a rat infection model. Inhaled doses of R-albuterol, administered 3 times daily for 7 days using concentrations of 0.01%, in the OVA sensitized guinea pig attenuated early and late phase reactions, the number of total inflammatory cells and eosinophils. Safety pharmacology studies for levalbuterol were submitted to and reviewed under NDA 20-837. The effects of R-albuterol are similar to those reported for racemic albuterol and include induction of tremors, tachycardia, and increased respiration.

The available data along with clinical experience indicate that levalbuterol will be effective for the proposed indication.

2.6.2.2 Primary pharmacodynamics

Mechanism of action: There is extensive information available related to the primary pharmacodynamics of levalbuterol and albuterol. Levalbuterol acts via the beta 2-adrenergic receptors resulting in activation of adenylate cyclase leading to increased levels of cyclic AMP (cAMP) and reduction of intracellular calcium levels resulting in relaxation of airway smooth muscle cells. Levalbuterol also has the potential to interact with beta₁ and beta₂ adrenergic receptors in the cardiovascular tissues to induce tachycardia, myocardial necrosis and sudden death.

The following pharmacology studies were recently conducted by the sponsor to further demonstrate the mechanism of action:

Study Number	Route	Species-Dose Range	Range
051-545: Effect of a Single Isomer and Racemic β_2 -Receptor Agonists on Neurogenic Inflammation in a Rat Model of Respiratory Syncytial Virus Bronchiolitis	In vivo, IH	Rat- R-albuterol- doses- 0.31-2.5 mg and racemic albuterol and S- albuterol- inhalation doses 2.5 and 5.0 mg	R-albuterol demonstrated anti-inflammatory effects by inducing significant decreases in vascular permeability in respiratory syncytial virus inoculated Fischer 344 rats at doses of 0.31 mg and higher. S-albuterol and racemic albuterol also demonstrated anti inflammatory effects but at higher doses, i.e. 2.5-5.0 mg. The anti-inflammatory effect dose not appear to be due to SP(NK1) expression

<p>051-561: Comparison of Albuterol, Salmeterol and Formoterol Activation of Human β_2 Adrenergic receptors.</p>	<p>In vitro</p>	<p>Human beta2 adrenergic receptors prepared from Spodoptera frugiperda cells infected with a recombinant baculovirus containing the human beta2 AR cDNA.</p>	<p>The ability of racemic and isolated isomers of albuterol, salmeterol and formoterol to bind to human beta 2 adrenergic receptors was assessed. The ligand binding and cAMP production are shown below:</p> <p>Ligand Binding and cAMP Production of β_2-Adrenergic Agonists</p> <table border="1"> <thead> <tr> <th>Drug</th> <th>K_D (nM)</th> <th>EC_{50} (nM)</th> <th>Intrinsic Binding</th> <th>cAMP Production</th> <th>Intrinsic activity</th> </tr> </thead> <tbody> <tr> <td>(±) Isoproterenol</td> <td>123 ± 20</td> <td>60</td> <td></td> <td></td> <td>1.00</td> </tr> <tr> <td>(R)-Albuterol</td> <td>357 ± 59</td> <td>183</td> <td></td> <td></td> <td>0.17</td> </tr> <tr> <td>(RS)-Albuterol</td> <td>421 ± 20</td> <td>124</td> <td></td> <td></td> <td>0.16</td> </tr> <tr> <td>(S)-Albuterol</td> <td>$37,000 \pm 2000$</td> <td>15,470</td> <td></td> <td></td> <td>0.02</td> </tr> <tr> <td>(R)-Salmeterol</td> <td>1.03 ± 0.16</td> <td>3.4</td> <td></td> <td></td> <td>0.13</td> </tr> <tr> <td>(RS)-Salmeterol</td> <td>0.62 ± 0.04</td> <td>0.6</td> <td></td> <td></td> <td>0.11</td> </tr> <tr> <td>(S)-Salmeterol</td> <td>15.5 ± 1.4</td> <td>ND</td> <td></td> <td></td> <td>0.04</td> </tr> <tr> <td>(R,R)-Formoterol</td> <td>2.1 ± 0.3</td> <td>1.1</td> <td></td> <td></td> <td>0.53</td> </tr> <tr> <td>(R,R/S,S)-Formoterol</td> <td>4.3 ± 0.4</td> <td>5.1</td> <td></td> <td></td> <td>0.65</td> </tr> <tr> <td>(S,S)-Formoterol</td> <td>$4,000 \pm 447$</td> <td>3774</td> <td></td> <td></td> <td>0.18</td> </tr> </tbody> </table> <p>These data also suggested that S-albuterol at a concentration of 0-2 mM antagonized R-albuterol (0.25 μM) stimulated c-AMP production by 90%.</p>	Drug	K_D (nM)	EC_{50} (nM)	Intrinsic Binding	cAMP Production	Intrinsic activity	(±) Isoproterenol	123 ± 20	60			1.00	(R)-Albuterol	357 ± 59	183			0.17	(RS)-Albuterol	421 ± 20	124			0.16	(S)-Albuterol	$37,000 \pm 2000$	15,470			0.02	(R)-Salmeterol	1.03 ± 0.16	3.4			0.13	(RS)-Salmeterol	0.62 ± 0.04	0.6			0.11	(S)-Salmeterol	15.5 ± 1.4	ND			0.04	(R,R)-Formoterol	2.1 ± 0.3	1.1			0.53	(R,R/S,S)-Formoterol	4.3 ± 0.4	5.1			0.65	(S,S)-Formoterol	$4,000 \pm 447$	3774			0.18
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Drug activity related to proposed indication:

Levalbuterol induces relaxation of smooth muscles in the pulmonary tissues resulting in the prevention and /or treatment of bronchospasm associated with reversible obstructive airway disease, i.e., asthma and COPD.

The following drug-related pharmacology studies were conducted to demonstrate drug activity related to proposed indication:

Study Number	Route-Dose Range	Species/ Model	Results
090-480 Effects of RS-,R-,and S-albuterol and RR, SS-RR- and SS –Formoterol on Tunor Necrosis Factor – Alpha Induced Hyperreactivity in Tracheal smooth Muscle of Guinea Pig in vitro	In Vitro	Guinea pig trachea	R- and RS-albuterol decreased Tumor Necrosis Factor- α (TNF- α) induced hyperactivity in tracheal smooth muscle. S, S-formoterol and S- albuterol had no effect on TNF- α on hyperreactivity in tracheal smooth muscle.
051-560- Effects of R-,S-, and RS-Albuterol on Basal Airway Reactivity and Allergen-Induced Early and Late Asthmatic Reactions. Airway Hyperreactivity and Inflammation	In Vivo, IH	Guinea pig, OA-sensitized	Inhaled R-albuterol (1.25 mM) and RS-albuterol (2.5 mM) significantly decreased basal airway reactivity to histamine by increasing PC100 3.7-fold. R-albuterol effect was longer in duration. Both also induced a significant reduction of allergen induced AHR. Neither of the drugs had any effect on late phase asthmatic reactions or the number of inflammatory cells and ciliated epithelial cells but both decreased the early response by 3.6 and 1.8-fold, respectively. S- albuterol had no effect on any of the parameters tested.
051-496- Effects of R-, S-, and RS-Albuterol on Allergic and Non-Allergic Airway Responses In Vivo, Ex Vivo, and In Vitro	In Vitro	Bovine tracheal smooth muscle cells	The ability of R-and RS-albuterol to decrease and S-albuterol to increase methacholine-induced calcium transients was investigated. The concentrations used were 0.1-10 μ M. At the 10 μ M concentration only, R- albuterol decreased and S-albuterol increased methacholine –induced calcium transients. RS-albuterol had no effect at any concentration.
051-498- Evaluation of the Effects of Albuterol Enantiomers on Mitogen-Stimulated Proliferation and Cytokine Production in Airway Smooth Muscle	In vitro	Human airway smooth muscle cells	The potential of R-, racemic and S- albuterol to inhibit GM-CSF production and DNA synthesis was investigated. R- and RS-albuterol at a concentration of 1 μ M inhibited the GM-CSF production (20-40%) and DNA synthesis (IC ₅₀ of 3-6.5 nM). R- and RS-albuterol at a concentration of 10 μ M completely inhibited the contractile response to carbachol. S-albuterol lacked significant effect on either of these experimental factors.
051-551-TheEffect of Formoterol and Albuterol Stereoisomers on Expression and Activity	In vitro	Human bronchial smooth muscle	The stereoisomers of albuterol at 10 - 1000 nM concentrations were investigated for their potential to activate Gi and Gs proteins, increase intracellular [free Ca ²⁺] and the pro-

of G-Protein, Intracellular free Ca ²⁺ and Nuclear Transcription Factor NF _κ B in Human Airway Smooth Muscle Cells		cells	inflammatory nuclear transcription factor NF _κ B. S-albuterol activated Gi proteins (2X) and activity in cells, increased intracellular [free Ca ²⁺] and decreases cAMP, and activates NF _κ B. No significant effects with R-albuterol.
051-567-The Effects of Enantiomers Albuterol on Indices of Remodeling in Human Lung Fibrosis	In Vitro	Human lung fibro-blasts	The effect of albuterol enantiomers on COL3A1 gene expression regulation was investigated at a concentration of 10 μm. R- and S-albuterol increased COL3A1 mRNA expression by 33% and 51%. The data suggest that repeated use of albuterol enantiomers could stimulate collagen production by airway fibroblasts and thus, contribute to airway remodeling.
051-568 The effects of Enantiomers of Albuterol non-Contracrile function of Human Airway Smooth Muscle	In Vitro	Human airway smooth muscle cells (HASM)	The potential of albuterol and formoterol enantiomers to inhibit GM-CSF production in was investigated using 10 nM to 20 μM concentrations. R-albuterol and RR-formoterol inhibited GM-CSF production (20-30%). S-albuterol and S, S-formterol increased GM-CSF production (5-20%).
051-577- Effects of Racemic Albuterol and Its Enantiomers on Cholinergic Twitch Contractions of Guinea Pigs and Bovine Trachea.	In Vitro	Guinea pig and bovine tracheal smooth muscle preparations	The objective of the study was to determine whether RS-, R- or S-albuterol was able to potentiate electrical field stimulation –induced cholinergic twitch contractions. Concentrations were 10 ⁻¹⁰ to 10 ⁻⁴ M. Neither albuterol nor its enantiomers increased basal cholinergic twitch contractions of guinea pig or bovine tissue.
051-555- S-Albuterol, But not Other β ₂ –Agonists has Stimulatory Effects on Mucin Secretion and Changes Gene Expression on Human Airway Epithelial.	In Vitro	Human lung tissue	The effect of albuterol and formoterol isomers to stimulate mucin secretion production or changes in gene expression in human tissue explants and tracheobronchial epithelial cells (TBE) was investigated in the presence of absence of 0.1 mM histamine. The drug concentrations were 0.001-100 μM for mucin secretion and 10 μM for gene expression. Neither albuterol nor formoterol isomers had any effect on mucin secretion production in the absence of histamine in either tissue explants or TBE cells; S-albuterol stimulated mucin secretion 25% in the presence of histamine. The results also show that S-albuterol, but not the other isomers elevated the expression of 13 genes belonging to signal

