

The Effect of a Thixotropic Nasal Gel on Nasal Symptoms and Inflammatory Biomarkers in Seasonal Allergic Rhinitis

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Keywords

Allergic rhinitis · Nasal gel · Allergen challenge chamber ·
Nonpharmacological treatment · Inflammatory biomarker

Abstract

Background: Drug-free viscous nasal applications have been shown to reduce nasal symptoms in individuals with seasonal allergic rhinitis (SAR). Nascum[®]-Plus (NP), a commercially available thixotropic gel, has been designed to reduce dryness and soreness of the nasal mucosa and prevent the absorption of small particles. **Objectives:** The aim of this study was to assess the efficacy of single-dose NP in treating nasal symptoms and secretion during challenge in an allergen challenge chamber (ACC). Furthermore, the effect of this treatment on biomarkers and immune cells of the allergic cascade were measured. **Methods:** This open-label, cross-over, sequence-randomized, monocentric trial randomized 18 adults with SAR and a positive skin prick test reaction to *Dactylis glomerata* pollen to receive NP or no treatment during two 4-h ACC sessions 3 weeks apart. On Day 1, 9 subjects were challenged for 4 h with treatment, the other 9 without treatment, and vice versa on Day 22. Nasal lavage fluid and nasal filter eluate samples were obtained pre, 2, and 18 h

post challenge in the ACC. **Results:** NP significantly reduced nasal symptoms, assessed by total nasal symptom score ($p < 0.001$), and minimized nasal secretion ($p = 0.047$), while no significant effect on biomarkers and immune cells in the nasal fluid was observed. The treatment was safe and well-tolerated. **Conclusions:** The physical barrier built by NP nasal gel can be safely applied in patients with allergic rhinitis. It reduces allergic nasal symptoms and secretion, but application of a single dose does not affect local inflammatory biomarkers.

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Introduction

Allergic rhinitis (AR) is one of the most common chronic diseases, with 100 million people affected in Europe [1] and 400 million worldwide [2]. Effective pharmacological treatment options comprise intranasal glucocorticosteroids, antihistamines, and cromoglicic acid. The side effects of these substances are mild, such as seda-

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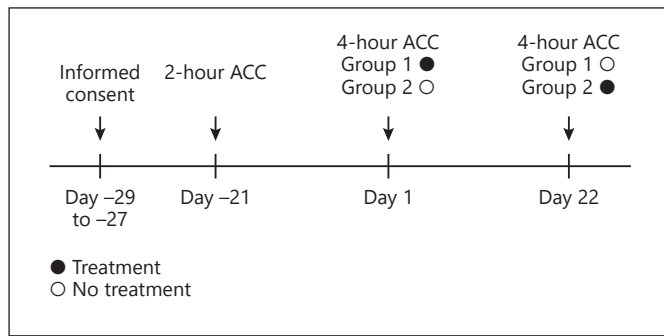


Fig. 1. Trial timeline. On Days 2 and 23, nasal lavage fluid and nasal filter eluate samples were obtained 18 h post challenge in the ACC (not depicted). ACC, allergen challenge chamber.

tion (antihistamines), irritation of the nasal mucosa (cro-moglicic acid), and rarely candidiasis (corticosteroids). Some patients with AR, however, prefer nonpharmacological therapies. These treatments are focused either on removing allergens and secretions from the nasal mucosa, such as saline nasal irrigation [3], or on reducing allergen contact ab initio. Since total avoidance is impractical in everyday life, there are several drug-free preparations commercially available that are meant to protect the nasal mucosa from allergen contact. These gels and ointments serve as a physical barrier and do not contain any pharmacologically active compounds. Some of these compositions have been investigated in clinical studies for efficacy in hay fever, for example, hypromellose [4–6], aliphatic long-chain hydrocarbons [7], ectoine [8], seawater gel [9], and xyloglucan [10]. All preparations studied have a high viscosity and are meant to entrap the pollen before they make contact with the nasal mucosa. Overall, the different products [4–10] were found to be effective in treating symptoms of AR, thus supporting the idea of a mechanical barrier against pollen. No study, however, examined the effects of these products in a controlled environment, nor their influence on biomarkers and cells of the allergic cascade.

