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Usefulness of Fractional Exhaled Nitric Oxide-Guided Treatment in Patients with Asthma-Chronic Obstructive Pulmonary Disease Overlap

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Abstract

Background: Some patients present clinical features of both asthma and chronic obstructive pulmonary disease (COPD), which has led to the recent proposal of asthma-COPD overlap (ACO) as a diagnosis. Fractional exhaled nitric oxide (FeNO) is a candidate biomarker to diagnose ACO. We assessed the effect of an add-on treatment with budesonide/formoterol (BUD/FM) combination in patients with ACO, which was diagnosed by FeNO. **Methods:** This was a prospective, single-arm, open-label, before and after comparison study. Subjects included 83 patients with COPD who attended outpatient clinics for routine checkups at Shizuoka General Hospital between June and November 2016. All patients fulfilled the GOLD definition of COPD and were receiving long-acting muscarinic antagonist (LAMA) or LAMA/long-acting β_2 agonist (LABA) combinations. After an 8-week run-in period, BUD/FM was added to the patients with FeNO levels of ≥35 ppb, defined as having ACO. For patients receiving LAMA/LABA, BUD/FM was added after the discontinuation of LABA. The modified British Medical Research Council (mMRC) score, COPD assessment test (CAT) score, spirometric indices, forced oscillation parameters, and FeNO were assessed before and after 8 weeks of BUD/FM add-on treatment. **Results:** Twenty-four patients (28.9%) had FeNO levels ≥ 35 ppb, and 17 patients completed the study (mean age: 73 years and GOLD I/II/III/IV, 5/10/1/1). The mean CAT scores significantly improved (9.2 to 5.4, $p = 0.015$) and 10 patients (58.8%) showed ≥2 points improvement, a minimal clinically important difference. The mean FeNO levels significantly decreased from 63.0 to 34.3 ppb ($p < 0.006$). However, there were no changes in mMRC

scores, spirometric indices, or forced oscillation parameters. **Conclusions:** FeNO-guided treatment with BUD/FM improves symptoms in patients with ACO.

Keywords

Asthma-COPD Overlap, Budesonide/Formoterol Combination, COPD Assessment Test, Fractional Exhaled Nitric Oxide

1. Introduction

Since some patients present clinical features of both asthma and chronic obstructive pulmonary disease (COPD), the asthma-COPD overlap (ACO) has recently been proposed as a diagnosis [1]. Clinically, it can be challenging to distinguish asthma from COPD, especially in smokers and older adults. ACO is heterogeneous and includes different phenotypes: COPD with eosinophilic airway inflammation is one such phenotype, accounting for 10% - 40% of patients with COPD [2] [3] [4].

Several reports have suggested that high sputum and blood eosinophil counts in patients with COPD are associated with specific clinical phenotypes defined by 1) more frequent exacerbations and 2) better response to inhaled corticosteroids (ICS) for exacerbation prevention [5] [6] [7] [8] [9]. In addition, ICS therapy may reduce the rate of decline of forced expiratory volume in 1 second (FEV1) in patients with high blood eosinophil counts [10]. Eosinophilic airway inflammation can be noninvasively measured through the airway using induced sputum analysis and fractional exhaled nitric oxide (FeNO) [11]. However, induced sputum is less repeatable and reliable than FeNO due to the complexities of sputum collection. The blood eosinophil count acts as an indirect biomarker of eosinophilic airway inflammation. Regarding treatment, there is no evidence-based consensus on ACO treatment since patients with COPD, complicated with asthmatic components, have been excluded from large-scale clinical trials.

Based on the evidence of benefit of ICS/long-acting β_2-agonist (LABA) combination in asthma, we hypothesized that ICS/LABA can also be utilized for managing the eosinophilic COPD phenotype of ACO. In the present study, we assessed the effects of add-on treatment with a budesonide/formoterol (BUD/FM) combination in patients with COPD with high FeNO levels who were receiving a long-acting muscarinic antagonist (LAMA) or LAMA/LABA.

2. Methods

2.1. Subjects

Outpatients with COPD at Shizuoka General Hospital, seen between June and November 2016 were enrolled in this study. All patients fulfilled the definition of GOLD, and the COPD grading was based on the GOLD classification [12]. The

inclusion criteria were as follows: 1) age over 40 years, 2) receiving LAMA or LAMA/LABA, and 3) smoking history of more than 10 pack-years. Exclusion criteria were as follows: 1) current smokers, 2) had asthma, 3) receiving ICS or had exacerbation of COPD in the 4 weeks preceding the study.

