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Loxapine Delivered as a Thermally Generated Aerosol Does Not Prolong QTc in a Thorough QT/QTc Study in Healthy Subjects

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Abstract

The objective of this study was to establish effects of inhaled loxapine on the QTc interval in this randomized, placebo-controlled, double-blind crossover study. Forty-eight healthy volunteers received a single inhaled placebo or 10 mg loxapine. Plasma concentrations of loxapine increased with a median Tmax of 1 minute and a mean Cmax of 312 ng/mL. After an initial rapid distribution phase, plasma concentrations of loxapine declined with a terminal half-life of 8 hours. Exposure to the active metabolite 7-OH-loxapine was 15% of the parent compound based on mean AUC_{inf} and its terminal half-life was 12 hours. Inhaled loxapine did not increase QT intervals, as demonstrated by the upper bound of the 1-sided 95% CIs placed on the point estimate of the placebo-subtracted change of QTcI ($\Delta\Delta$QTcI) being less than 10 milliseconds at all 11 post-dose times. The maximum $\Delta\Delta$QTcI occurred at 1 hour post-dose (LSmean 5.42 milliseconds, upper confidence bound 7.75 milliseconds). The study outcome was validated by the demonstrated assay sensitivity using the positive control moxifloxacin maximum $\Delta\Delta$QTcI occurred at 3 hour post-dose (LSmean 8.36 milliseconds, lower confidence bound 5.82 milliseconds). The analyses of QTc outliers, and the lack of emergent diagnostic findings for QTcI, QTcB, and QTcF; and simple mean placebo-subtracted changes of QTcI and QTcF supported the primary QT analysis conclusion that this is a negative finding and there is no apparent QT prolongation associated with the therapeutic dose of inhaled loxapine.

Keywords

inhalation delivery, loxapine, thorough QT/QTc, pharmacokinetics, pharmacodynamics

Loxapine, which was introduced more than 35 years ago in the US, Canada, and Europe, has a well-established efficacy and safety profile in the treatment of schizophrenia. Its antipsychotic effects are similar to those of other antipsychotics such as haloperidol, and are likely attributable to its action at dopamine D2 receptors.[1] There is limited evidence that loxapine shares some of its clinical effects with atypical antipsychotics, such as clozapine and olanzapine,[2] due to its unique binding profile, especially its action at 5-HT2A receptors.

Staccato Loxapine (inhaled loxapine) is a hand-held, single-use drug-device combination product using Alexza's proprietary Staccato delivery system. Oral inhalation through the mouthpiece initiates the controlled rapid heating of a thin film of excipient-free loxapine to form a thermally generated, highly pure drug vapor. The vapor condenses into aerosol particles with a particle size distribution appropriate for efficient delivery to the deep lung (for details see http://www.alexza.com/products). The rapid absorption of the drug provides peak plasma levels in the systemic circulation within minutes after administration.

Although loxapine has not been characterized as a risk for causing torsades de pointes (TdP), several of the antipsychotics are considered a known or possible TdP risk. Although loxapine was approved by the FDA more than 35 years ago, a thorough QT/QTc study has not been conducted for loxapine. The agency has taken the position that a thorough QT/QTc study will be required on any approved product, and required and approved this study in support of the approval of *Staccato* Loxapine.

This study was a Phase 1, single-center, double-blind, double-dummy, active and placebo-controlled, 3 period crossover study investigating the potential of single 10 mg doses of inhaled loxapine to delay cardiac repolarization in healthy volunteers. This thorough QT/QTc study was designed in accordance with the ICH E14 guideline.[3]

The dose used is the therapeutic dose approved for a single intermittent dose. This dose was also considered as the maximum tolerated dose in the single dose escalation

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study[4] and therefore no supratherapeutic dose was assessed. In addition, the IV-like inhaled absorption kinetics and early effects are relatively unaffected by metabolizing enzymes or transporters.

Methods

Study Design

This was a double-blind, double-dummy, active- and placebo-controlled, 3 period crossover study investigating single doses of inhaled loxapine (10 mg, the approved therapeutic dose), a positive control with known QT/QTc prolongation (oral moxifloxacin, 400 mg), and oral and inhaled placebos. Treatments were designated as A (oral placebo with inhaled loxapine), B (oral placebo with inhaled placebo), and C (oral moxifloxacin with inhaled placebo). Each healthy volunteer subject was randomized to 1 of 6 sequence groups and received all 3 treatments (A, B, and C), separated by a minimum 3-day washout period. Subjects were confined to the Covance Clinical Research Unit, Evansville, IN (CRU) during each treatment period. The study was reviewed and approved by the Independent Investigational Review Board, 6738 West Sunrise Blvd., Suite 102, Plantation, FL 33313. Written informed consent was obtained before any protocol specific procedures were performed.

Eligible subjects were healthy male and female volunteers between the ages of 18 and 65 years (inclusive) and with a body mass index between 21 and 30 (inclusive); and in good general health as determined by a detailed medical history, physical examination, 12-lead ECG, blood chemistry, hematology, and urinalysis, and in the opinion of the principal investigator.