Nascum[®]-Plus (NP, M et P Pharma AG, Emmetten, Switzerland) is a viscous, thixotropic, oily gel based on castor oil and colloidal silicon dioxide. Besides these ingredients, it does not contain any pharmacologically active compounds. The nasal gel has been designed to prevent and treat conditions of the nasal mucosa, such as dryness and soreness. Furthermore, it is meant to reduce aspiration of small particles. NP is inserted into the nose by means of a small, single-use applicator, using one for each nostril. By pressing the fingernail-sized mono-dose

device, 150 mg of the gel are released onto the nasal mucosa and subsequently need to be spread by gently massaging the nostrils. Preclinical studies have demonstrated a significant reduction of serum IgE-levels in NP-treated rats exposed to particulate matter (data on file, M et P Pharma AG). The aim of the present study was to assess the effect of a single-dose treatment with NP on nasal symptoms in patients with seasonal AR (SAR) under well-defined conditions in an allergen challenge chamber (ACC). This chamber provides a controlled environment for mechanistic and proof-of-concept studies in early-phase clinical trials, where antiallergic treatments can be tested with fewer subjects compared to field studies [11–20].

Materials and Methods

Study Design

This trial was conducted in an open-label, cross-over, sequence-randomized, monocentric design at the Fraunhofer Institute for Toxicology and Experimental Medicine ITEM (Fraunhofer ITEM, Hannover, Germany). Eighteen adults with SAR were exposed to grass pollen (*Dactylis glomerata*, Allergon AB, Ängelholm, Sweden) in 2 allergen chamber challenges 3 weeks apart (Fig. 1). On each exposure visit, one half of the subjects were challenged without any treatment, while the other half received NP. The concentration of grass pollen in the ACC was monitored continuously and kept stable for the duration of the challenges [19].

The study was performed between December 2018 and February 2019, before the beginning of the grass pollen season in Germany, which is usually in May. The screening included a 2-h challenge in the ACC to assess for eligibility, which required a total nasal symptom score (TNSS) of at least 6 out of 12. On Day 1, the 18 subjects were randomized: 9 subjects received 1 dose (150 mg) of NP into each nostril immediately before entering the ACC (Group 1), while the other 9 were challenged without treatment (Group 2). Three weeks later, on Day 22, the subjects of Group 1 underwent challenge in the ACC without treatment, while Group 2 received the nasal gel before the challenge. After the challenge, the subjects were given a salbutamol inhaler as a rescue medication and a peak flow meter. Subjects were instructed to inhale salbutamol, if the peak expiratory flow would drop by 20% or more until the following morning.

The study was approved by the Ethics Committee of the Hannover Medical School on November 29, 2018 and registered on ClinicalTrials.gov (NTC03772405). Before starting the screening procedures, written informed consent was obtained from each subject and the investigator evaluated the eligibility to participate.

Study Population

The subjects were 18–65 years old, with a history of SAR, and required a positive skin prick test reaction to *Dactylis glomerata* within 12 months prior to enrolment, a TNSS of at least 6 during a 2-h screening allergen challenge in the ACC, a normal lung function with a forced expiratory volume of at least 80% predicted, and they had to be nonsmokers or ex-smokers for at least 6

months prior to screening, with <10 pack years. During physical examination at the screening visit, abnormal nasal anatomy such as turbinate hypertrophy was excluded by nasal inspection. Subjects with asthma were eligible only if their asthma was mild and controlled.

The main exclusion criteria comprised rhinitis within 14 days prior to enrolment, specific immunotherapy within 2 years prior to the study, asthma requiring more than inhaled short-acting beta-2 agonists, an acute infection within 4 weeks prior to the informed-consent visit, any contraindication for the use of adrenaline such as therapy with beta-blockers or ACE inhibitors, treatment with systemic glucocorticoids within 8 weeks before allergen challenge, treatment with inhaled or intranasal glucocorticosteroids within 4 weeks before allergen challenge, treatment with loratadin or desloratadin within 1 week before allergen challenge, treatment with other systemic H1 antihistamines, leukotriene receptor antagonists, or topic antiallergic treatment within 72 h before allergen challenge; furthermore, participation in another clinical trial within 30 days prior to enrolment.