2.2. Study Design

This is a prospective, single-arm, open-label, before-and-after comparison study (**Figure 1**). At the first visit, after 8 weeks of a run-in period, eligible subjects underwent the measurement of FeNO. If the FeNO value was ≥35 ppb, BUD/FM 160/4.5 μg two inhalations bid was added to the medication regimen of the patients receiving LAMA and continued for 8 weeks. For patients receiving LAMA/LABA, BUD/FM was added after the discontinuation of LABA at the first visit without a washout period. The study doctors, nurses, or pharmacists checked the inhaler technique of each patient during the run-in period and after the add-on treatment was initiated. This study was conducted in accordance with the principles of the Declaration of Helsinki, this protocol was approved by the Institutional Review Board of Shizuoka General Hospital (SGHIRB #2015072), and written informed consent was obtained from all patients prior to the study.

2.3. Assessment of Symptoms

The modified Medical Research Council (mMRC) score was used to evaluate dyspnea in daily living, grading from 0 (only get breathless with strenuous exercise) to 4 (too breathless to leave the house or breathless when dressing) [13].

The COPD assessment test (CAT) (Japanese version, supplied by GlaxoSmithKline, Japan) consists of eight items (cough, phlegm, chest tightness, breathlessness going up hills/stairs, activity limitations at home, confidence leaving home, sleep, and energy) that assess and quantify the symptoms and the impact of COPD [14]. Each item is scored from 0 to 5, giving a total score range from 0 to 40, corresponding to the best and worst health statuses, respectively.

2.4. FeNO

FeNO was measured using an online method at a flow of 50 mL/s using an NO

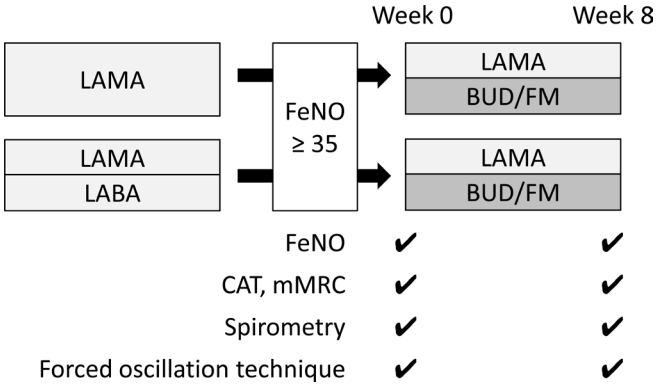


Figure 1. Study design.

analyzer (Sievers NOA 280i; GE Analytical Instruments, Boulder, CO, USA), in accordance with the American Thoracic Society/European Respiratory Society recommendations [15]. A FeNO of 35 ppb was selected as a cutoff point for high FeNO based on a previous study [16].

2.5. Forced Oscillation Technique (FOT) and Spirometry

Measurement of FOT and spirometry were performed in that order to avoid any influence of forced breathing. Broadband frequency FOT was performed using the MostGraph-01 (Chest M.I. Co. Ltd., Tokyo, Japan) in accordance with standard recommendations [17]. We used Rrs at 5 and 20 Hz (R5 and R20, respectively) and the difference between R5 and R20 (R5 - R20) as an indicator of the frequency dependence of Rrs. We also used Xrs at 5 Hz (X5), which reflects the elastic and inertial properties of the lung, resonant frequency (Fres) where Xrs crosses zero and the elastic and inertial forces are opposite and equal in magnitude, and a low-frequency reactance area (AX), which is an integral of Xrs at 5 Hz to Fres. Oscillatory indices were expressed as mean values during a respiratory cycle. All measured data were expressed as post-bronchodilator values.

Spirometry was performed using computerized equipment (model CHESTAC-8800; Chest M.I. Co. Ltd., Tokyo, Japan) in accordance with the American Thoracic Society (ATS)/European Respiratory Society (ERS) guidelines for spirometry [18] while patients were on daily medications. Measured data were expressed as post-bronchodilator values. Predicted values for pulmonary function tests were obtained from the Japanese Respiratory Society guidelines [19].

2.6. Study Outcomes

The primary outcome was the change in FEV1 between weeks 0 and 8. Secondary outcomes included changes in mMRC and CAT scores, forced vital capacity, forced oscillation parameters, and FeNO levels.

2.7. Long-Term Follow-Up

From 8 weeks until 12 months after the initiation of add-on treatment, patients attended outpatient clinics for routine checkups and the medication regimens were maintained as long as possible. Occurrences of exacerbation or pneumonia were assessed using medical records after 12 months.

