

Comparison of the efficacy and mechanisms of intranasal budesonide, montelukast, and their combination in treatment of patients with seasonal allergic rhinitis

Hui Chen, MD^{1,2,3}, Hongfei Lou, MD^{1,2}, Yang Wang, MBBS², Feifei Cao, MBBS³, Luo Zhang, MD, PhD^{1,2,3}  and Chengshuo Wang, MD, PhD^{1,2,3}

Background: Although intranasal steroids and anti-cysteinyl-leukotriene-receptor antagonists are efficacious in the treatment of seasonal allergic rhinitis (SAR), combinations of these agents have not unequivocally been demonstrated to be superior to the individual drugs. We aimed to compare the efficacy and potential mechanisms of budesonide nasal spray (BD), oral montelukast (MNT), and combination therapy comprising a half-dose of budesonide plus montelukast (hBD+MNT) in SAR patients.

Methods: We performed a single-center, randomized, open-label study in SAR subjects (n = 100). Participants were randomized to receive BD (256 μ g), MNT (10 mg), or hBD (128 μ g)+MNT for 14 days. Symptom severity scores, nasal cavity volume (NCV), fraction of exhaled nitric oxide (FeNO), eosinophil cationic protein (ECP), histamine and cysteinyl-leukotrienes (CysLTs), and T-cell subsets were assessed before and after treatment.

Results: All treatments significantly improved symptoms from baseline; however, hBD+MNT produced significantly greater improvements in nasal congestion compared with BD or MNT alone. The BD and hBD+MNT groups had fewer patients with uncontrolled symptoms and improved NCV to a greater level than the MNT group. FeNO was

decreased to a significantly greater extent from baseline after hBD+MNT treatment than after BD and MNT treatments. ECP, histamine, and CysLTs showed significantly greater decreases after BD and hBD+MNT treatments than after MNT treatment. BD decreased T-helper 1 (Th1) and Th2 cells and increased T-regulatory (Treg) cells in nasal mucosa and MNT decreased Th1 cells and increased Treg cells in peripheral blood, and this trend was reflected with hBD+MNT.

Conclusion: The hBD+MNT combination may have an overall better efficacy profile than BD and MNT monotherapy for treatment of SAR. © 2018 ARS-AAOA, LLC.

Key Words:

budesonide; combination therapy; eosinophil cationic protein; exhaled nitric oxide; montelukast; seasonal allergic rhinitis; T-cell subsets

How to Cite this Article:

Chen H, Lou H, Wang Y, et al. Comparison of the efficacy and mechanisms of intranasal budesonide, montelukast and their combination in treatment of patients with seasonal allergic rhinitis. *Int Forum Allergy Rhinol.* 2018;00:XXX-XXX.

¹Department of Otolaryngology–Head and Neck Surgery, Beijing TongRen Hospital, Capital Medical University, Beijing, China; ²Beijing Key Laboratory of Nasal Diseases, Beijing Institute of Otolaryngology, Beijing, China; ³Department of Allergy, Beijing TongRen Hospital, Capital Medical University, Beijing, China

Correspondence to: Luo Zhang, MD, PhD, Beijing Institute of Otolaryngology, No. 17, Hou Gou Hu Tong, Dong Cheng District, Beijing 100005, China; e-mail: dr.luozhang@139.com

L.Z. and C.W. and contributed equally to this study.

Funding sources for the study: the National Key R & D Program of China (2016YFC20160905200); the program for Changjiang scholars and Innovative Research Team (IRT13082); the National Natural Science Foundation of China (81420108009, 81400444, 81470678, and 81630023); the Capital Health Development Foundation (2016-1-2052); the Beijing Municipal

Allergic rhinitis (AR) is defined as symptoms of sneezing, rhinorrhea, nasal obstruction, and pruritus caused by immunoglobulin E (IgE)-mediated reactions against inhaled allergens and involving mucosal inflammation, which is driven by T-helper 2 (Th2) cells.¹ Epidemiologically, AR has become a global health problem that exacts a heavy social and economic burden on both the affected individual and society alike.²

Administration of Hospitals' Mission Plan (SML20150203); and the Capital Citizenry Health Program (z161100000116062).

Potential conflict of interest: None provided.

Received: 5 March 2018; Revised: 7 July 2018; Accepted: 26 July 2018

DOI: 10.1002/alr.22197

View this article online at wileyonlinelibrary.com.

Given the troublesome impact of AR, successful treatment of the condition is of particular importance. In clinical practice, the commonly used anti-inflammatory agents, such as intranasal steroids (INS), leukotriene-receptor antagonists, and antihistamines, are recommended by the Allergic Rhinitis and its Impact on Asthma (ARIA) guidelines as the first-line choice for treating AR.³ INS are undoubtedly effective and safe anti-inflammatory agents, which work by controlling the rate of protein synthesis through binding to a single glucocorticoid receptor.^{4,5} However, the potential adverse effects of long-term use, such as nasal burning, nasal dryness, sore throat and headache, epistaxis, cataracts and glaucoma, and effects on bone density and growth,^{5,6} may lead to lower compliance in their use as prescribed when compared with other drugs.⁶ A more recent study from China investigating the awareness of and phobias about topical corticosteroids in parents of infants with eczema has indicated that “steroid phobia” due to adverse effects may also contribute to lower compliance.⁷ The authors demonstrated that, although 95.7% of parents were very concerned about the side effects of topical steroids, 42% of them did not use a topical steroid ointment for recurrence of eczema because of concerns about use of these agents.⁷ Furthermore, INS have a slow onset of action and require 1–2 weeks to achieve maximum efficacy.⁸ In contrast to INS, leukotriene-receptor antagonists bind to cysteinyl-leukotriene receptors to block the effect of cysteinyl-leukotrienes secreted in the early and late phases of the AR process to attenuate nasal allergy symptoms, particularly nasal congestion. These agents were initially developed as a therapy for asthma, although they are now increasingly being used in the treatment of seasonal and perennial allergic rhinitis (SAR and PAR, respectively).⁹ Although studies comparing the efficacy of INS and anti-cysteinyl-leukotriene receptor antagonists have indicated INS to be more effective,^{10,11} some studies investigating the efficacy of a combination of these drugs have provided slightly conflicting efficacy data in comparisons with monotherapy.^{12,13} We have conducted an open-label study to compare the efficacy and safety of intranasal budesonide, oral montelukast, and a combination of the 2 drugs in mugwort-sensitized AR patients with moderate-to-severe seasonal AR during the pollen season, as mugwort is the most prevalent aeroallergen in northern China.¹⁴ However, in the present study, for the combination therapy, we employed only half the dose of budesonide in combination with another drug to improve patient compliance of the drugs prescribed, as this was an open-label study in outpatients, and to reduce the risk of adverse effects and “steroid phobia” among potential participants. In addition, we explored the putative underlying mechanisms of the different drugs by evaluating changes in the levels of inflammatory mediators in nasal secretions and T-cell subsets in the nasal mucosa and peripheral blood, before and after 2 weeks of treatment.

Patients and methods

Study design

One hundred patients (age, 18–60 years) with moderate to severe seasonal AR induced by mugwort, were recruited into this single-center, randomized, open-label study from the Outpatient Department at Beijing TongRen Hospital, Beijing, China, during the mugwort pollen season from July 2016 to September 2016. Severity of disease was classified according to ARIA guidelines,³ and sensitization to mugwort allergen was confirmed using the serum-specific IgE test (UniCAP; Phadia, Uppsala, Sweden). Atopy was defined as IgE levels >0.35 kU/L. No patient had received any therapy for AR or antibiotics for at least 7 days before the study. All subjects were nonsmokers and without asthma (based on patient history and pulmonary function testing results showing forced expiratory volume in 1 second [FEV₁]/predicted ≥80%, forced vital capacity [FVC]/predicted ≥80%, FEV₁/FVC ≥70%, and negative bronchodilator test results showing ≤12% and 200-mL improvement in FEV₁ from the pre-bronchodilator value after β -agonist administration) or any other chronic disease. Pregnant women were also excluded from the study.

