

Onset of Action of the Fixed Combination Intranasal Azelastine-Fluticasone Propionate in an Allergen Exposure Chamber



Jean Bousquet, MD, PhD^{a,b,c,d}, Eli O. Meltzer, MD^e, Peter Couroux, MD^f, Arkady Koltun, MD, PhD^g, Ferdinand Kopietz, PhD^h, Ullrich Munzel, PhD^h, Hans Christian Kuhl, PhD^h, Duc Tung Nguyen, PhD^h, Anne Marie Salapatek, PhD^e, and David Price, MD^{i,j,k} *Montpellier and Montigny-le Bretonneux, France; Brussels, Belgium; Berlin and Bad Homburg, Germany; Mississauga, Ontario, Canada; Canonsburg, Pa; Singapore; and Cambridge and Aberdeen, United Kingdom*

What is already known about this topic? Randomized controlled trials are insufficient to assess the onset of action of rhinitis medications. The 2 most common treatment strategies for uncontrolled rhinitis were not compared for efficacy or onset of action.

What does this article add to our knowledge? In a chamber study, single-device azelastine-fluticasone formulation is effective within 5 minutes and the effect persists over the 4-hour study period. A free combination of intranasal fluticasone propionate and oral loratadine is effective after 2 hours.

How does this study impact current management guidelines? Patients with allergic rhinitis want an effective rapid-onset treatment that is not reflected in the guidelines, which need to become closer to patients' preferences. The study may help physicians to decide which treatment to use.

^aMACVIA-France, Contre les Maladies Chroniques pour un Vieillessement Actif en France European Innovation Partnership on Active and Healthy Ageing Reference Site, Montpellier, France

^bINSERM U 1168, VIMA: Ageing and chronic diseases Epidemiological and public health approaches, Villejuif, Université Versailles St-Quentin-en-Yvelines

^cEuforea, Brussels, Belgium

^dCharité, Berlin, Germany

^eAllergy and Asthma Medical Group and Research Center, San Diego, Calif

^fInflammax Research, Mississauga, Ontario, Canada

^gMylan, Canonsburg, Pa

^hMeda Pharma GmbH & Co. KG (a Mylan Company), Bad Homburg, Germany

ⁱObservational and Pragmatic Research Institute, Singapore

^jOptimum Patient Care, Cambridge, UK

^kAcademic Centre of Primary Care, University of Aberdeen, Aberdeen, UK

This clinical study was funded by MEDA Pharma GmbH & Co KG (Bad Homburg, Germany). Graphical support for the development of figures for this article was provided by Ashfield Healthcare Communications (Middletown, Conn) and funded by Mylan, Inc (Canonsburg, Pa).

Conflicts of interest: J. Bousquet has received personal fees and other fees from Chiesi, Cipla, Hikma, Menarini, Mundipharma, Mylan, Novartis, Sanofi-Aventis, Takeda, Teva, and Uriach; and has received other fees from Kyomed, outside the submitted work. E. Meltzer has received provision of writing assistance, medicines, equipment, or administrative support from Ashfield Healthcare; has received consultancy fees from ALK, AstraZeneca, Boehringer Ingelheim, Circassia, GlaxoSmithKline, Glenmark, and Greer/Stallergenes; has received payment for lectures including service on speakers bureaus for ALK, GlaxoSmithKline, Glenmark, Mylan, Teva, and Greer/Stallergenes; and has received payment for developing educational presentations for ALK. A. Koltun is employed by Mylan. U. Munzel, H. C. Kuhl, and F. Kopietz are employed by MEDA Pharma GmbH & Co KG. A. M. Salapatek is employed by and has received fees for participation in review activities such as data monitoring boards, statistical analysis, end point committees, and the like from Inflammax Research in a

clinical contract research organization; Inflammax Research performed the clinical research and the statistical analyses for the research presented for this trial and money was sent to the institution. (Inflammax Research is a clinical contract research organization that was paid for the full conduct of the clinical research presented here including study design development, clinical conduct, data management, analysis, and reporting of the clinical outcomes.) D. Price has received other support from Aerocrine, Almirall, Amgen, AstraZeneca, Boehringer Ingelheim, Chiesi, Cipla, Merck, Mylan, Mundipharma, Napp, Novartis, Teva Pharmaceuticals, Almirall, GlaxoSmithKline, Kyorin, Pfizer, Skyepharma, Teva, Theravance, and Zentiva; has received grants from Aerocrine, AKL Research and Development Ltd, AstraZeneca, Boehringer Ingelheim, British Lung Foundation, Chiesi, Mylan, Mundipharma, Napp, Novartis, Pfizer, Respiratory Effectiveness Group, Teva Pharmaceuticals, Theravance, UK National Health Service, and Zentiva; has received nonfinancial support from Efficacy and Mechanism Evaluation program and Health Technology Assessment Commissioning Board, outside the submitted work; has stock/stock options from AKL Research and Development Ltd, which produces phytopharmaceuticals; and owns 74% of the social enterprise Optimum Patient Care Ltd (Australia, Singapore, and United Kingdom) and 74% of Observational and Pragmatic Research Institute Pte Ltd (Singapore). The rest of the authors declare that they have no relevant conflicts of interest.

Received for publication November 10, 2017; revised January 14, 2018; accepted for publication January 19, 2018.

Available online February 7, 2018.

Corresponding author: Jean Bousquet, MD, PhD, CHU Arnaud de Villeneuve, 371 Avenue du Doyen Gaston Giraud, 34295 Montpellier CEDEX 5, France. E-mail: jean.bousquet@orange.fr.

2213-2198

© 2018 American Academy of Allergy, Asthma & Immunology

<https://doi.org/10.1016/j.jaip.2018.01.031>

Abbreviations used

AEC	Allergen exposure chamber
AR	Allergic rhinitis
EEC	Environmental exposure chamber
ePDAT	electronic Patient Data Acquisition Tablet
FAS	Full analysis set
INCS	Intranasal corticosteroid
INFF	Intranasal fluticasone furoate
INFP	Intranasal fluticasone propionate
LORA	Oral loratadine
MCID	Minimal clinically important difference
MP-AzeFlu	Intranasal azelastine with intranasal fluticasone propionate
RCT	Randomized controlled trial
SPT	Skin prick test
T7SS	Total score of the 7 nasal and ocular symptoms (including TNSS and TOSS)
TNSS	Total nasal symptom score
TOSS	Total ocular symptom score
V1, V2, ..., V7	Visit 1, visit 2, ..., visit 7
VAS	Visual analog scale

BACKGROUND: A fixed-dose combination of intranasal azelastine hydrochloride and fluticasone propionate (MP-AzeFlu) is the most effective treatment of allergic rhinitis, but its onset of action requires further investigation.

OBJECTIVE: To compare the onset of action of MP-AzeFlu with the free combination of oral loratadine (LORA) and intranasal fluticasone propionate (INFP).

METHODS: In this single-center, randomized, placebo-controlled, double-blind, double-dummy, 3-period crossover trial, allergic rhinitis symptoms were induced in asymptomatic patients by ragweed pollen challenge in an allergen environmental exposure chamber. Patients received single-dose MP-AzeFlu, LORA/INFP, or placebo and were monitored for 4 hours. The primary outcome was onset of action measured by total nasal symptom score (TNSS). Secondary measures were total ocular symptom score (TOSS), total score of the 7 nasal and ocular symptoms (T7SS), and the global visual analog scale (VAS).

