

A Comparative Study of Type 2 Inflammatory Biomarkers Combined with Small Airway Parameters for Diagnosing Cough-Variant Asthma

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Background: The diagnosis of cough-variant asthma (CVA) traditionally relies on clinical manifestations and bronchial provocation test (BPT). However, the BPT can be costly and complex, and poses risks of severe bronchospasm. Most CVA patients exhibit type 2 airway inflammation and small airway dysfunction. Identifying practical biomarkers such as total immunoglobulin E (tIgE), percentage of eosinophil cell count in induced sputum (sEOS%), peripheral blood eosinophil count (PBEC), and fractional exhaled nitric oxide (FeNO) may enhance the diagnostic process for cough-variant asthma.

Objective: This study aims to evaluate and compare the diagnostic utility of T2 inflammatory biomarkers combined with small airway parameters for diagnosing CVA.

Methods: This was a single-center, cross-sectional diagnostic study including 211 patients with chronic cough. Participants underwent spirometry, FeNO measurement, and assessment of eosinophilic inflammation, including sEOS%, PBEC, and tIgE. Receiver operating characteristic (ROC) curve analysis was used to evaluate the diagnostic performance of individual parameters and combined models.

Results: CVA was diagnosed in 27.5% of patients. Compared with non-CVA patients, those with CVA showed significantly higher levels of T2 inflammatory biomarkers and reduced small airway function. Among individual biomarkers, FeNO demonstrated the highest diagnostic accuracy (AUC = 0.79), followed by sEOS% (AUC = 0.75), tIgE (AUC = 0.71), and PBEC (AUC = 0.66). Among small airway parameters, FEF_{50%} showed the best performance (AUC = 0.79). Combined models significantly improved diagnostic accuracy, with FEF_{50%} combined with FeNO or sEOS%, achieving the highest AUC (0.82). Correlation analysis revealed that higher inflammatory biomarker levels were associated with reduced small airway function.

Conclusion: The combination of T2 inflammatory biomarkers and small airway function parameters improves the diagnostic accuracy for CVA. This approach may provide a practical and non-invasive strategy for identifying CVA, particularly in settings where bronchial provocation testing is not readily available.

Keywords: chronic cough, cough-variant asthma, T2 inflammatory biomarkers, small airway

Introduction

Chronic cough (CC) in adults is defined as a cough lasting more than 8 weeks, with a global prevalence of approximately 10%.¹ Cough variant asthma (CVA) is a phenotype of asthma where the primary or sole symptom is a chronic cough without the classic symptoms of wheezing, chest tightness, or dyspnea. CVA is a significant cause of chronic cough, accounting for approximately 25% to 42% of cases.^{2,3} Currently, the diagnosis of CVA relies on the bronchial provocation test, which is expensive, complicated, and risks inducing severe bronchial spasms. Therefore, a more convenient diagnostic method for CVA is needed.

Asthma is a chronic inflammatory disorder of the airways characterized by episodes of wheezing, chest tightness, shortness of breath, and coughing.⁴⁻⁶ Although several studies have shown that CVA shares pathophysiological features with classic asthma (CA), including airway inflammation and airway hyperresponsiveness (AHR), CVA has been recognized as a type 2 (T2) inflammation-driven disorder with eosinophilic inflammation being central to its pathogenesis.^{7,8}

Biomarkers commonly used to assess airway inflammation in asthma include fractional exhaled nitric oxide (FeNO), sputum eosinophil count (sEOS%), peripheral blood eosinophil counts (PBEC), and serum total immunoglobulin E (tIgE). FeNO is a non-invasive surrogate marker of airway eosinophilic inflammation, and elevated FeNO levels predict a favorable response to inhaled corticosteroid (ICS) therapy.^{9,10} sEOS% is the gold standard for identifying eosinophilic airway inflammation and guiding corticosteroid therapy, especially in moderate-severe asthma. However, its use is limited by technical complexity and logistical challenges.¹¹ PBEC and tIgE are most accessible and valuable for phenotyping and predicting eligibility for biologic therapies, though their predictive accuracy for corticosteroid response is lower than FeNO or sEOS%. Blood eosinophil counts ≥ 300 cells/ μL and IgE ≥ 120 ng/mL are associated with poor asthma control, but these parameters are less specific and can be influenced by comorbid conditions.^{11,12}