Assessments

Nasal Symptoms

The primary endpoint was the change in the TNSS assessed during the pollen challenge after treatment with NP compared to no treatment. During the stay in the allergen chamber, subjects completed questionnaires to assess their nasal symptoms using the TNSS every 20 min. The TNSS is defined as the sum of nasal symptoms of the subscales congestion, rhinorrhea, itching, and sneezing, each of which is scored on a scale from 0 (no symptoms) to 3 (severe symptoms). This results in a TNSS number ranging from 0 to 12. In order to rank the resulting score, the TNSS was divided into 4 severity ranges: 0 = no symptoms; 1–4 = mild symptoms; 5–8 = moderate symptoms, and 9–12 = severe symptoms.

Nasal Secretion

A secondary endpoint was the change in nasal secretion during the pollen challenge after treatment with NP compared to no treatment. Before entering the ACC, subjects were handed out pre-weighed sachets with tissues, one sachet for each hour of the ACC stay. Each sachet was weighed afterwards and the pre-weight was subtracted from the corresponding post-weight in order to assess weight gain.

Biomarker and Cell Analysis

Further secondary endpoints were changes in local proinflammatory biomarkers in nasal secretion and the change in lactate dehydrogenase (LDH) levels in the blood. LDH, an enzyme, is an unspecific indicator of tissue injury. It was previously used in pre-clinical experiments as a cytotoxic marker from bronchoalveolar lavage fluid, enabling detection of the inflammatory response in the lungs of rats after exposure to particulate matter, and was, therefore, included in this study. The biomarkers were evaluated from nasal filter eluates pre and post allergen challenge [21]. The cytokines eotaxin, interleukin-3 (IL-3), IL-4, IL-5, IL-6, macrophage inflammatory protein-1 β , and tumor necrosis factor α were analyzed in the samples collected pre, 2 h post, and 18 h post challenge, using multiplex assays by Meso Scale Discoveries (Rockville, MD, USA). Histamine was analyzed in eluates obtained from the samples collected pre and 2 h post challenge, using ELISA systems by IBL International (Hamburg, Germany).

Additionally, changes in eosinophil, neutrophil, macrophage, lymphocyte, and monocyte concentrations in nasal lavage fluid pre, 2, and 18 h post challenge were measured. The cells in nasal lavage fluid were centrifuged, resuspended, and the cells of an aliquot stained with trypan blue were counted in a hemocytometer. Furthermore, cells were then differentiated using cytopins that were stained and counted by light microscopy.

Safety

Safety was assessed by documentation of adverse events and measurement of vital signs (noninvasive blood pressure and heart rate) before and immediately after the challenge in the ACC.

Statistical Analysis

The sample size was calculated based on the assumption that the mean increase in TNSS during allergen challenge in patients with AR is 5.4 with a SD of 1.4. By including 15 subjects and setting alpha to 0.05 (one-sided), it is possible to detect a difference in TNSS of 1.0 with a power of 0.84 and a difference in TNSS of 2.0 with a power of 0.99. To assure the minimum per-protocol size of 15 analyzable subjects, 18 subjects had to be randomized.

The alternative hypothesis for the primary endpoint was that application of NP prior to a challenge in the ACC could reduce allergic nasal symptoms assessed with the TNSS. This one-sided assumption was based on data of an unpublished clinical study conducted at Fraunhofer ITEM with another thixotropic gel that significantly reduced nasal symptoms during challenge in the ACC. For statistical analysis in the present study, the combined TNSS means of all subjects with and without NP were compared and checked for tendency using descriptive statistics. In case of a positive tendency, a one-sided paired *t* test was performed to check for statistical significance. The same procedure was applied to assess the change in nasal secretion. In order to evaluate the duration of the effect of NP, a two-sided *t* test was performed with the combined mean TNSS values of all subjects for each measurement time with and without treatment. To control the false discovery rate of the resulting 12 *t* tests, the Benjamini-Hochberg correction procedure for multiple testing was applied [22].

The alternative hypothesis for the secondary endpoints was that application of NP prior to a challenge in the ACC could alter the increase in proinflammatory biomarkers in nasal secretions and the immune cells in nasal lavage fluid. The change for each biomarker and cell type over time was evaluated using an ANOVA model with the factors period and treatment. This was followed by two-sided paired *t* tests with an alpha value of 5% for individual comparison of measurement times and groups.