Subjects were excluded for regular consumption of >5 cups of coffee/day; taking prescription or nonprescription medication within 5 days of initial treatment; having an acute illness within the last 5 days of initial treatment; smoking tobacco within the last year or having a positive cotinine test; a history within the past 2 years of drug or alcohol dependence or abuse as defined by the Diagnostic and Statistical Manual of Mental Disorders IV; a positive test for alcohol or a positive urine drug screen; an ECG abnormality (a normal ECG was a QTc for males of 450 milliseconds or less, and for females of 470 milliseconds or less; a consistent sinus rhythm, heart rate $\leq$99 and $\geq$40 beats per minute, a PR interval between 120 and 230 milliseconds, a QRS interval $\leq$110 milliseconds, no other conduction abnormalities, and QT intervals that could be consistently analyzed; hypotension (systolic $\leq$90 mm Hg, diastolic $\leq$50 mm Hg) or hypertension (systolic $\geq$140 mm Hg, diastolic $\geq$90 mm Hg); a history of unstable angina, syncope, coronary artery disease, myocardial infarction, congestive heart failure, transient ischemic attack, neurological disorder; or a history of asthma, chronic obstructive lung disease, or any use of an inhaler prescribed for wheezing or bronchospasm; or considered by the investigator, for any reason, to be an unsuitable candidate for receiving loxapine, or unable to use the inhalation device. Planned enrollment was 48 subjects.

Study Medication

The principal components of inhaled loxapine are:

- Breath sensor: The breath-activation mechanism that initiates actuation of the heat source
- Heat source (ie, heat package): The mechanism comprised of a reactant coating on the interior surface of a stainless steel substrate that generates heat to vaporize the drug and produce the aerosol
- Drug coating: The thin film of excipient-free loxapine free base on the exterior surface of the stainless steel substrate
- Airway: The medical-grade plastic housing surrounding the heat package; it controls and directs the airflow over the vaporizing drug

When activated, the heat source undergoes a controlled, gasless, oxidation-reduction (redox) reaction that liberates heat. The redox reaction is initiated by a battery-activated starter inserted into the heat package. Inhalation through the product is detected by the breath sensor, causing the starter to initiate the redox reaction with subsequent rapid heating of the substrate to approximately 400°C. Heat then transfers into the film of loxapine that is coated on the exterior surface of the substrate. The loxapine vaporizes in $<$1 second, thereby limiting thermal decomposition. The vapor cools in the airflow and condenses to form aerosol particles that are characterized by a mass median aerodynamic diameter in the range of 1.0–3.5 μm.

Inhaled placebo systems were identical in size and operation to inhaled loxapine except that inhaled placebo contained no drug coating. Inhaled placebo was used in the same manner and produced the same heat output as inhaled loxapine.

ECG Recording

Continuous 12-lead ECG recording (Mortara H12 + Digital Holter monitor) was used to capture ECG data with triplicate ECG recordings extracted at each sampling time point. During the sampling periods, subjects were supine and did not engage in other study activities.

ECG Analysis and Interpretation

ECGs used in the analysis were selected at predetermined time points and read centrally (Cardiocore, Bethesda, MD) using a high-resolution manual on-screen calipers in the semiautomatic mode with fiducial annotations over-read

and adjusted as necessary in the treatment-blinded environment. The ECG interval measurements were conducted in the composite 12-lead superimposed global view. Morphological analyses were performed with regard to the ECG waveform interpretation as defined by the central ECG laboratory.

Primary QT Analysis. Consistent with the ICH E14 guideline,[3] for a "thorough" QT/QTc study, the primary analysis consisted of constructing 1-sided upper 95% confidence bounds for the "true mean difference" in the differences between each inhaled loxapine 10 mg dose mean and the placebo mean at each time point. The primary outcome measure for the study was the time matched difference from the pre-dose baseline at each time point in the individual subject-corrected QT interval, QTcI.

The individual correction was based on the regression of QTc versus RR interval during the baseline day preceding the first dose of study medication. All QT/RR pairs from that day were used in determining QTcI for that subject. The number of intervals per subject was 42 for 45 subjects, 40, 39 and 9 for the other 3. The primary endpoint was based on least squares mean (LSmean) corrected for sequence, period, and pre-dose baseline according to the repeated measures model. Baseline was the mean of the 3 triplicates measured within 1 hour prior to each dosing. The primary hypothesis was tested by placing a 1-sided 95% upper confidence bound on the pre-dose-corrected LSmean difference between the inhaled loxapine 10 mg dose and the corresponding time-matched pre-dose-corrected placebo mean at each ECG time point. If none of the upper confidence bounds exceeded 10 milliseconds then the null hypothesis was rejected. An analysis to test for a differential drug effect on QTcI intervals between males and females was also performed; data from moxifloxacin were excluded from this analysis.