			transduction/transcription factors, surface proteins, apoptosis, proteases and differentiation markers by ~ 3-fold and down-regulated 30 other genes.
051-534- Effect of S-Albuterol on R-Albuterol Enhanced mucociliary Transport in Calf Trachea	In Vitro	Bovine trachea	The objective of this study was to study the effect of S-albuterol on R-albuterol mucociliary transport. Concentrations used were 10^{-9} to 10^{-4} . S-albuterol at concentrations of 10^{-6} and higher inhibited R-albuterol induced mucociliary transport ($EC_{50} = 3 \times 10^{-7}$ M).
051-563- The effects of Enantiomers of Albuterol on Human Monocyte and Macrophage Function	In Vitro	Human peripheral blood mononuclear cells	The objective of the study was to determine whether the release IL-10 was enhanced by incubation with albuterol and formoterol and their isomers. The concentrations used were 10 nM and 10 μ M. R-albuterol and R R-formoterol suppressed unstimulated TNF-a production by 60%; S-albuterol induced a less potent effect. IL-10 and GM-CSF were not affected by any enantiomer though when normalized for TNF-a release, R-albuterol enhanced the release of IL 10.
051-535- Study of Four Compounds on Various cytokine Secretion Using Basal Models.	In Vitro	Human PBMC and U-937 cells	R and S-albuterol and RR and SS formoterol were compared for their stimulatory effects on the basal secretion of IL-4, IL-5, IL-6 and TNF- α at 1 μ M concentrations. The compounds had negligible stimulatory effects after 48 hrs incubation on the release of IL-4, IL-5 and IL-6 and TNF-a. R-albuterol suppressed unstimulated TNF α production by 60% and stimulated TNF α production by 50 %.
051-538- Effect of RS, R and S-Albuterol on Sperm Motility	In vitro	Human semen cells	The aim of the study was to determine the effects of racemic, R and S-albuterol on sperm function after 15-30 min incubations. The drug concentrations were 300 to 0.003 μ g/mL. All 3 compounds at the highest concentration tested, elicited a small decrease in sperm linearity mirrored by a small but significant increase in average track speed. RS-albuterol also induced a slight increase in lateral amplitude.
051-546- Attenuation of Inflammatory-Induced Morphologic Changes and Apoptosis in Human	In Vitro	Human PMNs	The objective of this study was to determine whether albuterol or its isomers attenuate morphologic changes and apoptosis in human neutrophils. R-albuterol, but not S or RS –

Neutrophils by Albuterol is Isometric Specific			albuterol, attenuated PAF/fMLP induced increase in cell size by 1.5-fold. RS-albuterol attenuated PMN apoptosis % by ~ 33%. The data show that racemic and S-albuterol exhibited pro-inflammatory effects while R-albuterol attenuated these activities.
051-518- Differential Effects of Albuterol Enantiomers on Asthma-Like Syndrome in Rats	SC. 0.5 mg/kg/d, 14-d	Rats , post-viral asthma-like syndrome	The purpose of the study was to determine the effects of albuterol and its isomers on asthma-like syndrome. No conclusions could be drawn because the rats did not tolerate the pump implants well (.
051-531- Study of Four Compounds in Various Inflammatory Assays	In Vitro	Rat mast cells, Human HUVEC, HL-60, PBMC, U-937 cells	RR, and SS formoterol, and R and S-albuterol (10 µM) were assessed for their ability to inhibit stimulus-provoked secretion inflammatory mediators including IL-4, 5, and 6, PGD2, PGI2, LTC4 and TNF-α. None of the compounds had significant inhibitory activity on the secretion of any of the mediators tested with < 10% for LTC4 and PGI2 and 24-56% inhibition with the others.
051-574-Differential Effects of Racemic albuterol, R and S-Albuterol on Early and Late Phase Reactions, Airway Hyperresponsiveness and BAL Eosinophilia in Guinea Pig Model of asthma	In Vivo, IH, 7d (3x/day): 0.1 and 1% RS-albuterol; 0.01% S- and R-albuterol	Guinea Pig, antigen sensitized	Guinea pigs were sensitized with ovalbumin (OVA), placed in a plethysmographic chamber after being treated with racemic albuterol, or R or S-albuterol. Eosinophil infiltration was assessed by lavage, 24 hours after OVA challenge. R-albuterol attenuated the early and late phase reactions, with no significant effect noted for RS- or S-albuterol. R-albuterol attenuated the total number of inflammatory cells and eosinophils while S-albuterol increased these values.
051-516- Effect of Methacholine, RS, R and S-Albuterol and RR, SS and RR, SS-formoterol on Mucociliary Transport in Calf Trachea	In Vitro	Bovine trachea	The effect of methacholine, RS, R and S-albuterol and RR, SS and RR, SS formoterol was studied on mucociliary transport. Regarding the albuterol forms, R-albuterol was most potent in enhancing muciliary transport (EC50 – 4.1x10 ⁻⁹ M) followed by RS-albuterol (6x10 ⁻⁸ M) and then S-albuterol (1x10 ⁻⁷ M). A similar profile was observed in peak effect (110%, 102% and 56%).
051-486- Effects of Four compounds on Basal IL-5 Secretion By Human PBMC.	In Vitro, 10 ⁻¹⁰ to 3x10 ⁻⁶ M	Human PBMC	Racemic formoterol and albuterol and R and S- albuterol were studied for their stimulatory effect on IL-5 secretion by human PBM cells stimulated with concanavalin A. Cells were incubated with test compounds for 30 minutes prior to challenge. The results show that

			neither R or S-albuterol or racemic albuterol or formoterol had a stimulatory effect on concanavalin A induced IL-5 production.
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2.6.2.3 Secondary pharmacodynamics

There were no secondary pharmacodynamic studies submitted in this NDA. In studies in the dog, racemic albuterol causes increases in the rate and force of contraction of the heart. Albuterol also produces arrhythmias in the cat but at doses well above those required for bronchodilation. Albuterol has the potential to induce EKG changes in the dog. Prolonged tachycardia as well as increased force of contractions of the heart can result in myocardial necrosis. These effects are thought to be mediated mainly via beta 1 receptors. However, beta 2 receptors also play a role in these myocardial effects. Albuterol induces bronchodilation of the smooth muscle in the uterus. This effect plays a role in the formation of leiomyomas in the rat. Similar findings have been observed in toxicology studies with levalbuterol.

2.6.2.4 Safety pharmacology

No safety pharmacology studies were submitted to this NDA. However, safety pharmacology studies for levalbuterol were submitted to NDA 20-837. The effects of R-albuterol are similar to those reported for racemic albuterol.

Neurological effects:

R-albuterol had no locomotor effect in the mouse following IP dosing up to 199 mg/kg. R-albuterol was tremorigenic in mice in a dose-related fashion (32-128 mg/kg). Similar effects were noted with racemic and S-albuterol.

Cardiovascular effects:

R-albuterol induced dose-related (0.1 to 300 mg/kg, IV) decreases in blood pressure (35-50%) and concurrent increases in heart rate (up to 140% with 10% increase in QTc) in the dog. R-albuterol has been shown to induce myocardial necrosis in rats and dogs. R-albuterol induces decreases in plasma potassium as well as increases in plasma glucose. Similar effects were noted with racemic albuterol.

Pulmonary effects:

Albuterol is associated with relaxation of tracheobronchial smooth muscle, increased respiratory rate, volume, O₂ consumption and CO₂ production.

Abuse liability:

No abuse liability effects related to albuterol have been reported.

2.6.2.5 Pharmacodynamic drug interactions

No specific studies have been conducted with R-albuterol. However, pharmacodynamic drug interaction studies have been conducted with racemic albuterol and show that the concurrent use of beta 2 adrenergic agonists and phosphodiesterase inhibitors has been shown to exacerbate the myocardial toxicity in the rat and rabbits (Whitehurst, VE, et al: (1983), Cardiotoxic Effects in Rats and Rabbits Treated with Terbutaline Alone and in Combination with Aminophylline. J. Amer. Coll. Toxicol. 2, 147-153) and Joseph X et al: (1981), Enhancement of Cardiotoxic Effects of Beta -Adrenergic Bronchodilators by Aminophylline in Experimental Animals. Fundam. Appl. Toxicol. 1, 443-447). It has also

been reported that additive cardiotoxicity may occur in the asthmatic patient who is receiving theophylline and high doses of beta 2 agonists (Wilson, JD, et al: (1981), Has the Change to Beta Agonist Combination with Oral Theophylline Increased Cases of Fatal Asthma. Lancet 1, 1235-1237).

2.6.3 PHARMACOLOGY TABULATED SUMMARY

There was no pharmacology tabulated summary provided by the sponsor.

2.6.4 PHARMACOKINETICS/TOXICOKINETICS

2.6.4.1 Brief summary

Albuterol is rapidly absorbed following oral and inhalation administration. In newly submitted distribution studies, distribution of R-albuterol to brain tissue of rats following IV dosing was minimal while plasma and pituitary concentrations were similar. Protein binding of (R)-albuterol is weak (< 50%) and was not affected by the presence of (S)-albuterol. The Proventil HFA label indicates that albuterol-related material is transferred to the fetus following administration to pregnant rats. In studies conducted for NDA 20-837 the dog did not metabolize racemic albuterol after oral administration. However, rats, guinea pigs and rabbits metabolize oral racemic albuterol to a glucuronic acid conjugate. There was no evidence of stereo-selectivity. Published data suggests that racemic albuterol is stereo-selective in favor of R-albuterol in humans suggesting that man metabolizes albuterol differently than in studied animal species. The main metabolite in humans is thought to be albuterol-4-O-sulfate. Albuterol is primarily excreted through the urine.

The kinetics of R-albuterol were evaluated in 90-day inhalation studies in rats and dogs. In Wistar rats administered inhalation doses up to 6 mg/kg R-albuterol the results show that increases in R-albuterol levels were dose-related and increased in a generally proportional manner. The exposure to R-albuterol following inhalation of 6 mg/kg R-albuterol was similar to that observed following inhalation of 12 mg/kg racemic albuterol. The levels of R- and S-albuterol following inhalation of RS-albuterol were similar. There was no accumulation of R- or S-albuterol. In dogs, AUCs (0-24 hours) (ng.h/mL) on day 87 were similar in male and female dogs and ranged from 39 to 55 ng.h/mL. The systemic exposure of levalbuterol (AUCs) tended to decrease from day 1 to day 87 (8-30%). The C_{max} decreased by 2-3 fold. T_{max} was achieved 0.4 to 1.2 hours.

2.6.4.2 Methods of Analysis

See under individual study reviews.

2.6.4.3 Absorption

Racemic albuterol is rapidly absorbed in rodents and man after oral and inhalation administration. Maximum plasma concentrations were obtained 2 hours post dosing in

Sprague-Dawley rats following oral administration. Peak plasma levels were observed in rabbits and dogs 1-2.5 hours after oral administration.

2.6.4.4 Distribution

Five new studies were submitted to this NDA.

Study 051-504: Wistar male rats were administered a single IV dose of labeled 10 mg/kg (100 μ Ci/kg) R-, S- or RS-albuterol. Concentrations of radioactivity in plasma, brain and pituitary tissues were assessed. Plasma concentrations of radioactivity were similar regardless of the compound and remained fairly constant over the 1.5 hour sampling period for R and racemic albuterol. The S-albuterol radioactivity declined rapidly. The transfer of radioactivity matter across the blood:brain barrier was minimal (brain: plasma ratios < 0.082), however concentrations of radioactivity materials in the pituitary were similar to those in the plasma (tissue:plasma ratios ranged from 0.886 to 1.53).

051-554: Rat brain tissues were spike with varying levels of racemic albuterol for the purpose of determining R- and S-albuterol via an HPLC method. Pretreatment with propranolol produced significantly higher brain R-albuterol concentrations compared with saline pretreatment. No evidence of in vivo chiral inversion was observed in any brain tissues.

051-503: The study was designed to assess the in vitro blood to plasma partitioning, plasma protein binding of R-albuterol in male dogs and human, and to examine the effect of S-albuterol on protein binding of R-albuterol. The concentrations used were 0.5-500 ng/mL 3 H-(R)-albuterol. The effect of S-albuterol tested a 500 ng/ml concentration versus 0.5 and 100 ng/ml (R)-albuterol. The results show that the blood to plasma partitioning was constant over the range tested at 1.08-1.12. The fraction distributed to RBCs was 0.47. R-albuterol was weakly bound to plasma protein (5-47%) in both species with humans being slightly higher. The presence of S-albuterol had no significant effect of the plasma protein binding of R-albuterol.

