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Original article

Lung sound analysis can be an index of the control of bronchial asthma

Terufumi Shimoda [a, *], Yasushi Obase [b], Yukio Nagasaka [c], Hiroshi Nakano [a], Reiko Kishikawa [a], Tomoaki Iwanaga [a]

[a] Clinical Research Center, Fukuoka National Hospital, Fukuoka, Japan
[b] Second Department of Internal Medicine, School of Medicine, Nagasaki University, Nagasaki, Japan
[c] Kyoto Respiratory Center, Otowa Hospital, Kyoto, Japan

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Abbreviations:
FeNO, fractional exhaled nitric oxide; FEV_1, forced expiratory volume in one second; FVC, forced vital capacity; V_{50} and V_{25}, maximal expiratory flow at 50% and 25%; ICS, inhaled corticosteroid; LSA, lung sound analysis; LF, low frequency; PC20, provocative concentration of acetylcholine causing a 20 % fall in FEV_1; E/I LF, the expiration-to-inspiration sound power ratio in a low-frequency range

ABSTRACT

Background: We assessed whether lung sound analysis (LSA) is a valid measure of airway obstruction and inflammation in patients with bronchial asthma during treatment with inhaled corticosteroids (ICSs).

Methods: 63 good adherence patients with bronchial asthma and 18 poor adherence patients were examined by LSA, spirometry, fractional exhaled nitric oxide (FeNO), and induced sputum. The expiration-to-inspiration lung sound power ratio at low frequencies between 100 and 200 Hz (E/I LF) obtained by LSA was compared between healthy volunteers and bronchial asthma patients. Next, post-ICS treatment changes were compared in bronchial asthma patients between the good adherence patients and the poor adherence patients.

Results: E/I LF was significantly higher in bronchial asthma patients (0.62 ± 0.21) than in healthy volunteers (0.44 ± 0.12, $p < 0.001$). The good adherence patients demonstrated a significant reduction in E/I LF from pre-treatment to post-treatment (0.55 ± 0.21 to 0.46 ± 0.16, $p = 0.002$), whereas the poor adherence patients did not show a significant change. The decrease of E/I LF correlated with the improvement of FEV_1/FVC ratio during the ICS treatment ($r = -0.26$, $p = 0.04$). The subjects with higher pre-treatment E/I LF values had significantly lower FEV_1/FVC and $V_{50,\%pred}$ ($p < 0.001$), and significantly higher FeNO and sputum eosinophil percentages ($p = 0.008$ and $p < 0.001$, respectively).

Conclusions: The E/I LF measurement obtained by LSA is useful as an indicator of changes in airway obstruction and inflammation and can be used for monitoring the therapeutic course of bronchial asthma patients.

Introduction

Bronchial asthma is a chronic inflammatory disease characterized by airway infiltration of inflammatory cells, including eosinophils, mast cells, macrophages, neutrophils, and lymphocytes, as well as airway narrowing and increased bronchial hyperresponsiveness (BHR).[1]

Lung sound auscultation with a stethoscope is routinely performed in clinical practice.[2] Adventitious sounds that are audible on auscultation, such as rhonchi and wheezes, play an important role in the diagnosis of bronchial asthma.[3,4] However, wheezes due to airway obstruction are not as sensitive a diagnostic indicator as changes in lung sounds. When airway obstruction is mild, wheezes are often inaudible on auscultation with a stethoscope. Lung sound analysis (LSA) is useful for examining the pathophysiology of bronchial asthma because it is more sensitive than auscultation with a stethoscope. Furthermore, LSA is non-invasive and repeatable. We previously reported that the expiration-to-inspiration sound power ratio in a low-frequency range, between 100 and 195 Hz (E/I LF), was increased in bronchial asthma patients with airway inflammation and obstruction.[5]

* Corresponding author. Clinical Research Center, Fukuoka National Hospital, 4-39-1 Yakatabaru, Minami-ku, Fukuoka 811-1394, Japan.
E-mail address: t-shimoda@mfukuoka2.hosp.go.jp (T. Shimoda).
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In this study, we examined airway obstruction, inflammation, and BHR to evaluate whether E/I LF is useful for monitoring the effect of ICS therapy on the clinical course of bronchial asthma.