Secondary QT Analyses. Secondary endpoints included changes from the period-specific pre-dose values for QTcB and QTcF, and in heart rate. To assess the adequacy of the individual correction, repeated measures regressions of QT and each QTc by RR were shown for each correction method using only the data from the baseline day. The secondary endpoints were analyzed using the same statistical analysis methods as described for the primary analysis. The same linear model was employed and 2-sided 90% confidence intervals were constructed at each time-point for differences between inhaled loxapine 10 mg and placebo for QTcB and QTcF intervals. Categorical analyses of QTcI, QTcF, and QTcB outliers were also undertaken as secondary analyses. Categorical analyses are based on the numbers and percentages of subjects exceeding the following 3 upper limit values ($QTc > 450$ milliseconds, $QTc > 480$ milliseconds, and $QTc > 500$ milliseconds) for absolute QTcI, QTcF, and QTcB following administration of inhaled loxapine 10 mg and inhaled placebo, In addition, changes from the period-specific pre-dose baseline in QTcI, QTcF, and QTcB intervals after administration of inhaled loxapine 10 mg and inhaled placebo were determined, and the numbers and percentages of subjects exceeding the following 2 upper limits (QTc increase from baseline >30 milliseconds and QTc increase from baseline >60 milliseconds) were tabulated. Morphological findings, not present at the Day -1 baseline, were summarized for inhaled placebo and inhaled loxapine 10 mg.

Statistical Analysis

Determination of Sample Size. Based on a within subject standard deviation (residual standard deviation) of 8 milliseconds and 12 ECG time points, a sample size of 42 subjects was calculated to provide 90% power to reject the primary hypothesis under the assumption that the true difference from placebo is no more than 3 milliseconds. To account for potential dropouts, a total of 48 subjects were to be enrolled.

Analysis Populations. All randomized subjects who received at least placebo and 1 dose of active drug (inhaled loxapine 10 mg or moxifloxacin) and who had at least 1 paired set of placebo and active drug ECG assessments were included in QTc analyses. All randomized subjects who received at least placebo and 1 dose of active drug, and who had at least 1 paired set of placebo and active drug ECG assessments and associated active drug concentrations were included in concentration-QT analyses. All randomized subjects who received at least 1 dose of inhaled loxapine were included in PK analyses. All randomized subjects who received at least 1 dose of study drug were included in overall safety analyses.

Assay Sensitivity. Consistent with the ICH E14 guideline,[3] for a "thorough" QT/QTc study, the assay sensitivity of the study design and implementation was assessed by demonstrating statistically that oral moxifloxacin 400 mg, given at a dose expected to produce a QTcI prolongation, did, in fact, produce a QT prolongation. To do this, 2-sided 90% confidence intervals were constructed around the pre-dose-subtracted QTcI difference for moxifloxacin relative to the pre-dose-subtracted QTcI difference for placebo, using the statistical model described for the analysis of the primary endpoint. Suitable assay sensitivity was concluded if 1 or more of the 4 post-dose time points (1.5, 2, 2.5, and 3 hours) lower confidence bounds were above 5 milliseconds.

Concentration-QT Analysis. A linear mixed effects model was employed with the time-matched placebo-subtracted differences in pre-dose-subtracted QTcI intervals ($\Delta\Delta$QTcI) as the dependent variable and the corresponding log (base 10) inhaled loxapine 10 mg concentration as the independent variable. Similar analyses were done for its metabolite, 7–OH–loxapine, and for moxifloxacin.

Pharmacokinetic Analysis. For each subject, PK parameters were estimated based on noncompartmental analyses

(NCA) for inhaled loxapine and 7-OH-loxapine as follows: area under the concentration curve (AUC) from 0 to the last measurable value (AUC_{last}), AUC from 0 to 2 hours ($AUC_{0-2\,h}$), AUC from 0 to infinity (AUC_{inf}), maximum plasma concentration (C_{max}), time to maximum plasma concentration (T_{max}), elimination rate constant (k_e), and elimination half-life ($T_{1/2}$). Clearance/fraction absorbed (CL/F) was estimated for inhaled loxapine. Descriptive statistics involving sample mean, standard deviation, median, and range were used to characterize all plasma values.

Safety Analysis. Overall safety data were summarized by treatment group, and when appropriate, shift tables were prepared.

The assessment of overall safety was based on the frequency, intensity, and type of AEs, safety ECG measurements, and descriptive statistics for change from baseline in the clinical laboratory variables, vital signs, and urine electrolytes. Baseline values for clinical laboratory variables and urine electrolytes were the values obtained from samples collected on admission day of each treatment period. Baseline values for vital signs were the values obtained pre-dose within 1 hour of oral capsule administration during each treatment period. AEs that occurred during the study were attributed to the treatment received most recently. All AEs presented in this study report were treatment emergent.

Quantitative safety measures (QRS interval, heart rate and blood pressure) were examined with the 90% CI on the time-matched placebo-subtracted differences in pre-dose-subtracted values (ΔΔQRS, ΔΔHR, ΔΔSBP, ΔΔDBP).

Results

Subject Demographics and Disposition

Of the 132 healthy volunteer subjects that were screened, 48 were enrolled and 46 subjects completed the study. One subject was withdrawn by the investigator for alcohol ingestion prior to her third treatment, and a second subject was designated as an early withdrawal by his failure to appear at the CRU for a second treatment. This subject was lost to follow up, participated only in the first period, and received only placebo (Figure 1).