052-402: The study was designed to assess the in vitro blood to plasma partitioning and plasma protein binding of S-albuterol in dogs and humans and to examine the effect of R-albuterol on the protein binding of S-albuterol. The concentrations used were 0.5, 2.5, 25, 100 and 500 ng/mL 3 H-(S)-albuterol. The effect of R-albuterol tested a 500 ng/ml concentration versus 0.5 and 100 ng/ml (S)-albuterol. The results show that the blood to plasma partitioning declined with increasing concentration in humans (1.62 to 1.01) while it remained constant in dogs over the range tested at 1.09-1.12. The fraction distributed to RBCs was 0.66 to 0.44 in humans and 0.45-0.47 in dogs. S-albuterol was weakly bound to plasma protein (8-28%) in both species with humans being slightly higher. The presence of R-albuterol had no significant effect of the plasma protein binding of S-albuterol.

051-526: In vitro blood to plasma partitioning and plasma protein binding studies were conducted using fresh human blood and plasma. R-albuterol concentrations were 0.1-25

ng/mL. The fraction of 3H-R-albuterol distributed to blood cells was 0.46-0.5. The binding of R-albuterol to plasma proteins was low (5.4-13.5%) over the range tested. The presence of S-albuterol or formoterol or its isomers did not alter the protein binding of R-albuterol.

In studies in the rat, racemic albuterol was found in significant concentrations in the kidney, the liver and the plasma only 24 hours after oral dosing. There was no drug accumulation in the tissues of animals following repeated dosing. The Proventil HFA label indicates that albuterol-related material is transferred to the fetus following administration to pregnant rats.

2.6.4.5 Metabolism

In studies conducted for NDA 20-837 the dog did not metabolize racemic albuterol after oral administration. However, rats, guinea pigs and rabbits metabolize oral racemic albuterol to a glucuronic acid conjugate. There was no evidence of stereo-selectivity. Published data suggests that racemic albuterol is stereo-selective in favor of R-albuterol in humans suggesting that man metabolizes albuterol differently than in studied animal species.

The main metabolite in humans is thought to be albuterol-4-O-sulfate.

2.6.4.6 Excretion

Albuterol is excreted mainly in the urine in rats, dogs and humans. In the rat, urinary excretion accounted for 52-59% and fecal excretion 25-40%. In humans, approximately 60-70% is excreted in the urine.

2.6.4.7 Pharmacokinetic drug interactions

No drug interaction studies were conducted for this NDA.

2.6.4.8 Other Pharmacokinetic Studies

Two toxicokinetic studies were submitted to this NDA in support of toxicology studies and are reviewed below:

051-515: Toxicokinetics of R-and S-Albuterol during a Subchronic (90days) Repeat Dose Inhalation Toxicity Study in Rats:

Wistar rats (male and female; 5/sex/group) were administered inhalation doses of 0.06, 0.6, and 6 mg/kg R-albuterol and 12 mg/kg racemic albuterol for 86 days for 1 hr/day. Only C_{max} and t_{max} of R- and S-albuterol were evaluated. The results show that increases in R-albuterol levels were dose-related and increased in a generally proportional manner. The exposure to R-albuterol following inhalation of 6 mg/kg R-

albuterol was similar to that observed following inhalation of 12 mg/kg racemic albuterol. The levels of R- and S-albuterol following inhalation of RS-albuterol were similar. There was no accumulation of R- or S-albuterol. There were no gender differences in R-, S- or racemic-albuterol-treated rats. Plasma levels could not be detected for R or S albuterol 24 hours after dosing. S-albuterol was not detectable in the plasma of the rats after 86 day dosing.

PHARMACOKINETIC RESULTS					
Treatment/ Gender/ Dose (mg/kg/d)	(R)-Albuterol Results				
	C _{max} (ng/mL)		t _{1/2} (h)		
	Day 1	Day 86	Day 1	Day 86	
(R)-Albuterol					
Males					
0.06	4.44	3.86	Postdose*		Postdose
0.6	54.8	42.4	Postdose		Postdose
6	469	332	Postdose		Postdose
Females					
0.06	47.7	6.88	Postdose		Postdose
0.6	17.9	69.8	1		Postdose
6	454	494	Postdose		Postdose
(RS)-Albuterol					
Males					
12	360	256	Postdose		Postdose
Females					
12	458	318	Postdose		Postdose

Gender/ (RS)-Albuterol Dose (mg/kg/d)	(S)-Albuterol Results			
	C _{max} (ng/mL)		t _{1/2} (h)	
	Day 1	Day 86	Day 1	Day 86
Males				
12	370	337	Postdose	Postdose
Females				
12	460	371	Postdose	Postdose

*: post-dose defined as a sample taken immediately after the end of the 1-hr IH exposure.

051-578-Toxicokinetics of Levalbuterol During a 13 Week Inhalation Study in Dogs: Beagle dogs-40 actuations of aged vs fresh Levalbuterol HFA MDI. Levalbuterol was aged for 6 months. There were 3 dogs/sex/dose group. The drugs were given by inhalation daily for 90 days. Blood samples were collected 0.5, 1, 2, 4, 6, and 24 hours. The results show AUCs (0-24 hours) (ng.h/mL) on day 87 were similar in male and female dogs and ranged from 39 to 55 ng.h/mL. The systemic exposure of levalbuterol decreased day 1 to day 87. No S-albuterol was observed in the plasma samples. No gender differences were observed in this study.

051-578: Toxicokinetics of Levalbuterol During a 13 Week Inhalation Toxicity Study in Dogs

The objective of this study was to evaluate the TK of R- and S-albuterol in dogs following inhalation of fresh or aged Xopenex HFA. There were 3 dogs/sex/dose group. The drugs were given by inhalation daily for 90 days. Blood samples were collected 0.5, 1, 2, 4, 6, and 24 hours. The results show AUCs (0-24 hours) (ng.h/mL) on day 87 were similar in male and female dogs and ranged from 39 to 55 ng.h/mL (see table below). The systemic exposure of levalbuterol (AUCs) tended to decrease from day 1 to day 87 (8-30%) with the exception of males given unaged formulation. The C_{max} decrease by 2-3-fold. This decrease may have been at least partly due to lower dosing on a mg/kg basis

as the study proceeded as dosing was not adjusted once the study began. Tmax was achieved 0.4 to 1.2 hours. No S-albuterol was observed in the plasma samples. No gender differences were observed in this study.

Table 4. Mean (R)-Albuterol Pharmacokinetic Parameters Following the Inhalation Administration of Levalbuterol L-Tartrate via MDI or Aged MDI Canisters in Male and Female Dogs

Levalbuterol Treatment/ Evaluation Period	Male Dogs			Female Dogs		
	AUC (0-24h) (ng•h/mL)	C _{max} (ng/mL)	t _{max} (h)	AUC (0-24h) (ng•h/mL)	C _{max} (ng/mL)	t _{max} (h)
MDI						
Day 1	47.0±14.2	11.6±2.91	0.36±0.24	48.9±2.28	13.1±6.17	1.17±0.76
Day 87	55.6±20.1	7.59±4.43	1.00±0.00	45.5±7.51	7.69±3.23	1.17±0.76
Aged MDI						
Day 1	55.7±11.7	14.2±3.47	0.83±0.29	49.8±7.69	15.2±4.91	0.50±0.00
Day 87	38.9±8.98	4.65±1.25	0.83±0.29	46.0±3.72	7.38±3.35	0.83±0.29

2.6.4.9 Discussion and Conclusions

In vitro and *In vivo* studies reveal that absorption, metabolism, distribution and excretion of R-albuterol and racemic albuterol are similar in rats, rabbits, dogs and humans. Racemic albuterol is rapidly absorbed in rodents and man after oral and inhalation administration. In rats, rabbits and dogs, R- and racemic albuterol are metabolized via glucuronidation pathways. In studies in the rat, racemic albuterol was found in significant concentrations in the kidney, the liver and the plasma only 24 hours after oral dosing. Relatively low protein binding levels (< 50%) of R-albuterol were observed. There was no drug accumulation in the tissues of animals following repeated dosing of R-albuterol or racemic albuterol. R- and racemic albuterol are excreted mainly in the urine in rats, dogs and humans. R- and racemic albuterol crossed the blood-brain barrier at minimal levels. The ADME of S-albuterol could not be determined in *in vitro* and *in vivo* studies.

2.6.4.10 Tables and figures to include comparative TK summary

Not applicable.

2.6.5 PHARMACOKINETICS TABULATED SUMMARY

There were no pharmacokinetic tabulated summaries provided by the sponsor.

2.6.6 TOXICOLOGY

2.6.6.1 Overall toxicology summary

General toxicology:

Acute Toxicity

Two acute IV toxicity studies were submitted to and reviewed under NDA 20-837. Acute toxicity was compared for (R)-, (S)-, and racemic albuterol following a single intravenous administration in mice. Median lethal doses ranged from 66-70 mg/kg, IV. Deaths occurred within 15 minutes of dosing and were thought to be related to cardiovascular of CNS effects. Clinical signs included tremors and lethargy.

In a dose escalation study reviewed for this NDA, dogs received a single nebulized inhalation exposure of levalbuterol at estimated inhaled doses of 11-42 µg /kg Xopenex HFA. Treatment-related effects included tachycardia at all doses and tremors, hyperactivity and increased breathing rate at the highest dose.

Multidose Toxicity

Subchronic toxicity studies reviewed under NDA 20-837 were carried out in the rat and dog for up to 90 days and included oral and inhalation exposure with levalbuterol. These studies did not demonstrate any significant treatment-related findings that were not also observed with the appropriate comparative doses of racemic albuterol.

Three oral and three inhalation studies with (R)-, (S)-, and/or racemic albuterol were performed in rats. The oral dosing studies consisted of: racemic albuterol for 1 week using (R)-, (S)-, and racemic albuterol for 1 month ; and (R)- and racemic albuterol for 3 months. The highest oral doses in the longest treatment durations, (R)-albuterol at 2.5-25 mg/kg/day for 3 months, (S)-albuterol at 25 mg/kg/day for 1 month, and racemic albuterol at 50 mg/kg/day for 3 months, were tolerated by rats. The findings consisted of slight increases in body weights of animals that received racemic albuterol at 25 mg/kg/day and pathologic changes in spleen (granular surface, multifocal capsulitis, capsular thickening, and/or capsular cysts) in rats that received (R)- or (S)-albuterol at 2.5 mg/kg/day or racemic albuterol at 25 mg/kg/day. Following a 1-month recovery from the 3-month study, these changes were reversed or were in the process of recovery, and were not considered to be toxicologically relevant. The effects of (R)- and racemic albuterol at durations up to 3 months were comparable. After 3 months of oral dosing, no no-observed-adverse-effect level (NOAEL) was identified.

Three inhalation studies in rats using a nebulization delivery included 5-day with (R)- and (S)-albuterol and 1-month and 3-months with (R)- and racemic albuterol. After 3 months of exposure to (R)-albuterol at estimated doses of approximately 0.06, 0.61, and 6 mg/kg/day and racemic albuterol at an estimated dose of approximately 11.8 mg/kg/day followed by a 1-month recovery period, the treatment-related findings included transient clinical sign of salivation, increased body weights and food consumption, and/or increased heart weights that were correlated microscopically to cardiac hypertrophy from (R)-albuterol (~6 mg/kg/day) and racemic albuterol (~11.8 mg/kg/day). Additional pathologic findings in the heart included focal/multifocal myocardial necrosis/cytolysis in all treatment groups and increased incidence of myocardial inflammatory cell foci in the high-dose groups; these heart findings were partially reversible. The adverse findings

with (R)- and racemic albuterol were comparable. There were no NOAEL in the 3 months study because of the myocardial necrosis findings.