RESULTS: The full analysis set included 82 patients, of which 78 completed all treatments. TNSS was significantly reduced versus placebo from 5 minutes for MP-AzeFlu and 150 minutes for LORA/INFP onward (both $P < .05$) till the end of assessment (0–4 hours). MP-AzeFlu reduced TNSS to a greater extent at each time point from 5 to 90 minutes ($P < .05$) and over the entire assessment interval ($P \leq .005$) versus LORA/INFP or placebo. No statistically significant difference between LORA/INFP and placebo was observed over the assessment interval ($P = .182$). The onset of action of MP-AzeFlu assessed by TOSS, T7SS, and VAS was 10 minutes, 2 hours earlier than with LORA/INFP.

CONCLUSION: MP-AzeFlu had a more rapid onset of action (5 minutes) and was more effective than LORA/INFP. © 2018 American Academy of Allergy, Asthma & Immunology (J Allergy Clin Immunol Pract 2018;6:1726–32)

Key words: Allergic rhinitis; Azelastine; Environmental exposure chamber; Fluticasone propionate; Loratadine; Onset of action

INTRODUCTION

Most patients with allergic rhinitis (AR) have moderate/severe disease with persistent symptoms that are often insufficiently recognized by their physicians. Comedication is common, with patients self-medicating and doctors concurrently prescribing, most commonly, oral H_1 antihistamines and intranasal corticosteroids (INCSs) despite lack of evidence for this strategy.^{1–4} A large number of patients with AR use their treatment intermittently, and frequently expect a rapid onset of efficacy.

The US Food and Drug Administration (FDA) has proposed 3 study types to assess the onset of action of medications^{5,6}: the standard phase 3 double-blind randomized controlled trials (RCTs), park-setting studies, and allergen exposure chamber (AEC) studies.⁷ However, RCTs cannot provide a sufficient precision to assess the onset of efficacy because they cannot allow repeated timing over short periods of time (minutes). AECs offer some advantages over RCTs in assessing the onset of efficacy of medications because it can be demonstrated in minutes.⁷ AECs allow consistent allergen exposure. An AEC involves a manipulated *in vivo* procedure, whereas a park-setting study mirrors real-life exposure. Park-setting as well as AEC studies have not captured the early time. It appears that a crossover trial would be difficult with a park-setting study because of variations in allergen exposure between days. Nevertheless, an AEC cannot replace real-world allergen exposure but can only complement it. To date, AEC studies conducted have been monocentric and have followed protocols unique to each center. Because there are technical differences among AECs, it is not easy to compare the results obtained in different AECs.⁸ The environmental exposure chamber (EEC) used in this study has been used in the same manner for a number of onset of action studies and so provides an opportunity for comparison.^{9–17}

The aim of this study was to compare the onset of action of 2 treatment strategies for AR, a fixed drug combination of intranasal azelastine hydrochloride and intranasal fluticasone propionate (MP-AzeFlu) versus the sequential monotherapies of oral loratadine (LORA) and intranasal fluticasone propionate (INFP), in an Ontario EEC in treating the nasal and ocular symptoms of seasonal AR induced by a ragweed pollen challenge.

METHODS

Study design

The study was a single-center, randomized, active- and placebo-controlled, double-blind, double-dummy, and 3-period crossover trial (William's design¹⁸). It was divided into 3 visit categories: screening (V1), priming (V2, V4, and V6), and treatment (V3, V5, and V7)⁷ (see the [Methods](#) section in this article's Online Repository at www.jaci-inpractice.org). The primary outcome was the onset of action on nasal symptoms (total nasal symptom score [TNSS]). Secondary outcomes were the onset of action on (1) ocular symptoms (total ocular symptom score [TOSS]), (2) the total 7 nasal and ocular symptoms (T7SS), (3) the 7 individual symptoms, and (4) a global visual analog scale (VAS). Other secondary outcomes included the overall efficacy (0–4 hours after dosing) on (5) TNSS, (6) TOSS, (7) and T7SS, and (8) the comparison of time with relevant response to therapy (30% and 50% reductions in TNSS) to investigate a clinically meaningful response.¹⁹

Setting

The study was monocentric and was conducted at an EEC in Inflammax Research (Mississauga, Ontario).⁹⁻¹⁷

Participants

To be eligible for the study, a subject had to comply with all the inclusion and exclusion criteria detailed in this article's Online Repository at www.jaci-inpractice.org. Briefly, subjects (1) were in the age range of 18 to 55 years, (2) had a history of seasonal AR during the 2 previous ragweed pollen seasons, and (3) had a positive skin prick test (SPT) response to ragweed pollen extract.

The study conformed to good clinical practices and was approved by the ethical committee of IRB Services, Ontario, Canada. All subjects provided written informed consent.

Allergen challenge

The study was performed outside of the ragweed pollen season (January 7-March 16, 2017). The allergen challenge followed the schematic representation of a typical EEC study design.⁷ The details are reported in the Methods section. Two days before each treatment period, subjects were primed by exposure to airborne ragweed allergen at an average concentration of 3500 ± 500 grains/m³ for 2 hours (V2, V4, and V6). On the treatment days (V3, V5, and V7), subjects were exposed to the same allergen concentration in the EEC for 6.5 hours (see [Figure E1](#) in this article's Online Repository at www.jaci-inpractice.org).

To be eligible for V2 before the first allergen challenge, the subjects had to be asymptomatic or with mild symptoms (TNSS $\leq 3/12$). Before EEC entry and during a 2-hour priming ragweed EEC session, subjects recorded their individual nasal and ocular symptoms and their overall allergy symptoms on a VAS using an electronic diary (ePDAT, Inflammax Research, Toronto, Canada).

To be randomized and included in the treatment phase (V3), primed subjects had (1) to report a TNSS of 6/12 or more and a rhinorrhea score of 2/3 or more on at least 2 time points during the 2-hour allergen challenge at V2 and also during the first 2-hour allergen challenge at V3 and (2) to show no evidence of complete nasal blockage on anterior rhinoscopy performed within 30 minutes before dosing. If subjects met the inclusion criteria, they were randomized to a treatment sequence of MP-AzeFlu (1 spray per nostril MP-AzeFlu for a total dose of 274 mcg azelastine/100 mcg fluticasone propionate + oral administration of a placebo tablet), LORA/INFP (1 spray per nostril of generic fluticasone propionate for a total dose of 100 mcg fluticasone propionate and 1 tablet of 10mg loratadine), and placebo (1 spray per nostril of placebo nasal spray and oral administration of a placebo tablet). They received a single dose of the treatment approximately 2.5 hours after entering the EEC. The EEC session was continued over the next 4 hours with subjects recording symptoms and VAS using the same ePDAT. Time points for assessment were 5, 10, 15, 30, 60, 90, 120, 150, 180, 210, and 240 minutes after dosing.

Subjects returned to the study site after a period of at least 12 days (+ 5 days). The procedures at V4 and V5 (period 2) and V6 and V7 (period 3) were identical to those at V2 and V3, respectively, except that subjects did not need to meet a minimum threshold value for TNSS and rhinorrhea.

Outcomes

Symptoms Severity Rating Scale assessment. Severity scores for 7 individual AR signs/symptoms (nasal congestion/stuffiness, sneezing, rhinorrhea/nasal discharge, nasal pruritus, eye itching,

eye redness, and eye tearing) were recorded by the subject using ePDAT at each time point. Each sign/symptom was scored on a scale of 0 to 3 (0 = none and 3 = severe). The TNSS is the sum of the ratings for the individual nasal scores, the TOSS is the sum of the ratings for the individual ocular scores, and the T7SS is the sum of the ratings for the individual nasal and ocular scores.

Overall allergy symptoms. VAS rating was recorded at each time point using ePDAT. Scores ranged from not at all bothersome (0 mm) to very bothersome (100 mm).^{20,21}

Safety. Vital signs and adverse events were recorded.

Randomization. Subjects were randomized in a 1:1:1 ratio to the 6 treatment sequences by means of a computer-generated randomization schedule.