LDH was assessed as baseline measurement at the screening visit and measurements were repeated before and 2 h after allergen challenge on each challenge visit. The measured parameters were statistically analyzed using a two-sided paired *t* test.

Results

Study Population

In total, 18 subjects were randomized to receive study medication, 6 of them female and 12 male, aged 22–65 years (mean age 46 years). The demographics of the study

Table 1. Demographics and allergen sensitizations

Randomization number	Demographics		Asthma history	Allergen sensitizations					Dermatophagoides (house dust mite); nd, not done, summary information for age and body weight is given as mean ± SD.										
	gender	age, years		body weight, kg	dactylis glomerata	grass mix	rye	early blooming trees	late blooming trees	mugwort	ribwort	ragweed	D. pteronyssinus	D. farinae	dog	cat	horse	fungi	
101	Female	37	81	+	+	+	+	+	+	+	-	-	-	+	-	-	-	-	
102	Male	65	72	-	+	+	+	+	-	-	+	-	-	-	+	+	-	+	
103	Male	58	83	-	+	+	+	+	-	-	+	-	-	-	+	+	-	-	
104	Male	36	102	-	+	+	+	-	nd	-	-	-	-	+	-	-	-	-	
105	Male	56	74	-	+	+	+	+	+	+	+	+	-	-	-	-	-	-	
106	Male	60	75	-	+	+	+	+	+	+	+	+	+	+	-	-	-	-	
107	Male	59	79	-	+	+	+	-	-	-	+	-	-	-	-	+	-	-	
108	Female	53	70	+	+	+	+	-	-	-	+	+	-	-	+	+	-	-	
109	Male	42	95	-	+	+	+	+	nd	+	+	+	-	-	-	-	-	-	
201	Male	49	86	-	+	+	+	+	+	-	+	+	-	-	-	-	-	-	
202	Male	50	83	-	+	+	+	+	nd	-	-	-	-	-	-	+	-	-	
203	Male	28	88	-	+	+	+	+	+	-	+	+	+	-	-	+	+	+	
204	Female	22	70	-	+	+	+	+	+	-	+	+	-	+	+	+	-	+	
205	Female	23	76	-	+	+	+	nd	nd	nd	nd	nd	nd	+	+	+	nd	nd	
206	Female	45	73	+	+	+	+	+	-	-	+	+	-	+	+	-	-	-	
207	Female	40	66	-	+	+	+	+	+	+	-	-	-	-	-	-	-	-	
208	Male	42	76	-	+	+	+	+	+	-	-	-	-	+	+	+	-	-	
209	Male	54	83	-	+	+	+	+	+	+	+	+	+	+	-	-	-	-	
n (range)	Male 12 Female 6	46±12 (22–65)	79.6±8.9 (66–102)	3	18	18	18	17	13	10	5	12	4	5	3	7	9	0	3

D, Dermatophagoides (house dust mite); nd, not done, summary information for age and body weight is given as mean ± SD.

Table 2. Combined TNSS means of all participants, ranked in ascending order for individual *p* value for each measurement time, with Benjamini-Hochberg correction procedure for multiple testing [22]

Test no	Time	Without gel	With gel	Individual <i>p</i> value	Critical value	Significance
1	80	6.6	4.2	0.00048	0.0042	Yes
2	160	6.9	4.1	0.00098	0.0083	Yes
3	180	6.9	4.1	0.00098	0.0125	Yes
4	40	5.2	3.0	0.00210	0.0167	Yes
5	200	7.0	4.7	0.00413	0.0208	Yes
6	120	6.8	4.4	0.01159	0.0250	Yes
7	100	6.9	4.7	0.01215	0.0292	Yes
8	60	5.8	4.3	0.01520	0.0333	Yes
9	140	6.4	4.4	0.01520	0.0375	Yes
10	240	6.3	5.1	0.05125	0.0417	No
11	20	3.7	2.8	0.07217	0.0458	No
12	220	6.5	5.2	0.11916	0.0500	No

Critical value for each individual *p* value = (i/m)Q. i: rank of individual *p* value, m: total number of tests, Q: false discovery rate.