Methods

Subjects

Our study included 79 patients with mild persistent bronchial asthma who were examined before and after 6 months of ICS therapy (800 μg/day as CFC-BDP equivalent). Of these patients, 63 (82%) used ICS regularly as prescribed (90–100% adherence; according to the concordance rate between the asthma diary and the inhalation usage) were classified in the good adherence group. 14 subjects (18%) used ICS irregularly (30–50% adherence) were classified in the poor adherence group. There were 2 patients with 50–90% adherence and no patients with less than 30% adherence. Bronchial asthma was diagnosed according to the Global Initiative for Asthma Guidelines.[1] All subjects had to present values less than or equal to 8000 mcg/mL in a test of BHR to inhaled acetylcholine, and they had to have a history of wheezing and/or dyspnea. At the initial visit, 80% of the subjects showed positive reversibility (reversible with at least 12% and 200 mL improvements in FEV_1 after bronchodilator therapy), whereas the remaining 20% had negative reversibility with normal respiratory function at the visit and were diagnosed with bronchial asthma based on positive BHR and medical history.

At the enrollment, no patients were treated with inhaled or systemic corticosteroids. The use of anti-asthma drugs, including bronchodilators, was discontinued for at least 24 h prior to the examination. Subjects with a history of chronic obstructive pulmonary disease, any cardiovascular diseases, or with a current viral or bacterial infection were excluded from the study. The healthy control subjects ($n = 27$) had no respiratory symptoms, no overt illnesses, and exhibited no abnormalities in their lung function tests and chest radiographies. The ethics committee of Fukuoka National Hospital approved the study protocol (protocol No. 20-12), and all participants received verbal and written study information before providing their informed consent.

Lung sound analysis

Lung sounds were recorded for ≥30 s over the base of the left lung using a hand-held microphone. The patients took a deep breath through a disposable mouthpiece to synchronize their breath cycles while the breath sounds were recorded. The recording system consisted of an electro-stethoscope containing a wide-range audio sensor that was adhered inside a diaphragm (Bio-Sound Sensor BSS-01; Kenz Medico, Saitama, Japan), a signal processing system, and a personal computer. The sensor had a band-pass filter range of 40–2500 Hz and a sound-collecting ability in the 40–2000 Hz range. The recorded sounds were analyzed using a sound spectrometer (Easy LSA-2008; Nakano, Fukuoka National Hospital). The recorded sounds were re-sampled to 10,000 Hz and analyzed by 1024-point fast Fourier transformation (FFT) with 60% overlap into adjacent segments, using a Hanning data window. The results are presented as a sound spectrograph with frequencies in Hz on the vertical axis and time on the horizontal axis. On the display, the vertical axis displays the cycles per minute in kilohertz, and the horizontal axis displays the time in seconds. The intensity (dBSPL) of the sound is depicted by the color and brightness (Fig. 1). The recording system was calibrated with a reference sound pressure (1 kHz; 94 dB [0 dB = 20 μPa]). We defined low frequencies (LF) as frequencies of 100–200 Hz, and we determined inspiration sound pressure, expiration sound pressure, and inspiration-to-expiration sound pressure ratio in the low-frequency range (E/I LF). E/I LF data were converted from logarithmic values (dBSPL) to real values.[5]

Measurement of flow–volume curves

Lung function was measured using a spirometer (Chest Graph HI-701, Chest M.I., Tokyo, Japan). The results are expressed as a

