
Onset of action of a topical antihistamine as assessed by histamine challenge–induced plasma exudation responses

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Background: Although usually administered orally, antihistamines are available also for topical use in allergic rhinitis. Information on onset of action of these drugs is incomplete.

Objective: To examine onset of action of topical cetirizine-dinitrate on plasma exudation evoked by repeated nasal histamine challenges.

Methods: A liposome formulation of cetirizine-dinitrate (2.44 mg per nasal cavity) was delivered via a nasal spray device as 2 consecutive actuations per nasal cavity in a placebo-controlled design. The nasal mucosal surface was challenged and lavaged with a histamine solution (100 $\mu\text{g/mL}$) 5, 15, 25, and 55 minutes after each treatment. In addition, the mucosa was lavaged with saline before each treatment. The lavage fluid levels of α_2 -macroglobulin were measured as an index of mucosal exudation (luminal entry) of plasma.

Results: Histamine produced significant increases in nasal lavage fluid levels of α_2 -macroglobulin at all observation points (5 through 55 minutes after treatment). Nasal cetirizine-dinitrate significantly inhibited this response at 5 and 15 minutes after treatment.

Conclusions: The effect of topical cetirizine-dinitrate, as established by histamine challenge–induced mucosal exudation of plasma, has an early onset (ie, within 5 to 10 minutes).

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INTRODUCTION

Antihistamines have been used in the treatment of allergic airway disease for approximately 50 years. Usually these drugs are given orally, but a few antihistamines are available also for topical use in allergic rhinitis. The onset of action is a major aspect of such drugs, particularly since they often are used as on-demand treatment rather than as prophylaxis.

Onset of action of antihistamines has been evaluated at natural allergen exposure¹ and in allergen challenge models,^{2–6} with a focus on symptoms of allergic rhinitis. Thus, oral antihistamines have been shown to exert their symptom-reducing effects within 1 hour,^{1,3,5,6} whereas single observations available for a topical antihistamine (levocabastine) suggest an earlier symptom-reducing onset for topical drugs (10–15 minutes).^{2,4} Further information on the onset of action of topical antihistamines is warranted, particularly focusing on their specific pharmacologic ability to counteract effects of histamine and on time points earlier than 10 minutes.

We and others have previously demonstrated that repeated nasal histamine challenges evoke reproducible plasma exu-

dation responses (extravasation and luminal or nasal entry of bulk plasma).^{7,8} Hence, repeated histamine-induced mucosal exudation of plasma can be used as a method for exploration of the pharmacologic action of antihistamines. Using this method, we have previously determined the duration of action of such drugs.⁸ Furthermore, we have shown that topical azelastine and oral cetirizine attenuate nasal histamine challenge–induced plasma exudation within 1 hour after their administration.⁸

In the present study, which involved healthy subjects, we determined the onset of action of nasal cetirizine-dinitrate using luminal entry of plasma induced by repeated histamine challenges as index. The effects of cetirizine-dinitrate have been compared with placebo.

METHODS

Study Design

The present study was of a randomized, placebo-controlled, double-blind, crossover design. Subjects were treated on 2 occasions, once with active nasal cetirizine-dinitrate and once with placebo nasal sprays. The washout time between the 2 treatments was 1 week. Combined histamine challenges and lavages were performed 5, 15, 25, and 55 minutes after each treatment. In addition, a saline lavage was performed 10 minutes before each treatment. The concentrations of α_2 -macroglobulin in lavage fluids were determined as index of mucosal exudation (luminal entry) of plasma.

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Subjects

Twelve healthy subjects 18 to 28 years of age participated in the study. The subjects had no history of allergy or recent nasal disease and no history of recent drug treatment for allergic rhinitis. The study was approved by the regional ethics committee, and informed consent was obtained.

Drug Treatment and Challenges and Lavages

Placebo and the study drug (Biolipox AB, Stockholm, Sweden) were given as 2 consecutive actuations per nasal cavity. In the study drug run, the nasal spray device delivered 110 μ L of a liposome formulation of cetirizine-dinitrate (11.1 mg/mL), resulting in a total dose 4.88 mg (2.44 mg per nasal cavity). Using a nasal pool device,^{8,9} a histamine (100 μ g/mL) solution was administered as a combined challenge and lavage solution 5, 15, 25, and 55 minutes after each treatment. The dose of histamine was chosen based on previous dose response data to produce mild to moderate plasma exudation and no or only minor symptoms.⁸ In addition to the histamine lavages, a saline lavage was performed 10 minutes before each treatment. In the present study, the volume of the nasal pool fluid was 14 mL. All solutions were maintained in the nasal cavity for 5 minutes. The lavage fluids were centrifuged (325g, 10 minutes, 4°C), and samples were obtained from the supernatant and frozen (−20°C), awaiting analysis of α_2 -macroglobulin.