The mean age (±SD) of randomized subjects was 40.9 years (±13.5) years and 50% were male (Table 1). The majority of subjects (85.4%) were Caucasian; 6 subjects were Black and 1 subject was Hispanic. Most subjects

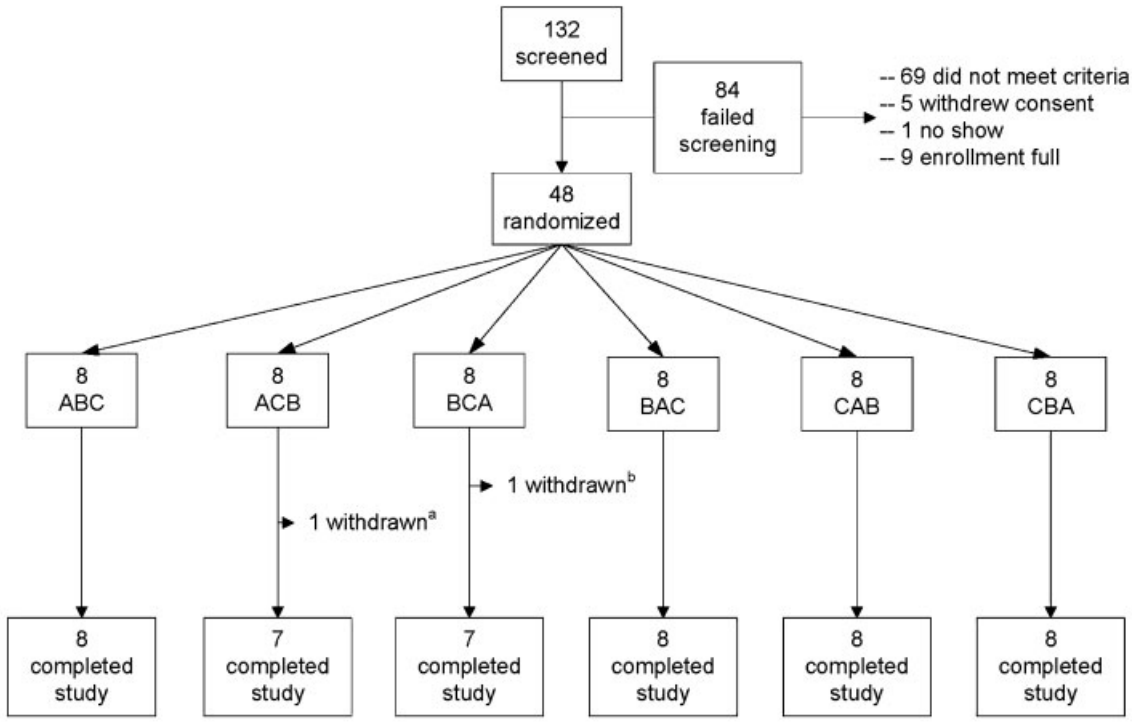

Figure 1. Subject disposition (safety population).

Table 1. Demographics and Baseline Characteristics (Safety Population)

Demographic or baseline characteristic	Loxapine 10 mg (N = 47)	Placebo (N = 47)	Oral moxifloxacin 400 mg (N = 47)
Sex, N (%)			
Female	24 (51.1%)	23 (48.9%)	24 (51.1%)
Male	23 (48.9%)	24 (51.1%)	23 (48.9%)
Age (years)			
Mean (SD)	41.1	41.3	41.1
Median	46.0	46.0	46.0
Min, max	18.0, 65.0	18.0, 65.0	18.0, 65.0
Race, N (%)			
Caucasian	41 (85.4%)	41 (85.4%)	41 (85.4%)
Black	6 (12.8%)	6 (12.8%)	6 (12.8%)
Hispanic	1 (2.1%)	1 (2.1%)	1 (2.1%)
Smoking history, N (%)			
Never smoked	37 (78.7%)	37 (78.7%)	37 (78.7%)
Current smoker	0	0	0
Ex-smoker	11 (21.3%)	11 (21.3%)	11 (21.3%)

(79.2%) had no history of smoking. Of the 10 ex-smokers in the study, none had smoked since 2006.

QT Data

Most (2715 of 2727) QT intervals were analyzed as triplicates. Due to missing data 6 were duplicates and 6 were singletons.

Analyses of QT versus RR interval clearly demonstrated an increase in QT with increasing RR interval, demonstrating the need to correct QT for heart rate. In contrast, Analyses of QTcI verified an appropriate correction of QTc based on RR intervals.

Effect of Moxifloxacin on QTc

Assay sensitivity was demonstrated by the lower 1-sided 95% confidence bounds placed on point estimates of ΔΔQTcI being greater than 5 milliseconds at 2 (2.5 and 3 hours) of the 4 times post-dose, chosen a priori (1.5, 2, 2.5, and 3 hours), and the expected time course of the moxifloxacin response. The moxifloxacin maximum ΔΔQTcI occurred at 3 hour post-dose (LSmean 8.36 milliseconds, lower confidence bound 5.82 milliseconds) (Figure 2).

Effect of Loxapine on QTc

QT Primary Endpoint Results. inhaled loxapine at a dose of 10 mg did not increase QT intervals, as demonstrated by the upper bound of the 1-sided 95% CIs placed on the point estimate of the placebo-subtracted change of QTcI (ΔΔQTcI) being less than 10 milliseconds at all post-dose times. The maximum ΔΔQTcI occurred at 1 hour post-dose (LSmean 5.42 milliseconds, upper confidence bound 7.75 milliseconds). No important differences were seen between males and females in their ΔΔQTcI results (Figure 3).