Four inhalation toxicology studies were performed with (R)-albuterol in dogs. Two 1-week studies and two 3-month studies were performed using nebulizers. In 1-week dog nebulizer studies with (R)-albuterol at estimated doses of 0.001, 0.01, 0.1, 0.13, 0.4, 0.55, and 2.73 mg/kg/day, treatment-related effects were limited to transient tachycardia and generalized flushing. The electrocardiographic evaluation indicated a possible myocardial hypoxia that was secondary to tachycardia. The pathologic evaluation exhibited a focal myocardial degeneration in the dorsal papillary muscle in the 0.55 and 2.73 mg/kg/day groups. The NOAEL was 0.4 mg/kg.

In a 3-month nebulization study achieved (R)-albuterol doses of 0.002, 0.01, and 0.29 mg/kg/day and the achieved racemic albuterol dose of 0.52 mg/kg/day were used. The NOAEL in this study was 0.29 mg/kg for R-albuterol. Tachycardia was noted at all doses. There was no myocardial necrosis reported in this study.

Subsequently, a 3-month study was performed using the intended commercial MDI device (fresh versus aged formulations) as a part of the leachable qualification of Xopenex MDI. In these multiple-dose inhalation studies, findings were limited to known pharmacologic effects of beta-adrenoceptor agonists. This study is reviewed in the Toxicology section.

Genetic toxicology

Two new in vitro studies were conducted to determine the genotoxic potential of R-albuterol as well as S- and RS-albuterol in a CHO cell chromosome aberration assay and an in vivo mouse micronucleus assay. All three compounds were negative in the mouse micronucleus assay under the conditions tested. Although none of the compounds produced positive findings in the CHO cell assay under the conditions tested, the assay was not valid for R- or S-albuterol at the 3 hr time point since the maximum doses used were not adequate and a confirmatory assay was not conducted for S-albuterol. Therefore, this assay should be repeated should the sponsor wish to include the results with R-albuterol in the product label. An Ames/ Salmonella-E-coli reverse mutation and CHO/HPRT mammalian forward gene mutation assays were reviewed under NDA 20-837. R-albuterol was considered to be negative in these assay under the conditions tested. In addition negative genotoxicity findings for racemic albuterol have previously been established (as per the product labeling for Proventil). These data show that albuterol sulfate was not mutagenic in the Ames test with and without metabolic activation. No forward mutation was seen in yeast strain *S. cerevisiae* S9 nor any mitotic gene conversion in yeast strain *S. cerevisiae* JD1 with and without metabolic activation. Fluctuation assays in *S.typhimurium* TA98 and *E.coli* WP2, both with metabolic activation, were negative. Albuterol sulfate was not clastogenic in the human peripheral lymphocyte assay or in an AH1 strain mouse micronucleus assay..

Carcinogenicity:

No specific carcinogenicity studies have been conducted with R-albuterol. However, carcinogenicity studies have been conducted with albuterol sulfate and are described in the product labels for Xopenex and Proventil. Albuterol sulfate was not carcinogenic in an 18 month oral (diet) chronic/carcinogenicity study in CD1 mice at doses up to 500 mg/kg. In a 2 year oral (feeding) study in the CD rat, albuterol sulfate at doses of 2, 10 and 50 mg/kg induced significant, dose-related increases in smooth muscle tumors, leiomyomas, in the mesovarium at all doses; an effect that was blocked by the coadministration of propranolol, a nonselective beta-adrenergic antagonist. A 99 week (696 days) oral oncogenic study was carried out in the female Syrian hamster. The albuterol sulfate doses were 10 and 50 mg/kg. There was no tumorigenicity in this study.

Reproductive toxicology:

No new studies were conducted for this NDA. The following section describes the findings summarized in the approved Xopenex package insert.

Studies to assess fertility have not been performed with levalbuterol. Reproduction studies in rats using racemic albuterol sulfate demonstrated no evidence of impaired fertility at oral doses up to 50 mg/kg.

A maternal and fetal developmental study in rabbits was performed with R- albuterol at oral gavage doses of 0.5, 5, and 25 mg/kg/day. There were no maternal or fetal developmental toxicities.

Segment II studies were conducted with racemic albuterol in rabbits and mice. A segment II study was carried out in the Wistar rat using oral albuterol doses of 0.0.5, 2.32, 10.75 and 50 mg/kg, days 1-19 of pregnancy. No teratogenic effects were observed. Segment II studies in the Stride Dutch rabbit demonstrated cranioschisis following oral doses of 50 mg/kg. Subcutaneous injection of racemic albuterol to CD-1 mated mice resulted in cleft palate at doses of 0.25 mg/kg or greater; no effects were observed at 0.025 mg/kg. Cleft palate also occurred in 22 of 72 (30.5%) fetuses from females treated subcutaneously with 2.5 mg/kg of isoproterenol (positive control).

Based upon the results of the aforementioned studies, albuterol sulfate has been given a Pregnancy Category C designation.

Special toxicology:

An acute dermal irritation study of levalbuterol and _____ and an acute eye irritation study of levalbuterol and _____ studies were conducted in albino rabbits. The Primary Irritation Indices for levalbuterol and _____ were 0.2 and 0.0, respectively in the dermal test. In the eye irritation test, the group mean scores for corneal opacities, iritis, conjunctival redness and swelling for levalbuterol were 0, 0, 1.2, and 0.3, respectively, indicating that levalbuterol was a mild ocular irritant.

2.6.6.2 Single-dose toxicity

Xopenex HFA Pressured Metered Dose Inhaler: Dose Ranging-Finding Study in the Dog (Study Number 051-824).**Methods:**

The objective of the study was to determine a suitable levalbuterol inhalation dose level in the dog for use in a 3 month repeat-dose inhalation study. Two beagle dogs/sex were included in this study. Levalbuterol MDI was delivered through the mouth via a face mask. The test article was delivered using an escalating dose sequence of 0.11, 0.16, 0.21 and 0.42 mg/kg (inhalation dose); actuations per session ranged from 20-40 and 1 dosing session was used for each group except the highest dose in which animals were dosed in 2 sessions. Each animal was dosed on each day either to placebo or to levalbuterol; 1/sex was exposed to either. There was a day clearance between doses. To deliver a uniform dose, the dosing unit was shaken prior to each actuation and was actuated within 5 seconds of shaking during the inspiratory breath of the dog. Plasma samples were taken for toxicokinetics but later discarded.

Results:

All the dogs survived. There were no drug-related effects on body weights, food consumption or blood pressure. Clinical signs included tremors, hyperactivity, increased breathing rate and dilated pupils at the highest dose of 0.42 mg/kg. Levalbuterol-related tachycardia was observed in the dogs treated with 0.11 mg/kg and higher. All the symptoms were reversible. No gross or microscopic pathology was conducted. Based on the above results, a procedure of 40 actuations (target delivery of 0.21 mg/kg/d) was recommended in the 13-week study.

2.6.6.3 Repeat-dose toxicity

Study title: Xopenex HFA Pressurized metered Dose Inhaler: 13 Week Inhalation Qualification study in the Dog.

Key study findings:

- The objective of this study is to assess the comparative toxicity of fresh and aged (6 months) formulations of Xopenex HFA in order to qualify the proposed specifications of certain leachables.
- Levalbuterol, aged and fresh, induced increased body weights, tachycardia, fibrosis/mineralization in the heart, increased plasma glucose and lower serum potassium levels.
- A NOAEL was not identified but all findings were expected based on the known toxicity profile of levalbuterol.
- There was no significant difference in the responses to the unaged or aged formulations indicating that increases in potential leachates from the aged product did not affect the overall toxicity profile.

Study no.: 051-825A2

Volume #, and page #: Electronic submission

Conducting laboratory and location: —

Date of study initiation: December 17, 2002

GLP compliance: Yes

QA report: yes (x) no ()

Drug, lot #, and % purity: Xopenex HFA/020636 (fresh, stored at 25 degrees C and 65% RH, valve up)/ 2B285 (aged for 6 months, stored at 40 degrees C and 75% RH)/

Methods

Doses: Target doses were 0.2118 mg/kg based on 40 actuations/daily dose session and 45 µg/actuation and assumed body weight of 8.5 kg.

Achieved inhalation doses: Fresh: 0.125 mg/kg; Aged: 0.128 mg/kg administered daily for 13 weeks.

Achieved dose levels were calculated using the following formula:

$$\text{Dose (mg/kg/day)} = \frac{C \times F \times N \times 10^3}{\text{BW}}$$

Where:

Dose = Estimated test article administered to the animal (mg/kg/day); C = Group mean weight loss from the canisters (g); F = the fraction of the MDI formulation delivered as levalbuterol base to the animal (0.000623); N = Number of actuation (40); 10³ Converts from grams to milligrams; BW = Group mid study bodyweight (kg)

Assuming a 25% pulmonary deposition dose for dogs, the doses were 31 µg/kg (unaged) and 32 mg/kg (aged).

Group 1: MDI placebo control

Group 2: Levalbuterol MDI unaged valve up

Group 3: Levalbuterol MDI aged valve down

Group 4: Air (sham) control

The aged canisters contained a — s. (Table on the levels of leachables) A Xopenex HFA placebo (Batch 01-005-01-042) and an air control group were also used. The number of actuations per session was not adjusted for increasing group mean body weight.

The dosage was selected after consideration of the dose level required to elicit the significant clinical pharmacologic signs (i.e., tachycardia) noted in the previous dose range-finding study (— study number 1487/3). In the dose range-finding study, it was apparent that 40 actuations/session was the maximum that could be tolerated by the dog in a repeated dosing scheme in terms of feasibility when the study dosing procedure was used.

Species/strain: Beagle dog

Number/sex/group or time point (main study): 3/sex/dose group

Route, formulation, volume, and infusion rate: Oral inhalation through face mask; nostrils were occluded. Inhalation by actuators and canisters / suspension/each actuation was 45 micrograms of levalbuterol base

Satellite groups used for toxicokinetics or recovery: NA

Age: 28-33 weeks

Weight: Males: 8.98-11.89 kg and females: 8.89-11.56 kg

Sampling times: blood samples were on day 1 and during week 13. Blood was collected at 5, 30 60 minutes, 2, 4, 6 and 24 hours after dosing.

Unique study design or methodology (if any): NA

Observations and times:

Achieved doses:

Mortality: Observed daily;

Clinical signs: Observed daily;

Body weights: Weighed weekly and before necropsy.

Food consumption: Assessed weekly;

Ophthalmoscopy: Examined pre-study and week 12;

EKG: Pre-test, days 2, 8, 15, 22, 29, weeks 9 and 13; heart rate readings only were made pre-dose and at 30 minutes and 2, and 23 hours after dosing.

Hematology: Assessed Pre-test and week 13

Clinical chemistry: Assessed Pre-test and week 13

Urinalysis: assessed pre-test and week 12

Gross pathology: Termination of the study

Organ weights (specify organs weighed if not in histopath table): adrenals, kidneys, spleen, liver, lung, heart, salivary glands, brain, pituitary, thyroids, parathyroids, prostate, testes, epididymides, ovaries, uterus

Histopathology: Adequate Battery: yes (), no (x)—explain

Only specific tissues from all dogs were evaluated. The tissues include the lung, heart, stomach, liver, kidney, pituitary, nasal cavity, Duodenum, epididymis, parathyroid, trachea, tonsils, thyroid, Oral cavity, cecum, thymus, prostate, salivary gland, tongue, skin, pancreas, muscle, optic nerve, mammary gland, spleen, spinal cord, sternum, adrenals and gall bladder

Peer review: yes (x), no ()

Toxicokinetics: Blood samples taken on days 1 and during week 13 within 30 minutes prior to dosing and then at 5, 30, 60 minutes, 2, 4, 6 and 24 hours after dosing. Samples were dispatched to _____ for analysis of R- and S-albuterol levels by LC/MS/MS method.