2.8. Statistical Analyses

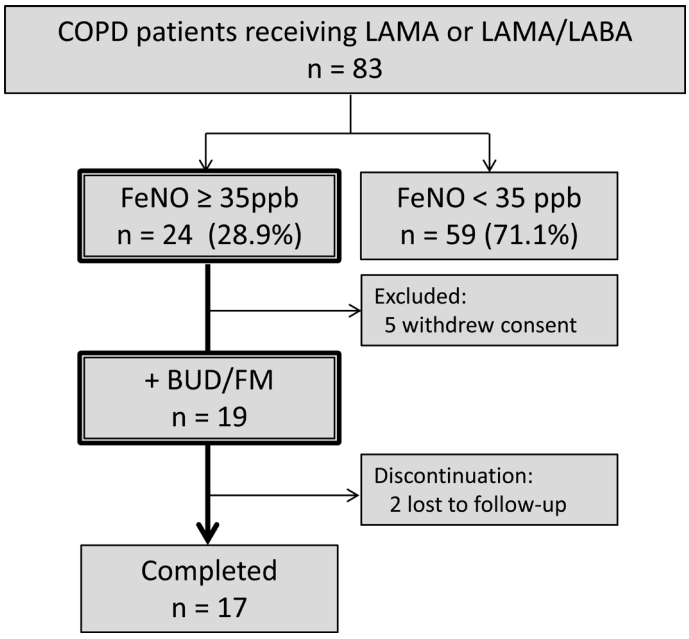
The quantitative data were summarized as means with standard deviations and were compared using *t*-tests. The categorical data were summarized as counts and compared using a Fisher's exact test. Paired *t*-tests were used to compare changes between weeks 0 and 8. The patients were classified into two groups (improvement and non-improvement groups) based on a minimal clinically important difference of improvement of two or more points of their CAT scores.

Receiver operator characteristic (ROC) analyses were performed and the area under the curve (AUC) was calculated to estimate the predictive capability of CAT improvement. The statistical significance for all analyses was set at $p < 0.05$ (two-tailed). All analyses were performed using EZR version 1.27 (Saitama Medical Center, Jichi Medical University, Saitama, Japan) [20].

3. Results

The study flow and the clinical characteristics of the subjects at baseline are shown in Figure 2 and Table 1. Of the 83 patients receiving LAMA or LAMA/LABA, 24 patients (28.9%) had FeNO levels ≥ 35 ppb. Patients with high FeNO levels had higher blood eosinophil counts, lower respiratory resistance and ALX, and less negative X5. However, there were no differences in airway reversibility or spirometry between the two groups. There was a significant, moderate correlation between FeNO and blood eosinophil counts in 83 patients with COPD ($r = 0.399$, $p < 0.001$).

Of the 24 patients with high FeNO levels, 19 patients received add-on treatment with BUD/FM, and 17 patients completed the study. After 8 weeks of add-on treatment, their CAT scores significantly improved, including total, cough, phlegm, and chest tightness (Table 2 and Figure 3), and 10 patients (58.8%) showed two or more points of improvement. The FeNO levels significantly decreased from 63.0 to 34.3 ppb ($p = 0.006$) and FEV1 tended to improve after intervention ($p = 0.091$), although the changes did not rise to the level of statistical significance (Figure 3). However, there was no significant change observed in FOT. There were no significant differences in CAT score improvements, FeNO,



Abbreviations: BUD/FM, Budesonide/formoterol; FeNO, fraction of exhaled nitric oxide; LABA, long-acting β_2-agonists; LAMA, long-acting muscarinic antagonists.

Figure 2. Patients' flow diagram.

Table 1. Characteristics of the subjects.