Recent studies have highlighted the increasing complexity and heterogeneity of CVA, emphasizing the need for more precise diagnostic approaches. A recent study has identified distinct clinical and molecular phenotypes of CVA, suggesting that airway inflammation and functional impairment may vary significantly among patients.¹³ In parallel, emerging research has explored novel biomarkers, including metabolomic signatures and transcriptomic profiles, to improve disease characterization; however, these approaches remain complex and are not readily applicable in routine clinical practice.¹⁴

Despite these advances, most existing studies have focused on either inflammatory biomarkers or airway function parameters in isolation. Therefore, there remains a need for practical, non-invasive, and clinically accessible strategies that integrate multiple dimensions of CVA pathophysiology. However, previous studies have typically focused on a limited number of biomarkers or single functional parameters. The present study expands on this by systematically evaluating a broader panel of T2 inflammatory biomarkers in combination with small airway function parameters to improve diagnostic accuracy for CVA.

Methods

Subjects

Chronic cough patients who attended the Department of Pulmonary and Critical Care Medicine of Tongji Hospital between November 2022 and October 2023 were enrolled. Their clinical data were collected and analyzed after pulmonary function test, bronchial provocation test, FeNO, tIgE, sEOS%, PBEC, and other examinations (see Figure 1). This study was conducted in accordance with the ethical principles of the Declaration of Helsinki and its later amendments. The procedure was approved by the Ethics Committee of Tongji Hospital (K-2022-028) and registered with the Chinese Clinical Trials Register (<http://www.chictr.org.cn>) number ChiCTR2300067906. Written informed consent was obtained from all patients.

Patients who met all of the following criteria were eligible: (1) over 18 years old; (2) cough lasting no less than 8 weeks and with a normal chest radiograph; (3) forced expiratory volume in 1s (FEV₁) / forced vital capacity (FVC) $>70\%$, predicted FEV₁ $>80\%$.

Patients were excluded if they met one of the following criteria: (1) Current smokers or ex-smokers for ≤ 2 years; (2) Pregnant or lactating patients; (3) A history of acute upper respiratory tract infection within 8 weeks; (4) Use of inhaled or oral corticosteroid within 4 weeks, or use of montelukast and long-acting β_2 -agonists in the previous week; (5) Patients with severe cardiac insufficiency, severe liver and kidney insufficiency, mental and cognitive dysfunction, hearing and communication impairment; (6) Multiple causes of chronic cough.

According to Chinese national guidelines on the diagnosis and management of cough (2021),¹⁵ the patient who met the following criteria was diagnosed with CVA: (1) chronic cough, often accompanied by a significant night cough; (2) Positive bronchial provocation test, or positive bronchial dilation test; (3) positive to antiasthma treatment.