Each eligible participant was allocated to 1 of the 3 treatment groups according to a random number sequence set ranging from 1 to 100, which was generated by computer using SPSS version 22 (IBM SPSS Corp, Armonk, NY). The patients were assigned a random sequence number consecutively at recruitment and then treated accordingly for 2 weeks with either: (i) 2 sprays of intranasal budesonide (Rhinocort; AstraZeneca AB, Cambridge, UK) at 64 μ g per spray twice daily (total 256 μ g budesonide per day; BD group); (ii) 1 tablet of 10 mg montelukast (Merck Sharp & Dohme Australia Pty, Ltd, NSW, Australia) in the evening (MNT group); or (iii) a combination of 2 sprays of BD in the morning (1 spray per nostril, total 128 μ g BD per day) and 1 tablet of 10 mg MNT in the evening (hBD+MNT group). At the beginning and end of treatment, the severity of nasal symptoms (including nasal congestion, rhinorrhea, itching, and sneezing) and eye itching was assessed using a visual analog scale (VAS); nasal cavity volume (NCV) was assessed by acoustic rhinometry; fractional exhaled nitric oxide (FeNO) from lower airway was assessed using a nitric oxide (NO) analyzer; and samples of nasal secretions, nasal exfoliative cells, and peripheral blood were collected for assessment of eosinophil cationic protein (ECP), histamine, and cysteinyl-leukotrienes (CysLTs) using enzyme-linked immunoassay (ELISA), and T-cell subsets using flow cytometry (Fig. 1).

The study was approved by the medical ethics committee of Beijing TongRen Hospital, and written informed consent was obtained from each participant before enrollment in the study.

Visual analog scale

All the eligible subjects were asked by an investigator to complete a questionnaire detailing nasal and eye symptoms

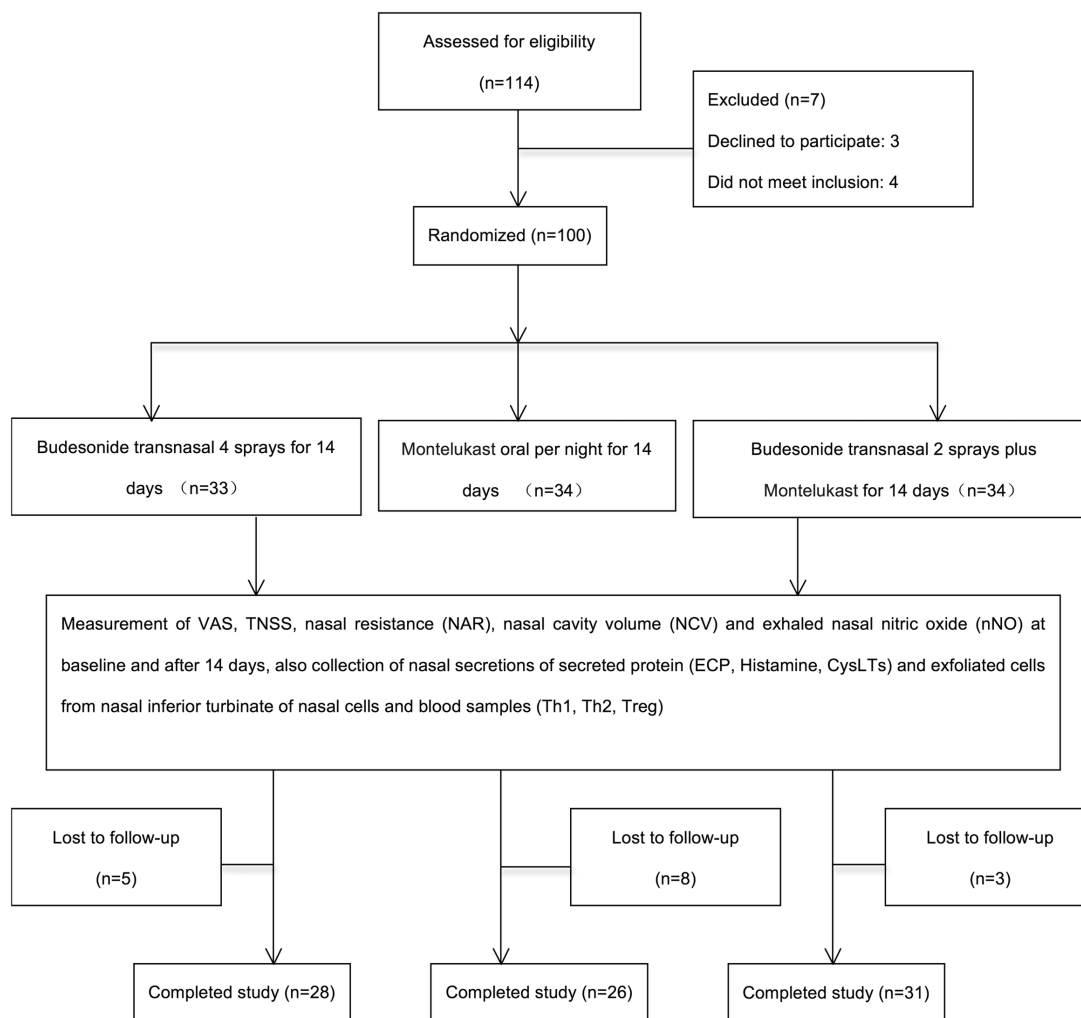


FIGURE 1. Study flowchart.

(including congestion, rhinorrhea, itching, sneezing, and eye itching), and to score the severity of each symptom on a VAS of 0–10 cm (0 = no symptoms and 10 = most severe and bothersome symptoms). Individual and mean of total symptom scores were recorded at baseline and again after 2 weeks of treatment. Any subject with a mean score of <4 (VAS <4 cm) at baseline was excluded from the study.

Acoustic rhinometry

Acoustic rhinometry (Hood Labs, Pembroke, MA) was used to measure the total nasal cavity volume (NCV) according to standardized recommendations.¹⁵ The following parameters were recorded for each nostril: nasal volume from the first 2 cm (V2), from the the first 4 cm (V4), from the first 6 cm (V6), from the segment between 0 and 5 cm (V0-5), and from the segment between 0 and 7 cm (V0-7) of the nose; and minimal cross-sectional areas between 0 and 2.2 cm (min1) and between 2.2 and 5.4 cm (min2). All measurements were performed 3 times by the same operator, and nasal volumes were calculated as the sum of both nostrils.¹⁵

Fractional exhaled nitric oxide

Exhaled NO concentration was measured using a NO analyzer (Niox; Aereocrine, Solna, Sweden) at a constant expiratory flow rate of 50 mL/s, according to American Thoracic Society/European Respiratory Society (ERS/ATS) guidelines.¹⁶ Before testing, the participants rinsed their mouths with water and refrained from eating nitrate-rich food for 2 hours, as doing so could affect the FeNO test results. Each subject was seated in a comfortable position, and instructed on the breathing technique during the test. Briefly, NO-free air was aspirated at a flow rate of 50 mL/s through the nasal cavity, via a central cavity nasal olive fitted with a side port. The participant was guided to exhale against the air resistance, resulting in an intraoral pressure of approximately 10 cmH₂O to close the velum, and nasal gas from the circuit was continuously routed via the side port directly into the FeNO analyzer. The level of FeNO was recorded from a plateau lasting for at least 3 seconds, and the test was repeated 3 times to obtain a mean concentration of FeNO (parts per billion, ppb).

Nasal secretions

Nasal secretions were collected and processed according to the method of Watelet and colleagues.¹⁷ Briefly, nasal secretions were obtained by inserting a sponge pack into both sides of the nasal cavity for 5 minutes, and the quantity of secretions collected in each sponge was determined by calculating the difference in the weight of the sponge before and after insertion into the nasal cavity. After weighing, 1.5 mL of 0.9% sodium chloride was added to each sponge and all sponges were stored at 4°C in a refrigerator for at least 2 hours, before being transferred to a 5-mL BD syringe. The bulk of the nasal secretions were forced out of each sponge using the syringe piston, and the secretions were centrifuged at 1500g for 10 minutes at 4°C. The supernatants were separated and stored in eppendorf tubes at -80°C until analysis for the levels of secreted proteins by commercial ELISA kits, including ECP (Cusabio, Barksdale, DE), histamine (USCN Life Science, Wuhan, China), and CysLTs (Abcam, Cambridge, UK).