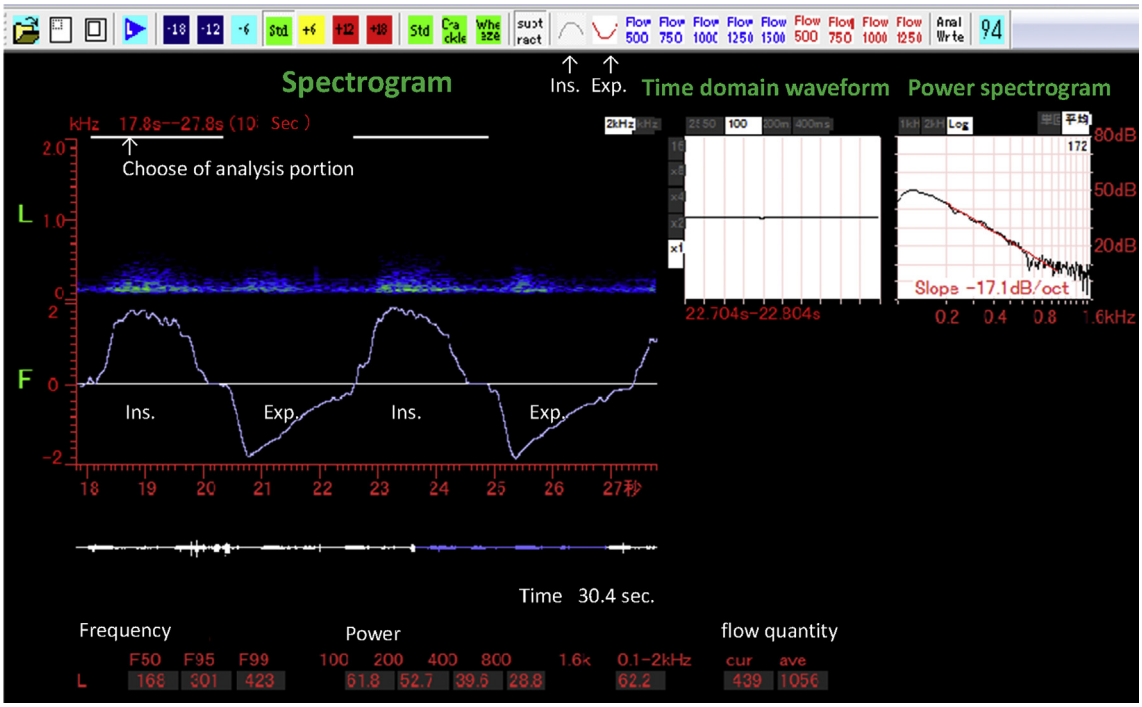

Fig. 1. Sound spectrogram display of lung sounds in a patient (Easy LSA). The recorded sound was analyzed by fast Fourier analysis and displayed as a spectrograph with the frequency shown in Hz on the vertical axis and time on the horizontal axis. The sound intensity is presented by color and brightness. Selected portion of the analysis. Selected range of inspiratory or expiratory position. Calculation of the average sound pressure level (dBSPL) at a low frequency (100–200 Hz).

percentage of the predicted values based on the relevant reference standards.[6]

Measurement of the fractional exhaled nitric oxide (FeNO) concentration

Following the guidelines published by American Thoracic Society (ATS), FeNO was measured using the online single-breath method and a fast response (0.02 s) chemiluminescence analyzer (Sievers Nitric Oxide Analyzer NOA 280i, GE Analytical Instruments, Boulder, CO, USA).[5,7] All measurements were obtained using a mouth pressure of 16 cmH_2O, corresponding to an expiratory flow of 50 mL/s. The FeNO concentrations were recorded as the average of 3 FeNO values.

Measurement of airway hyperresponsiveness to acetylcholine (Ach)

The challenge test was performed using standardized methodology. The subjects inhaled isotonic saline first for 2 min from a hand-held nebulizer (PARI BOY 038; PARI GmbH, Starnberg, Germany). They then inhaled progressive doublings of the Ach concentration from 39 to 20,000 mcg/mL. The test was continued until the FEV1.0 had decreased by >20%. BHR was expressed as the provocative concentration of acetylcholine causing a 20% fall in FEV_1 (PC20). Subjects with a PC20 < 8000 mcg/mL were considered to have positive BHR.[8]

Sputum induction and processing

The participants inhaled 5 mL 3% NaCl solution through an ultrasonic nebulizer to induce sputum collection.[9] The subjects were asked to cough during and after the inhalation and to expectorate into empty containers. Sputum was induced over a 20-min period.