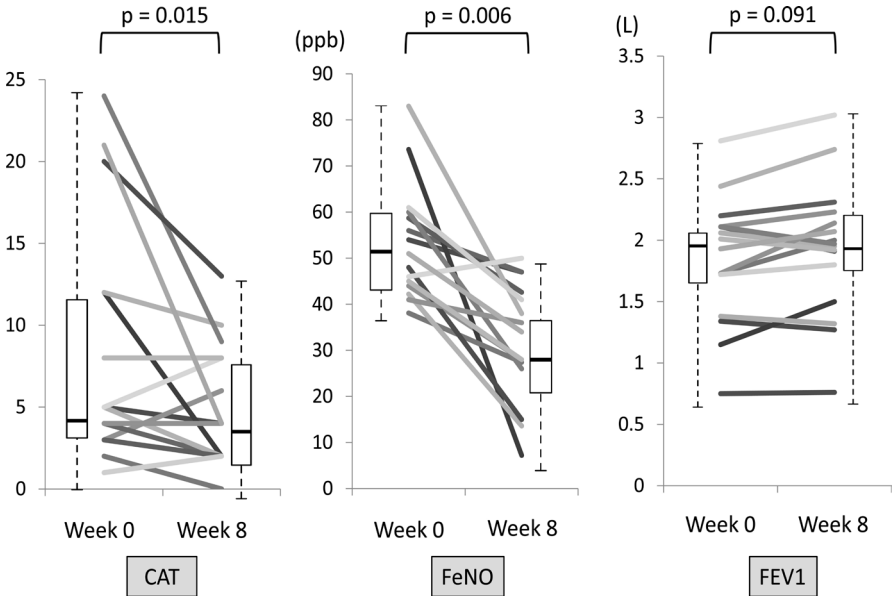
	All subjects $n = 83$	High FeNO $n = 24$	Low FeNO $n = 59$	p-value
Age, years	72.6 ± 6.3	73.6 ± 6.8	72.7 ± 6.1	0.341
Gender, male/female	76/7	23/1	53/6	0.667
Body mass index, kg/m^2	21.9 ± 3.8	21.7 ± 4.2	22.0 ± 3.7	0.738
Pack-years	56.9 ± 30.6	63.5 ± 36.7	54.3 ± 27.7	0.218
History of allergic rhinitis, *n*	9	2	7	0.999
History of asthma, *n*	5	2	3	0.624
Atopy, *n*	25	9	16	0.999
Treatment, LAMA/LAMA+LABA	28/55	9/15	19/40	0.798
FeNO, ppb	29.7 ± 26.4	58.7 ± 32.7	18.0 ± 8.6	<0.001
Blood eosinophil counts, /μL	209.6 ± 126.1	287.6 ± 166.2	181.4 ± 87.8	<0.001
Blood eosinophil counts > 300/μL, *n*	16	11	5	<0.001
IgE, IU/ml	269.2 ± 520.3	401.2 ± 675.3	199.9 ± 410.3	0.153
Atopy, *n*	25	9	16	0.999
mMRC 0/1/2/3/4, *n*	10/45/21/7/0	5/12/5/2/0	5/32/16/5/0	–
CAT	10.6 ± 7.3	10.5 ± 7.9	10.6 ± 7.1	0.972
GOLD 1/2/3/4, *n*	20/38/15/10	5/15/2/2	15/23/13/8	–
%FVC, %	93.1 ± 23.2	93.3 ± 26.9	93.0 ± 21.7	0.967
%FEV1, %	61.6 ± 22.4	63.9 ± 18.4	60.7 ± 23.9	0.547
FEV1/FVC, %	52.6 ± 14.4	56.1 ± 14.8	51.2 ± 14.1	0.159
IC, L	2.01 ± 0.61	2.14 ± 0.68	1.95 ± 0.57	0.191
Reversibility				
>12% and >200 ml, *n*	5	2	3	0.639
>12%, *n*	9	2	7	0.466
Absolute change, ml	72.1 ± 86.6	74.7 ± 106.7	70.8 ± 75.9	0.874
% change, %	5.9 ± 8.4	5.0 ± 8.1	6.4 ± 8.6	0.732
R5, cmH_2O/L/s	3.57 ± 1.55	2.98 ± 1.42	3.81 ± 1.55	0.027
R20, cmH_2O/L/s	2.64 ± 0.98	2.30 ± 0.94	2.78 ± 0.96	0.041
R5 - R20, cmH_2O/L/s	0.92 ± 0.64	0.69 ± 0.53	1.02 ± 0.67	0.034
X5, cmH_2O/L/s	−1.33 ± 1.30	−0.83 ± 0.84	−1.55 ± 1.41	0.024
Fres, Hz	12.45 ± 5.63	10.67 ± 4.51	13.20 ± 5.91	0.065
ALX, cmH_2O/L/s × Hz	9.60 ± 11.90	5.19 ± 6.81	11.40 ± 13.10	0.031

Values are shown as mean ± SD. Abbreviations: ALX, low-frequency reactance area; CAT, COPD assessment test; FeNO, fraction of exhaled nitric oxide; FEV1, forced expiratory volume in 1 s; Fres, resonant frequency; FVC, forced vital capacity; IC, inspiratory capacity; LABA, long-acting β_2-agonists; LAMA, long-acting muscarinic antagonists; mMRC, modified medical research council; R5 and R20, respiratory system resistance at 5 and 20 Hz, respectively; X5, respiratory system reactance at 5 Hz.