Flow-cytometric analysis of T cells in nasal mucosa and peripheral blood

Nasal cells were collected from the inferior turbinate of each patient with a cytology brush (ThinPrep®; Hologic, Marlborough, MA), by a few backward-forward and rotatory movements under direct visualization of nasal endoscopic display. Each brush was placed in a 15-mL centrifuge tube (Thermo Fisher Scientific, Waltham, MA) containing 1 mL of distilled water. The brush was then agitated gently to release the adhering sample into the solution, which was subsequently centrifuged at 1500g for 10 minutes at 4°C to separate the cells for further analysis.

Peripheral venous blood samples (2 mL) were drawn from each subject, and immediately transferred to sterile heparin tubes.

Samples from the nasal mucosa and peripheral blood were incubated with cell stimulation (eBioscience, San Diego, CA) for 4 hours at 37°C. Lysing solution was added only to the blood samples (BD Bioscience, San Jose, CA) for 8 minutes in the dark followed by washing with isotonic phosphate buffer. Before washing with the isotonic phosphate buffer, both the nasal mucosa and peripheral blood were incubated with permeabilizing solution (BD Bioscience) for 10 minutes. Cells in both nasal mucosa and peripheral blood were incubated with T-cell subset antibodies (eBioscience) and evaluated by flow cytometry (Attune NxTFlow Cytometer; Thermo Fisher Scientific), according to the manufacturer's instructions. T-cell subsets were selected for detailed phenotypic analysis as follows: (1) Th1 cells were labeled as interferon-gamma-positive (IFN- γ^+)interleukin-4-negative (IL-4)⁻CD4⁺ T cells; (2) Th2 cells were labeled as IFN- γ^- IL-4⁺CD4⁺ T cells; and (3) Treg cells were labeled as CD4⁺CD25⁺CD127⁻ T cells.

TABLE 1. Demographic characteristics of subjects of different treatment groups at baseline

Groups	Gender ^a (male/female)	Age ^b (years)
BD	16/12	34.10 ± 2.36
MNT	14/12	34.28 ± 1.98
hBD+MNT	14/17	38.38 ± 2.09

^aData expressed as number.

^bData expressed as mean ± standard error of the mean.

BD = budesonide; hBD = half-dose budesonide; MNT= montelukast.

Safety assessment

The safety of treatment was based on the incidence and severity of self-reported adverse events by the patients over the course of the treatment period.

Statistical analyses

Sample size was established on the basis of preliminary findings of mean (standard deviation, SD) values of VAS in the BD and MNT treatment groups at 5.190 (1.903) and 3.350 (1.891), respectively. The number of subjects in each group was estimated according to the formula: $N = [(\mu_\alpha + \mu_\beta) \sigma / \delta]^2 (1 + 1/k)$, where $\alpha = 0.05$, $\beta = 0.1$, $\mu_\alpha = 1.96$, $\mu_\beta = 1.28$, $\sigma = [(S_1^2 + S_2^2) / 2]^{0.5} = 1.89$, $\delta = X_1 - X_2 = 5.19 - 3.35 = 1.84$, and $k = 1$. Thus, the number of subjects was calculated to be $N = 23$, and an extra 20% of subjects were added to the group to allow for uncontrollable situations, providing a total of 28 subjects in each treatment group. Based on a ratio of 1:1:1 for each treatment group (BD, MNT, and hBD+MNT), a sample size of at least 84 would be necessary for assessment of statistically significance differences between groups.

Statistical analysis was performed using SPSS version 22.0. Data are expressed as mean and standard error of the mean (SEM). The sex ratio and number of patients with uncontrolled symptoms after each treatment were calculated using the chi-square test. Comparisons within groups of differences between baseline and after therapy were performed by paired-samples *t* test. Differences between baseline values and posttreatment values in VAS, NCV, FeNO, ECP, histamine, CysLTs, and T-cell subsets from nasal mucosa and blood samples for the 3 treatment groups were compared by one-way analysis of variance (ANOVA) and Bonferroni's *t* test if $p < 0.05$. A value of $p < 0.05$ (two-tailed) was considered statistically significant.

Results

Clinical characteristics of patients

Overall, 100 subjects met the inclusion criteria and were randomized to receive 1 of 3 different treatments for 2 weeks. The demographic characteristics of the treatment groups are shown in Table 1, and indicate that the groups were not significantly different with respect to age and sex distribution of patients. A total of 15 patients (5 in the BD

TABLE 2. Baseline values of study parameters for different treatment groups*

Characteristic	BD (n = 28)	MNT (n = 26)	hBD+MNT (n = 31)	p value
Sneezing	6.821 ± 0.367	7.038 ± 0.326	7.000 ± 0.467	0.922
Rhinorrhea	5.286 ± 0.454	6.192 ± 0.456	6.597 ± 0.425	0.106
Nasal block	5.375 ± 0.502	6.673 ± 0.384	6.145 ± 0.441	0.138
Nasal itching	5.250 ± 0.476	5.519 ± 0.559	5.613 ± 0.475	0.867
Eye itching	4.893 ± 0.612	5.481 ± 0.538	5.436 ± 0.581	0.732
Average of total symptoms	5.525 ± 0.259	6.181 ± 0.304	6.100 ± 0.313	0.241
NCV (cm ³)	7.804 ± 0.164	7.213 ± 0.292	8.092 ± 0.197	0.105
FeNO (ppb)	42.00 ± 6.363	42.92 ± 5.034	49.68 ± 5.233	0.557
ECP (ng/mL)	19.44 ± 3.582	17.33 ± 2.434	13.31 ± 1.844	0.279
Histamine (ng/mL)	0.612 ± 0.085	0.723 ± 0.091	0.584 ± 0.076	0.474
Cysteine leukotrienes (ng/mL)	1017 ± 131.5	1279 ± 177.7	951.4 ± 145.2	0.288

*Data expressed as mean ± SD.

BD = budesonide; ECP = eosinophil cationic protein; hBD = half-dose budesonide; MNT = montelukast; SD = standard deviation.

group, 8 in MNT group, and 2 in the hBD+MNT group) did not fully adhere to treatment and therefore did not complete the study. The mean (SD) baseline values of all study parameters for the remaining 85 patients in the 3 treatment groups are shown in Table 2. The groups were not significantly different in any respect at baseline.

Clinical responses

All symptoms of AR were significantly improved from baseline after treatment with BD, MNT, and hBD+MNT (Fig. 2). However, assessment of improvements in symptoms between groups demonstrated that BD provided greater improvements in sneezing, nasal block, nasal itching, and average of total symptoms than MNT (Fig. 2A, C, D, and F), but was not significantly better than MNT for improving symptoms of rhinorrhea and eye itching (Fig. 2B and E). Although combination therapy with hBD+MNT demonstrated better efficacy in improving AR symptoms than BD or MNT alone, apart from nasal obstruction (Fig. 2C), improvements in the other symptoms were not significant.

Based on the classification by Hellings and colleagues,¹⁸ in the present study we considered any subject with a symptom score of >5 (based on VAS >5 cm) after 2 weeks of treatment as an uncontrolled case. In this regard, the number of patients with uncontrolled symptoms was found to be significantly greater after treatment with MNT than after treatment with BD ($p = 0.012$) or hBD+MNT ($p = 0.01$), with no significant difference between the BD and hBD+MNT treatments ($p = 0.521$; Table 3).

Objective examinations of clinical nasal function

Objective assessments of nasal function demonstrate that all treatments significantly improved NCV and reduced FeNO concentrations from baseline (Fig. 3A and B). The

improvements in NCV were significantly greater in patients receiving hBD+MNT (1.529 ± 0.262) and BD (1.405 ± 0.199) than in patients receiving MNT (0.677 ± 0.261); however, improvements in NCV were not significantly different between the hBD+MNT and BD treatment groups ($p = 0.712$; Fig. 3A).