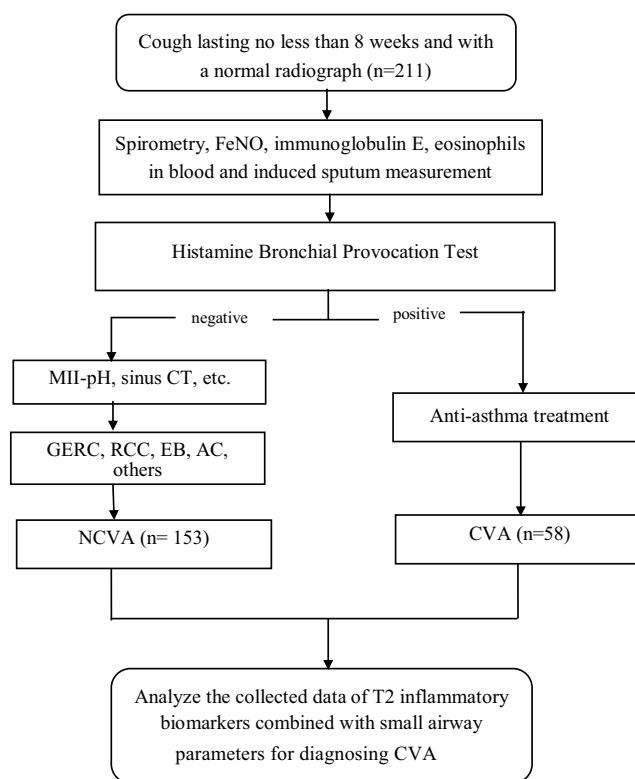


Figure 1 CONSORT (Consolidated Standards of Reporting Trials) flow diagram of the study.

FeNO Measurement

Using an Ncoulomb expiratory analyzer (Sunvou-CA2122, China), FeNO was measured following the American Thoracic Society/European Respiratory Society (ATS/ERS) guidelines and product operating instructions.¹⁶ Subjects were instructed to inhale NO-free air and exhale via a mouthpiece at a consistent rate (50 mL/s) for 10 seconds. FeNO levels were measured before spirometry and bronchial provocation tests.

Peripheral Blood Eosinophil Count

For the peripheral blood examination, 2–3 mL of venous blood was collected and analyzed by the laboratory department using an automated hematology analyzer.

Sputum Induction Measurement

After baseline Spirometry according to the ATS/ERS standard criteria,¹⁷ subjects were pretreated with inhaled salbutamol (400 µg by metered-dose inhaler) and 10 minutes later were asked to inhale a hypertonic saline (4.5%) solution during three periods of 7 min each at most using an ultrasonic nebulizer. Nebulization would be discontinued if one of the following symptoms occurred: wheezing, chest tightness, or moderate-to-severe dyspnea.

Detection of Total Serum Immunoglobulin E

Venous blood samples (2 mL) were collected from all participants and centrifuged at 3,500 rpm for 10 minutes at room temperature to obtain serum. tIgE levels were measured using a fully automated enzyme immunoassay system (AP22-speedy, China) in accordance with the manufacturer's instructions.

Spirometric Measurement

Spirometric measurements were performed using the Master Screen Impulse Oscillometry System (IOS) (Jaeger, Hoechberg, Germany) in accordance with the American Thoracic Society/European Respiratory Society (ATS/ERS) guidelines.¹⁸

Histamine Bronchial Provocation Test

Histamine is indeed used as a provocative agent in bronchial provocation testing according to the criteria of the ATS, which was performed using the Jaeger APS Pro system with a Medic-Aid sidestream nebulizer. Task Force on the Standardization of Lung Function Testing.¹⁹ A 20% decrease in FEV₁ was noted as a provocative dosage, and bronchial hyperresponsiveness (BHR) was considered present if the PD₂₀-FEV₁ ratio was less than 7.8 μmol.

Statistical Analysis

The data were analyzed using the statistical software SPSS 27, and MedCalc 23.1.3 was used for ROC curve analysis and the DeLong test. The correlation was tested with Spearman correlation test and was plotted through the “corrplot” R package using R software Version 4.4.3. The Kolmogorov–Smirnov normality test was used to determine the normality of the distributions of all continuous variables. Normally distributed variables were shown as mean ± standard deviation, whereas nonnormally distributed variables were represented by median (interquartile range). The *T*-test and Mann–Whitney *U*-tests were used to compare the differences between the CVA and NCVA groups. The receiver operating characteristic (ROC) curve and measuring the area under the curve (AUC) were used to analyze the diagnostic values of single indicators and combined indicators for CVA. The optimal cut-off point, $P < 0.05$, was considered statistically significant.