The sputum samples were processed within 30 min according to a method described by Metso *et al.*[9] The sputum cells were separated by centrifugation at 2000 *g* for 10 min. The cell suspension was cytocentrifuged (Cytospin 3; Shandon, Astmoor, UK) onto microscope slides at 450 rpm for 6 min. The cytospin products from the sputum were air-dried for 30 min and then stained using the Giemsa staining method (Muto Pure Chemicals Co., Ltd., Tokyo, Japan). At least 400 non-squamous cells were counted differentially, including eosinophils, neutrophils, lymphocytes, macrophages, and ciliated epithelial cells. The results are expressed as the percentages of the total non-squamous cell counts.

Statistical analysis

The data analysis was performed using JMP (Statistical Discovery™ from SAS Institute, Cary, NC, USA). The Student's t-test was used to compare E/I LF between healthy subjects and bronchial asthma patients. The Tukey–Kramer HSD test was used to compare E/I LF among the group with sputum eosinophils <3%, the group with sputum eosinophils ≧3%, and healthy volunteers. To examine changes by ICS treatment, Student's t-test was applied to compare pre-treatment and post-treatment values between the good adherence and poor adherence groups. The correlation between the E/I LF change and the other factors were also evaluated. A suitable E/I LF cutoff value was determined using receiver operating characteristic analysis.[10] The good adherence group was stratified into two subgroups based on the cutoff value, and a t-test was performed to compare the subgroup with a pre-treatment E/I LF higher than or equal to the cutoff to the subgroup with a pre-treatment E/I LF below the cutoff value. To analyze major factors influencing the pre-treatment E/I LF value, variable selection was conducted in a stepwise manner with a combination of forward and backward selection techniques; a predictive model was then constructed, and a nominal logistic regression analysis was performed. Finally, to investigate factors associated with ICS-unresponsive E/I LF in the good adherence group, subjects were stratified based on E/I LF reduction after ICS therapy and compared using Student's t-test.

Results

Background characteristics of bronchial asthma patients and healthy subjects before treatment

Age, body mass index (BMI), and sex were not significantly different between bronchial asthma patients and healthy subjects. However, $FEV_{1.0}/FVC$ (%), $FEV_{1.0,\%pred}$ and $V_{50,\%pred}$ were significantly reduced in bronchial asthma patients compared with healthy controls ($p < 0.001$, $p = 0.002$, and $p < 0.001$, respectively) (Table 1).

E/I LF was significantly higher in bronchial asthma patients (0.55 ± 0.20) compared with healthy subjects (0.44 ± 0.12, $p = 0.0006$) (Table 1). When bronchial asthma patients were stratified into two subgroups according to sputum eosinophil percentages (<3% vs. ≥3%) and compared with the healthy group, E/I LF was significantly higher in the bronchial asthma patient subgroup with sputum eosinophils ≥3% (0.62 ± 0.21) than in the subgroup with sputum eosinophils <3% (0.45 ± 0.16) or the healthy subject group ($p < 0.001$). No significant differences were noted between the bronchial asthma subgroup with eosinophils <3% and the healthy group (Fig. 2).

Table 1
Patient characteristics.

	BA ($n = 77$)	HV ($n = 27$)	p-Value (BA vs. HV)
	Mean ± SD	Mean ± SD	
Age (yr)	42.7 ± 13.8	38.5 ± 13.6	0.09
BMI	22.8 ± 3.4	22.0 ± 4.0	0.36
Male/Female	32/45	9/18	0.45
Duration (yr)	5.0 ± 7.8		
Atopic/non-atopic	53/29		
Non-smoker/ex- smoker/smoker	42/18/18	15/5/7	0.91
$FEV_1/FVC\%$	75.9 ± 9.4	84.9 ± 7.1	<0.0001
FEV_1,%pred.	92.1 ± 14.1	101.2 ± 11.3	0.0015
V_{50},%pred.	66.6 ± 24.9	88.1 ± 17.2	<0.0001
E/I LF	0.55 ± 0.20	0.44 ± 0.12	0.0006