The concentration of FeNO was decreased to a significantly greater extent from baseline after treatment with hBD+MNT compared with after treatment with BD and MNT ($p = 0.0025$ and $p = 0.0098$; Fig. 3B), with no significant difference between the MNT and BD treatment groups (difference between means: -1.635 ± 3.990 ; $p = 0.684$).

Inflammatory mediators in nasal secretion

Data were not available for nasal secretions of all subjects because relatively small amounts of individual samples were obtained in some patients. In this respect, overall, 16 samples were available for evaluation in each treatment group, and 12, 10, and 15 samples were not available in the BD, MNT, and hBD+MNT groups, respectively. In the evaluable samples, concentrations of ECP were significantly decreased from baseline in all treatment groups after 2 weeks (Fig. 4A). Similarly, the concentrations of histamine and leukotrienes were significantly decreased from baseline after 2 weeks in the BD and hBD+MNT treatment groups, but not in the MNT treatment group (Fig. 4B and C). Moreover, the change in all mediators was significantly greater in the BD treatment group than in the MNT treatment group, but not significantly different from that in the hBD+MNT treatment group (difference between means: -1.635 ± 3.990 ; $p = 0.684$). In the BD treatment group, concentrations of ECP, histamine, and leukotriene were decreased from baseline by -11.87 ± 3.223 ng/mL, -0.029 ± 0.062 , and -636.1 ± 139.2 pg/mL, respectively;

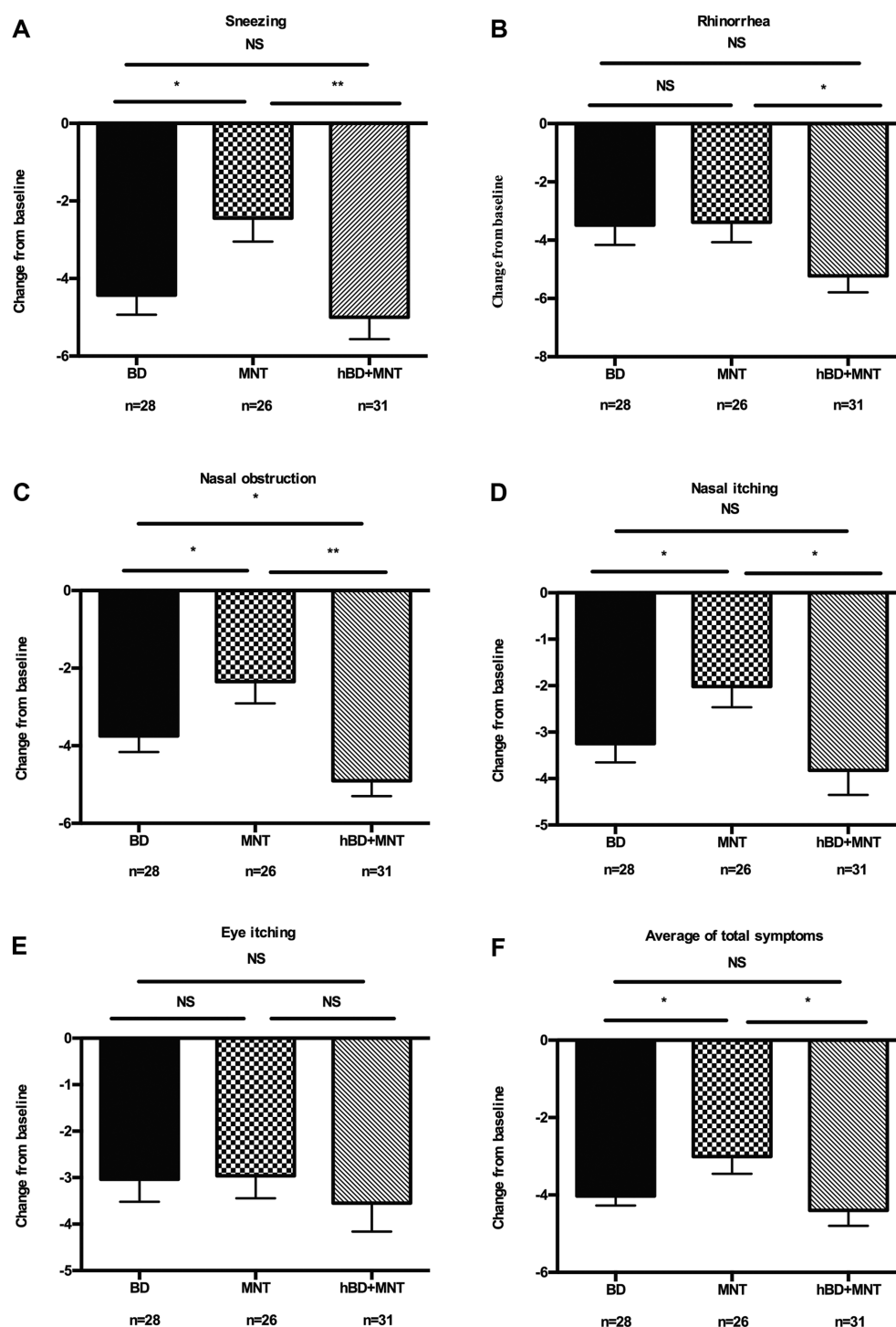


FIGURE 2. Change from baseline in individual and total scores (mean $\pm$ standard error of the mean) for major symptoms of allergic rhinitis after treatment for 2 weeks with BD, MNT, and hBD+MNT. (A) Sneezing. (B) Rhinorrhea. (C) Nasal obstruction. (D) Nasal itching. (E) Eye itching. (F) Average of total symptoms. * $p < 0.05$; ** $p < 0.01$; NS, not significant. BD = budesonide; hBD = half-dose budesonide; MNT = montelukast.

and by -3.501 ± 1.700 ng/mL, 0.027 ± 0.102 ng/mL, and -200.6 ± 153.1 pg/mL, respectively, in the MNT treatment group.

T-cell subsets in nasal cavity and peripheral blood
Topical inferior turbinate exfoliative cells and peripheral

blood were collected for analysis of changes in T-lymphocyte subsets. Th1 cells and Th2 cells were significantly decreased and Treg cells significantly increased from baseline in turbinate samples from patients treated with BD and hBD+MNT (Fig. 5A-C). In contrast, the numbers of these cell types were not altered from baseline by treatment with MNT.

TABLE 3. Number of patients with uncontrolled symptoms in the different treatment groups after 2 weeks*

	BD (n = 28)	MNT (n = 26)	hBD+MNT (n = 31)	p value
Sneezing	1 (3.6%)	11 (42%)	2 (6.5%)	0.000
Rhinorrhea	2 (7.1%)	5 (19%)	2 (6.5%)	0.255
Nasal block	2 (7.1%)	8 (30.7%)	0 (0%)	0.001
Nasal itching	2 (7.1%)	4 (15%)	1 (3.2%)	0.246
Eye itching	3 (7.1%)	3 (12%)	1 (3.2%)	0.431
Total	7 (25%)	16 (62%)	5 (16%)	0.001

*Data expressed as number (%).

BD = budesonide; hBD = half-dose budesonide; MNT = montelukast.

Assessment of T-cell subsets in blood samples demonstrated that BD treatment did not significantly alter the numbers of Th1, Th2 and Treg cells from baseline values (Fig. 6A-C). Although MNT or hBD+MNT treatment also did not significantly alter the numbers of Th2 cells from baseline (Fig. 6B), both of these treatments significantly decreased Th1 cells (Fig. 6A) and increased Treg cells in the peripheral blood (Fig. 6C).

Side effects

No side effects were reported by patients over the course of the 2-week treatment period.

Discussion

Anti-inflammatory agents, such as corticosteroids and leukotriene-receptor antagonists, are recommended by ARIA guidelines³ as the first-line choice for treating SAR. However, it is still unclear whether combination therapy with these drugs is superior to therapy with individual drugs. In the present study, we compared the efficacy of BD, a nasal steroid, and MNT, a cysteinyl-leukotriene receptor antagonist, both individually and in combination, by assessing symptoms severity, nasal function, and inflammatory mediatory indices in patients with SAR during the pollen season.