QT Secondary Endpoint Results. The maximum of all upper 1-sided 95% confidence bounds for ΔΔQTcF occurred at 1 hour post-dose (LSmean 6.41 milliseconds, upper confidence bound 8.77 milliseconds). The maximum of all upper 1-sided 95% confidence bounds for ΔΔQTcB occurred at 1 hour post-dose (LSmean 11.0 milliseconds,

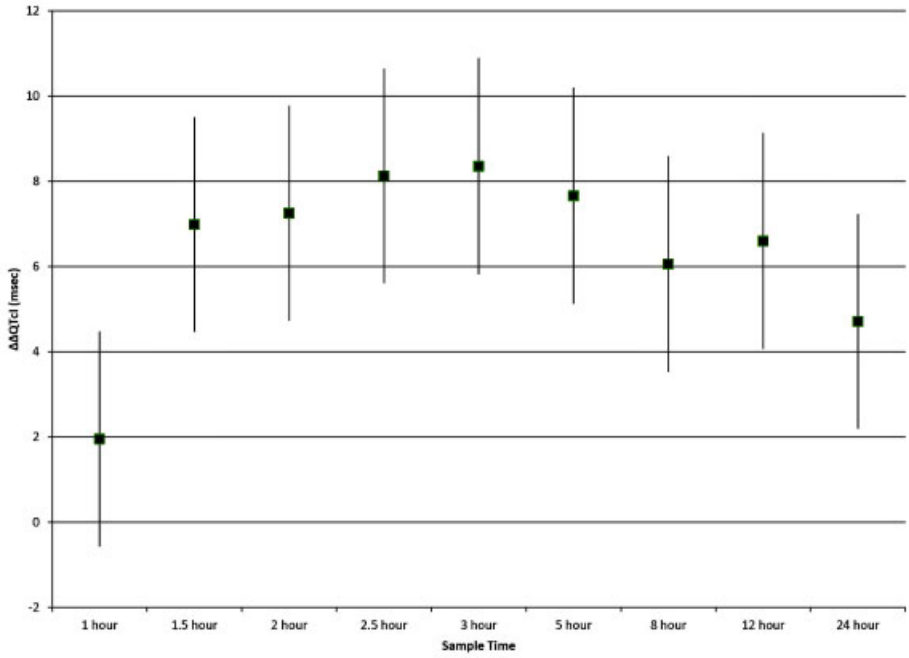

Figure 2. Moxifloxacin QTcI, LSmean differences from placebo in change from baseline and 90% CI, primary analysis model (QT population).

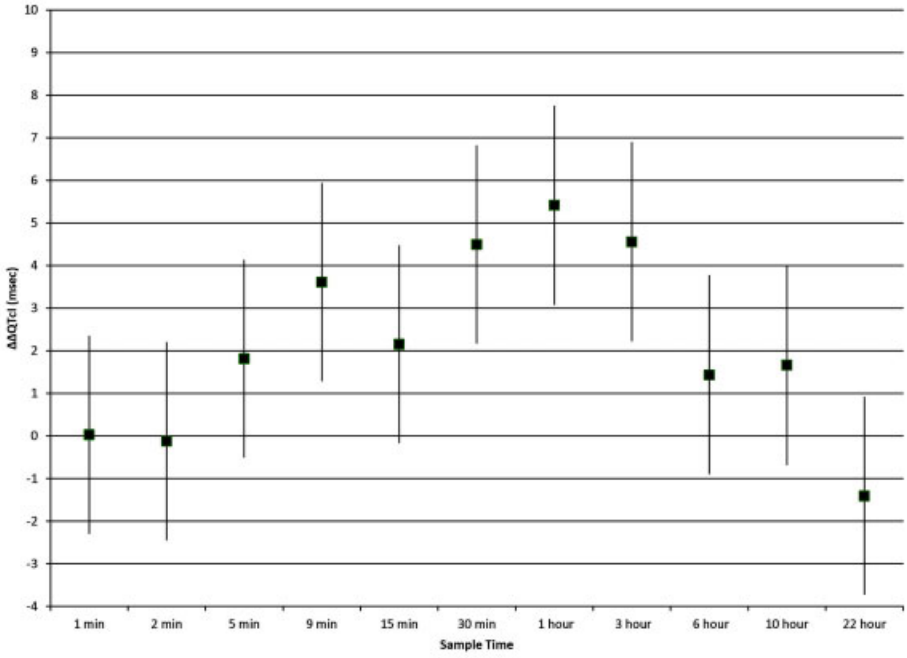

Figure 3. Staccato Loxapine QTcI, LSmean differences from placebo in change from baseline and 90% CI, primary analysis model (QT population).

upper confidence bound 14.2 milliseconds); however, with the addition of change from pre-dose baseline in heart rate to the analysis model, the maximum occurred at 1 hour post-dose (LSmean 6.07 milliseconds, upper confidence bound 8.55 milliseconds). Computation of simple mean placebo-subtracted changes of QTcI and QTcF supported the primary QT analysis conclusion.