Results

Demographic Characteristics

A total of 211 chronic cough subjects with a single cause were included in this study. 58 (27.5%) subjects were eventually diagnosed with CVA. The Non-CVA (NCVA) group involved 153 patients: 43 (28.1%) patients with gastroesophageal reflux-related chronic cough (GERC), 38 (24.8%) patients with eosinophilic bronchitis (EB), 20 (13.07%) patients with refractory chronic cough (RCC), 16 (10.5%) patients with upper airway cough syndrome (UACS), 12 (7.8%) patients with atopic cough (AC) and 24 (15.3%) patients with other causes such as angiotensin-converting enzyme inhibitor (ACEI)-related cough, psychogenic cough, postinfectious cough, and so on. There were no significant differences in age, gender, Leicester cough questionnaire (LCQ) score, and cough symptom score between the two groups (Table 1).

Comparison of Pulmonary Function and T2 Inflammatory Biomarkers Between CVA and NCVA

Comparing pulmonary function and T2 inflammatory biomarkers between the two groups, CVA subjects have significantly lower FEV₁, FEV₁/FVC, FEF_{50%}, FEF_{75%}, and MMEF than NCVA subjects (all $P < 0.01$), indicating reduced small airway function. FeNO, sEOS%, tIgE, and PBEC were significantly higher in CVA subjects (all $P < 0.01$) (Table 2).

ROC Curves of Single Measurement for CVA

Significant differences in small airway function parameters and T2 inflammatory biomarkers were observed between CVA and NCVA subjects, suggesting that these could be potential indicators for diagnosing CVA in patients with chronic cough. The ROC curve was used to evaluate the diagnostic performance of these indicators, and the AUC was calculated to quantify their ability to identify CVA cases. FeNO and FEF_{50%} exhibited the highest AUC values, indicating they are the most accurate indicators for identifying CVA. MMEF showed an AUC of 0.74, which was comparable to FEF_{75%} ($P = 0.07$) but slightly lower than FEF_{50%} ($P < 0.05$). There was no significant difference between the AUC of FeNO and sEOS% ($P = 0.08$). The AUC of tIgE and PBEC was significantly lower than that of FeNO (all $P < 0.01$), indicating limited diagnostic value (Table 3).

Table 1 Demographic Characteristics of CVA and NCVA Subjects

	CVA	NCVA	p-value
N (%)	58 (27.5)	153 (72.5)	–
Age, years	50.12 ±17.71	51.76 ±15.83	0.13
Gender			0.48
Male	27 (46.6%)	63 (41.2%)	
Female	31 (53.4%)	90 (58.8%)	
LCQ	13.10±3.00	13.78±3.20	0.16
Cough Symptom Score			
Daytime	3 (2)	4 (1)	0.08
Nighttime	4 (2)	3 (2)	0.12

Abbreviations: CVA, cough-variant asthma; IQR, interquartile range; LCQ, Leicester cough questionnaire; NCVA, non-cough-variant asthma; SD, standard deviation. Data were presented as mean ± SD or median (IQR).

Table 2 Comparison of Pulmonary Function and T2 Inflammatory Biomarkers Between CVA and NCVA

	CVA (n = 58)	NCVA (n = 153)	p-value
FEV ₁ , %predicted	86.46±18.15	94.61±19.19	< 0.01*
FEV ₁ /FVC, %	74.42±12.83	79.81±12.05	< 0.01*
FEF _{50%} , %predicted	59.37±17.55	84.24±29.53	< 0.01*
FEF _{75%} , %predicted	69.47±19.72	88.19±24.33	< 0.01*
MMEF, %predicted	57.10±18.50	79.63±30.04	< 0.01*
FeNO, ppb	31.00 (19.00)	16.00 (15.00)	< 0.01*
sEOS, %	5.00 (9.20)	2.00 (3.30)	< 0.01*
tlgE, IU/mL	61.35 (147.30)	17.50 (55.10)	< 0.01*
PBEC, 10 ⁹ /L	0.24 (0.23)	0.16 (0.22)	< 0.01*

Abbreviations: CVA, cough-variant asthma; FEF_{50%}, forced expiratory flow at 50% of forced vital capacity; FEF_{75%}, forced expiratory flow at 75% of forced vital capacity; FENO, fractional exhaled nitric oxide; sEOS%, percentage of eosinophil differential cell count in induced sputum; PBEC, peripheral blood eosinophil counts; tlgE, total serum immunoglobulin E; FEV₁, forced expiratory volume in 1 s; FVC, forced vital capacity; IQR, interquartile range; MMEF, maximal mid-expiratory flow; 10⁹/L, billion per liter; IU/mL international units per milliliter; ppb, parts per billion; SD, standard deviation. Data were presented as mean ± SD or median (IQR). *P < 0.05.