We found that both BD and MNT significantly improved nasal symptoms compared with baseline, and that BD-mediated improvements in symptoms, apart from rhinorrhea and eye itching, were significantly greater than MNT-mediated improvement. These findings are in accordance with those of Jindal and colleagues,¹⁰ who demonstrated that treatment with intranasal fluticasone propionate was significantly more effective than oral MNT in improving asthma as well as rhinitis symptom scores in patients with concomitant asthma and rhinitis.

Several studies have made a case for use of combination therapy by demonstrating better efficacy than monotherapy.^{11,12,19,20} It is noteworthy that, in the present

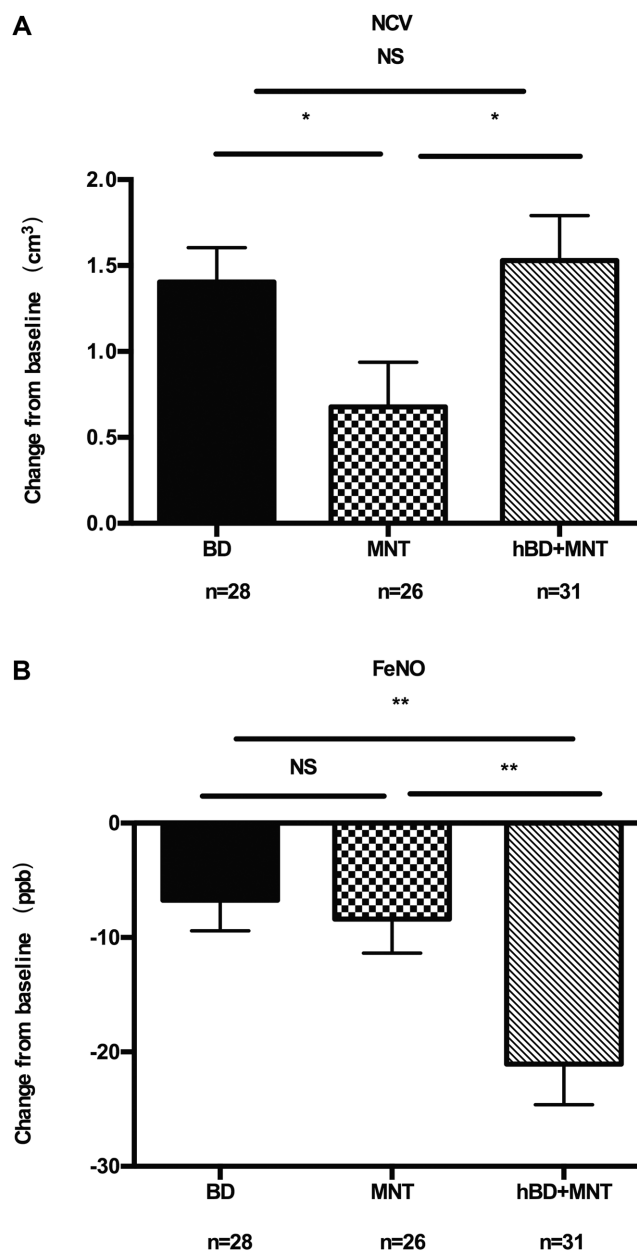


FIGURE 3. Change from baseline in objective clinical nasal function tests (mean $\pm$ standard error of the mean) after treatment for 2 weeks with BD, MNT, and hBD+MNT. (A) NCV. (B) FeNO. * $p < 0.05$, ** $p < 0.01$, NS, not significant. BD = budesonide; FeNO = fractional exhaled nitric oxide; hBD = half-dose budesonide; MNT = montelukast; NCV = nasal cavity volume.

study, we used only half the recommended dose of BD (hBD 128 μ g) in combination with the recommended 10-mg dose of MNT to explore the potential advantage of combination therapy over individual drug therapy. We found that this hBD+MNT combination was significantly more effective than BD or MNT alone in attenuating the nasal obstruction, but not other nasal symptoms of AR. It is possible that this effect on nasal congestion may be a consequence of MNT inhibiting mucus production and mucosal swelling brought about by increased vascular permeability and blood flow to the mucosa, as reported previously for

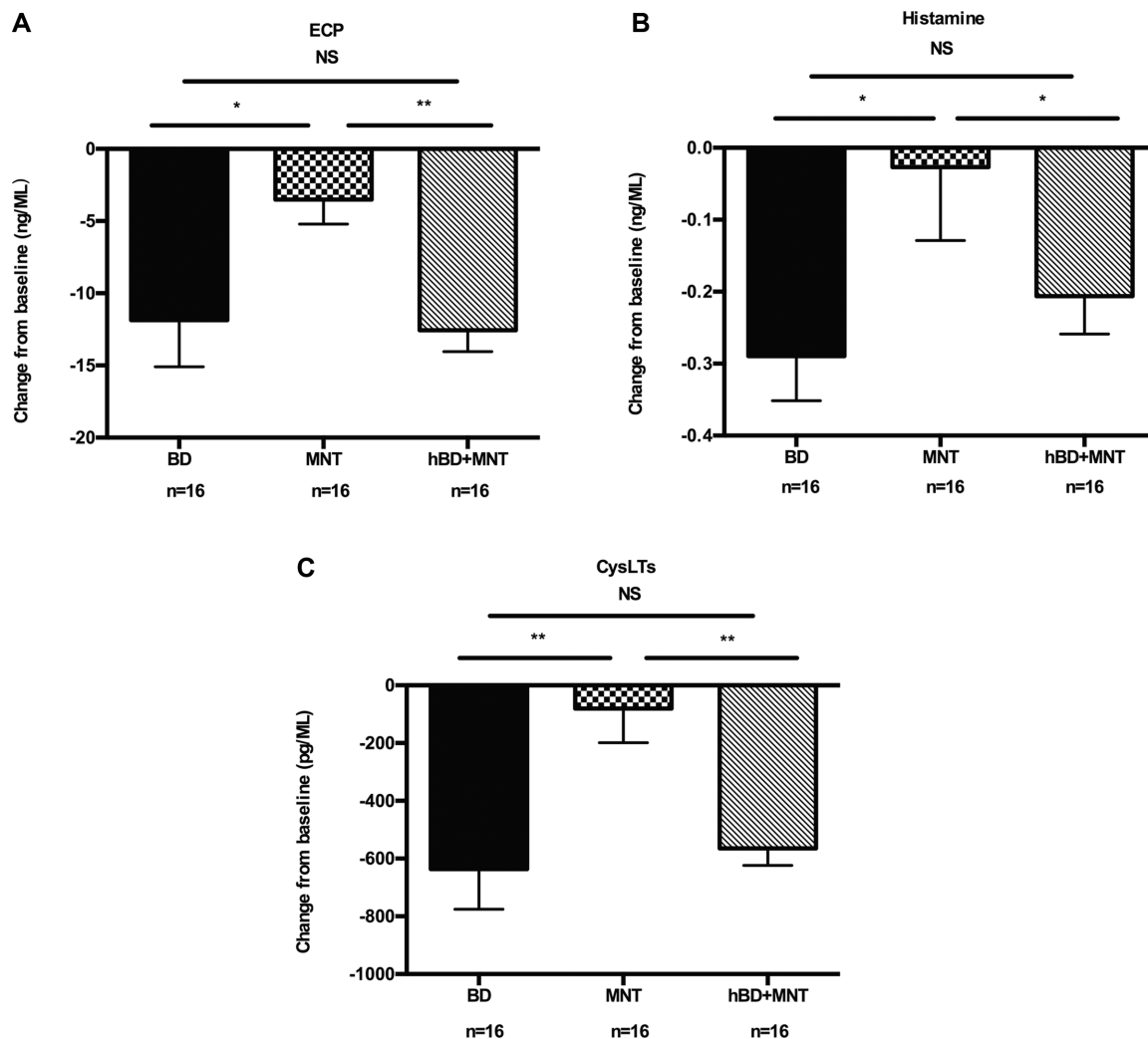


FIGURE 4. Change from baseline in concentrations of inflammatory mediators in nasal secretions after treatment for 2 weeks with BD, MNT, and hBD+MNT. * $p < 0.05$; ** $p < 0.01$; NS, not significant. (A) ECP. (B) Histamine. (C) CysLTs. BD = budesonide; CysLTs = cysteinyl-leukotrienes; ECP = eosinophil cationic protein; hBD = half-dose budesonide; MNT = montelukast.

cysteinyl-leukotrienes.^{21,22} In the present study, objective and quantitative assessment of nasal patency by measurement of NCV, an objective measurement of mucosal congestion and structural abnormalities,²³ was also undertaken to verify subjective findings for the improvement of nasal blockage. In this regard, our study has confirmed subjective findings that both BD and MNT expanded the nasal cavity significantly after 2-week treatments and the hBD+MNT combination therapy was better than either monotherapy.