Sample size. Sample size considerations have been based on a previous study of intranasal azelastine versus oral mometasone.¹⁰ Scenarios based on these results have been considered with modifications on applied doses. Assuming a one-sided alpha level of 2.5% and a power of about 90% for the primary comparison of MP-AzeFlu against placebo at the 15-minute time point, as well as a power of 80% for comparison against the free combination, a total of 80 randomized subjects were considered to be sufficient in several of these scenarios.

Statistical analysis. Time courses were displayed graphically. For all end points, baseline is defined as average of the last 2 assessments in the EEC before dosing with study treatment.

By time point. Baseline-adjusted crossover analysis of covariance with fixed-effects treatment, period, and sequence was used for analysis of changes from baseline for efficacy end points. The covariance matrix over treatment periods was left unspecified. Degrees of freedom were determined by Satterthwaite approximation as implemented in SAS 9.2 PROC MIXED (SAS Institute, Cary, NC).

Overall efficacy. For all time points (0-4 hours after dosing) together, the same analysis of covariance model described earlier was used with the addition of time point as fixed effect.

For both active treatments and for TNSS, TOSS, and T7SS as well as individual nasal and ocular symptoms, the onset of action was defined as the first time point after initiation of treatment when the product demonstrates a greater change from baseline in the efficacy end point compared with the placebo treatment that proves durable till the last assessed time point.

Clinical relevance. Clinical relevance was assessed from both individual and population perspectives. Although responsiveness and minimal clinically important difference (MCID) threshold estimation may be related, the 2 concepts are not interchangeable.³

To investigate a clinically meaningful response from an individual perspective, Kaplan-Meier curves were derived for time response analysis. Cox proportional hazard models were used to evaluate and compare the time to relevant response (30% and 50%) between the treatment groups.

For population means, Barnes et al²² developed and tested anchor- and distribution-based models to determine MCID thresholds for AR treatments. The anchor-based MCID threshold value for a TNSS scale of 0 to 12 points ranged from 0.23 to 0.28 points. The distribution-based model applied commonly used

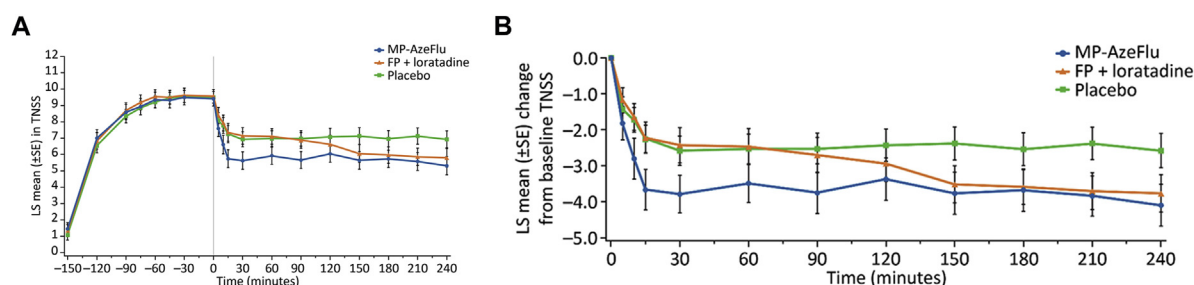


FIGURE 1. (A) TNSS for 6.5 h in the EEC (pre- and postdosing; FAS). **(B)** Change from baseline in TNSS (0-12) for 4 h postdosing (FAS). FP, Fluticasone propionate.

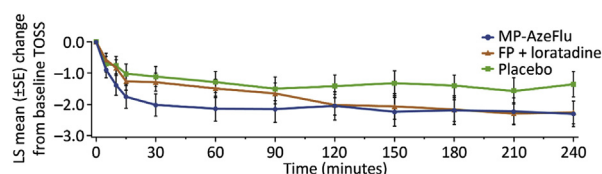


FIGURE 2. Change from baseline in TOSS (0-9) for 4 h postdosing (FAS). FP, Fluticasone propionate.

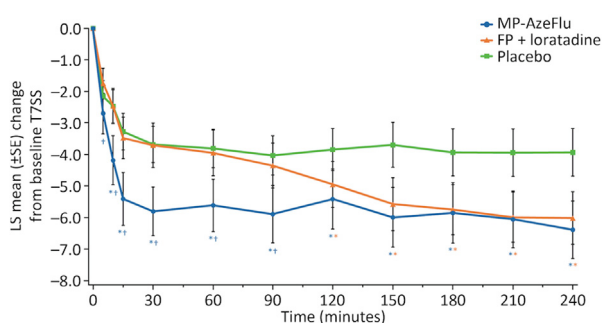


FIGURE 3. Change from baseline in T7SS (0-21) for 4 h postdosing (FAS). FP, Fluticasone propionate.

statistical methods for determining effect size by measuring the distance between 2 treatment means in relation to the baseline SD of the samples (Hedges' *g* and Cohen *d*). According to Cohen, a difference of 0.2 SD is regarded as a small relevant effect and one of 0.5 SD as a medium effect.

Populations for statistical analysis

The safety population included all subjects who administered at least 1 dose of study treatment. The full analysis set (FAS) population included all subjects in the study who had at least 1 evaluation of efficacy after treatment administration of 1 of the study treatments. Missing second or third treatment periods did not lead to exclusion. The statistical models were computed without these periods.

RESULTS

Characteristics of the subjects

A total of 82 subjects were included in the safety population and in the FAS, of which 78 completed all 3 periods/treatments of the crossover design as specified in the protocol. The mean age in the FAS was 41.6 ± 9.05 years and ranged from 20 to 55

years, and 53.7% ($n = 44$) of the subjects were women. Among the study participants, 43% were black, 28% white, 22% Asian, 6% multiple, and 1% Indian (see Table E1 in this article's Online Repository at www.jaci-inpractice.org). The disposition of subjects is presented in Figure E2 in this article's Online Repository at www.jaci-inpractice.org.

Primary outcome

In the TNSS evaluation, the significant effect of MP-AzeFlu versus placebo is documented from 5 minutes onward till the end of assessment (0-4 hours) ($P \leq .04$), resulting in an onset of action of 5 minutes for MP-AzeFlu (Figure 1, A and B; see also Table E2 in this article's Online Repository at www.jaci-inpractice.org). The onset of action of LORA/INFP lies at 150 minutes. A direct comparison of MP-AzeFlu and LORA/INFP shows that from minute 5 to minute 90 the effect of MP-AzeFlu on TNSS is significantly higher at each time point ($P < .05$). The treatment effect of LORA/INFP increases in the range of 120 minutes. In the overall comparison over the entire assessment interval (0-4 hours), MP-AzeFlu shows a better effect than LORA/INFP ($P = .005$) and placebo ($P < .001$) but LORA/INFP was not superior to placebo ($P = .182$).

Secondary objectives

Similar results are seen for other (secondary) efficacy end points: MP-AzeFlu onset of action assessed by TOSS, T7SS, and VAS is 10 minutes, which is about 2 hours earlier than the onset of LORA/INFP. For TOSS and T7SS, a direct comparison of MP-AzeFlu versus LORA/INFP shows superiority of MP-AzeFlu from minute 5 to minute 90 and also for the entire assessment period ($P < .05$). LORA/INFP was not superior to placebo for TOSS and T7SS over the entire assessment period (Figures 2 and 3).

For VAS, the superiority of MP-AzeFlu over LORA/INFP was shown from minute 5 to minute 60 ($P < .05$) (see Figure E3 in this article's Online Repository at www.jaci-inpractice.org).

Moreover, all individual nasal and ocular symptoms contributed to the overall effects and reached first statistical significance at early time points (see Tables E3 and E4 in this article's Online Repository at www.jaci-inpractice.org).