ROC Curves of T2 Inflammatory Biomarkers Combined with Small Airway Function Parameter (FEF_{50%}) for Diagnosing CVA

According to the results in [Table 3](#), we selected the small airway function parameter with the most accurate AUC to combine with T2 inflammatory biomarkers. The combining model was assessed using ROC curve analysis ([Figure 2](#)). Across all the models tested, the combination of FeNO or sEOS% with FEF_{50%} showed higher AUC values compared to each marker assessed individually. In most comparisons, these improvements were statistically significant ([Table 4](#)).

Correlation Between T2 Inflammatory Biomarkers and Small Airway Parameters in Chronic Cough Subjects with CVA

Correlation analysis ([Figure 3](#)) revealed strong positive associations among small airway function parameters. FEF_{50%} showed a very strong correlation with MMEF ($r = 0.905$, $p < 0.001$) and a strong correlation with FEF_{75%} ($r = 0.767$, $p < 0.001$). MMEF was also strongly correlated with FEF_{75%} ($r = 0.681$, $p < 0.001$), confirming the consistency of these parameters in reflecting peripheral airway function. Among inflammatory biomarkers, sEOS% showed moderate negative correlations with

Table 3 Optimal Cutoff Values for Predicting CVA

Parameter	Cutoff	AUC	Sensitivity (%)	Specificity (%)	PPV (%)	NPV (%)	PLR	NLR
FEV1	97.20	0.66	79.31	53.59	39.3	87.2	1.71	0.39
FEV1/FVC	77.21	0.67	56.90	73.20	44.60	81.80	2.12	0.59
FEF _{50%}	77.80	0.79	96.55	62.75	50.0	98.0	2.64	0.05
FEF _{75%}	89.70	0.78	96.50	55.66	44.30	95.50	2.09	0.12
MMEF	76.20	0.74	89.66	56.86	44.10	93.50	2.08	0.18
FeNO	23	0.79	81.03	73.20	53.40	91.10	3.02	0.26
sEOS%	2.5	0.75	77.60	61.44	43.30	87.90	2.01	0.36
tIgE	19.8	0.71	84.48	54.90	41.50	90.30	1.87	0.28
PBEC	0.29	0.66	44.83	86.93	56.50	80.60	3.43	0.63

Abbreviations: AUC, area under the curve; CVA, cough-variant asthma; FEF_{50%}, forced expiratory flow at 50% of forced vital capacity; FEF_{75%}, forced expiratory flow at 75% of forced vital capacity; FENO, fractional exhaled nitric oxide; sEOS%, percentage of eosinophil differential cell count in induced sputum; PBEC, peripheral blood eosinophil counts; tIgE, total serum immunoglobulin E; FEV₁, forced expiratory volume in 1 s; FVC, forced vital capacity; MMEF, maximal mid-expiratory flow; NLR, negative likelihood ratio; NPV, negative predictive value; PLR, positive likelihood ratio; PPV, positive predictive value. Cutoff points of FEV₁, FEV₁/FVC, FEF_{50%}, FEF_{75%}, and MMEF are in percentage predicted; the cutoff point of FeNO is in ppb; the cutoff point of sEOS is in percentage; the cutoff point of tIgE is in IU/mL; the cutoff point of PBEC is in billion per liter.