Bousquet and colleagues²⁴ reported that, although the majority of patients present with controlled symptoms during treatment, many remain uncontrolled, as indicated by a cutoff value of ≥ 5 cm on a 0–10 cm VAS for symptoms of severity. Based on this criterion, our study has shown that groups treated with hBD+MNT combination therapy and BD had significantly fewer patients with uncontrolled symptoms and lower dropout rates than the group treated with MNT alone. This finding suggests that, in addition to being more efficacious, adherence and therefore compliance

with hBD+MNT combination therapy and BD alone was also probably greater than adherence/compliance to MNT alone.

The anti-inflammatory effects of different SAR therapies have been investigated in several studies, as a variety of inflammatory mediators have been shown to be increased in allergic disease. Exhaled NO (eNO) is considered a marker of lower airway inflammation and increased in asthmatic patients.²⁵ Similarly, in AR patients, eNO is increased during the pollen season.²⁶ In accordance with other studies,^{27,28} our study has also demonstrated that FeNO concentration was significantly decreased from baseline after treatment with oral MNT, as expected. However, to our surprise, we found that the topically applied BD nasal spray also decreased FeNO concentration of the lower airway. Although the mechanism by which intranasal BD decreases NO in the lower airways is currently unknown, this result nevertheless suggests that INS may be suitable as adjunctive treatment for asthma patients as well

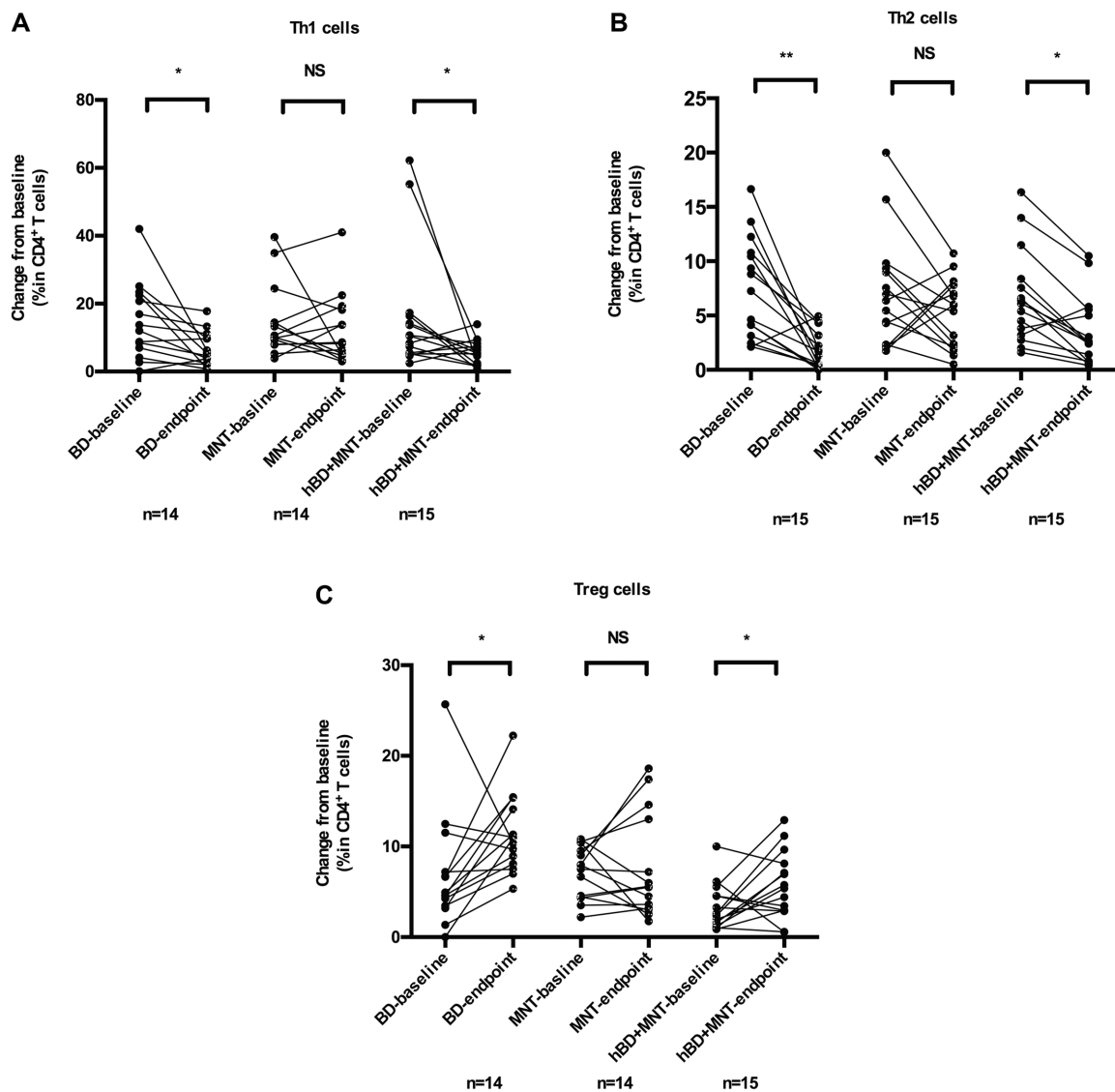


FIGURE 5. Change from baseline in T-cell subsets in nasal turbinate mucosa after treatment for 2 weeks with BD, MNT, and hBD+MNT. (A) Th1 cells. (B) Th2 cells. (C) Treg cells. * $p < 0.05$, ** $p < 0.01$, NS, not significant. BD = budesonide; hBD = half-dose budesonide; MNT = montelukast; Th cells = T-helper cells; Treg cells = T-regulatory cells.

as AR patients with concomitant asthma. Moreover, we have demonstrated that the hBD+MNT combination treatment was significantly better at decreasing FeNO concentrations than BD and MNT treatments alone. This finding is in accordance with the findings of Ghiri and colleagues,²⁹ who investigated the effect of adding MNT to maintenance therapy with inhaled corticosteroids (ICS) for 3 weeks in asthmatic children, and demonstrated that there was a significantly greater reduction in FeNO values in children receiving MNT+ICS therapy, compared with children receiving ICS alone.

ECP, released by activated eosinophils, has also been shown to be a major mediator of allergic inflammation. LTD4 was able to induce eosinophil chemotaxis.³⁰ In this regard, the present study has demonstrated that MNT and BD treatments both significantly reduced the

concentration of ECP in nasal secretions. Again, these findings are in accordance with the findings of investigators.^{31,32} However, the BD-induced attenuation of ECP was significantly greater than MNT-induced attenuation, and similar to hBD+MNT combination-induced attenuation of ECP. As LTD4 has been shown to promote eosinophil chemotaxis,³⁰ it is possible that the observed effect of MNT, and probably BD, in the present study is a consequence of the attenuation of eosinophil chemotaxis and activation.