Categorical Analysis

QTcI and QTcF both exceeded 450 milliseconds $\times 1$ among subjects after inhaled loxapine and $\times 1$ after placebo, QTcB $\times 3$ after inhaled loxapine and $\times 4$ after placebo. No QTcI, QTcF, or QTcB exceeded 480 or 500 milliseconds. The QTcI after inhaled loxapine was 429 milliseconds predose and 461 milliseconds at 3 hours, and $\times 1$ after placebo was 422 milliseconds predose and 452 milliseconds at 3 hours.

QTcI and QTcF both increased >30 milliseconds from baseline x 1 among subjects after inhaled loxapine and $\times 1$ after placebo, QTcB $\times 6$ after inhaled loxapine and $\times 6$ after placebo. No QTcI, QTcF, or QTcB increased >60 milliseconds from baseline. The QTcI increase after inhaled loxapine was 31 ms at 3 hours and after placebo loxapine was 30 milliseconds at 3 hours.

All 4 QTcI events were reported for subject 36, a 52 year-old female.

The only morphological finding not present at baseline which was more frequent following inhaled loxapine than after placebo was ectopic atrial rhythm ($\times 1$ after inhaled loxapine $\times 0$ after placebo).

QRS, Heart Rate and Blood Pressure

For the subjects receiving inhaled loxapine, all of the 90% CI on the 11 time-matched placebo-subtracted differences in pre-dose-subtracted QRS values ($\Delta\Delta$QRS) included 0. The minimum lower 90% CI was -1.47% and the maximum was 1.60% of baseline QRS. For the heart rate, the minimum and maximum of the 11 $\Delta\Delta$HR 90% CIs ranged from -6.76% to 12.7%.

For systolic and diastolic BP measured at 9 post-dose time points, the 90% CI ranges were -7.34% to 3.89% for $\Delta\Delta$SBP, and -7.53% to 4.98% for $\Delta\Delta$DBP.

Pharmacokinetics of Loxapine

After administration of inhaled loxapine, plasma concentrations of loxapine increased with a median T_{max} of 1 minute and a mean C_{max} of 312 ng/mL. After an initial rapid distribution phase, plasma concentrations of loxapine declined with a terminal half-life of 8 hours. The clearance uncorrected for bioavailability (CL/F) of loxapine was 52 L/h.

Exposure to active metabolite 7-OH-loxapine ranged from 12% to 15% of the parent compound based on mean AUC_{last} and AUC_{inf} values, respectively. The mean C_{max} of 1.6 ng/mL occurred at median T_{max} 3 hours. The terminal half-life of 7-OH-loxapine was 12 hours.

Relationship Between QT Interval and Loxapine Concentrations

The relationship between loxapine concentrations and $\Delta\Delta$QTcI was nonlinear and downwardly parabolic (eg,

inverted-U in shape), indicating that there was no positive concentration-response relationship between inhaled loxapine blood levels and QT intervals. The median observed loxapine concentration (32.1 ng/mL) was associated with a mean of 4.25 milliseconds and upper confidence bound of 5.62 milliseconds. The relationship between 7-OH-loxapine concentrations and ΔΔQTcI was likewise nearly flat such that the maximum observed 7-OH-loxapine concentration (2.87 ng/mL) was associated with a mean of 3.66 milliseconds and upper confidence bound of 6.06 milliseconds (Figure 4).

Safety and Tolerability Results

All AEs reported in this study were judged as mild or moderate and resolved without sequelae. No subject discontinued because of an AE, and there were no