TNSS, total nasal symptom score.

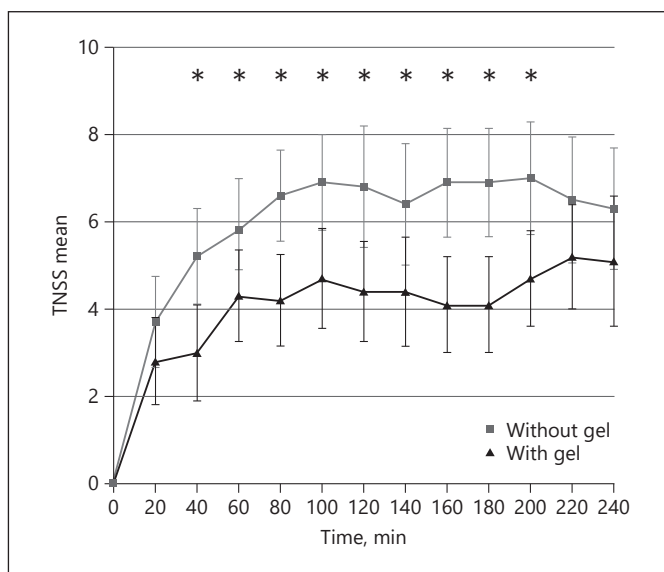


Fig. 2. Combined TNSS means of all participants for each measurement time. Asterisks indicate a significant difference compared with no treatment. Whiskers display 95% CI for each mean. TNSS, total nasal symptom score.

participants and details on their sensitizations are depicted in Table 1. All 18 randomized subjects completed the study. There were no major protocol deviations during the trial.

Nasal Symptoms

The TNSS mean was 6.3 (95% CI 5.2–7.4) without NP and 4.3 (95% CI 3.4–5.2) with NP, indicating that the gel reduced allergic nasal symptoms ($p = 0.00052$). Overall, NP treatment reduced the TNSS by 32%. One participant experienced worsening of nasal symptoms (TNSS increase by 1.6 with gel), one did not report any difference between NP treatment and no treatment. Thus, 89% of all participants (16/18) experienced an improvement with NP.

The gel had a statistically significant effect at all times, except for the assessments at 20, 220, and 240 min (Table 2). As can be seen in Figure 2, the 2 curves converge between 180 and 240 min. The decreasing difference between the TNSS means with and without treatment indicates that the effect of the gel begins to wear off after 3 h.

As for the TNSS subscales (nasal congestion, rhinorrhea, itching, and sneezing), NP improved all symptoms significantly compared to no treatment. Exemplary analysis of the rhinorrhea subscale revealed that the percentage of subjects who reported rhinorrhea when treated was never above 50%. On the contrary, without treatment, always >50% of subjects experienced rhinorrhea (apart from the 20-min assessment).

Figure 3 demonstrates the distribution of symptom severity for each measurement time. With NP treatment, moderate or severe symptoms were experienced less frequently during the allergen challenge (45%), while the reverse was observed without treatment (72%).