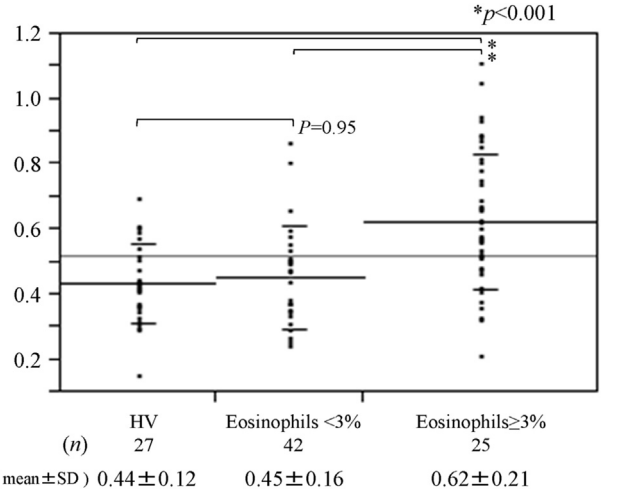

Fig. 2. Comparison of E/I LF among healthy subjects, bronchial asthma with sputum eosinophils <3% and bronchial asthma with sputum eosinophils ≥3%. E/I LF is significantly higher in bronchial asthma with sputum eosinophils ≥3% than in bronchial asthma with sputum eosinophils <3% or in healthy subjects.

Changes in spirogram, PC_{20}, FeNO, and sputum cell counts in the good and poor adherence groups during ICS therapy

No significant differences were observed in all parameters before treatment between the good adherence and poor adherence groups. In the good adherence group, $FEV_{1.0}/FVC$ (%), $FEV_{1.0,\%pred}$, $V_{50,\%pred}$, and $logPC_{20}$ values were significantly increased ($p < 0.0001$), and FeNO and the sputum eosinophil percentage were significantly decreased after ICS therapy ($p < 0.0001$). However, the poor adherence group did not show noticeable changes in any of these parameters after the study treatment (Table 2).

Changes in E/I LF values by ICS therapy in the good adherence and poor adherence groups

There were no significant differences in E/I LF values before treatment between the poor and good adherence groups ($p = 0.574$). After ICS therapy, E/I LF values were significantly reduced in the good adherence group ($p = 0.002$) but not in the poor adherence group ($p = 0.345$) (Fig. 3).

The correlations of the E/I LF changes with other factors by the ICS treatment in good adherence patients

The change of E/I LF was correlated with the improvement of FEV1.0/FVC ($r = -0.26$, $p = 0.04$) (Table 3).

Comparison of background factors, spirogram, PC_{20}, FeNO, and sputum cell counts between subgroups with pre-treatment E/I LF ≥0.50 vs. <0.50 in the good adherence group

A receiver operating characteristic analysis was performed using the pre-treatment data from healthy subjects and bronchial asthma patients with airway inflammation to investigate the discrimination power and cutoff of E/I LF. Based on the results, when the area under the curve was 0.76 and the E/I LF point on the ROC curve closest to (0, 1) was 0.50 with a sensitivity of 0.71 and a specificity of 0.69, then the E/I LF cutoff of 0.50 was established for further analysis. In the good adherence group, the subgroup with a pre-treatment E/I LF ≥ 0.50 included more males than the subgroup with a pre-treatment E/I LF < 0.50 ($p = 0.049$). The higher E/I LF subgroup also had a significantly lower FEV_1/FVC (%) and $V_{50,\%pred}$

Table 2
Comparison of data between before treatment and after treatment.