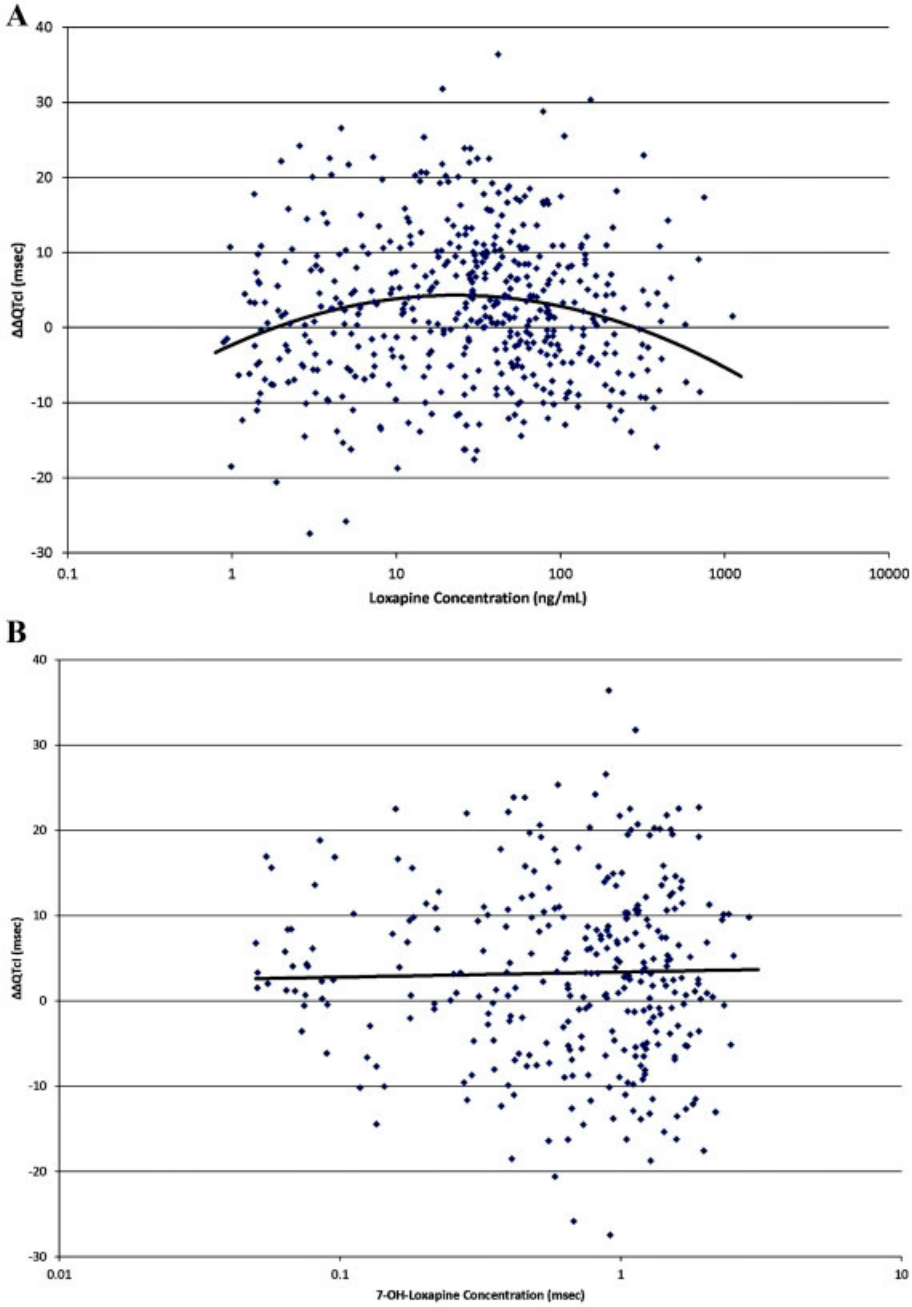

Figure 4. Placebo-subtracted changes from baseline of QTcI (milliseconds) versus Loxapine and 7-OH loxapine concentrations (ng/mL) and fitted regressions (QT population). (A) Regression: $\Delta\Delta QTcI = -2.308 + 9.730$ [log(loxapine)] $- 3.575$[log(loxapine)]2. (B) Regression: $\Delta\Delta QTcI = 3.391 + 0.595$[log(7-OH-lox)], Slope (90% CI) $= 0.595$ (-1.902, 3.093).

serious adverse events (SAEs) or deaths. The percentage of subjects with any AE was 80.9% in the inhaled loxapine and 40.4% in the placebo control group. The percentage with treatment-related AEs 78.7% in the inhaled loxapine group and 27.7% in the placebo control group (Table 2). The most common treatment-related AEs reported were somnolence, dizziness, dysgeusia, and cough. Somnolence and dizziness are known effects of oral loxapine, and dysgeusia and cough are common with orally inhaled drugs. There were no reports of hypotension, dyspnea, wheezing, or bronchospasm in any of the subjects. There was 1 report of an extrapyramidal event (moderate akathisia) 8 hours after inhaled loxapine treatment that resolved within 3.3 hours and was judged to be possibly treatment-related. There were no clinically significant changes from baseline within any treatment group in ECGs, laboratory tests, physical examinations, or vital signs, nor were there clinically significant differences among groups. Overall, inhaled loxapine was well tolerated in this study.

Discussion

The assessment of a drug's potential to delay cardiac repolarization, as assessed by the QT/QTc interval, is now a required part of the development of ethical pharmaceutical compounds.[3] This is the first QT/QTc study of loxapine by any route. This study provided a rigorous assessment of the potential for inhaled loxapine to prolong ventricular repolarization in human subjects at the intended therapeutic dose.

This was a double-blind, double-dummy, active- and placebo controlled, 3-period crossover study investigating a single dose of Staccato Loxapine (10 mg, the approved therapeutic dose), a positive control known to prolong QT/QTc (oral moxifloxacin, 400 mg), and oral and inhaled placebos.

ECGs in this study were recorded on a 12-lead digital Holter and analyzed at a central ECG facility using state-of-the-art techniques of cardiologist executed ECG interval measurement.

Assay sensitivity was demonstrated by the lower 90% CI for ΔΔQTcI being greater than 5 milliseconds after administration of the moxifloxacin active control, validating the study size and methodology as well as the outcome of the study.

Staccato Loxapine at a dose of 10 mg met the primary QTcI endpoint. Secondary outcome measures, QTcF and QTcB secondary endpoints supported the primary endpoint. No important differences were seen between males and females in their ΔΔQTcI results. Simple mean placebo-subtracted changes of QTcI and QTcF also supported the primary endpoint as did the negative analyses of outliers for absolute QTc and change of QTc from baseline, and by the lack of significant emergent diagnostic findings for all QTc analyses (QTcI, QTcB, and QTcF).