Analysis of α_2 -Macroglobulin

The lavage fluid levels of α_2 -macroglobulin were measured using a radioimmunoassay sensitive to 7.8 ng/mL.⁸ Rabbit anti-human α_2 -macroglobulin (Dakopatts, Copenhagen, Denmark) was used as anti-serum and human serum (Behringwerke Diagnostica, Marburg, Germany) as standard. Human α_2 -macroglobulin (Cappel-Organon Teknika, Turnhout, Belgium) was iodinated using the lactoperoxidase method. Tracer and standard (or sample) were mixed with anti-serum before adding goat anti-rabbit anti-serum (Astra Zeneca, Lund, Sweden). The bound fraction was measured using a gamma counter (Pharmacia, Uppsala, Sweden). The intra-assay and interassay coefficients of variation were 3.8% to 6.0% and 3.1% to 7.2%, respectively.

Statistical Analysis

Differences in histamine-induced nasal lavage fluid levels of α_2 -macroglobulin between the placebo and the antihistamine treatment were analyzed by the Wilcoxon signed rank test. Differences in levels of α_2 -macroglobulin within each treatment group were analyzed by the Friedman test and the Wilcoxon signed rank test (when appropriate). $P \leq .05$ was considered statistically significant.

RESULTS

In the placebo run, histamine (100 μ g/mL) produced significant plasma exudation responses at all observation points compared with control saline challenge (Friedman test, $P < .001$) (Fig 1). In contrast, this effect was not seen in the topical cetirizine-dinitrate run (Friedman test, $P = .07$). In the

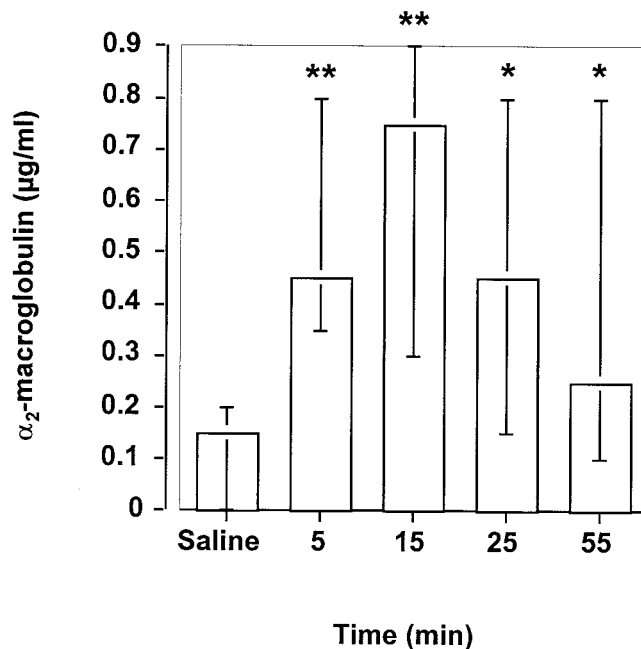


Figure 1. Levels of α_2 -macroglobulin in a saline lavage performed before placebo treatment and in histamine (100 μ g/mL) lavages performed 5, 15, 25, and 55 minutes thereafter. Histamine produced a significant plasma exudation response. Data are presented as medians and interquartile ranges (error bars). Comparisons between observations 5, 15, 25, and 55 minutes after treatment, respectively, and the observation before treatment: * $P < .05$, ** $P < .01$.

placebo run, the response to repeated histamine challenges decreased with time, but this reduction did not reach statistical significance (Friedman test, $P < .05$; Wilcoxon signed rank test, $P = .59$, .42, and .15, respectively, for comparisons between the observation at 5 minutes after treatment and the following 3 observations).

The lavage fluid levels of α_2 -macroglobulin were similar in the control saline lavages performed before each treatment ($P = .32$) (Fig 2). Cetirizine-dinitrate reduced the histamine challenge-induced mucosal exudation of plasma compared with placebo at all time points. There was a statistically significant effect within 5 to 10 minutes after treatment ($P < .01$), which was maintained through the 15-minute observation point ($P < .05$) but not thereafter (Fig 2).

DISCUSSION

The present study, which involved healthy subjects, demonstrates that a single-dose treatment with cetirizine-dinitrate nasal spray significantly reduces histamine challenge-induced mucosal exudation of plasma (α_2 -macroglobulin) within 5 to 10 minutes after its topical administration. Our results indicate that the pharmacologic effect of topical cetirizine-dinitrate has a rapid onset of action and that histamine challenge-induced exudative responses can be used to monitor this effect. With such rapid onset of action, cetirizine-