Results

Mortality: There were no deaths in the study.

Clinical signs: There were no clinical signs induced by aged or unaged Xopenex HFA.

Body weights: All the dogs in the study gained weight. Body weight gains were greater in the Beta 2 agonist treated groups than in the air or placebo dose groups. For males, body weight gain in the unaged, and aged formulation groups were increased by 64 and 66%, respectively, versus the placebo formulation and 23 and 24%, respectively, versus the air control group. In females, body weight gain in the unaged and aged formulation groups increased by 78-93% versus the placebo and air control groups.

Food consumption: There were no drug-related effects on food consumption.

Ophthalmoscopy: There were no test article effects on the eyes.

EKG: Expected increases in heart rate (up to 30-40%) were observed in the drug-treated groups, usually in the first 0.5 to 2 hours after dosing.

Hematology: The minor changes observed in the hematology parameters in the drug-treated groups were considered to reflect routine variation in animals and were not related to treatment.

Clinical chemistry: At week 13, glucose plasma levels were decreased by 2-10% versus placebo and air controls and serum potassium levels were increased by 7-10% versus both control groups. The increases in glucose and decreases in potassium were very small, approximately 4-5% in each dose group. A 25-30% increase in creatine versus placebo control was also observed in the drug-treated groups.

Urinalysis: There were no treatment-related changes in urinalyses parameters.

Gross pathology: There were no treatment-related macroscopic findings.

Organ weights: Absolute organ weights were not affected by drug treatment. The heart and liver weights (relative to body weight) were reduced slightly in the beta 2 treated males versus placebo and air controls. The decreases were approximately 13-15% in the heart and 7-14% in the liver. The sponsor mentions adrenals and lung organ weight decreases in males and females, however the decreases were very small.

Histopathology:

Microscopic examination of the tissues revealed fibrosis /mineralization characterized by focal collagen fiber accumulation containing basophilic mineralized granules within the papillary muscles of the heart. The findings are shown below:

		Group incidence of selected microscopic findings in the heart							
		Males				Females			
Tissue and finding	Level (mg/kg/day)	1M	2M	3M	4M	1F	2F	3F	4F
				0	*	*	0	0	*
Heart	No. examined:	3	3	3	3	3	3	3	3
Fibrosis/mineralisation	Grade -	3	2	2	3	3	1	2	3
	1	0	0	0	0	0	0	1	0
	2	0	1	1	0	0	2	0	0

*Estimated dose levels: Group 2: 125 mg/kg/day unaged formulation, Group 3: 128 mg/kg/day aged formulation.
Pathology severity key: "*" = finding not present, 1 = minimal, 2 = slight

There were no other significant microscopic findings due to the test articles.

Toxicokinetics:

The results show AUCs (0-24 hours) (ng.h/mL) on day 87 were similar in male and female dogs and ranged from 39 to 55 ng.h/mL (see table below). The systemic exposure of levalbuterol (AUCs) tended to decrease from day 1 to day 87 (8-30%) with the exception of males given unaged formulation. The C_{max} decrease by 2-3 fold. This decrease may have been at least partly due to lower dosing on a mg/kg basis as the study proceeded as dosing was not adjusted once the study began. T_{max} was achieved 0.4 to 1.2 hours. No S-albuterol was observed in the plasma samples. No gender differences were observed in this study.

Table 4. Mean (R)-Albuterol Pharmacokinetic Parameters Following the Inhalation Administration of Levalbuterol L-Tartrate via MDI or Aged MDI Canisters in Male and Female Dogs

Levalbuterol Treatment/ Evaluation Period	Male Dogs			Female Dogs		
	AUC (0-24h) (ng•h/mL)	C _{max} (ng/mL)	t _{max} (h)	AUC (0-24h) (ng•h/mL)	C _{max} (ng/mL)	t _{max} (h)
MDI						
Day 1	47.0±14.2	11.6±2.91	0.36±0.24	48.9±2.28	13.1±6.17	1.17±0.76
Day 87	53.6±20.1	7.59±4.43	1.00±0.00	45.5±7.51	7.69±3.23	1.17±0.76
Aged MDI						
Day 1	55.7±11.7	14.2±3.47	0.83±0.29	49.8±7.69	15.2±4.91	0.50±0.00
Day 87	38.9±8.98	4.65±1.25	0.83±0.29	46.0±3.72	7.38±3.35	0.83±0.29

2.6.6.4 Genetic toxicology

Study title: Chromosomal Aberration in Chinese hamster Ovary (CHO) Cells With R-Albuterol, S-Albuterol and RS-Albuterol

Key findings:

- (RS)-albuterol was negative for inducing chromosomal aberrations in CHO cells with and without metabolic activation under the conditions tested.
- R-albuterol and S-albuterol did not produce aberrations consistently under the conditions tested. However, the tests for these compounds were not considered to be valid since dosing under 3 hr treatments for both did not achieve adequate cellular toxicity and did not use an adequate maximum dose for the assay. Further, a confirmatory assay was not conducted for S-albuterol.
- The sponsor concluded that the compounds were negative in the assay.

Study no.: 051-822

Volume #, and page #: Electronic submission

Conducting laboratory and location: —

Date of study initiation: August 18, 2000

GLP compliance: Yes

QA reports: yes (x) no ()

Drug, lot #, and % purity: R-albuterol/004-0008/ purity not reported; S-albuterol/080698A/purity not reported; and RS-albuterol/245764/ —

Methods

Strains/species/cell line: Chinese hamster ovary cells

Doses used in definitive study: R-albuterol: Trial I: 0.555, 1.11, 2.21, 4.42, 10.6, 21.2, 42.3, 84.5, 169, 338, 676, 966, 1380, 1970 and 2810 mcg/mL with and without metabolic activation; Trial IA: 168, 336, 671, 959, 1370, 1960, 2800 µg/ml with and without metabolic activation. Trial II: 31.3, 62.5, 125, 250, 500, 1000, 1500, 2000, 2400, 2800 µg/ml.

S-albuterol: Trial I: 0.660, 1.32, 2.63, 5.25, 10.5, 21.0, 42.0, 84.0, 168, 336, 671, 1370, 1960 and 2800 mcg/mL without metabolic activation and 0.555, 1.11, 2.21, 4.42, 10.6, 21.2, 42.3, 84.5, 169, 338, 676, 966, 1380, 1970, and 2800 µg/mL were tested with metabolic activation for 3.0 hours. A confirmatory assay was not conducted due to limited supply of S-albuterol.

RS-albuterol: Trial I: 600, 1200, 1720, 2450, 3500 and 5000 mcg/mL with and without metabolic activation -1720, 2450, 3500, 5000 mcg/mL for chromosomal aberration. Trial IA: 300, 600, 1200, 1720, 2450, 3500, and 5000 µg/mL were tested with and without metabolic activation. Trial II: 31.3, 62.5, 125, 250, 500, 1000, 1500, 2000, 2500, 3750, and 5000 µg/mL without activation

Basis of dose selection: The high concentration for RS-albuterol is the highest recommended concentration for this assay. The high concentrations for R- and S-albuterol were noted as being the “recommended high dose” for this assay; rationale was not provided.

Negative controls: Cell culture-grade water and McCoy 5a-culture medium

Positive controls: Mitomycin C (MMC) for the nonactivation series and cyclophosphamide (CP) in the metabolic activation series. In the chromosomal aberrations assays, two concentrations of MMC (0.750 and 1.50 µg/mL, for the 3-hour treatment, 0.200 and 0.400 µg/mL, for 17.5-hour treatment) and CP (5.00 and 10.0 µg/mL) were used to induce chromosomal aberrations in the CHO cells. One of the concentrations was analyzed in each of the aberration assays. Both MMC and CP were dissolved in water.

Incubation and sampling times:

Replicate cultures were used at each concentration and the vehicle and negative controls, and for each of two concentrations of the positive control. A single ~ 20.0-hour harvest time without and with S9 was conducted. This harvest time corresponds to 1.5 times a cell cycle time of approximately 13 hours (Galloway *et al.*, 1994).

Aberrations Assay Without Metabolic Activation Cultures were initiated by seeding approximately 0.3×10^6 cells per 25 cm² flask into 5 mL of complete McCoy's 5a medium

(Trial I) or initiated by seeding approximately 1.2×10^6 cells per 75 cm² flask into 10 mL of complete McCoy's 5a medium (Trial IA and II). One day after culture initiation, the cells were incubated at 37 degrees C with the test article at predetermined concentrations for 3.0 hours or 17.5 hours. The cultures were then washed with buffered saline. In the initial assay, the cells were refeed with complete McCoy's 5a medium and incubated for the rest of the culture period up to the time of harvest with 0.1 µg/mL Colcemid present during the last 2.0 hours of incubation. In the confirmatory assay, the cells were refeed with complete McCoy's 5a medium with 0.1 µg/mL Colcemid and cultured for 2.0 additional hours. The cultures were then harvested.

Aberrations Assay With Metabolic Activation Cultures were initiated by seeding approximately 0.3×10^6 cells per 25 cm² flask into 5 mL of complete McCoy's 5a medium (Trial I) or initiated by seeding approximately 1.2×10^6 cells per 75 cm² flask into 10 mL of complete McCoy's 5a medium (Trial IA). One day after culture initiation, the cultures were incubated at 37°C for 3.0 hours in the presence of the test article and the S9 reaction mixture in McCoy's 5a medium without FBS. The cells were then washed twice with buffered saline and the cells were refeed with complete McCoy's 5a medium. The cells were incubated for the rest of the culture period up to the time of harvest with 0.1 µg/mL Colcemid present during the last 2.0 hours of incubation. The cultures were then harvested.

Harvest Procedure. Prior to the harvest of the cultures, visual observations of cytotoxicity were made. These observations included an assessment of the percent confluence of the cell monolayer within the culture flasks. The cultures were also evaluated for the presence of mitotic (large rounded cells) or dead cells floating in the medium. The cultures were then trypsinized and centrifuged to collect mitotic and interphase cells and a sample of cells were taken for evaluating the number of cells/mL (Trial IA and II only). The cell pellet was resuspended and swollen with 75 mM KCl, fixed in methanol: glacial acetic acid (3:1, v/v) fixative.

One hundred cells, if possible, from each replicate culture from at least four concentrations of the test articles and controls. At least 25 cells were analyzed from those cultures that had greater than 25% of cells with one or more aberrations. Mitotic index was evaluated from the vehicle control and a range of concentrations by analyzing the number of mitotic cells in 1000 cells and the ratio expressed as a percentage of mitotic cells. Percent polyploidy and endoreduplication were also analyzed by evaluating 100 metaphases, if available, and tabulated.

Results

Study validity: The study used an acceptable number of replicates; positive controls were acceptable; methodology and dose selection for RS-albuterol were acceptable, and acceptable criteria for a positive response (a test article was considered positive for inducing chromosomal aberrations if a significant increase (the difference was considered significant when $p < 0.01$) in the number of cells with chromosomal aberrations is observed at one or more concentrations. Statistical evaluation of the percentage of cells with more than one aberration provided an indication of the severity of the positive response observed. The linear trend test evaluated the dose responsiveness. A dose-response should be observed if a significant increase was seen at one or more concentrations.).

The testes for R- and S-albuterol were not valid since the maximum concentrations tested did not achieve acceptable toxicity or the maximum concentration recommended for the test. Also, no confirmatory test was done for S-albuterol.

Positive and vehicle controls produced expected results.

Study outcome:

Test articles were soluble at concentrations up to 5000 µg/ml.