Table 2. Changes in clinical parameters after add-on treatment.

	Week 0	Week 8	*p*-value
CAT-total	9.2 ± 7.3	5.4 ± 3.7	0.015
Q1 (cough)	1.4 ± 1.5	0.7 ± 0.7	0.011
Q2 (production of phlegm)	1.8 ± 1.8	0.8 ± 0.9	0.004
Q3 (chest tightness)	1.4 ± 1.2	0.8 ± 1.0	0.034
Q4 (breathlessness)	2.3 ± 1.3	1.9 ± 1.4	0.110
Q5 (activity limitation)	0.2 ± 0.4	0.2 ± 0.4	0.999
Q6 (confidence)	0.4 ± 0.9	0.2 ± 0.4	0.265
Q7 (sleep)	0.9 ± 1.7	0.3 ± 0.6	0.085
Q8 (energy)	0.8 ± 1.1	0.6 ± 0.7	0.361
mMRC	0.94 ± 0.83	0.65 ± 0.70	0.056
FeNO, ppb	63.0 ± 37.8	34.3 ± 14.0	0.006
FVC, L	3.46 ± 0.81	3.52 ± 0.73	0.326
%FVC, %	100.6 ± 23.3	102.1 ± 21.2	0.310
FEV1, L	1.87 ± 0.50	1.95 ± 0.54	0.091
%FEV1, %	67.9 ± 17.4	70.6 ± 18.0	0.124
IC, L	2.33 ± 0.58	2.34 ± 0.57	0.827

Values are shown as mean ± SD. Abbreviations: CAT, COPD assessment test; FeNO, fraction of exhaled nitric oxide; FEV1, forced expiratory volume in 1 s; FVC, forced vital capacity; IC, inspiratory capacity; mMRC, modified medical research council.



After 8 weeks of add-on treatment, CAT scores and FeNO levels improved significantly and FEV1 tended to improve. The line across the box is the median, the top and bottom portions of the box represent the 25th and 75th percentiles, respectively, and the whiskers indicate the maximum and minimum values at week 0 and 8. Abbreviations: CAT, COPD assessment test; FeNO, fraction of exhaled nitric oxide; FEV1, forced expiratory volume in 1 s.

Figure 3. Changes in CAT, FeNO, and FEV1.

spirometric indices, or FOT between patients treated with LAMA and LAMA/LABA at baseline.

Next, we stratified the patients into two groups based on a minimal clinically important improvement of two or more points on their CAT scores. Table 3 shows the characteristics of groups with and without improvement. The group with improved CAT scores had higher CAT scores at baseline than those without improvement. There were no differences noted in blood eosinophil counts, FeNO levels, IgE, or lung function. The ROC analysis for predicting CAT score improvement revealed an AUC of 0.836 for baseline CAT with the best cutoff value of 10 points at baseline (Figure 4).

During long-term follow-up, until 12 months after initiation of add-on treatment, one of the 14 patients developed pneumonia but none experienced exacerbations. Three patients quit BUD/FM during the follow-up period.

4. Discussion

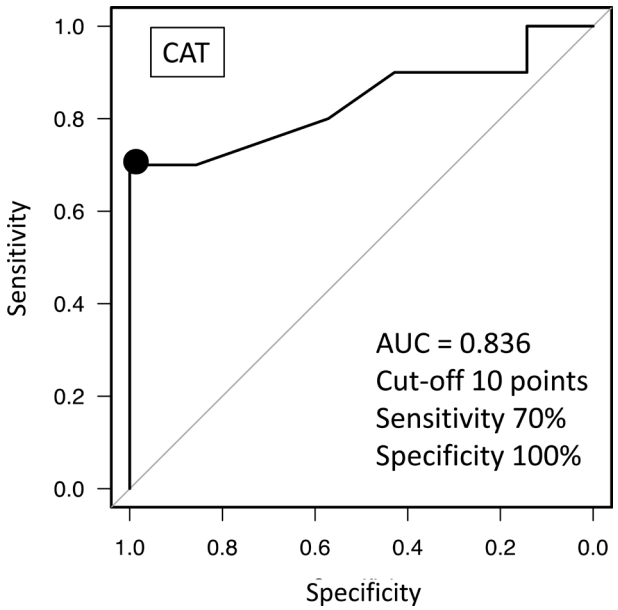
We prospectively assessed the effects of an add-on treatment with BUD/FM in patients with an eosinophilic COPD phenotype of ACO. The results revealed that the CAT scores, including total, cough, phlegm, and chest tightness showed significant improvement, and 10 out of 17 patients (58.8%) showed ≥ 2 points improvement, a minimal clinically important difference. The ROC analysis for predicting CAT score improvement revealed the best cutoff value of 10 points at baseline.