	Good adherence				Poor adherence				
		Before treatment	After treatment			Before treatment	After treatment		†
	n	mean ± SD	mean ± SD	*p*	*n*	mean ± SD	mean ± SD	*p*	
FEV_1/FVC(%)	63	75.8 ± 9.7	79.3 ± 6.4	<0.0001	14	76.6 ± 7.7	79.1 ± 7.0	0.12	0.34
FEV_1,%pred	63	93.1 ± 14.4	99.1 ± 11.6	<0.0001	14	87.9 ± 12.3	91.7 ± 12.5	0.19	0.42
V_{50},%pred	63	65.9 ± 24.2	75.9 ± 22.8	<0.0001	14	69.8 ± 28.8	70.3 ± 15.2	0.93	0.36
$logPC_{20}$	63	2.83 ± 0.50	3.33 ± 0.61	<0.0001	14	2.85 ± 0.80	3.22 ± 0.67	0.053	0.83
FeNO	58	71.1 ± 70.1	31.1 ± 24.0	<0.0001	13	83.1 ± 75.4	71.9 ± 65.1	0.56	0.67
Macrophages (%) in sputum	49	38.4 ± 22.3	48.0 ± 24.0	0.003	10	40.8 ± 23.6	45.5 ± 20.8	0.69	0.87
Neutrophils (%) in sputum	49	38.4 ± 22.3	44.0 ± 23.6	0.12	10	35.0 ± 23.6	38.3 ± 16.6	0.69	0.98
Eosinophils (%) in sputum	49	17.6 ± 22.7	3.7 ± 9.0	<0.0001	10	18.1 ± 29.1	13.1 ± 15.6	0.65	0.96
E/I LF	63	0.55 ± 0.21	0.46 ± 0.16	0.002	63	0.55 ± 0.18	0.49 ± 0.17	0.35	0.92

† Comparison of before treatment values at poor adherence and good adherence group.

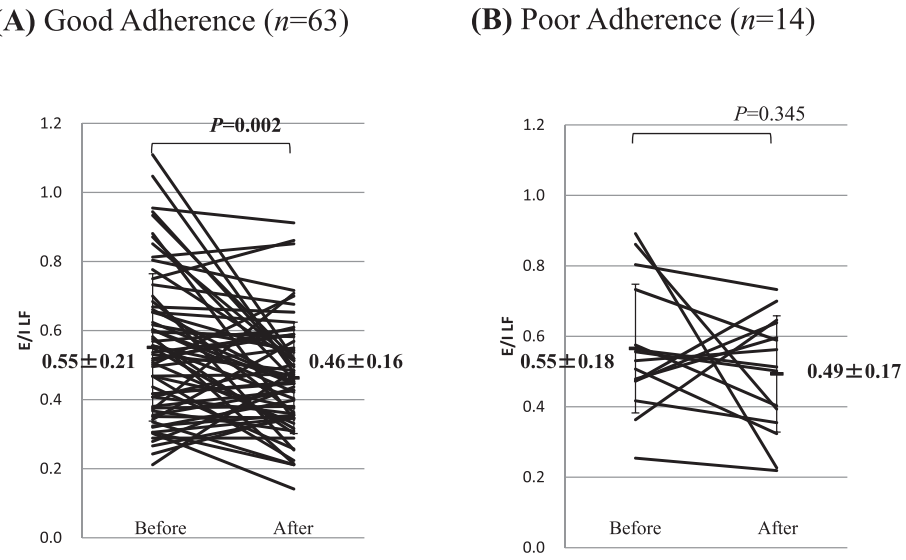

Fig. 3. Change in E/I LF after ICS therapy. **(A)** In the good adherence group, post-treatment E/I LF values are significantly reduced compared with pre-treatment levels. **(B)** In the poor adherence group, post-treatment E/I LF values are not significantly different from pre-treatment values.

Table 3
The relation between the change of E/I LF and the changes of other factors.

ΔE/I LF		
vs. factors	*r*	*p* Values
ΔFEV1.0/FVC(%)	−0.26	0.04
$\Delta FEV_{1.0}$,%pred	−0.12	0.36
ΔV_{50},%pred	−0.18	0.16
ΔV_{25},%pred	−0.07	0.58
$\Delta logPC_{20}$	0.09	0.50
ΔFeNO	0.01	0.95
ΔMacrophage	−0.02	0.90
ΔNeutrophils	0.10	0.51
ΔEosinophils	−0.07	0.61

($p < 0.001$), as well as a significantly higher FeNO and sputum eosinophil percentage ($p = 0.008$ and $p < 0.001$, respectively) (Table 4A).