When assessing individual relevance of effects, the time to response analyses showed that for a reduction of either 30% or 50% TNSS (Figure 4), only MP-AzeFlu was significantly different from placebo. Moreover, MP-AzeFlu was significantly different from LORA/INFP (Table 1), whereas LORA/INFP could virtually not be distinguished from placebo.

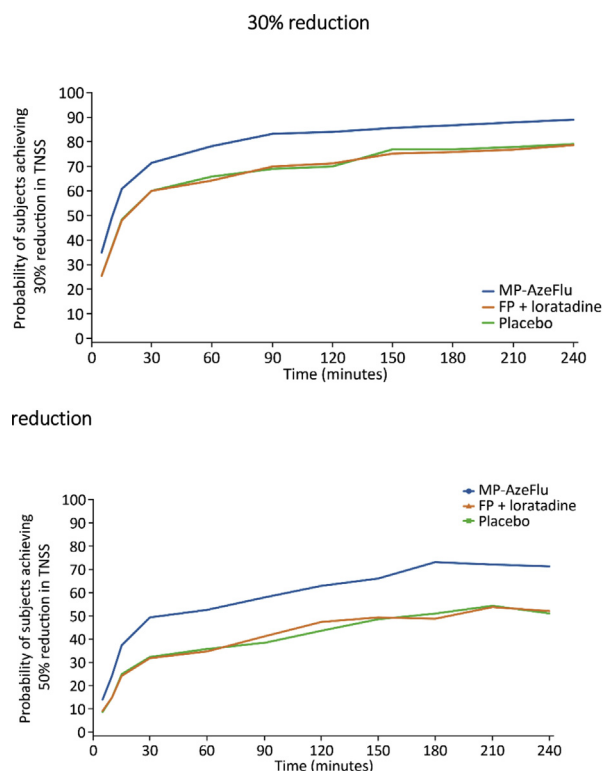


FIGURE 4. Percentage of subjects with a reduction in TNSS. FP, Fluticasone propionate.

Assessment of population relevance (MCID). In the present study, baseline SD was 2.43, 2.25, and 1.93 for MP-AzeFlu, LORA/INFP, and placebo, respectively. A conservative approach to the Hedges' *g* uses a sample size-adjusted pooled estimate of the 2 larger SDs leading to a *g* of 2.34. Hence, a difference of 0.47 TNSS score points would correspond to a small effect and one of 1.17 to a medium effect. This means that the differences observed at 5 minutes after dosing between MP-AzeFlu and placebo (0.5 score points) as well as between MP-AzeFlu and LORA/INFP (0.72 score points) are relevant. Moreover, the differences observed at 15 minutes after dosing between MP-AzeFlu and placebo (1.41 score points) as well as between MP-AzeFlu and LORA/INFP (1.45 score points) are medium effect sizes and therefore relevant. LORA/INFP reaches a relevant effect size versus placebo at 150 minutes (difference 1.11) and medium relevant effect size at 210 minutes (difference 1.28).

Safety

No serious adverse events occurred during the study (see this article's Online Repository at www.jaci-inpractice.org).

DISCUSSION

This is the first study to assess the onset of action of MP-AzeFlu, a fixed combination of an intranasal antihistamine, azelastine, and an INCS, fluticasone propionate, in a state-of-the-art design and to compare it with another treatment strategy in an AEC. It was found that MP-AzeFlu provided patients with AR with moderate to severe symptoms significant symptom relief 2 hours earlier than the sequential dosing of the monotherapies of an oral H₁ receptor antihistamine (loratadine) and an INCS (fluticasone propionate).

The latter is a very common strategy to use to manage AR in these highly symptomatic patients. The results are consistent for all outcomes addressed including nasal and ocular symptoms. This study is the first AEC study that determined clinically meaningful difference between these regimens (MP-AzeFlu vs LORA/INFP).

Strengths and weaknesses

The Ontario EEC has wide experience in assessing the onset of action of medications and has been validated over a number of years. It has the advantages to allow for a standardized intrasubject comparison.

The present study was not meant to compare 2 medications but rather 2 therapeutic strategies and therefore it was not necessary to use additional treatment arms of LORA and INFP as monotherapies.

The choice of medications may be questioned. MP-AzeFlu is the only marketed intranasal combination of an intranasal H₁ antihistamine and an INCS. We selected INFP because it is one among the most widely used globally and it is the INCS used in AzeFlu. Cetirizine could have been used instead of LORA but there is no large study assessing cetirizine with an INCS. Moreover, LORA and cetirizine are the most widely used oral H₁ antihistamines. LORA was found to be less effective than cetirizine or levocetirizine in the Ontario EEC and the onset of action was delayed.⁹ Cetirizine or levocetirizine has an onset of action 1 hour after dosing.^{23,24} Another study showed that the onset of action of cetirizine was just 15 minutes earlier than that of LORA.²⁵ Fluticasone propionate was the comparator used, whereas intranasal fluticasone furoate (INFF) is a newer molecule. RCTs comparing the 2 medications found that treatment with once-daily INFF was effective and noninferior to twice-daily INFP in reducing nasal symptoms. However, the onset of action for INFF was observed only before some hours, as well.²⁶ Moreover, in recent guidelines, all INCS or oral H₁ antihistamines have been considered as a class.²⁷ Thus, the data obtained in this study are generalizable to all medications of the same class.

Interpretation of the results

In the recent FDA guidance, "the definition of the onset of action of an AR product is the point at which subjects might reasonably expect to see a meaningful decrease in their allergic rhinitis symptoms. For the purposes of AR, it is the first time point after initiation of treatment when the product demonstrates a greater change from baseline in the primary efficacy outcome compared to the placebo treatment that proves durable from this point until the end of the proposed dosing interval."⁶ In the 2000 FDA guidance, a statistically significant difference was requested.⁵ Clinical phase 3 studies showed an onset of action of MP-AzeFlu at 30 minutes after dosing²⁸ and that the effect was maintained until the end of the dosing interval. Hence, symptoms monitoring until the end of dosing interval was not required for the present study that focused on an onset of action faster than 30 minutes. The results are in line with both guidances and confirm a very fast onset of action of MP-AzeFlu evidenced by a significant symptom relief already seen at 5 minutes after dosing and maintained until the last assessed time point. The differences seen between placebo and the active control LORA/INFP are in line with expectations and suggest that the study was sensitive to detect treatment differences but was not overpowered.

TABLE I. Rate of change in log of the hazard for reduction in TNSS (0-12): FAS population

	Rate of change, mean $\pm$ SEM	Z value	P value	Hazard ratio (95% CI)
30% reduction in TNSS				
Baseline TNSS	0.03 $\pm$ 0.04			
MP-AzeFlu vs INFP/LORA	0.38 $\pm$ 0.13	2.96	.003	1.46 (1.14-1.87)
MP-AzeFlu vs placebo	0.38 $\pm$ 0.15	2.43	.015	1.46 (1.08-1.97)
INFP/LORA vs placebo	-0.01 $\pm$ 0.13	-0.01	.99	1.00 (0.77-0.30)
50% reduction in TNSS				
Baseline TNSS	-0.04 $\pm$ 0.04			
MP-AzeFlu vs INFP/LORA	0.55 $\pm$ 0.16	3.37	<.001	1.74 (1.26-2.41)
MP-AzeFlu vs placebo	0.56 $\pm$ 0.18	3.11	.002	1.75 (1.23-2.50)
INFP/LORA vs placebo	0.01 $\pm$ 0.18	0.04	.97	1.01 (0.70-1.44)

TABLE II. Comparison of the time of onset of action using the same EEC

Drug (dose)	Formulation	Onset of		Source
		action	Parameter	
Azelastine (548 μ g)	Nasal spray	15 min	TNSS	10
Olopatadine (0.4% and 0.6%)	Nasal spray	90 min	TNSS	13
Ciclesonide (200 μ g)	Nasal spray	60 min	TNSS	14
Budesonide (64 μ g)	Nasal spray	8 h	TNSS	15
Budesonide (64 μ g) and azelastine (274 μ g)	Nasal spray	0.33 h		
CDX-313 (solubilized budesonide [64 μ g] and azelastine [274 μ g])	Nasal spray	0.33 h		
Levocetirizine (5 mg)	Tablet	2.5 h	MSC	30

MSC, Major symptom complex.