MMEF ($r = -0.356$, $p = 0.006$), FEF_{50%} ($r = -0.386$, $p = 0.003$), and FEF_{75%} ($r = -0.447$, $p < 0.001$). Similarly, PBEC showed moderate negative correlations with MMEF ($r = -0.417$, $p = 0.001$), FEF_{50%} ($r = -0.421$, $p = 0.001$), and FEF_{75%} ($r = -0.340$, $p = 0.009$). FeNO was moderately negatively correlated with small airway parameters, including MMEF ($r = -0.284$, $p = 0.031$) and FEF_{50%} ($r = -0.295$, $p = 0.025$), while its correlation with FEF_{75%} did not reach statistical significance ($r = -0.247$, $p = 0.061$). tIgE showed no significant correlation with small airway function parameters, including MMEF ($r = -0.057$, $p = 0.672$), FEF_{50%} ($r = -0.120$, $p = 0.371$), and FEF_{75%} ($r = -0.106$, $p = 0.430$). Additionally, tIgE was not significantly correlated with FeNO ($r = 0.042$, $p = 0.756$) or PBEC ($r = 0.077$, $p = 0.567$). sEOS% was positively correlated with FeNO ($r = 0.273$, $p = 0.038$), whereas PBEC showed no significant association with FeNO ($r = 0.064$, $p = 0.631$).

Discussion

CVA is a phenotype of asthma in which the primary, and often sole, symptom is chronic cough. CVA is one of the leading causes of chronic cough cases. More than 30% to 40% of adults with CVA who do not receive appropriate treatment may progress to CA. Our study explores the diagnostic potential of combining T2 inflammatory biomarkers with small airways function parameters, enhancing their ability to identify CVA.

In this study, CVA was identified in 27.5% of all patients with chronic cough. Comparing pulmonary function and T2 inflammatory biomarkers between the CVA and NCVA groups, we observed that CVA subjects exhibited significantly reduced pulmonary function, whereas T2 inflammatory biomarkers were notably higher.

Although CVA is often associated with normal baseline spirometry, subtle small airway dysfunction may already be present. These changes may not affect FEV₁ or FEV₁/FVC but can be detected using more sensitive parameters such as FEF_{50%}, FEF_{75%}, and MMEF. Our findings suggest that CVA is associated with early small airway impairment, consistent with a continuum toward classic asthma.

T2 inflammatory biomarkers are key clinical indicators for identifying T2 asthma and airway inflammation.^{12,20,21} FeNO is primarily produced in the bronchial epithelium by the enzyme inducible nitric oxide synthase (iNOS), which is upregulated in response to T2 inflammatory cytokines such as IL-4 and IL-13. Elevated FeNO levels are indicative of increased type 2 inflammation.^{22,23} Measuring FeNO at a flow rate of 50 mL/s (FeNO₅₀) is indeed a well-established method for assessing central airway inflammation.^{10,24} Furthermore, the study by Lehtimäki et al highlights that FeNO₅₀ is sensitive to changes in the NO dynamics in the central airways, making it a useful tool for differentiating between central and peripheral inflammation.¹⁰ Eosinophilic inflammation is a hallmark of asthma, particularly in the context of T2 immune responses. Elevated levels of eosinophils in serum and sputum further support the presence of eosinophilic inflammation in CVA.^{25,26} Sputum eosinophilia in CVA is not only indicative of ongoing airway inflammation but also serves as a predictor for the potential progression to CA. PBEC, while less specific than