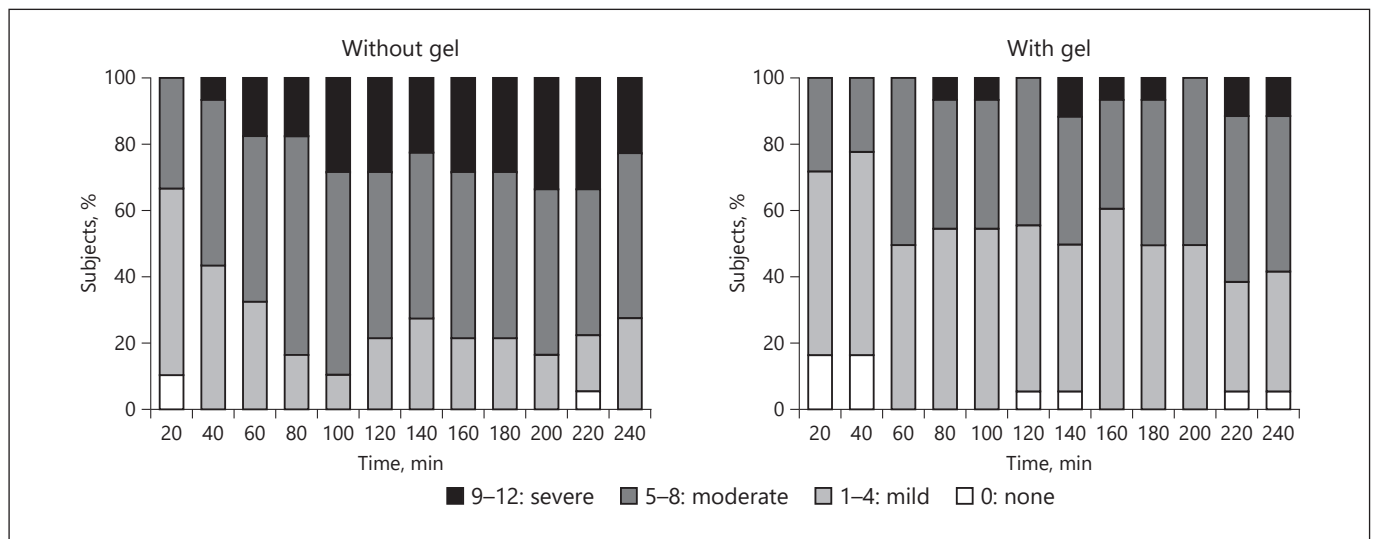


Fig. 3. Distribution of TNSS assessments to 4 severity ranges for each measurement time.

Nasal Secretion

Regarding nasal secretion, the combined mean was 3.1 g (95% CI 0.81–5.39 g) without NP and 2.2 g (95% CI 0.47–3.93 g) with NP. Thus, NP significantly reduced the amount of nasal discharge ($p = 0.047$).

Biomarker Analysis

The majority of nasal biomarkers were found to be significantly increased at 2 h post challenge, both with and without treatment. When looking at the subgroup of untreated subjects (i.e., without gel), IL-4 did not change over time. At 18 h post challenge, most nasal biomarkers were still significantly elevated but numerically lowered compared with 2 h post challenge. Importantly, the analysis showed no significant difference between NP treatment and no treatment for any biomarker at 2 or 18 h after allergen challenge (Table 3). In contrast, IL-5 and tumor necrosis factor α were found not to be significantly increased at 2 h post challenge in the subjects treated with NP (i.e., with gel). The increase for these 2 biomarkers was significantly different at 18 h post challenge. Also, when both treatment groups were compared regarding the post-challenge measurement times, no significant difference was detectable, making a meaningful difference with gel treatment unlikely.

Cell Analysis

In most nasal lavage samples, lymphocytes and monocytes were virtually absent, while eosinophils and macro-

phages were low but clearly measurable at baseline. Neutrophils were the dominant cell type at baseline. Eosinophil granulocytes increased significantly after allergen challenge, but neutrophil granulocytes and macrophages did not change. There was no significant difference between the treated and untreated groups 2 and 18 h after the allergen challenge (Table 4).

Lactate Dehydrogenase

LDH in the blood was measured at the screening visit as well as pre and 2 h post challenge in the ACC. Compared with baseline, no significant difference was detected 2 h after challenge with ($p = 0.523$) nor without gel ($p = 0.488$). Furthermore, there was no difference in LDH levels between treated and nontreated subjects at 2 h post challenge ($p = 0.298$).

Safety

During the study, 7 adverse events were reported by 5 of 18 subjects. There were no serious adverse events. Two subjects experienced a headache on 2 ACC days. One of them had a moderate headache late into the night of the last ACC day and took analgesic medication, which resolved the symptoms. This was the only adverse event that needed treatment; all other symptoms reported did not require any intervention. Two subjects reported prolonged running nose late into the evening after the allergen challenge (one with and one without gel).