Moreover, the effect of MP-AzeFlu appears to be clinically relevant from both individual and population perspectives. Although responsiveness and MCID threshold estimation may be related, the 2 concepts are not interchangeable.³ In the present study, the difference observed at 5 minutes after dosing between MP-AzeFlu and placebo as well as between MP-AzeFlu and LORA/INFP already exceeded the MCID on the basis of Hedges' *g*. Hence, MP-AzeFlu reaches clinically relevant effect sizes versus placebo and LORA/INFP after 5 minutes and medium relevant effect sizes after 15 minutes, whereas LORA/INFP reaches relevant effect sizes versus placebo at 150 minutes, that is, more than 2 hours later.

Furthermore, the clinical relevance of the results from an individual perspective was assessed using a responder analysis described in regulatory guidance documents to approach this issue¹⁹ and that was reported in an RCT.²⁹ Independent from the chosen response criteria (30% or 50% TNSS reduction from baseline), the results showed that MP-AzeFlu was statistically significantly superior to LORA/INFP and placebo ($P < .05$), whereas there was no difference between LORA/INFP and placebo. Thus, the rapid onset of action should be noticed by the subjects.

It is noteworthy that all outcomes measured showed the same trend. As expected, nasal symptoms were more rapidly improved (5 minutes) than ocular ones (10 minutes) by MP-AzeFlu as reflected by TNSS, TOSS, and T7SS. VAS levels were improved similarly to T7SS because this scale also incorporates both nasal and ocular symptoms.²¹ However, the regimen of sequential

monotherapies of LORA followed by INFP was significantly effective only after 2 (TOSS, T7SS, and VAS) to 2.5 (TNSS) hours. For both MP-AzeFlu and LORA/INFP, the efficacy was maintained. All individual symptoms including ocular symptoms contributed to the overall effects. However, it can be expected that nasal symptoms are more rapidly controlled with an intranasal formulation. The faster symptom relief after MP-AzeFlu was also seen for the most bothersome rhinitis symptoms such as nasal congestion, itchy nose, and itchy eye.

Comparison with other medications shows that using an AEC, none of the INCS tested was effective before a few hours⁷ (Table II). A combination between intranasal budesonide and azelastine were effective only after 30 minutes and the effect was not sustained.¹⁵ Budesonide and fluticasone propionate have different potencies and the lack of sustained effect may be related to these properties. Intranasal azelastine showed a statistically significant improvement in the TNSS at 15 minutes compared with placebo.¹⁰ The effect was durable at each time point during the 8-hour study. Intranasal azelastine was also significantly more effective than mometasone at each time point. In a study comparing olopatadine and mometasone nasal sprays, the onset of action was 30 minutes for olopatadine and 150 minutes for mometasone. Interestingly, olopatadine was superior to both placebo nasal spray and mometasone in patient satisfaction.¹¹ Using the Vienna Challenge Chamber, another well-characterized AEC, treatment with INFP/levocabastine fixed-dose combination and levocabastine treatment led to a rapid (15 minutes) reduction of symptoms, but the statistical significance of the onset of action was not tested.³¹ These studies indicate a fast onset of action of intranasal H₁ antagonists, alone or in combination with INCS. However, a symptom relief faster than 15 minutes was never shown for any AR medication. For the first time, there is an AR medication that has been proved to improve AR symptoms within minutes in a state-of-the-art design.

CONCLUSIONS

The results of the present study are of importance in treatment algorithms and guidelines.⁴ It is clear that patients with AR often self-medicate, stop their treatment when they have few or no symptoms, and restart their treatment when they are uncontrolled. They, therefore, need a rapid-onset effective treatment. INCS are effective but their onset is slow. Even when they are added with an oral H₁ antihistamine, the onset is not greatly improved. MP-AzeFlu appears to be a useful drug in patients who desire a rapid onset of symptom improvement including

those who are not compliant to treatment. The results of the present study are in line with the recent recommendations of the Joint Task Force on Practice Parameters.^{27,32,33}