Effects of loxapine administration on ECG were evaluated in several prospective studies and no anomalies

Table 2. Treatment-Related Adverse Events Reported More Frequently in Staccato Loxapine Then Placebo in Any Treatment Group (Safety Population)

System organ class adverse event	*Staccato* Loxapine10 mg (N = 47)	Placebo[a] (N = 47)	Oral moxifloxacin 400 mg (N = 47)
No. (%) of subjects with any treatment-related AE	37 (78.7%)	13 (27.7%)	8 (17.0%)
Dysgeusia	9 (19.1%)	1 (2.1%)	2 (4.3%)
Dysphagia	1 (2.1%)	0 (0.0%)	0 (0.0%)
Injury, poisoning, and procedural complications			
Excoriation	1 (2.1%)	0 (0.0%)	0 (0.0%)
Nervous system disorders			
Akathisia	1 (2.1%)	0 (0.0%)	0 (0.0%)
Dizziness	17 (36.2%)	2 (4.3%)	4 (8.5%)
Somnolence	29 (61.7%)	6 (12.8%)	2 (4.3%)
Psychiatric disorders			
Euphoric mood	1 (2.1%)	0 (0.0%)	0 (0.0%)
Mood altered	1 (2.1%)	0 (0.0%)	1 (2.1%)
Respiratory, thoracic, and mediastinal disorders			
Cough	7 (14.9%)	1 (2.1%)	0 (0.0%)
Nasal congestion	1 (2.1%)	0 (0.0%)	0 (0.0%)
Vascular disorders			
Flushing	1 (2.1%)	0 (0.0%)	0 (0.0%)

All AEs presented in this study report were treatment emergent. AEs tabulated above were judged by the investigator to be possibly or probably related to study treatment. Subjects with more than 1 occurrence of a specific AE are counted only once.

[a]Placebo includes exposure to oral placebo prior to inhalation exposure and post inhalation exposure when both oral and inhalation were placebo.

in heart rhythm or ECG intervals except sinus tachycardia were reported.[5–13] Some studies described non-specific ECG changes after loxapine administration, which may have been drug related.[14–18]

Prolongation of QT interval has not been reported in prospective studies evaluating effects of loxapine on ECG. QT prolongation was, however, reported in 3 out of 10 patients following loxapine overdose of 450 to 2750 mg,[19] but ventricular arrhythmias have not been reported in loxapine overdose.

As is the case for most other antipsychotics, loxapine weakly blocks the hERG (human Ether-à-go-go Related Gene) channel—an indicator of potential risk for QT interval prolongation in humans with the associated risk of fatal arrhythmias. To explore the potential interaction of loxapine with hERG channels, Alexza conducted a non-GLP in vitro study to evaluate the effects of loxapine on hERG current expressed in stably transfected human embryonic kidney (HEK-293) cells. Loxapine dose-dependently blocked the hERG current with an IC50 value of 1800 nM (or 590 ng/mL unbound). For comparison, IC50 values for blocking hERG channel current for other antipsychotics include: sertindole 3 nM, droperidol 32 nM, risperidone 167 nM, ziprasidone 169 nM, thioridazine 191 nM, perphenazine 1003 nM, chlorpromazine 1561 nM, quetiapine 5765 nM, olanzapine 6013 nM.[20,21] This invitro study suggested relatively low risk for QT prolongation after loxapine at therapeutic doses and this was bourne out in the current in vivo study.

Although inhaled loxapine was associated with small mean increases in heart rate, this did not affect the ΔΔQTcI because the individual correction adequately corrected for heart rate.

The relationship between Staccato Loxapine concentrations and ΔΔQTcI was nonlinear and downwardly parabolic, indicating that there was no positive concentration-response relationship between Staccato Loxapine blood levels and QT intervals. The pharmacokinetics of Staccato Loxapine were comparable to those observed in previous Staccato Loxapine studies, demonstrating rapid absorption, elimination, and clearance.

Because of the short duration of the study, the withdrawal rate was low. Sufficient subjects completed the study to achieve the objectives. Men and women were represented equally in the study population, and no important differences were seen between males and females in their ΔΔQTcI results.

A limitation of this study was that the highest approved inhaled loxapine dose of 10 was studied. Supratherapeutic doses were not evaluated because of tolerability concerns in healthy subjects. The inclusion of only healthy subjects is likewise a limitation of this study. There may also be other factors that contribute to the development of torsade's de pointes in vivo, such as hypokalaemia, hypomagnesaemia and organic heart disease,[22] which cannot be mimicked in a healthy subject study.

The findings of this study lead to the following conclusions:

- This is a negative Thorough QT/QTc Study as defined in the ICH E14 guideline, that is, the upper bound of the 95% 1-sided confidence interval for the largest time-matched mean effect of loxapine on the QTcI interval excluded 10 milliseconds. The study outcome was validated by the demonstrated assay sensitivity using the positive control moxifloxacin.
- There was no positive concentration-response relationship between loxapine no 7-OH loxapine blood concentrations and QT intervals.
- Inhaled loxapine at the therapeutic dose of 10 mg was safe and well tolerated in these healthy volunteers. All AEs were mild or moderate and resolved without intervention. No clinically meaningful, treatment-related changes were observed in vital signs, safety ECG, laboratory, or physical examination results.

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Declaration of Conflicting Interests

D.A.S. was an employee during execution of this study and is currently a paid consultant of Alexza Pharmaceuticals. P.V. and E.R.H. have been paid consultants of Alexza Pharmaceuticals. J.-V.C. is an employee of Alexza Pharmaceuticals.

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