Table 3. Biomarker combined means with and without gel with *p* values for measurement times and treatment groups

Biomarker	Application	Pre	2 h post	<i>p</i> value (pre/2 h post)	<i>p</i> value 2 h post (with and without)	18 h post	<i>p</i> value (pre/18 h post)	<i>p</i> value 18 h post (with and without)
Histamin, ng/mL	Without gel With gel	2.27 2.77	26.23 23.02	0.010 0.010	0.3697	– –	– –	–
Eotaxin, pg/mL	Without gel With gel	4.59 4.93	18.02 22.99	0.015 0.001	0.343	13.74 14.32	0.012 0.001	0.861
IL-3, pg/mL	Without gel With gel	0.51 0.53	4.51 3.74	0.001 0.034	0.357	0.58 0.61	0.145 0.037	0.684
IL-4, pg/mL	Without gel With gel	0.51 0.51	1.21 1.45	0.129 0.127	0.542	0.53 0.53	0.331 0.331	0.331
IL-5, pg/mL	Without gel With gel	0.99 1.06	35.23 42.19	0.034 0.142	0.596	7.31 5.83	0.004 0.033	0.518
IL-6, pg/mL	Without gel With gel	3.23 2.54	8.18 10.19	0.002 0.004	0.399	6.3 5.13	0.018 0.042	0.371
MIP-1 β , pg/mL	Without gel With gel	7.2 7.53	33.41 49.09	0.048 0.001	0.178	35.17 24.68	0.018 0.003	0.350
TNF- α , pg/mL	Without gel With gel	0.97 0.93	2.01 1.8	0.023 0.102	0.434	1.17 1.06	0.283 0.083	0.540
LDH, U/L	Without gel With gel	164.22 170.39	162.17 167.44	0.488 0.523	0.298	– –	– –	–

IL, interleukin; MIP-1 β , macrophage inflammatory protein-1 β ; TNF- α , tumor necrosis factor α ; LDH, lactate dehydrogenase.

Table 4. Combined means of relative percentages of cells in nasal lavage fluid with and without gel, with *p* values for measurement times and treatment groups

Cell type, %	Application	Pre	2 h post	<i>p</i> value (pre/2 h post)	<i>p</i> value 2 h post (with and without gel)	18 h post	<i>p</i> value (pre/18 h post)	<i>p</i> value 18 h post (with and without gel)
Eosinophil Granulocytes	Without gel With gel	1.06 0.74	10.19 14.37	0.01 0.001	0.276	10.10 6.25	0.003 0.05	0.411
Neutrophil Granulocytes	Without gel With gel	4.59 4.93	18.02 22.99	0.015 0.001	0.340	13.74 14.32	0.012 0.001	0.860
Macrophages	Without gel With gel	0.51 0.53	4.51 3.74	0.001 0.034	0.360	0.58 0.61	0.145 0.037	0.680
Lymphocytes	Without gel With gel	0.11 0.05	0.06 0.09	0.240 0.664	0.388	0.13 0.53	0.757 0.397	0.418
Monocytes	Without gel With gel	0.01 0	0.17 0.092	0.005 0.042	0.30	0.05 0.05	0.140 0.025	0.904

The investigator did not judge any of the reported adverse events to be related to the investigational medication. Rather, these are common side effects of the prolonged allergen exposure.

Noninvasive blood pressure and heart rate were measured before and immediately after allergen challenge. No deviations >30% in systolic or diastolic blood pressure and heart rate were observed. Both with and without gel, 3 subjects experienced changes of blood pressure or heart rate by between 20 and 30% (clinically not significant).

As for the assessment of lung function parameters, no subject experienced a drop of peak expiratory flow by >20% during and after the allergen challenge until the following morning.

Discussion/Conclusion

This 23-day open-label, cross-over, sequence-randomized, monocentric trial assessed the effect of a single-dose nasal application of NP on nasal symptoms, local biomarkers, and immune cells before, during, and after grass pollen challenges in an ACC.

We found NP to significantly reduce nasal symptoms, assessed by the TNSS, during *Dactylis glomerata* pollen exposure. Analysis of TNSS data over time indicated that under ACC conditions, the effect of the gel might begin to wear off after 3 h (cf. Table 2). Overall, moderate and severe symptoms were experienced less frequently with treatment. These observations are in line with the results of trials investigating other gel preparations [4, 5, 7, 9, 10]. Furthermore, we found NP to significantly reduce nasal secretions, assessed by pre- and post-weight comparison of tissue-filled sachets. This reduction in an objective outcome measure supports the earlier described reduction of clinical symptoms as subjective patient-reported outcome measure assessed by the TNSS. While the gel reduced subjective and objective outcome measures for symptoms, it did not significantly change the concentration of inflammatory biomarkers in nasal secretions, nor the number of immune cells in nasal lavage fluid, nor LDH in the blood 2 and 18 h after challenge in the ACC, compared to no treatment.