REFERENCES

- Anolik R. Mometasone Furoate Nasal Spray with Loratadine Study Group. Clinical benefits of combination treatment with mometasone furoate nasal spray and loratadine vs monotherapy with mometasone furoate in the treatment of seasonal allergic rhinitis. *Ann Allergy Asthma Immunol* 2008;100:264-71.
- Ratner PH, van Bavel JH, Martin BG, Hampel FC Jr, Howland WC III, Rogenes PR, et al. A comparison of the efficacy of fluticasone propionate aqueous nasal spray and loratadine, alone and in combination, for the treatment of seasonal allergic rhinitis. *J Fam Pract* 1998;47:118-25.
- Meltzer EO, Wallace D, Dykewicz M, Shneyer L. Minimal clinically important difference (MCID) in allergic rhinitis: Agency for Healthcare Research and Quality or anchor-based thresholds? *J Allergy Clin Immunol Pract* 2016;4:682-688.e6.
- Brozek JL, Bousquet J, Agache I, Agarwal A, Bachert C, Bosnic-Anticevich S, et al. Allergic Rhinitis and its Impact on Asthma (ARIA) guidelines—2016 revision. *J Allergy Clin Immunol* 2017;140:950-8.
- Food and Drug Administration; Center for Drug Evaluation and Research. Draft guidance for industry: allergic rhinitis: clinical development programs for drug products; 2000. Available from: <http://www.fda.gov/cder/guidance/index.htm>. Accessed February 26, 2018.
- US Department of Health and Human Services; Food and Drug Administration; Center for Drug Evaluation and Research. Allergic rhinitis: developing drug products for treatment. Guidance for industry; 2016. Available from: <https://www.fda.gov/downloads/drugs/guidances/ucm071293.pdf>. Accessed February 26, 2018.
- Katyal RK, Salapatek AM, Patel P. Establishing the onset of action of intranasal corticosteroids: Is there an ideal study design? *Allergy Asthma Proc* 2009;30:595-604.
- Pfaar O, Calderon MA, Andrews CP, Angjeli E, Bergmann KC, Bonlokke JH, et al. Allergen exposure chambers: harmonizing current concepts and projecting the needs for the future—an EAACI position paper. *Allergy* 2017;72:1035-42.
- Day JH, Briscoe M, Rafeiro E, Chapman D, Kramer B. Comparative onset of action and symptom relief with cetirizine, loratadine, or placebo in an environmental exposure unit in subjects with seasonal allergic rhinitis: confirmation of a test system. *Ann Allergy Asthma Immunol* 2001;87:474-81.
- Patel P, D'Andrea C, Sacks HJ. Onset of action of azelastine nasal spray compared with mometasone nasal spray and placebo in subjects with seasonal allergic rhinitis evaluated in an environmental exposure chamber. *Am J Rhinol* 2007;21:499-503.
- Patel D, Garadi R, Brubaker M, Conroy JP, Kaji Y, Crenshaw K, et al. Onset and duration of action of nasal sprays in seasonal allergic rhinitis patients: olopatadine hydrochloride versus mometasone furoate monohydrate. *Allergy Asthma Proc* 2007;28:592-9.
- Salapatek AM, Patel P, Gopalan G, Varghese ST. Mometasone furoate nasal spray provides early, continuing relief of nasal congestion and improves nasal patency in allergic patients. *Am J Rhinol Allergy* 2010;24:433-8.
- Patel P, Roland PS, Marple BF, Benninger PJ, Margalias H, Brubaker M, et al. An assessment of the onset and duration of action of olopatadine nasal spray. *Otolaryngol Head Neck Surg* 2007;137:918-24.
- Patel P, Patel D, Kunjibettu S, Hall N, Wingertzahn MA. Onset of action of ciclesonide once daily in the treatment of seasonal allergic rhinitis. *Ear Nose Throat J* 2008;87:340-53.
- Salapatek AM, Lee J, Patel D, D'Angelo P, Liu J, Zimmerman RO Jr, et al. Solubilized nasal steroid (CDX-947) when combined in the same solution nasal spray with an antihistamine (CDX-313) provides improved, fast-acting symptom relief in patients with allergic rhinitis. *Allergy Asthma Proc* 2011;32:221-9.
- Couroux P, Patel D, Armstrong K, Larche M, Hafner RP. Fel d 1-derived synthetic peptide immuno-regulatory epitopes show a long-term treatment effect in cat allergic subjects. *Clin Exp Allergy* 2015;45:974-81.
- Patel D, Couroux P, Hickey P, Salapatek AM, Laidler P, Larche M, et al. Fel d 1-derived peptide antigen desensitization shows a persistent treatment effect 1 year after the start of dosing: a randomized, placebo-controlled study. *J Allergy Clin Immunol* 2013;131:103-109.e1-7.
- William E. Experimental designs balanced for the residual effects of treatment. *Aust J Chem* 1949;2:149-68.
- European Medicines Agency. Guideline on the clinical development of medicinal products for the treatment of allergic rhinoconjunctivitis; 2004. Available from: http://www.ema.europa.eu/docs/en_GB/document_library/Scientific_guideline/2009/09/WC500003554.pdf. Accessed February 23, 2018.
- Bousquet J, Hellings PW, Agache I, Bedbrook A, Bachert C, Bergmann KC, et al. ARIA 2016: care pathways implementing emerging technologies for predictive medicine in rhinitis and asthma across the life cycle. *Clin Transl Allergy* 2016;6:47.
- Klimek L, Bergmann K, Biederman T, Bousquet J, Hellings P, Jung K, et al. Visual analogue scales (VAS): Measuring instruments for the documentation of symptoms and therapy monitoring in cases of allergic rhinitis in everyday health care: Position Paper of the German Society of Allergology (AeDA) and the German Society of Allergy and Clinical Immunology (DGAKI), ENT Section, in collaboration with the working group on Clinical Immunology, Allergology and Environmental Medicine of the German Society of Otorhinolaryngology, Head and Neck Surgery (DGHNOKHC). *Allergo J Int* 2017;26:16-24.
- Barnes ML, Vaidyanathan S, Williamson PA, Lipworth BJ. The minimal clinically important difference in allergic rhinitis. *Clin Exp Allergy* 2010;40:242-50.
- Day JH, Briscoe M, Widlitz MD. Cetirizine, loratadine, or placebo in subjects with seasonal allergic rhinitis: effects after controlled ragweed pollen challenge in an environmental exposure unit. *J Allergy Clin Immunol* 1998;101:638-45.
- Day JH, Briscoe MP, Rafeiro E, Ratz JD. Comparative clinical efficacy, onset and duration of action of levocetirizine and desloratadine for symptoms of seasonal allergic rhinitis in subjects evaluated in the Environmental Exposure Unit (EEU). *Int J Clin Pract* 2004;58:109-18.
- Ellis AK, Zhu Y, Steacy LM, Walker T, Day JH. A four-way, double-blind, randomized, placebo controlled study to determine the efficacy and speed of azelastine nasal spray, versus loratadine, and cetirizine in adult subjects with allergen-induced seasonal allergic rhinitis. *Allergy Asthma Clin Immunol* 2013;9:16.
- Okubo K, Nakashima M, Miyake N, Komatsubara M, Okuda M. Comparison of fluticasone furoate and fluticasone propionate for the treatment of Japanese cedar pollinosis. *Allergy Asthma Proc* 2009;30:84-94.
- Dykewicz MS, Wallace DV, Baroody F, Bernstein J, Craig T, Finegold I, et al. Treatment of seasonal allergic rhinitis: an evidence-based focused 2017 guideline update. *Ann Allergy Asthma Immunol* 2017;119:489-511.e41.
- Carr W, Bernstein J, Lieberman P, Meltzer E, Bachert C, Price D, et al. A novel intranasal therapy of azelastine with fluticasone for the treatment of allergic rhinitis. *J Allergy Clin Immunol* 2012;129:1282-1289.e10.
- Meltzer E, Ratner P, Bachert C, Carr W, Berger W, Canonica GW, et al. Clinically relevant effect of a new intranasal therapy (MP29-02) in allergic rhinitis assessed by responder analysis. *Int Arch Allergy Immunol* 2013;161:369-77.
- Patel P, Patel D. Efficacy comparison of levocetirizine vs montelukast in ragweed sensitized patients. *Ann Allergy Asthma Immunol* 2008;101:287-94.
- Murdoch RD, Bareille P, Ignar D, Miller SR, Gupta A, Boardley R, et al. The improved efficacy of a fixed-dose combination of fluticasone furoate and levocabastine relative to the individual components in the treatment of allergic rhinitis. *Clin Exp Allergy* 2015;45:1346-55.
- Wallace DV, Dykewicz MS. Seasonal allergic rhinitis: a focused systematic review and practice parameter update. *Curr Opin Allergy Clin Immunol* 2017;17:286-94.
- Wallace DV, Dykewicz MS, Oppenheimer J, Portnoy JM, Lang DM. Pharmacologic treatment of seasonal allergic rhinitis: synopsis of guidance from the 2017 Joint Task Force on Practice Parameters. *Ann Intern Med* 2017;167:876-81.

APPENDIX

Methods

Study design. The study was a single-center, randomized, active- and placebo-controlled, double-blind, double-dummy, and 3-period crossover trial (William's design).

The study consisted of a screening visit (V1), which took place on day -28 to day -1. A priming/screening visit was performed at V2 (day -1), where subjects recorded their individual nasal and ocular symptoms and their overall allergy symptoms on a VAS using ePDAT before EEC entry and during a 2-hour ragweed EEC session. V1 and V2 may have taken place on the same day if subjects met the eligibility criteria to continue to V2. At V2, if subjects met a minimum threshold TNSS of 6 or more out of a possible of 12 at least twice, with at least 1 occurring during the last 2 time points, as well as a score of at least 2/3 for runny nose at least twice, with at least 1 occurring during the last 2 time points, during hours 0 to 2 in the EEC, then they were eligible to attend V3.

At V3 (day 1), subjects recorded their individual nasal and ocular symptoms and their overall allergy symptoms on a VAS using ePDAT before EEC entry and during the EEC session for the first 2 hours. Subjects who met the predetermined minimum TNSS of 6 or more out of a possible 12 at least twice, with at least 1 occurring during the last 2 time points, as well as a score of at least 2/3 for runny nose at least twice, with at least 1 occurring during the last 2 time points, during hours 0 to 2 in the EEC and with nasal passages that were not completely blocked on either 1 or both sides as assessed by the investigator within 30 minutes before dosing, were randomized to a treatment sequence. They received a single dose of the first study treatment approximately 2.5 hours after entering the EEC. The EEC session was continued over the next 4 hours with subjects recording their individual nasal and ocular symptoms and overall allergy symptoms on a VAS using ePDAT (see [Tables E1-E3](#)).