Recent evidence pertaining to type 2 inflammation indicates that FeNO is primarily driven by IL-13 and IL-4 and its levels often correlate with eosinophilic

Table 3. Comparison between patients with CAT improvement and non-improvement.

	CAT		*p*-value
	Improvement $n = 10$	Non-improvement $n = 7$	
Age, years	73.0 ± 5.5	72.4 ± 8.5	0.868
Body mass index, kg/m^2	22.7 ± 3.0	23.2 ± 3.8	0.781
Blood eosinophil count, /μL	260 ± 161	235 ± 151	0.747
FeNO, ppb	55.9 ± 14.8	73.1 ± 57.3	0.373
Reversibility, %	5.1 ± 6.2	8.0 ± 9.6	0.480
IgE, IU/ml	287 ± 611	288 ± 529	0.996
CAT	12.7 ± 7.6	4.1 ± 2.2	0.012
%FVC, %	96.2 ± 27.7	106.9 ± 14.9	0.364
%FEV1, %	66.3 ± 20.9	70.1 ± 12.0	0.678
IC, L	2.27 ± 0.67	2.41 ± 0.45	0.624

Improvement means two or more point improvement, a minimal clinically important difference, of CAT scores. Values are shown as mean ± SD. Abbreviations: CAT, COPD assessment test; FeNO, fraction of exhaled nitric oxide; FEV1, forced expiratory volume in 1 s; FVC, forced vital capacity; IC, inspiratory capacity.



The ROC analysis for predicting CAT score improvement showing the AUC of 0.836 with the best cutoff value of 10 points. Abbreviations: AUC, area under the curve; CAT, COPD assessment test; ROC, receiver operator characteristic.

Figure 4. The ROC analysis for predicting improvement in CAT.

airway inflammation driven by IL-5 [21]. We detected IL-13- or IL-4-related eosinophilic airway inflammation using FeNO measurements in patients with COPD. We selected 35 ppb as a cutoff point for high and low FeNO levels in accordance with s previously reported research on severe asthma [16]. Based on these criteria, we diagnosed 24 out of 83 patients with COPD (28.9%) as having ACO. Researchers in a previous report investigated the prevalence rate of ACO in 331 patients with COPD and identified 54 patients (16.3%) by using 35 ppb as the cutoff point [22]. A possible explanation for the difference may be that 40% of the patients were receiving ICS or oral corticosteroids in the previous study. In this study, we compared patients with high FeNO and those with low FeNO and found that the former had higher blood eosinophil counts than the latter. However, the correlation between FeNO and blood eosinophil counts was modest, which suggests that the patients with high FeNO in this study could not be selected based on blood eosinophil counts. Neither could the patients with high FeNO be selected based on their history of asthma. They presented no asthmatic symptoms, including paroxysmal dyspnea or wheeze. FeNO measurements are useful in detecting asymptomatic asthmatic components. In contrast to spirometric indices, patients with high FeNO had milder FOT parameters than those with low FeNO, suggesting that FOT may be useful in detecting the ACO phenotype of COPD.

There was no significant change in FEV1 after 8 weeks of add-on treatment, whereas the CAT scores significantly improved, including total, cough, phlegm, and chest tightness. Importantly, 10 patients (58.8%) reached a minimal clinically important difference. BUD/FM was added on to LAMA alone or after

withdrawal of LABA without a washout period. It seems that there was no room for improvement of spirometric indices after initiation of the add-on treatment. In contrast, CAT score improvement is explained by the necessity of not only LABA but also ICS. Together with the improvement in FeNO, ICS was necessary for the patients with high FeNO to improve in this study. Previous research reported the usefulness of BUD/FM in the management of COPD [23] [24]. However, patients with ACO were historically excluded from such studies. Currently, there is no consensus on ACO treatment [25]. Whether ICS/LABA, ICS/LAMA, or triple therapy is beneficial for patients with ACO requires further research.

5. Conclusion

In conclusion, add-on treatment with BUD/FM improves symptoms in patients with ACO, as determined by high FeNO levels.

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