In a model search performed to predict factors that affect the pre-treatment E/I LF value, the sputum eosinophil percentage had the most profound effect on the pre-treatment E/I LF value ($p = 0.034$), followed by FEV_1/FVC(%) ($p = 0.035$) (Table 4B).

Investigation of factors responsible for ICS-unresponsive E/I LF in the good adherence subgroup with a pre-treatment E/I LF ≥ 0.50

When the subjects in the good adherence group with a pre-treatment E/I LF ≥ 0.5 were stratified into subjects whose E/I LF decreased after ICS treatment (responders) and subjects without E/I LF reduction (non-responders), the non-responders showed a significant elevation in sputum neutrophil percentages after ICS therapy compared with the responders ($p = 0.049$) (Table 5).

Discussion

We found that the LSA may be useful to deduce the patient ICS adherence and the lung function improvement, however, it is not a specific index to the airway eosinophilic inflammation change.

Lung sounds are generated in areas closer to large airways. The pitch and strength of normal lung sounds are proportional to the square of the airflow velocity.[11] Lung sounds attenuate in proportion to the distance squared from the sound source, and high-pitched tone sounds are not transmitted in alveolar areas that are located far from the sound source. In asthmatic patients, the frequency and strength of lung sounds are modified due to fluctuations in respiratory flow and airway damage caused by inflammation.[12] In broncho-provocation tests, acute airway narrowing causes an increase in the frequency and intensity of lung sounds.[12–17] Fiz *et al.* reported a greater decrease in the frequency of wheezing during maximal forced exhalation after bronchodilation in asthmatics compared with normal controls.[18] Shreur *et al.* has suggested LSA can detect differences in the pathophysiology of airway narrowing in asthma.[19]

We observed higher E/I LF values in bronchial asthma patients with obvious airway inflammation compared with healthy people and asthmatic patients without airway inflammation. Our previously proposed LSA, which uses E/I LF as an index of inflammation and obstruction in asthmatic patients, is useful, simple and noninvasive.[4] Habukawa *et al.* has reported that LSA is useful for evaluating the impact of medications on bronchial asthma control.[20] Improving treatment adherence is a complex task.[21] In the present study, a significant improvement in airway obstruction with reduced airway inflammation and hyperresponsiveness was observed in the good adherence group but not in the poor adherence group. Similarly, evaluation of the changes in E/I LF after ICS therapy confirmed a reduction in E/I LF in the good adherence group but not in the poor adherence group and the decrease of E/I LF was related with the improvement of FEV1/FVC during the ICS medication.

The subgroup with a higher pre-treatment E/I LF had a higher degree of airway obstruction and eosinophilic airway inflammation. In the good adherence group, 79% of subjects with higher pre-treatment E/I LF values demonstrated E/I LF reduction after ICS treatment. However, the remaining 21% did not show a reduction in

Table 4A
Patient characteristics of good adherence group (Divided into >0.50 and ≤0.50 pre-treatment E/I LF values).

	E/I LF ≥ 0.50	E/I LF < 0.50	*p* Values
	(*n* = 37)	(*n* = 26)	
Age (yr)	40.0 ± 11.2	45.8 ± 14.3	0.076
BMI	22.7 ± 2.8	23.3 ± 4.0	0.53
Male/Female	19/18	7/19	0.049
Duration (yr)	4.3 ± 7.5	4.5 ± 7.2	0.92
Atopic/non-atopic	22/15	17/9	0.633
Non-smoker/ex-/smoker	21/9/7	14/5/7	0.725
$FEV_{1.0}$/FVC%	72.3 ± 10.5	80.7 ± 5.9	0.0002
$FEV_{1.0}$,%pred.	90.5 ± 16.4	96.7 ± 10.1	0.072
V_{50},%pred.	57.0 ± 22.6	78.5 ± 20.7	0.0003
$logPC_{20}$	2.76 ± 0.48	2.93 ± 0.52	0.19
FeNO	99.7 ± 78.1	44.0 ± 73.8[†]	0.008
Macrophages (%) in sputum	31.2 ± 17.6[‡]	49.7 ± 20.7[§]	0.002
Neutrophils (%) in sputum	37.5 ± 21.7[‡]	35.1 ± 24.0[§]	0.717
Eosinophils (%) in sputum	27.6 ± 26.2[‡]	7.4 ± 13.8[§]	0.0006

[†] *n* = 23.
[‡] *n* = 32.
[§] *n* = 21.