In the initial chromosomal aberrations assays, the treatment period was for 3.0 hours with and without metabolic activation. Two trials were actually performed in which 5 mL cultures (Trial I) and 10 mL cultures (Trial IA) were used. For unknown reasons, the use of 5 mL cultures led to highly variable toxicity measurements, and this variability was eliminated by repeating the test conditions using 10 mL cultures. The cultures were harvested 20.0 hours from the initiation of treatment. In the confirmatory chromosomal aberrations assay, only 10 mL cultures were used (Trial II). The treatment period was for 17.5 hours without metabolic activation and all cultures were harvested 20.0 hours from the initiation of treatment.

(R)-albuterol

In Trial I (5.0 mL cultures), cultures treated with concentrations of 966, 1380, 1970, and 2810 µg/mL without metabolic activation and 338, 676, 966, 1380, 1970, and 2810 µg/mL with metabolic activation were analyzed for chromosomal aberrations. Culture with metabolic activation exhibited slightly unhealthy monolayers and a reduction in mitotic cells at the 4 highest concentrations. A significant increase in cells with chromosomal aberrations with no dose-relationship was observed only in the cultures treated with 966, 1970, and 2810 µg/mL (% cells with chromosomal aberrations 18, 13 and 7.5%, respectively vs 1.5% in vehicle control) with metabolic activation. In neither assay was adequate toxicity achieved at the highest concentrations tested. Also, the high concentration was not the maximum recommended (5000 µg/ml).

In a repeat assay using 10 mL cultures, concentrations of 168-2800 µg/mL were tested with and without metabolic activation using a 3 hr treatment (Trial IA). No visible signs of toxicity were noted. Cultures treated with concentrations of 959, 1370, 1960, and 2800 mcg/mL with and without metabolic activation were analyzed for chromosomal aberrations. No significant increase in cells with chromosomal aberrations was observed in the cultures analyzed, showing that the increase observed in Trial I was not repeatable.

In a confirmatory assay using 17.5 hr treatment and 10 mL cultures (Trial II), concentrations of 31.3- 2800 µg/mL were tested without metabolic activation. Unhealthy monolayers and a severe reduction in visible mitotic cells were observed in the cultures treated with 2400 and 2800 µg/mL. Slightly unhealthy monolayers and a reduction in visible mitotic cells were observed at 2000 µg/mL. A slight reduction in visible mitotic cells were observed in the cultures treated with 1000 and 1500 µg/mL. Reductions of 11%, 23%, 63%, 82%, and 89% were observed in the mitotic indices of the cultures treated with 1000, 1500, 2000, 2400, and 2800 g/mL, respectively. Cultures treated with concentrations of 500,

100, 1500, and 2000 µg/mL were analyzed for chromosomal aberrations. No significant increase in cells with chromosomal aberrations was observed in the cultures analyzed.

(S)-albuterol

Using 5 mL cultures (Trial I), concentrations of 0.660-2800 µg/mL were tested without metabolic activation and 0.555-2800 µg/mL were tested with metabolic activation after 3 hr treatments. Without activation, reductions in mitotic indices of up to 42% were observed at the 3 highest concentrations; reductions up to 30% (not dose-related) were noted with activation. Cultures treated with concentrations at 959, 1370, 1960, and 2800 µg/mL without metabolic activation and 676, 966, 1380, 1970, and 2810 µg/mL with metabolic activation demonstrated no significant increase chromosomal aberrations. As with R-albuterol, insufficient cell toxicity was noted at the highest dose tested which was below the maximum recommended dose.

(RS)-albuterol

Concentrations of 1.18, 2.35, 4.70, 9.40, 18.8, 37.5, 75.0, 150, 300, 600, 1200, 1720, 2460, 3520, and 5030 µg/mL were tested without metabolic activation and 1.18, 2.35, 4.70, 9.40, 18.8, 37.5, 75.0, 150, 300, 600, 1200, 1720, 2460, 3510, and 5020 µg/mL were tested with metabolic activation (Trial I; in 5.0 mL cultures; 3 hr treatment). Without activation, very unhealthy monolayer and no visible mitotic cells were observed at 3520 and 5030 µg/ml. With activation, mitotic indices were up to 49% at the highest concentrations. Cultures treated with concentrations of 600, 1200, 1720, and 2460 µg/mL without metabolic activation and 1720, 2460, 3510, and 5020 µg/mL with metabolic activation were analyzed for chromosomal aberrations. A significant increase in the % cells with chromosomal aberrations was observed only in one of the cultures treated with 3510 (24) and 5020 µg/mL (20.8) with metabolic activation, while the duplicate culture had no evidence of a significant increase (4 and 0, respectively). The respective mean values were 12.6 and 15.8%. The vehicle control had a mean % of 1.5.

In a repeat assay using 10 mL cultures, concentrations of 300, 600, 1200, 1720, 2450, 3500, and 5000 µg/mL were tested with and without metabolic activation (Trial IA, 3 hr treatment). No significant cellular toxicity was observed. Cultures treated with concentrations of 1720, 2450, 3500, and 5000 µg/mL with and without metabolic activation were analyzed for chromosomal aberrations. No significant increase in cells with chromosomal aberrations was observed in the cultures analyzed.

In a confirmatory assay using 10 mL cultures (Trial II, 17.5 hr treatment), concentrations of 31.3, 62.5, 125, 250, 500, 1000, 1500, 2000, 2500, 3750, and 5000 µg/mL were tested without metabolic activation. Significant cellular toxicity was noted at the 4 highest concentrations (mitotic indices reduced by 73-86%). Cultures treated with concentrations of 500, 1000, 1500, and 2000 µg/mL were analyzed for chromosomal aberrations. No significant increase in cells with chromosomal aberrations was observed in the cultures analyzed.

The study shows that under the conditions tested, RS-albuterol was negative in the chromosome aberration assay for clastogenic potential. R-albuterol was negative up to the concentrations tested but the study was not valid as dosing did not achieve the maximum recommended concentration. The test for S-albuterol was also not valid due to inadequate dosing and the lack of a confirmatory assay. According to the study report, the test articles, (R)-albuterol, (S)-albuterol, and (RS)-albuterol, were considered negative for inducing chromosomal aberrations in CHO cells with and without metabolic activation. However, the assay is not considered to be valid for R-and S-albuterol because the highest target concentration tested in the assay was 2810 µg/mL for (R) and (S)-albuterol. The assay for racemic albuterol is considered to be valid. The maximum concentration tested was 5030 µg/mL for this assay, the highest concentration recommended for this assay.

Study title: In Vivo Mouse Micronucleus Assay**Key findings:**

The test articles, (R)-albuterol, (S)-albuterol and (RS)-albuterol, did not induce a statistically significant increase in the frequency of micronucleated PCEs and are considered negative in the mouse bone marrow micronucleus assay under the conditions of this assay.

Study no.: 051-823

Volume #, and page #: Electronic Submission

Conducting laboratory and location: _____

Date of study initiation: August 21, 2000

GLP compliance: Yes

QA reports: yes (x) no ()

Drug, lot #, and % purity: R-albuterol/004 0008/not reported; S-albuterol/080698A/not reported; and racemic albuterol/245764/ _____

Methods

Strains/species/cell line: Male and female mice of the _____ :CD-1. Due to lack of gender specific toxicity in the range finding assay, only males were tested in the definitive assay.

Doses used in definitive study:

Dosing Scheme for Micronucleus Assay					
Target Treatment (mg/kg)	Route of Administration	Dosing Volume (mL/kg)	Males/ Harvest Timepoint		Replac- ment Males ^a
			24-Hour	48-Hour	
Vehicle Control, 0.5% HPMC	Oral gavage	10	6	6	-
Positive Control, Cyclophosphamide, 80.0	Oral gavage	10	6	-	-
500	Oral gavage	10	6	-	-
1000 (R)-albuterol	Oral gavage	10	6	-	-
2000	Oral gavage	10	6	6	3
500	Oral gavage	10	4	-	-
1000 (S)-albuterol	Oral gavage	10	4	-	-
2000	Oral gavage	10	4	4	-
500	Oral gavage	10	6	-	-
1000 (RS)-albuterol	Oral gavage	10	6	6	-
2000	Oral gavage	10	6	6	3

^a The animals in the secondary group were dosed as potential replacements for the original high-dose group. Animals not used as replacements were euthanized at the completion of the trial.

Basis of dose selection: Dose ranging study in 3 mice/sex/group using 500, 1000 and 2000 mg/kg, PO (gavage).

Negative controls: Vehicle control, 0.5% hydroxyl propylmethylcellulose (HPMC); 6 males tested

Positive controls: Cyclophosphamide-8 mg/mL; 6 males tested

Incubation and sampling times:

Sampling times were approximately 24 and 48 hours after dosing. Marrow was flushed from the tibia and slides were prepared and scored for the PCE to NCE ratio. The micronucleus frequency was determined by the number of micronucleated PCEs from at least 2000 PCEs per animal. The frequency of PCE:NCE ratio was determined by scoring the number of PCEs and NCEs observed in the optic fields while scoring at least the first 500 erythrocytes on the slide.

Results

Study validity:

The study was considered valid as dose selection and methodology were acceptable. The appropriate numbers of cells were assessed for PCEs. The positive and negative controls produced the expected results. The criteria for a positive response (detection of a statistically significant increase in micronucleated PCEs for at least one dose level, and a statistically significant dose-related response; a test article that did not induce both of these responses was considered negative. Statistical significance was not the only determinant of a positive response; the Study Director also considered the biological relevance of the results in the final evaluation) were also acceptable.

Study outcome:

Range finding assay: 3/6 animals administered R-albuterol demonstrated labored respiration after dosing while S-albuterol also induced hyperactivity and signs of rough coat. Similar findings were note with RS-albuterol with the addition of squinting eyes. Based on these findings, a high dose of 2000 mg/kg was considered tolerable in the definitive study.

Definitive assay:

Mortality was noted in one secondary 2000 mg/kg (R)-albuterol-treated animal ~ 1 day after dosing. All (S)-albuterol and (RS)-albuterol-treated animals survived until their assigned harvest timepoint. Clinical signs were similar to those observed in the range finding study.

The test articles, (R)-abuterol, (S)-albuterol and (RS)-albuterol, induced no statistically significant decrease in the PCE:NCE ratio, demonstrating that the test article was not cytotoxic to the bone marrow. (R)-abuterol (S)-albuterol, and (RS)-albuterol did not induce a statistically significant increase in the frequency of micronucleated PCEs and are considered negative in the mouse bone marrow micronucleus assay under the conditions of this assay. This conclusion is in agreement with that of the sponsor.

2.6.6.5 Carcinogenicity:

Carcinogenicity studies have not been conducted with levalbuterol. Carcinogenicity studies have been conducted with racemic albuterol. The results as described in the package insert for Proventil are shown below:

Albuterol sulfate was not carcinogenic in an 18 month oral (diet) carcinogenicity study in CD1 mice. The doses were 0, 50, 150 and 500 mg/kg. In a 2 year oral (feeding) study in the SD rat, albuterol sulfate at doses of 0, 2, 10 and 50 mg/kg induced significant, dose-related increases in smooth muscle tumors, leiomyomas, in the mesovarium at all doses. This effect was blocked by the coadministration of propranolol, a nonselective beta-adrenergic antagonist. A 99 week (696 days) oral oncogenic study was carried out in the Golden hamster. The albuterol sulfate doses (dietary) were 10 and 50 mg/kg. There was no tumorigenicity in this study.

2.6.6.6 Reproductive and developmental toxicology

No new studies were conducted for this NDA. The following section describes the findings summarized in the approved Xopenex package insert.

Studies to assess fertility have not been performed with levalbuterol. Reproduction studies in rats using racemic albuterol sulfate demonstrated no evidence of impaired fertility at oral doses up to 50 mg/kg.

A maternal and fetal developmental study in rabbits was performed with R- albuterol at oral gavage doses of 0.5, 5, and 25 mg/kg/day. There were no maternal or fetal developmental toxicities.