Histamine and CysLTs have been shown to be inflammatory mediators secreted in early- and late-phase allergic reactions, respectively. Similar to the findings for effects of treatment on ECP release in the present study, BD-induced attenuation of histamine and the CysLTs was significantly greater than MNT-induced attenuation, and

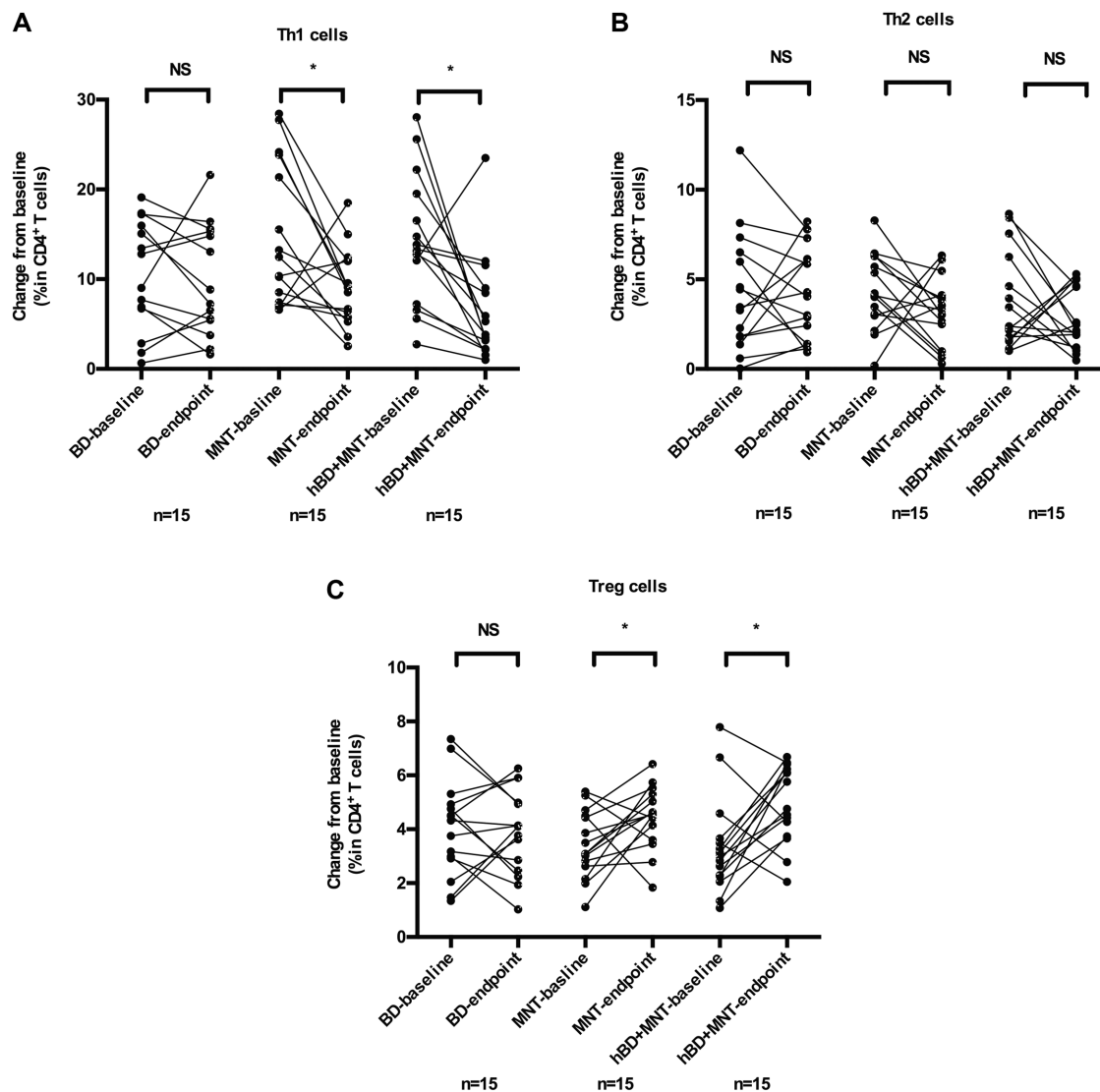


FIGURE 6. Change from baseline in T-cell subsets in peripheral blood after treatment for 2 weeks with BD, MNT, and hBD+MNT. (A) Th1 cells. (B) Th2 cells. (C) Treg cells. * $p < 0.05$; ** $p < 0.01$, NS, not significant. BD = budesonide; hBD = half-dose budesonide; MNT = montelukast; Th cells = T-helper cells; Treg cells = T-regulatory cells.

similar to hBD+MNT-combination-induced attenuation of these mediators, indicating that BD played a major role in the suppression of these mediators in local secretions. These findings are in accordance with the findings of Di Lorenzo and colleagues,³³ who demonstrated that combination therapy with fluticasone propionate aqueous nasal spray (FPANS) plus MNT did not offer a substantial advantage over FPANS monotherapy for reducing symptoms or eosinophil numbers/ECP in patients affected by seasonal AR.

Th1, Th2, and Treg cells represent 3 major functional T-lymphocyte subtypes, which play important roles in the etiology of AR. Several studies have elucidated the prevalence of the T-cell subsets after treatments by INS or MNT, based on changes in particular cytokines.^{34–37} However, as different cells have the ability to synthesize particular inflammatory cytokines released by the different

T-cell subsets, in the present study we investigated the effect of treatment on T-cell subsets by directly detecting the number of these T-cell subtypes in the nasal mucosa and peripheral blood using flow cytometry. Analysis of Th1, Th2, and Treg cells has demonstrated that BD treatment decreased Th1 and Th2 cell numbers and elevated Treg-cell numbers in the nasal mucosa of SAR patients. This is in accordance with findings of Wang and colleagues³⁸ in patients with eosinophilic chronic rhinosinusitis with nasal polyps. In contrast, our study has demonstrated that MNT treatment did not significantly alter the numbers of T-cell types in the nasal mucosa, but significantly decreased the Th1 cells and increased the Treg cells in peripheral blood of SAR patients. Moreover, the changes in nasal mucosa and peripheral blood were also reflected by hBD+MNT combination treatment, thus highlighting differing mechanisms and efficacies of BD and MNT in influencing T-cell types

in the nasal mucosa and peripheral blood. Indeed, Spinozzi and colleagues³⁹ found that MNT reduced proliferative response of T cells to T-cell receptor activation and led to apoptosis at minimal concentrations of MNT in vitro, suggesting that the biologic effects of MNT are complex and involve gene activation and repression, probably induced by cysteine-leukotriene-receptor antagonist blockade.

This investigation is limited in that it was an open-label study, conducted with a relatively small number of patients and without a placebo group, which may have led to some bias. Our study is also somewhat limited by not having adequate nasal secretion samples for assessment from all participants. However, this design was used primarily for observational purposes, particularly as there have been no studies conducted in outpatients on the effect of using a combination of half the normal recommended dose of BD with MNT in patients with moderate-to-severe SAR induced by mugwort allergen in Chinese subjects. Consequently, these findings need to be confirmed in a well-controlled, double-blind, randomized, placebo-controlled investigation involving larger numbers of patients and control subjects. This is

especially so because, to our knowledge, this is the first study to document the usefulness of half the recommended dose of intranasal BD, which may be important for safety in long-term use in treating patients with moderate to severe SAR.