Subjects returned to the study site after a period of at least 12 days (+ 5 days). The procedures at V4 and V5 (period 2) and V6 and V7 (period 3) were identical to those at V2 and V3, respectively, except that subjects did not need to meet a minimum threshold value for TNSS. Subjects were assessed for complete nasal blockage within 30 minutes before dosing. Any evidence of complete nasal blockage on 1 or both sides before dosing should have been documented in an electronic case report form. Subjects received the alternate study treatment in the second and third periods. Throughout the study, the investigator was to adequately treat and follow up on adverse events.

Inclusion and exclusion criteria

Inclusion criteria. To be eligible for the study, a subject had to comply with all of the following criteria:

1. Provided written informed consent.
2. Male or female subjects (childbearing and nonchildbearing potential, defined as females with no menstruation for a minimum of 12 months before screening) aged 18 to 55 years (inclusive) at screening.
3. History of seasonal AR to ragweed pollen for at least the previous 2 ragweed pollen seasons.
4. Positive SPT response to ragweed pollen (allergen-induced wheal diameter at least 3 mm larger than that achieved with the negative control). A test performed in the previous 12 months may have been used to qualify the subject.

To be eligible for V2 EEC, a subject had to additionally comply with the following criteria:

5. Asymptomatic or showed mild symptoms during the recording of symptoms before start of the screening EEC (V2):
 - TNSS of 3/12 or less, with the score for each symptom being less than 2.

To be eligible for V3, a subject had to additionally comply with the following criteria during V2 EEC:

6. Demonstrate adequate symptomology:
 - TNSS of 6/12 or more on at least 2 ePDAT time point assessments during hours 0 to 2 in the EEC (V2), with at least 1 occurring during the last 2 time points. In addition, subjects were required to meet a score of at least 2/3 for runny nose at least twice during hours 0 to 2 in the EEC, with at least 1 occurring during the last 2 time points.

To be eligible for randomization (V3), a subject had to additionally comply with the following criteria:

7. Demonstrated adequate symptomology:
 - TNSS of 6/12 or more on at least 2 ePDAT time point assessments during hours 0 to 2 in the EEC (V3), with at least 1 occurring during the last 2 time points. In addition, subjects were required to meet a score of at least 2/3 for runny nose at least twice during hours 0 to 2 in the EEC, with at least 1 occurring during the last 2 time points.
 - No evidence of complete nasal blockage on either 1 or both sides on anterior rhinoscopy within 30 minutes before dosing.

Exclusion criteria. The following were the exclusion criteria:

1. History of allergic reaction to fluticasone propionate, azelastine hydrochloride, loratadine, or one of the excipients of the study treatments (eg, benzalkonium chloride, phenyl-ethyl alcohol, and microcrystalline cellulose) or a component of the container.
2. History of anaphylaxis, cardiovascular, pulmonary, hepatic, renal, gastrointestinal, hematological, endocrine, metabolic, psychiatric, neurological, or other disease at screening that may affect subject safety during the study or evaluation of the study end points at the discretion of the investigator and/or designee.
3. Subjects with a current diagnosis of asthma or subjects with measured FEV₁ of less than 75% of the predicted value.
4. Pregnant, breast-feeding, or planning a pregnancy during the study and women of childbearing potential not using adequate contraception. Women of childbearing potential not abstinent or using highly effective methods of birth control—defined as those that result in a low failure rate (ie, <1% per year) when used consistently and correctly such as implants, injectables, combined oral contraceptives, hormonal intrauterine devices, tubal ligation, or vasectomized partner—started at least 4 weeks before screening.
5. Previous and concomitant treatments: use of prohibited therapies as specified in [Table E5](#); use of any medication considered to have an influence on the outcome of the study during the EEC session, at the discretion of the investigator and/or designee.

6. Subjects with (expected) clinically relevant symptoms at the timing of the scheduled EEC assessments because of concomitant sensitization, that is, positive SPT result (mean wheal diameter at least 3 mm larger than that achieved with the negative control) and a history of allergic response to the causative allergen, at the discretion of the investigator. Subjects with a positive SPT result for cat and/or dog allergens were acceptable if the subjects avoided cats and/or dogs for the duration of the study.
 7. Concomitant diseases: abnormalities during the screening visit that might interfere with study results as determined by the investigator and/or designee.
 8. Presence of a severely deviated septum, septal perforation, structural nasal defect, or large nasal polyps causing obstruction as determined by the investigator.
 9. Acute conditions: any acute illness within 7 days before the screening visit, including acute conjunctivitis or any other ocular infection.
 10. History of increased ocular pressure, glaucoma, and/or cataracts.
 11. Tuberculosis, untreated local or systemic fungal or bacterial infections, systemic viral or parasitic infections, or ocular herpes simplex.
 12. Recent nasal ulcers, mucosal erosion, nasal surgery, or nasal trauma, as judged by the investigator.
 13. Exposure to chicken pox or measles within 4 weeks before the screening visit or during the study.
 14. History of chronic sinusitis or no history of AR.
 15. Exposure to another investigational product within the last 30 days before screening.
 16. History of malignancy within the past 5 years, except for basal cell skin carcinomas that have been treated with no recurrence for at least 3 months.
 17. Neurological or psychiatric disease or drug or alcohol abuse that would interfere with the subject's proper completion of the protocol assignment. Subjects with a positive urine drug screen will be excluded.
 18. Subjects undergoing surgical procedures with general anesthesia within 90 days before screening or who plan to undergo surgery/hospitalization during the study.
 19. Vulnerable subjects (such as persons kept in detention).
- ### Safety
1. Five out of 82 subjects had in total 8 treatment emergent adverse events (defined as onset within 120 hours after dosing).
 - MP-AzeFlu (each once, mild and resolved): headache (likely), epistaxis (likely), hematuria (unlikely), abdominal pain (unlikely);
 - INFP/LORA (each once, mild and resolved): hyperhidrosis (likely), feeling hot (likely), paresthesia (likely);
 - Placebo (once, moderate and resolved): ocular hyperemia (unlikely).
 2. Seven out of 82 subjects had in total 9 other adverse events that could not be attributed to a special treatment (onset more than 120 hours after dosing in the crossover design). All adverse events were assessed with relationship "unlikely."
 3. One subject discontinued the study because of an adverse event (gastroenteritis, unlikely).
 4. No serious adverse events occurred during the study.

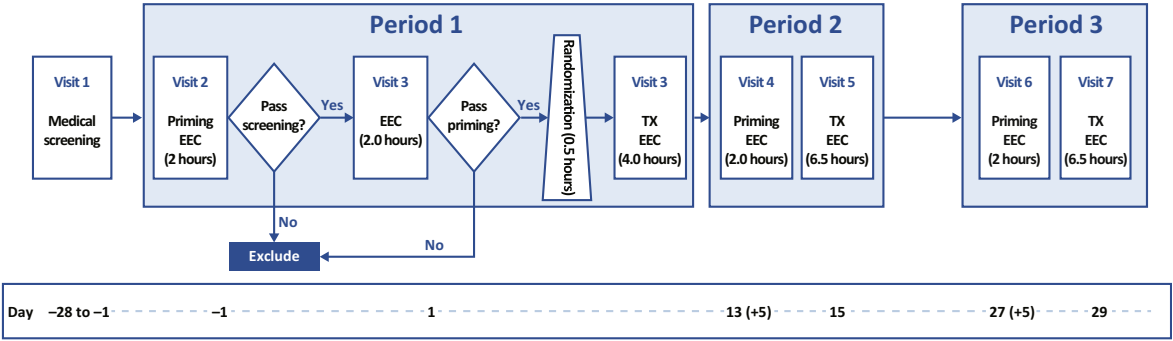


FIGURE E1. Study design.