Segment II studies were conducted with racemic albuterol in rabbits and mice. A segment II study was carried out in the Wistar rat using oral albuterol doses of 0.0.5, 2.32, 10.75 and 50 mg/kg, days 1-19 of pregnancy. No teratogenic effects were observed. Segment II studies in the Stride Dutch rabbit demonstrated cranioschisis following oral doses of 50 mg/kg. Subcutaneous injection of racemic albuterol to CD-1 mated mice resulted in cleft palate at doses of 0.25 mg/kg or greater; no effects were observed at 0.025 mg/kg. Cleft palate also occurred in 22 of 72 (30.5%) fetuses from females treated subcutaneously with 2.5 mg/kg of isoproterenol (positive control).

Based upon the results of the aforementioned studies, albuterol sulfate has been given a Pregnancy Category C designation.

2.6.6.7 Local tolerance

No studies were conducted for this NDA.

2.6.6.8 Special toxicology studies

Study title: Acute Dermal Irritation Study of Levalbuterol and _____ in Albino Rabbits

Key study findings: There were no deaths in the study. Levalbuterol induced slight erythema on three rabbits (3/6) which completely subsided within 72 hours. There were no other dermal findings. The Primary Irritation Indices for levalbuterol and _____ were calculated as 0.2 and 0.0, respectively.

Study no.: 051-820

Volume #, and page #: Electronic submission

Conducting laboratory and location: _____

Date of study initiation: May 5, 1998

GLP compliance: Yes

QA reports: yes (x) no ()

Drug, lot #, and % purity: Levalbuterol/003 0002/ _____ and _____ /H40761/ _____

Formulation/vehicle: Levalbuterol/deionized water (0.2 ml) and _____ and deionized water (0.6 ml)/ deionized water

Methods

Doses: 0.5 g

Study design:

On the day prior to dosing, the hair was removed from the backs and flanks of the rabbits using an — small animal clipper. Each 0.5-g dose was applied to an area of skin approximately 2.5 cm x 2.5 cm under a secured two-ply gauze patch that was overwrapped with a gauze binder and secured with Dermiform® tape for the three-minute, one-hour and four-hour semi-occluded exposures. Approximately 0.2 and 0.6 ml of deionized water was added for moistening Levalbuterol and — respectively, to form a paste. Plastic restraint collars were applied to prevent ingestion of the test material and/or wrappings for the one-hour and four-hour exposure periods. At the end of the exposures, the collars (if applicable) and bandages of each rabbit were removed and the exposure sites were wiped with disposable paper towels moistened with deionized water.

Six rabbits, New Zealand white, young adults weighing 3588-4235 gram were used for each compound

Results: There were no deaths in the study. Levalbuterol induced slight erythema on three rabbits which completely subsided within 72 hours. There were no other dermal findings. There were no remarkable body weight changes in the study. The Primary Irritation Indices for levalbuterol and — were 0.2 and 0.0, respectively.

Study title: Acute Eye Irritation Study of Levalbuterol and — in Albino Rabbits

Key study findings: There were no deaths or significant body weight changes in the rabbits treated with the experimental compounds. Levalbuterol induced redness and swelling in the eye while — caused moderate corneal opacities, iridal irritation and conjunctival redness and swelling. The results suggested that levalbuterol was a mild ocular irritant.

Study no.: 051-821

Volume #, and page #: Electronic submission

Conducting laboratory and location: —

Date of study initiation: May 11, 1998

GLP compliance: Yes

QA reports: yes (x) no ()

Drug, lot #, and % purity: Levalbuterol/003 0002/ — and — H40761/

Formulation/vehicle: Levalbuterol and — powder

Methods and Study design:

The test materials were dosed based on their weight equivalent volume to 0.1 ml. Three one-milliliter allotments of each test material were weighed and the mean divided by 10 to derive the weight (gently packed down to remove any possible air pockets) equivalent volume of 0.1 ml. It was calculated that 41 mg Levalbuterol and 37 mg of — were equivalent to 0.1 ml. Individual 41-mg or 37-mg doses of the test materials were weighed on tared weigh papers that were folded closed and transported to the animal room for dosing.

Route and Rationale of Test Material Administration:

The route of test material administration was direct conjunctival instillation. This route is standard for assessment of local ocular irritative potential.

Method of Test Material Administration:

The test materials were placed directly into the cupped lower conjunctival sac of the rabbit's right (test) eye. The eyelid was held closed for approximately one second after instillation. The left eye was manipulated in an identical manner to simulate the dosing of the right eye.

Dose Level(s)/Group(s)/Treatment Regimen:

The dose was 41 mg Levalbuterol/right eye for Group 1 and 37 mg — right eye for Group 2. There were six rabbits in each group. Each rabbit received a single, unwashed exposure.

There were 6 young female New Zealand white rabbits in each dose group. The eyes were examined for ocular reactions with the method of Draize at approximately 1, 24, 48, and 72 hours.

Doses: Single doses levalbuterol- 41 mg and — 37 mg

Results: There were no deaths or significant body weight changes. Levalbuterol induced redness (6/6) and swelling (4/6) in the eye while — caused moderate corneal opacities (2/6), iridal irritation (3/6), conjunctival redness (6/6) and swelling (6/6). The group mean scores for corneal opacities, iritis, conjunctival redness and swelling for levalbuterol were 0, 0, 1.2, and 0.3 indicating that levalbuterol was a mild ocular irritant.

Summary of the Special Studies:

An acute dermal irritation study of levalbuterol and — and an acute eye irritation study of levalbuterol and — studies were conducted in albino rabbits. The dose applied in the dermal test was 0.5 gram. The results of the study show that levalbuterol induced slight erythema in three rabbits which completely subsided within 72 hours. The primary irritation indices for levalbuterol and — were 0.2 and 0.0, respectively. In the eye irritation test, single doses were 41 mg for levalbuterol and 37 mg for — Levalbuterol induced redness and swelling in

the eye while caused moderate corneal opacities, iridal irritation and conjunctival redness and swelling. The group mean scores for corneal opacities, iritis, conjunctival redness and swelling for levalbuterol were 0, 0, 1.2, and 0.3 indicating that levalbuterol was a mild ocular irritant.

2.6.7 TOXICOLOGY TABULATED SUMMARY

There was no toxicology tabulated summary provided by the sponsor.

OVERALL CONCLUSIONS AND RECOMMENDATIONS

The submission is a NDA for Xopenex HFA (levalbuterol tartrate HFA) Inhalation Aerosol for the treatment or prevention of bronchospasm in adults, adolescents and children, 4 years of age and older with reversible obstructive airway disease. Levalbuterol is the R-isomer of the commonly used beta 2 adrenergic agonist, racemic albuterol. Levalbuterol has been widely used since the approval of Xopenex Inhalation Solution (NDA 20-837) on March 25, 1999.

There are no non-clinical issues for Xopenex HFA. The NDA is referencing data from other NDAs as follows:

The following data are being incorporated by reference, based on prior agreement with the Agency: (1) The long-term clinical safety data for Proventil[®] HFA Inhalation Aerosol, provided to FDA in NDA 20-503, are incorporated into this application based on a direct right-of-reference for this purpose granted by 3M Pharmaceuticals; and (2) all relevant toxicology, human safety, human efficacy, and other data provided to FDA in NDA 20-837 (submitted and owned by Sepracor Inc.) for Xopenex[®] Inhalation Solution are incorporated by reference into this application. Additionally, carcinogenicity data for racemic albuterol provided to FDA in NDA 19-243 for Proventil[®] Inhalation Solution are relied upon under 21 USC 355(b)(2) based on a prior agreement with the Agency.

In addition, the sponsor submitted a number of pharmacology, ADME/pharmacokinetic studies, a 90-day toxicology study in dogs for the purpose of leachables qualification, as well as two special toxicity studies and two genotoxicity studies. The pharmacology studies indicated that the activity of albuterol is primarily related to the presence of R-albuterol. The ADME studies demonstrated minimal distribution of albuterol to brain tissue and relatively low protein binding. The 90-day dog toxicology study confirmed the previously demonstrated toxicity profile which is largely cardiovascular. The genotoxicity studies include an in vivo mouse micronucleus assay and a chromosomal aberration in Chinese hamster ovary (CHO) cells and both produced negative results. The chromosomal aberration assay, however, is not considered to be valid because the highest target concentration tested in the assay was inadequate for R- and S-albuterol; only 2810 µg/mL for (R)-albuterol and (S)-albuterol with no significant cytotoxicity observed at the 3 hour assessment. The assay for racemic albuterol is considered to be valid. The maximum concentration tested was 5030 µg/mL for this assay, the highest concentration

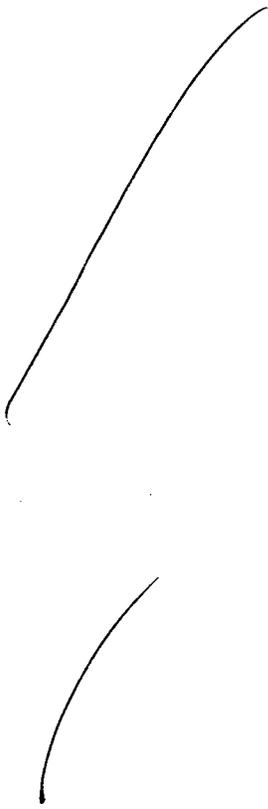
recommended for this assay.

In conclusion, the sponsor has provided adequate nonclinical safety data via studies submitted to this NDA, studies reviewed under NDA 20-837 or via reference to non-clinical data in package inserts for approved products to assess the safety of Xopenex HFA. The toxicology of levalbuterol was adequately characterized under NDA 20-837 and the sponsor has right of reference to information related to the safety of the HFA propellant. An additional 3-month study in dogs with the HFA formulation did not identify any unexoected adverse findings. Thus, the proposed use of Xopenex HFA in this NDA is approvable from a non-clinical perspective.

Unresolved toxicology issues (if any): None

Recommendations: The NDA is approvable from a non-clinical perspective pending the recommended revised labeling.

Suggested labeling:



3 Page(s) Withheld

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§ 552(b)(5) Deliberative Process

§ 552(b)(4) Draft Labeling

APPENDIX/ATTACHMENTS

Drug:		Xopenex HFA						
	age	mg/dose	# daily doses	mg/day	kg	mg/kg	factor	mg/m ²
Pediatric dose	4	0.045	12	0.54	16	0.03	25	0.84
Adult dose	>12	0.045	12	0.54	50	0.01	37	0.40

	route	mg/kg/day	factor	mg/m ²	Dose Ratio		Rounded Dose Ratio	
					Adults	Children	Adults	Children
<u>Carcinogenicity:</u>								
rat	dietary	2	6	12	30.03	14.22	30	15
mouse	dietary	500	3	1500	3753.75	1777.78	3800	1800
hamster	dietary	50	4	200	500.50	237.04	500	240
extra			---	---	---	---	---	---
extra			---	---	---	---	---	---
<u>Reproduction and Fertility:</u>								
rat	oral	50	6	300	750.75	N/A	750	N/A
extra			---	---	---	N/A	---	N/A
extra			---	---	---	N/A	---	N/A
extra			---	---	---	N/A	---	N/A
<u>Teratogenicity:</u>								
rabbit	oral	25	12	300	750.75	N/A	750	N/A
mouse	SC	0.25	3	0.75	1.88	N/A	2	N/A
mouse	SC	2.5	3	7.5	18.77	N/A	20	N/A
mouse	SC	0.025	3	0.075	0.19	N/A	1/5	N/A
rabbit	oral	50	12	600	1501.50	N/A	1500	N/A
<u>Overdosage:</u>								
mouse	IV	66	3	198	495.50	234.67	500	230
rat	IV	60	6	360	900.90	426.67	900	430
dog	IH	2.73	20	54.6	136.64	64.71	140	65
extra			---	---	---	---	---	---
<u>Other:</u>								
extra	teratogenicity		---	---	---	---	---	---
extra			---	---	---	---	---	---
extra			---	---	---	---	---	---
extra			---	---	---	---	---	---
extra			---	---	---	---	---	---

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/s/

Virgil Whitehurst
2/23/05 01:47:53 PM
PHARMACOLOGIST

Timothy McGovern
2/23/05 01:53:30 PM
PHARMACOLOGIST
I concur.