Hence, there is a discrepancy between the findings of the clinical assessments (TNSS, nasal secretion), which demonstrated a reduction of symptoms, and the inflammatory biomarker assessments, which did not find any significant differences between treatment and no treatment. A considerable factor might be the time of sampling in relation to the effect duration of NP. The TNSS

data showed that the effect of NP begins to wear off 3 h after the start of challenge in the ACC. The first laboratory samples were taken 6 h after the start of the allergen exposure in the ACC (2 h after the end of each challenge), at a time when the nasal mucosa was likely to have had contact with pollen and the allergic cascade was in full progress. Had early-phase biomarkers such as histamine or eotaxin been measured during the first half of the challenge in the ACC, differences between treatment and no treatment might have been captured. Unfortunately, nasal lavage and nasal filter sampling would have had a profound impact on the integrity of the nasal gel treatment, and it was deemed inappropriate to collect biospecimens during allergen exposure. Therefore, the above-described hypothesis cannot be tested in this setting. Also noteworthy, the study was not designed and powered to detect differences in biomarkers but based on clinical outcome measures (TNSS), which showed low intra- and intersubjective variability compared with biomarkers [23]. Thus, potential effects of NP on biomarkers would need additional studies for a conclusive evaluation.

A major limitation of this study was the lacking placebo. Since NP does not contain any pharmacologically active compound, we refrained from using a placebo. Worth noting, some other studies investigating comparable drug-free nasal applications did apply a placebo-controlled design. For example, Schwetz et al. [7] used a water-based placebo ointment with carboxymethyl cellulose against a pollen blocker cream. The placebo significantly reduced nasal symptoms, although less pronounced than the verum. The group assumed that carboxymethyl cellulose actually had a barrier effect, thus it might not have been a real placebo. Creating an adequate placebo for a drug-free viscous gel is not possible in our opinion.

Another limitation of our study was the rather small sample size. However, as the ACC has excellent reproducibility [23], the study design with intraindividual cross-over was able to provide a power of 84% in detecting a TNSS treatment difference of 1 and a power of 99% for a TNSS treatment difference of 2 with only 15 subjects. Therefore, this can also be regarded as a strength in detecting treatment differences with small subject numbers. Another strength of the study design might be the option to include biomarker sampling under well-standardized conditions.

Finally, similar to other investigated nonpharmacological AR treatments [4–7, 9, 10], NP was safe and well tolerated. Adverse events were mild/moderate in intensity and not regarded to be related to study treatment.

Overall, NP appears to have a positive risk-benefit ratio and might be a nonpharmacological treatment alter-

native for reducing AR symptoms. Future research on physical pollen barrier preparations should be conducted under standardized and controlled conditions to make inter-study results comparable. Also, objective clinical and laboratory assessments should be included to enhance the findings of self-reported symptoms and enlighten about the anti-inflammatory potential of physical barrier gel preparations.

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Statement of Ethics

This study was approved by the local competent authority and the Ethics Committee of the Hannover Medical School (Hannover, Germany, Vote No. 8101_MPG_OG_S_2018). It was conducted in accordance with the Declaration of Helsinki (Brazil Amendment, 2013), the European Union Medical Devices Directive 93/42/EEC (June 1993), the German Medical Devices Act (§§20 and 21, August 2, 1994), the European Standard EN ISO 14155:2011 + AC: 2011 (Clinical investigation of medical devices for human subjects – Good Clinical Practice), and the Guideline for Good Clinical Practice E6(R2) EMA/CHMP/ICH/135/1995.

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Author Contributions

N.S. contributed to the study conduct, statistical analysis, and writing of the manuscript. P.B. contributed to the design and conduct of the study and writing of the manuscript. C.M. contributed to conception and design of the study, reviewed the article, and gave final approval of the version to be published. U.M. contributed to conception and design of the study and gave final approval of the version to be published. J.M.H. contributed to study design, interpretation of results, and writing of the manuscript.

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