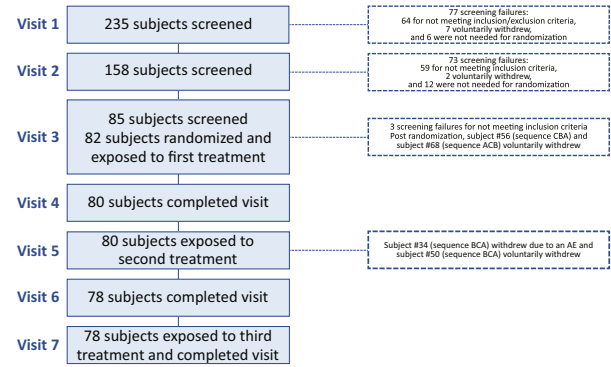


FIGURE E2. Flowchart showing the disposition of subjects.

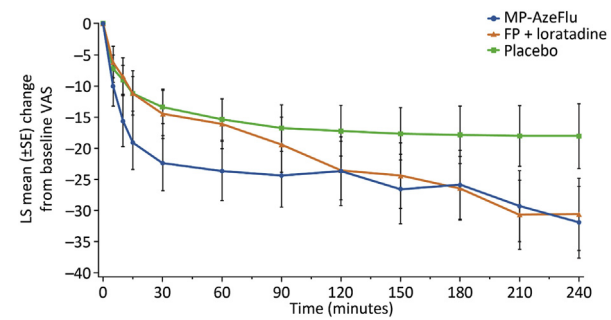


FIGURE E3. Change from baseline in VAS (0-100) for 4 h post-dosing (FAS).

TABLE E1. Demographic characteristics

Parameter	FAS (N = 82)
Sex, n (%)	
Female	44 (53.7)
Male	38 (46.3)
Age (y)	
Mean $\pm$ SD	41.6 $\pm$ 9.05
Median	42.0
Range	20-55
Racial designation, n (%)	
America Indian or Alaska Native	1 (1.2)
Asian	18 (22.0)
Black or African American	35 (42.7)
Multiple	5 (6.1)
White	23 (28.0)
Ethnicity, n (%)	
Hispanic or Latino	8 (9.8)
Not Hispanic or Latino	74 (90.2)
Height (cm)	
Mean $\pm$ SD	167.85 $\pm$ 10.38
Median	168.80
Range	138.9-191.6
Weight (kg)	
Mean $\pm$ SD	78.56 $\pm$ 16.83
Median	78.20
Range	41.4-142.7
BMI (kg/m ²)	
Mean $\pm$ SD	27.72 $\pm$ 4.65
Median	27.60
Range	19.5-48.7

BMI, body mass index.

TABLE E2. Onset of action for all treatments

Comparison	<i>P</i> value										
	5 min	10 min	15 min	30 min	60 min	90 min	120 min	150 min	180 min	210 min	240 min
TNSS											
MP-AzeFlu and INFF/LORA	.0031	<.001	<.001	<.001	<.001	<.001	.1149	.3906	.6567	.515	.1803
MP-AzeFlu and placebo	.0385	<.001	<.001	<.001	.0025	<.001	.005	<.001	<.001	<.001	<.001
INFP + LORA and placebo	.3567	.9649	.8791	.5606	.7399	.6972	.2028	.001	.0031	<.001	<.001
TOSS											
MP-AzeFlu and INFF/LORA	.0172	<.001	.0209	<.001	.0012	.0096	.6233	.2861	.8253	.9759	.4235
MP-AzeFlu and placebo	.2385	<.001	<.001	<.001	<.001	.003	.0059	<.001	.0024	.0049	<.001
INFF/LORA and placebo	.2163	.96	.2946	.5784	.5097	.6971	.0223	.0085	.0044	.0051	<.001
T7SS											
MP-AzeFlu and INFF/LORA	.001	<.001	<.001	<.001	<.001	<.001	.261	.273	.707	.624	.228
MP-AzeFlu and placebo	.051	<.001	<.001	<.001	<.001	<.001	.002	<.001	<.001	<.001	<.001
INFF/LORA and placebo	.198	.929	.805	.886	.92	.626	.049	<.001	.001	<.001	<.001
VAS											
MP-AzeFlu and INFF/LORA	.0469	.0017	.0015	<.001	.0045	.0747	.9271	.3336	.8183	.5918	.7232
MP-AzeFlu and placebo	.1256	.0026	.0012	<.001	.0015	.0048	.0249	<.001	.0068	<.001	<.001
INFF/LORA and placebo	.6424	.8879	.9596	.7052	.7268	.2877	.0301	.0155	.0031	<.001	<.001

Bold indicates first time statistical significance ($P < .05$).

TABLE E3. First significant effect for individual symptoms

Sign/symptom	Time for $P < .05$
Nasal symptoms	
Itchy nose	5 min
Runny nose	15 min
Sneezing	10 min
Congestion	10 min
Ocular symptoms	
Watery eyes	10 min
Eye redness	15 min
Itchy eyes	10 min

TABLE E4. Overall LS mean difference between treatments on change from baseline in TNSS, TOSS, T7SS, and VAS (FAS)

Parameter	Comparison	LS, mean $\pm$ SE	95% CI	P value
TNSS	MP-AzeFlu vs placebo	-0.67 ± 0.160	-0.99 to -0.36	<.001
	INFP + LORA vs placebo	-0.21 ± 0.159	-0.53 to 0.10	.182
	MP-AzeFlu vs INFP + LORA	-0.46 ± 0.160	-0.78 to -0.15	.005
TOSS	MP-AzeFlu vs placebo	-0.29 ± 0.091	-0.47 to -0.12	.001
	INFP + LORA vs placebo	-0.01 ± 0.090	-0.19 to 0.17	.914
	MP-AzeFlu vs INFP + LORA	-0.28 ± 0.091	-0.46 to -0.11	.002
T7SS	MP-AzeFlu vs placebo	-0.99 ± 0.218	-1.42 to -0.56	<.001
	INFP + LORA vs placebo	-0.31 ± 0.216	-0.74 to 0.11	.149
	MP-AzeFlu vs INFP + LORA	-0.67 ± 0.217	-1.10 to -0.24	.002
VAS	MP-AzeFlu vs placebo	-4.60 ± 1.160	-6.89 to -2.31	<.001
	INFP + LORA vs placebo	-2.50 ± 1.156	-4.78 to -0.22	.032
	MP-AzeFlu vs INFP + LORA	-2.10 ± 1.161	-4.39 to 0.19	.072

LS, least squares; SE, standard error.

TABLE E5. Prohibited treatments

Prohibited medication	Before screening	During study
Antihistaminic agents, all presentations	1 wk	Not allowed
Theophylline, all presentations	1 wk	Not allowed
Cromolyn sodium, all forms Nedocromil sodium	24 h	24 h before each allergen challenge
Corticosteroids (inhaled, oral, and intravenous)	4 wk	Not allowed
Topical corticosteroids (ocular and intranasal)	2 wk	Not allowed
Corticosteroids (intramuscular or intraarticular)	12 wk	Not allowed
Decongestants, all forms	24 h	Not allowed
Immunotherapy	6 mo	Not allowed
Systemic antibiotics	1 wk	Within 1 wk before day of visit
Any cytochrome P450 3A4 inhibiting or inducing drug (eg, ritonavir, ketoconazole, itraconazole, erythromycin, cimetidine, rifampicin, and St. John's wort [<i>Hypericum perforatum</i>])	14 d	Not allowed
Tricyclic antidepressants and MAO inhibitors*	2 wk	Not allowed

MAO, Monamine oxidase.

*Antidepressants devoid of anticholinergic effects: Fluoxetine (fluoxetine), Seroxat (paroxetine), and Trevilor (venlafaxine) are permitted during the study period provided the subject has been on a stable dose for 4 wk before screening and there is no change in the dose or regimen during the entire study.