2.6 PHARMACOLOGY/TOXICOLOGY REVIEW

2.6.1 INTRODUCTION AND DRUG HISTORY

NDA number: NDA 21-730

Chemistry consult # 1

Sequence number/date/type of submission: 000/May 11, 2004/original NDA submission

Information to sponsor: Yes (x) No ()

Sponsor and/or agent: Sepracor Inc

Manufacturer for drug substance: 3 M Pharmaceuticals, 19901 Nordhoff Street, Northridge, CA 91324-3298

Reviewer name: Virgil Whitehurst, Ph.D.

Division name: Division of Pulmonary and Allergy Drug Products

HFD #: HFD 570

Review completion date: February 15, 2005

Drug:

Trade name: Xopenex™ (levalbuterol tartrate) HFA Inhalation Solution

Generic name: Levalbuterol tartrate

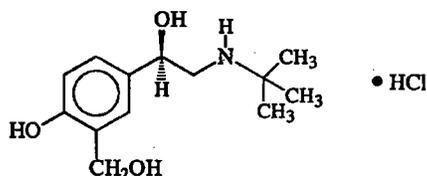
Code name: NA

Chemical name: (R)- α 1-[[[(1,1-dimethylethyl)amino]methyl]-4-hydroxy-1,3-benzenedimethanol hydrochloride

CAS registry number: 50293-90-8

Molecular formula/molecular weight:

Structure:



Relevant INDs/NDAs/DMFs: IND 47,363, Levalbuterol HCl Inhalation Solution;
NDA 20-837, Xopenex (levalbuterol HCl) Inhalation Solution; DMF —
DMF — DMF — , DMF — DMF —

Drug class: Beta adrenergic agonist

Intended clinical population: Treatment or prevention of acute bronchospasm in patients with reversible obstructive airway disease (asthma) in adults and children 4 year and older.

Clinical formulation:

Components	Function	Amount/Actuation	Amount/Canister
Levalbuterol tartrate	Active ingredients		
Oleic acid			
Dehydrated alcohol			
HFA-134a	Propellant		
Total	-----	60 mg	15.00 g

* equivalent to — of levalbuterol free base (ex-valve), to deliver 45 µg levalbuterol free base/59 µg of levalbuterol tartrate (ex-actuator)/actuation. Each canister provides 200 inhalations.

Route of administration: Oral inhalation- the maximum recommended daily dose is 2 actuations (45 µg/actuation) every 4 to 6 hours (maximum of 12 actuations per day and 540 µg/day) or 10.8 µg/kg for a 50 kg person or 33.8 µg/kg for a 16 kg person 4 years of age.

Introduction and History:

This is a chemistry consult in response to consult requests from Dr. Suong Tran (written requests dated July 23 and September 14, and a verbal request of December 21, 2004) to evaluate the safety of the proposed acceptance criteria for leachables in the drug product specification and the safety of the extractables in the container-closure system specification. Also, Dr. Tran wanted an evaluation of the safety of the proposed acceptance criteria for particulate matter in the drug product. The sponsor stated that the acceptance criteria for leachables are based on release and stability data, characterization studies described in section 10.2.22 and toxicological considerations (the sponsor submitted a report entitled “Assessment of potential risks associated with exposures to extractable and leachable compounds from administration of levalbuterol via metered dose inhaler”; report # 051-826 produced by —). The sponsor also submitted a 13-week study titled “Xopenex HFA Pressurised Metered Dose Inhaler: 13 Week Inhalation Qualification Study in the Dog” to assist in the qualification of certain materials. This study is reviewed formally in the original NDA review and is summarized in this consult.

Listed below are the leachables in the drug product with their associated proposed specification, the acceptable daily intake (ADI) based upon this review, and a determination as to whether or not the compound has been adequately qualified.

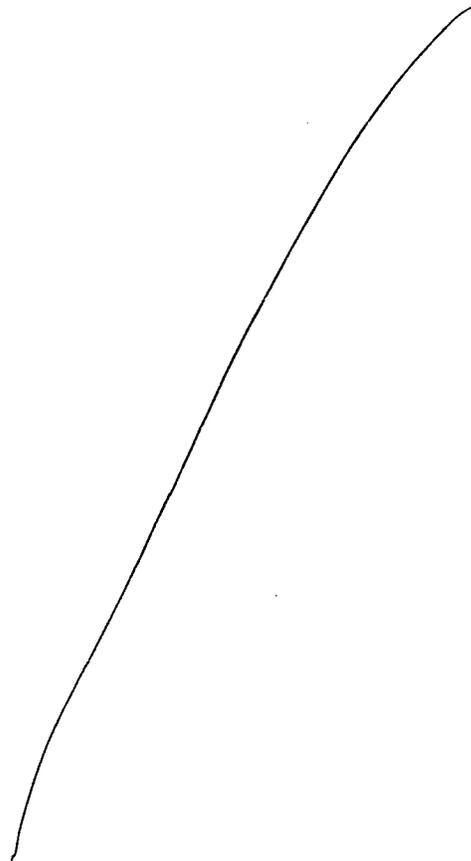
Leachables in Xopenex Inhalation Solution			
Leachable, Proposed Specification	Max Daily Exposure (µg/kg)*	ADI (µg/kg)	Qualified
/	/	/	Yes
			Yes

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§ 552(b)(5) Deliberative Process

§ 552(b)(4) Draft Labeling



Conclusion on leachables:

The specifications for _____

are qualified based either on the Division's safety qualification threshold of _____ g/kg (or _____ day), which is based on an assessment of the US EPA HEAST data base in which no systemic toxicities were observed for compounds at exposure levels below that stated, or based on other animal and human toxicity/exposure study data.

The specifications for _____ are not qualified and the sponsor should lower their proposed specifications or provide adequate qualification data to support the proposed specifications. In the absence of adequate qualification data, the specifications for _____ should be lowered to approximately _____ can (_____ day) due to data suggestive of genotoxic potential of these compounds. Similarly, the specification for _____ should be reduced to _____ can (_____ day) due to a lack of adequate qualification data.

Particulate Matter:

The sponsor's proposed specifications for particulate matter is as follows:

The maximum allowable level for particulate matter is $0.1 \mu\text{g/day}$ (reference: NDA 21-433 –Fluticasone Aerosol- Chemistry consult dated April 16, 2004). The acceptance level of $0.1 \mu\text{g/day}$ is based on a USEPA particulate matter (PM_{10}) standard of $50 \mu\text{g}/\text{m}^3$ and an assumed 20 m^3 of air breathed over a 24 hour period.

The focus of this evaluation is on the number of particles $<10 \mu\text{m}$ since they are the respirable particles. In determining the weight of the daily exposure, it is assumed that the particles are spherical. The volume of 1 particle is determined from the formula for a sphere: Volume of sphere: $\frac{4}{3}\pi r^3$. To achieve this and the weight of the particles, it is assumed that 100% of the particles were $10 \mu\text{m}$ in diameter and their density is $1 \text{ g}/\text{cm}^3$.

Conclusion: The proposed acceptance criteria which equates to $0.1 \mu\text{g/day}$ for particulate matter exposure is acceptable since it is significantly below (approximately $1/500$) the EPA PM_{10} standard.

Related Extractables from the Canister:

The table below summarizes the sponsor's proposed criteria for extractables in the container closure system:

Related Extractables From the Canister				
Related Extractables***	Proposed Criteria (µg/canister)	Maximum Daily Exposure (µg/kg)	ADI** (µg/kg)	Qualified
				Yes
				No
				Yes
				No
				Yes

Reviewer Assessment:

All but 3 of the above listed extractables have not been structurally identified. Thus, it is not possible to determine the presence of structural alerts for genotoxic/carcinogenic potential. Assuming that there is no structural alert present, the proposed specifications for the unidentified extractables from the canister to be used for Xopenex are acceptable. The acceptance of these extractables is based on the Division's assessment of the US EPA HEAST data base and determination of a safety qualification threshold of — /kg or — day. Dr Tran did not mention that any of these extractables had a structural alert. In addition, the proposed specification for — is acceptable as the maximum expected human exposure is below the determined acceptable inhalation dose (see assessment of leachables above).

The proposed specifications for — are not acceptable due to data indicating the potential for genotoxic/carcinogenic potential (see assessment of leachables above). Based on the positive findings in multiple genotoxicity assays, and

According to current Division practices, the general acceptable qualification threshold of 10^{-4} day is not applicable for the compound. Therefore, the sponsor should lower the specification to that which will result in a maximum possible human inhalation exposure of 10^{-4} g/day or less (10^{-4} /can) or provide adequate qualification information to demonstrate that the Division's concern regarding the genotoxic/carcinogenic is unwarranted. If this concern is alleviated, the sponsor's proposed specification of 10^{-4} can would be acceptable as it would result in a maximum daily exposure of $< 10^{-4}$ μ g/day, the Division's qualification threshold for compounds with no structural alerts.

Related Extractables From The Actuator/Dust Cap

Related Extractables From the Actuator/Dust Cap				
Related Extractables	Proposed Criteria (μ g/canister)	Maximum Daily Exposure (μ g/kg)	ADI (μ g/kg)	Qualified
				Yes

Reviewer Assessment:

All of the proposed specifications for the extractables from the actuator/dust cap to be used for Xopenex are acceptable. The specs for compounds " " are Ok as they are below the Div safety TH. The acceptance of these extractables is based on the Division's assessment of the US EPA HEAST data base and the determination of a safety qualification threshold of 10^{-4} /kg or 10^{-4} day for compounds with no structural alerts for genotoxic and/or irritation potential.

OVERALL CONCLUSIONS AND RECOMMENDATIONS

This is a chemistry consult based on requests by Dr. Suong Tran to evaluate the safety of the proposed acceptance criteria for leachables and particles in the drug product specification and the safety of the extractables in the container-closure system specification. Also Dr. Tran wanted an evaluation the safety of the proposed acceptance criteria for particulate matter in the drug product. The sponsor submitted a review of the available toxicity data on the copmpounds based on published literature as well as the

results of a 13 week toxicology and toxicokinetic studies in the dog using an aged Xopenex formulation.

Conclusions:

The proposed specifications for _____ are qualified based either on the Division's safety qualification threshold of _____ g/kg (or _____ /day) or on other animal toxicity data or other human exposure data.

The proposed specifications for _____ are not acceptable. Available data indicates that _____ have genotoxic potential. Therefore, in the absence of adequate data to refute the genotoxic or carcinogenic potential of these compounds, the specifications should be reduced to a level which would result in a daily exposure < _____ µg/day. A specification of _____ can should achieve this level of daily exposure.

The specification for _____ should be reduced to a level that would result in a maximum daily exposure of _____ day since there is no adequate data to support the safety of this compound. A specification of _____ can should achieve this level of daily exposure. Alternately, the sponsor can conduct a study (3 months in duration via the inhalation route) to support their proposed specification.

The proposed acceptance criteria for particle matter exposure is acceptable.

The proposed specifications for the extractables from the canister to be used for Xopenex are acceptable with the exception of _____ for the reasons stated above. Limiting the level of the leachables as recommended above would control the patient exposure to these compounds.

Additionally, the proposed specifications for the extractables from the actuator/dust cap to be used for Xopenex are acceptable.

Reviewer Signature _____

Supervisor Signature _____ Concurrence Yes ___ No ___

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/s/

Virgil Whitehurst
2/17/05 08:35:31 AM
PHARMACOLOGIST

Timothy McGovern
2/17/05 10:15:57 AM
PHARMACOLOGIST
I concur.