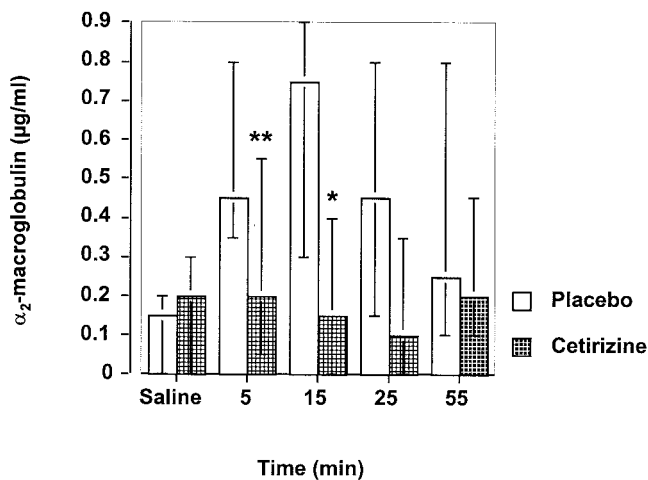


Figure 2. Effects of topical cetirizine on histamine challenge-induced plasma exudation (ie, nasal lavage fluid levels of α_2 -macroglobulin). Histamine challenges were performed 5, 15, 25, and 55 minutes after each treatment. Cetirizine reduced the histamine challenge-induced plasma exudation already after 5 to 10 minutes. Data are presented as medians and interquartile ranges (error bars). Comparisons between topical cetirizine and placebo: * $P < .05$, ** $P < .01$.

dinitrate may have potential as an on-demand treatment of allergic rhinitis. However, clinical studies evaluating symptoms are needed to explore this possibility.

In allergic rhinitis, the acute response to allergen is critically dependent on histamine, and treatment with antihistamines has marked effects in allergen challenge models.¹⁰ Histamine challenges, similar to acute allergic airway conditions, produce mucosal exudation (luminal entry) of plasma, and in the present study this effect was verified. The exudative responsiveness (to histamine) was somewhat reduced by the repeated challenges, but this feature did not reach statistical significance. Such a reduction has been observed previously⁸ and likely reflects a degree of tachyphylaxis at the level of the microvascular permeability regulating endothelial cells.¹¹ Because of this feature, the primary comparisons in the present study were made between cetirizine-dinitrate and placebo at each point after treatment.

The effect parameter in the present study was the histamine-induced plasma exudation response. Histamine acts directly on the endothelial cells of postcapillary venules to increase the macromolecular permeability of the microcirculation.¹² When this response is evoked in the superficial microcirculation of the airway mucosa, the extravasated plasma distributes in the lamina propria and then moves up between epithelial cells and produces paracellular pathways for its clearance into the airway lumen.¹³ Since the luminal entry is a prompt event and a major clearance route for exuded plasma, the extravasation process in the airways can be monitored by analyzing the concentrations of plasma proteins in airway mucosal surface liquids. It is likely that the antipermeability effects of the antihistamines, as verified by

the present study, reflect actions on histamine₁-receptors on the permeability regulating endothelial cells of the subepithelial microcirculation.¹⁴

In the present study, onset of an antiexudative action of cetirizine-dinitrate was observed within 5 to 10 minutes after its topical application. This is earlier than any onset reported for oral antihistamines that focus on symptoms of allergic rhinitis.^{1,3,5,6} It is also somewhat earlier than the 10- and 15-minute onsets that have been reported for topical levocabastine (focusing on symptoms),^{2,4} although no observations were included before these observation points. Our present findings indicate that the pharmacologic onset of action of topical cetirizine-dinitrate is rapid. Additional studies are required to determine the onset of action of topical cetirizine-dinitrate in regard to clinical efficacy. In the present study, the histamine challenge only produced limited symptoms, and it was not possible to use this parameter to evaluate effects of the treatment.

The antiexudative effect of nasal cetirizine-dinitrate was not statistically significant 25 and 55 minutes after treatment (compared with placebo). Likely, this feature of the present study does not reflect any major lack of efficacy of the study drug, since there clearly were no exudative effects of histamine in the cetirizine-dinitrate run compared with the saline challenge performed before the treatment. Rather, it may be speculated that the present low dose of histamine, which produces small to moderate plasma exudation responses, and the tendency toward tachyphylaxis to histamine, albeit not statistically significant, reduced the power to detect antiexudative effects late into the present observation period. The fact that the antiexudative effect of nasal cetirizine-dinitrate was not statistically significant compared with placebo 25 and 55 minutes after treatment likely did not correlate to any lack of clinical efficacy of topical cetirizine, since this treatment has been demonstrated to be efficacious in (perennial) allergic rhinitis at natural allergen exposure when administered 3 times daily.¹⁵

Focusing on specific histamine antagonism, onset of action of oral antihistamines has previously been examined using histamine-induced skin responses. Thus, antihistamines have been shown to attenuate histamine-induced wheal-and-flare responses, reflecting plasma exudation and vasodilation, respectively, with onset from 1 to 10 hours after administration.^{16,17} Although skin models produce specific data and may be useful, the present model is more relevant to topical antihistamines intended for allergic rhinitis, since the mechanism is studied in the relevant target organ. We suggest that the present model is useful for comparative studies of the onset of the pharmacologic action of antihistamines.

We conclude that nasal cetirizine-dinitrate reduces histamine-induced mucosal exudation of plasma in human nasal airways and that the onset of action of this effect is rapid (within 5–10 minutes). The present study confirms that histamine-induced mucosal exudation (luminal entry) of plasma, as monitored using lavage techniques, may be a useful ex-

perimental method to determine onset of action of the pharmacologic effect of antihistamines.

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