Table 4B
Logistic regression analysis (Divided into >0.50 and ≤0.50 pre-treatment E/I LF values).

	Estimate	SE	Chi square	*p* (Prob > ChiSq)
Age	−0.008	0.033	0.06	0.808
Sex [F]	0.605	0.448	1.82	0.177
Smoking (Pack-years)	0.033	0.032	1.09	0.297
FEV_1/FVC%	0.125	0.059	4.44	0.035
FEV_1,%pred.	−0.029	0.035	0.68	0.410
$logPC_{20}$	0.250	0.953	0.07	0.793
Eosinophils (%) in sputum	−0.067	0.032	4.49	0.034

Table 5
Comparison of the background by E/I LF > 0.50.

	Responder	Non-responder	*p* Values
	(*n* = 29)	(*n* = 8)	
Age (yr)	45.2 ± 14.8	48.0 ± 13.3	0.618
BMI	22.7 ± 2.9	22.9 ± 2.7	0.881
Male/Female	13/16	6/2	0.13
Duration (yr)	4.3 ± 7.8	4.3 ± 6.7	0.98
Atopic/non-atopic	19/10	3/5	0.153
Non-smoker/ex-/smoker	18/6/5	3/3/2	0.45
ΔFEV1.0/FVC%	5.16 ± 8.65	6.45 ± 5.50	0.616
ΔFEV1.0,%pred.	6.82 ± 12.89	11.16 ± 6.77	0.213
ΔV50,%pred.	11.39 ± 18.27	13.64 ± 15.86	0.737
ΔlogPC20	0.46 ± 0.61	0.41 ± 0.71	0.958
Δlog FeNO	−0.32 ± 0.32[†]	−0.46 ± 0.25	0.244
Δ Macrophages (%) in sputum	12.22 ± 23.64[‡]	8.580 ± 35.67[§]	0.821
Δ Neutrophils (%) in sputum	2.13 ± 26.92[‡]	24.50 ± 20.28[§]	0.049
Δ Eosinophils (%) in sputum	−15.07 ± 14.60[‡]	−32.33 ± 33.91[§]	0.056

Responder: E/I LF is decrease by treatment, Non-responder: E/I LF is not decrease by treatment.
Δ, Amount of changes.
[†] *n* = 28.
[‡] *n* = 23.
[§] *n* = 6.

E/I LF. We found that the responder group had a reduced sputum eosinophil percentage, whereas the non-responder group had a reduced sputum eosinophil percentage and an increased neutrophil percentage also. This reason may be that the neutrophilic airway inflammation has been shown to respond poorly to steroids.[22]

The present findings indicate that E/I LF increases in both eosinophilic and neutrophilic types of airway inflammation. Among bronchial asthma patients with high E/I LF values who received ICS treatment, those who showed a reduction in E/I LF were likely to have experienced an improvement in eosinophilic airway inflammation that did not involve neutrophils. Table 5 suggests the E/I LF decrease was also caused by neutrophilic inflammation decrease. The patients with high E/I LF before treatment had high level of both of eosinophilic and neutrophilic inflammation. E/I LF may be possible to be an index of the bronchial inflammation and obstruction level in asthmatic patients, but is not specified to the eosinophilic inflammation.

In conclusion, E/I LF is useful for evaluating the efficacy of ICS therapy in patients with mild persistent bronchial asthma. We anticipate that E/I LF could be applied as a new noninvasive indicator method of bronchial asthma control level.

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Conflict of interest

The authors have no conflict of interest to declare.

Authors' contributions

TS designed the study and wrote the manuscript. YO supported the writing of the manuscript and the data analysis. YN participated in the data analysis. HN participated in the design of the study and the data analysis. RK assisted in the data interpretation. TI coordinated the recruitment of patients.

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