Conclusion

The results of this study demonstrate that BD had better efficacy than MNT for AR patients in improving nasal symptoms, nasal potency, and nasal inflammatory level over a short treatment period. BD and MNT significantly regulated T-cell subsets in nasal mucosa and peripheral blood, respectively. Compared with BD and MNT monotherapy, the combination therapy of hBD+MNT was generally better, especially in controlling nasal obstruction. Moreover, in view of differing mechanisms and efficacies of BD and MNT in influencing particular T-cell subsets in the nasal mucosa and peripheral blood, it is likely that a combination of these agents will provide superior overall efficacy than either drug alone in the treatment of AR with or without concomitant asthma. ☉

References

- Bousquet J, Khaltaev N, Cruz AA, et al. Allergic Rhinitis and its Impact on Asthma (ARIA) 2008 update (in collaboration with the World Health Organization, GA (2) LEN and Allergen). *Allergy*. 2008;63(suppl 86):8–160.
- Meltzer EO, Blaiss MS, Derebery MJ, et al. Burden of allergic rhinitis: results from the Pediatric Allergies in America survey. *J Allergy Clin Immunol*. 2009;124(suppl):S43–S70.
- Brożek JL, Bousquet J, Baena-Cagnani CE, et al. Allergic Rhinitis and its Impact on Asthma (ARIA) guidelines: 2010 Revision. *J Allergy Clin Immunol*. 2010;126:466–476.
- Barnes PJ. Molecular mechanisms of steroid action in asthma. *J Allergy Clin Immunol*. 1996;97:159–168.
- Bensch GW. Safety of intranasal corticosteroids. *Ann Allergy Asthma Immunol*. 2016;117:601–605.
- Gupta R, Fonacier LS. adverse effects of nonsystemic steroids (inhaled, intranasal, and cutaneous): a review of the literature and suggested monitoring tool. *Curr Allergy Asthma Rep*. 2016;16:44.
- Li Y, Han T, Li W, Li Y, Guo X, Zheng L. Awareness of and phobias about topical corticosteroids in parents of infants with eczema in Hangzhou, China. *Pediatr Dermatol*. 2018;35:463–467.
- Lund VJ. International consensus report on the diagnosis and management of rhinitis. International rhinitis management working group. *Allergy*. 1994;49(suppl 19):1–34.
- Nayak A, Langdon RB. Montelukast in the treatment of allergic rhinitis. *Drugs*. 2007;67:887–901.
- Dempsey OJ, Wilson AM, Sims EJ, et al. A comparison of topical budesonide and oral montelukast seasonal allergic rhinitis and asthma. *Eur Respir J*. 1999;14(suppl 30):S30s.
- Jindal A, Suriyan S, Sagadevan S, et al. Comparison of oral montelukast and intranasal fluticasone in patients with asthma and allergic rhinitis. *J Clin Diagn Res*. 2016;10:OC06–10.
- Cingi C, Gunhan K, Gage-White L, et al. Efficacy of leukotriene antagonists as concomitant therapy in allergic rhinitis. *Laryngoscope*. 2010;120:1718–1723.
- Florinescu-Cugheorghe NA, Popescu F, Alexandru DO. Treatment evaluation with mometasone furoate, alone or in combination with desloratadine/montelukast in moderate severe allergic rhinitis. *Acta Med Marisensis*. 2014;60:106–108.
- Lou H, Ma S, Zhao Y, et al. Sensitization patterns and minimum screening panels for aeroallergens in self-reported allergic rhinitis in China. *Sci Rep*. 2017;7:9286.
- Clement P, Gordts F. Consensus report on acoustic rhinometry and rhinomanometry. *Rhinology*. 2005;43:169–179.
- American Thoracic Society/European Respiratory Society. ATS/ERS recommendations for standardized procedures for the online and offline measurement of exhaled lower respiratory nitric oxide and nasal nitric oxide. *Am J Respir Crit Care Med*. 2005;171:912–930.
- Watalet JB, Gevaert P, Holtappels G, et al. Collection of nasal secretions for immunological analysis. *Eur Arch Otorhinolaryngol*. 2004;261:242–246.
- Hellings P, Fokkens W, Akdis C, et al. Uncontrolled allergic rhinitis and chronic rhinosinusitis: where do we stand today? *Allergy*. 2013;68:1–7.
- Tatar EŞ, Sürenoglu UA, Ozdek A, et al. The effect of combined medical treatment on quality of life in persistent allergic rhinitis. *Indian Otolaryngol Head Neck Surg*. 2013;65(suppl 2):333–337.
- Pinar E, Eryigit O, Oncel S, et al. Efficacy of nasal corticosteroids alone or combined with antihistamines or montelukast in treatment of allergic rhinitis. *Auris Nasus Larynx*. 2008;35:61–66.
- Kanaoka Y, Boyce JA. Cysteinyl leukotrienes and their receptors: cellular distribution and function in immune and inflammatory responses. *J Immunol*. 2004;173:1503–1510.
- Peters-Golden M, Henderson WR Jr. The role of leukotrienes in allergic rhinitis. *Ann Allergy Asthma Immunol*. 2005;94:609–618.
- Mamikoglu B, Houser SM, Corey JP. An interpretation method for objective assessment of nasal congestion with acoustic rhinometry. *Laryngoscope*. 2002;112:926–929.
- Bousquet PJ, Bachert C, Canonica GW, et al. Uncontrolled allergic rhinitis during treatment and its impact on quality of life: a cluster randomized trial. *J Allergy Clin Immunol*. 2010;126:666–8.e1–5.
- Kharitonov SA, Yates D, Robbins RA, et al. Increased nitric oxide in exhaled air of asthmatic patients. *Lancet*. 1994;343:133–135.
- Henriksen AH, Sue-Chu M, Lingaas HT, et al. Exhaled and nasal NO levels in allergic rhinitis: relation to sensitization, pollen season and bronchial hyperresponsiveness. *Eur Respir J*. 1999;13:301–306.
- Montuschi P, Mondino C, Koch P, et al. Effects of montelukast treatment and withdrawal on fractional exhaled nitric oxide and lung function in children with asthma. *Chest*. 2007;132:1876–1881.
- Biggaard H, Loland L, Oj JA, et al. NO in exhaled air of asthmatic children is reduced by the leukotriene receptor antagonist montelukast. *Am J Respir Crit Care Med*. 1999;160:1227–1231.
- Ghiro L, Zancanato S, Rampon O, et al. Effect of montelukast added to inhaled corticosteroids on fractional exhaled nitric oxide in asthmatic children. *Eur Respir J*. 2002;20:630–634.
- Fregonese L, Silvestri M, Sabatini F, et al. Cysteinyl leukotrienes induce human eosinophil locomotion and adhesion molecule expression via a CysLT₁ receptor-mediated mechanism. *Clin Exp Allergy*. 2002;32:745–750.
- Pizzichini E, Leff JA, Reiss TF, et al. Montelukast reduces airway eosinophilic inflammation in asthma: a randomized, controlled trial. *Eur Respir J*. 1999;14:12–18.
- Benson M, Strannegård IL, Strannegård O, et al. Topical steroid treatment of allergic rhinitis decreases nasal fluid TH2 cytokines, eosinophils, eosinophil cationic protein, and IgE but has no significant effect on IFN- γ , IL-1 β , TNF- α , or neutrophils. *J Allergy Clin Immunol*. 2000;106:307–312.
- Di Lorenzo G, Pacor ML, Pellitteri ME, et al. Randomized placebo-controlled trial comparing fluticasone aqueous nasal spray in mono-therapy, fluticasone plus cetirizine, fluticasone plus montelukast and cetirizine plus montelukast for seasonal allergic rhinitis. *Clin Exp Allergy*. 2004;34:259–267.
- Ciprandi G, Tosca MA, Cirillo I, et al. The effect of budesonide on the cytokine pattern in patients with perennial allergic rhinitis. *Ann Allergy Asthma Immunol*. 2003;91:467–471.
- Scaife A, Miller D, Spiteri-Cornish D, et al. Inhibitory effects of montelukast on mediator release by nasal epithelial cells from asthmatic subjects with or without allergic rhinitis. *Respir Med*. 2013;107:1859–1865.
- Ciprandi G, Frati F, Marcucci F, et al. Nasal cytokine modulation by montelukast in allergic children: a pilot study. *Eur Ann Allergy Clin Immunol*. 2003;35:295–299.
- Malmhäll C, Bossios A, Pullerits T, et al. Effects of pollen and nasal glucocorticoid on FOXP3+, GATA-3+ and T-bet+ cells in allergic rhinitis. *Allergy*. 2007;62:1007–1013.
- Wang C, Lou H, Wang X, et al. Effect of budesonide transnasal nebulization in patients with eosinophilic chronic rhinosinusitis with nasal polyps. *J Allergy Clin Immunol*. 2015;135:922.
- Spinozzi F, Russano AM, Piattoni S. Biological effects of montelukast, a cysteinyl-leukotriene receptor-antagonist, on T lymphocytes. *Clin Exp Allergy*. 2004;34:A1876–82.