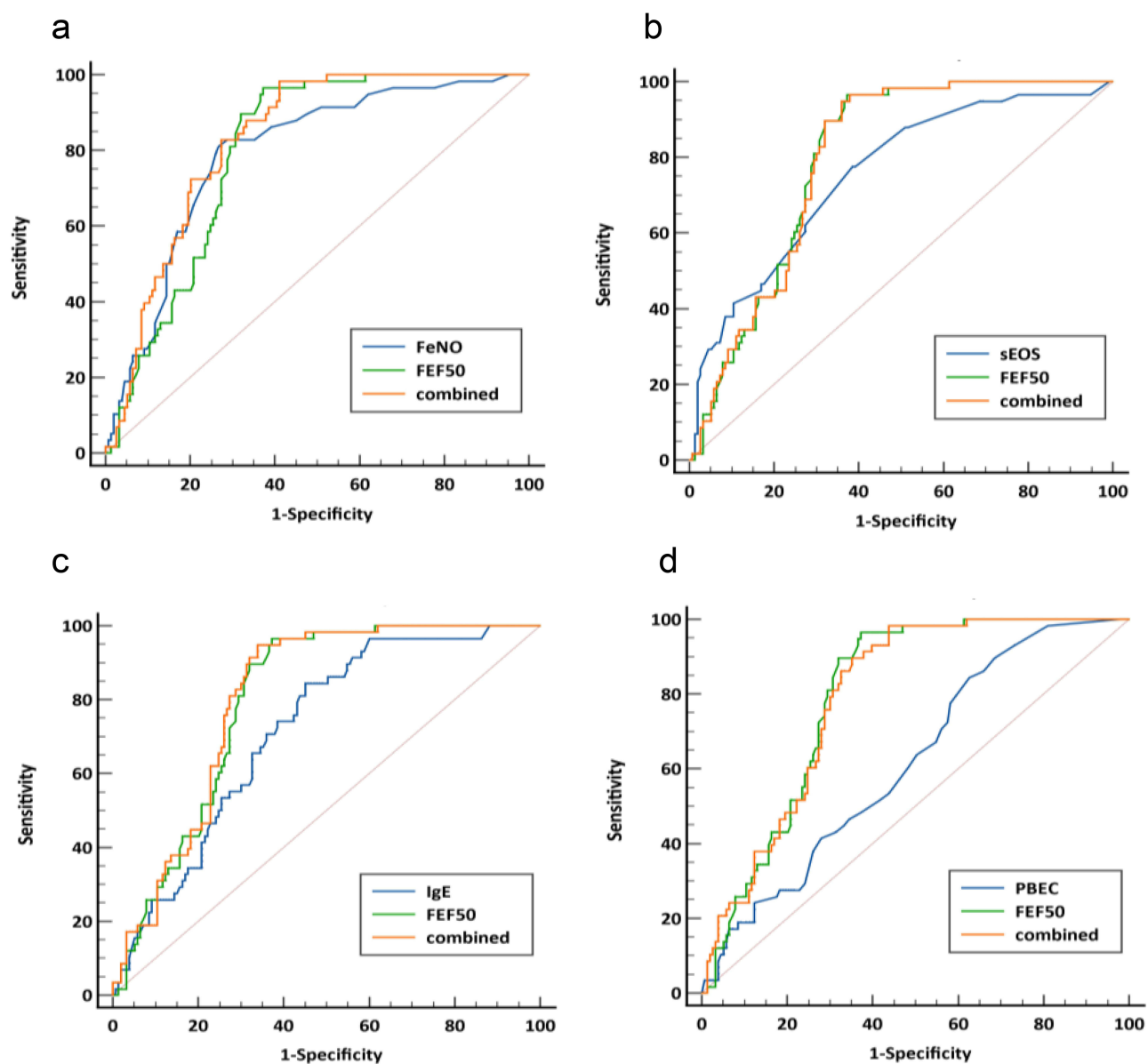


Figure 2 ROC curves of: (a) FeNO combined with FEF_{50%}, (b) sEOS% combined with FEF_{50%}, (c) tIgE combined with FEF_{50%}, (d) PBEC combined with FEF_{50%}, for diagnosing CVA.

sEOS%, also reflect systemic eosinophilic activity and correlate with disease severity in asthma, including CVA.²⁵ IgE is indeed a key mediator of T1 hypersensitivity reactions, which are central to the pathogenesis of allergic asthma.^{27,28} Sunyer et al reported that total serum IgE levels were associated with asthma independently of specific IgE levels to common aeroallergens.²⁹

Consistent with previous studies, FeNO showed the best diagnostic accuracy among individual biomarkers with an AUC of 0.79. Compared with sEOS% (AUC of 0.75), PBEC (AUC of 0.66), and tIgE (AUC of 0.71). sEOS% appears to be a more consistent marker of eosinophilic airway inflammation in CVA than PBEC. tIgE reflects allergic sensitization but has moderate specificity, as it may also be elevated in other allergic conditions.

In clinical practice, MMEF, FEF_{50%}, and FEF_{75%} are key spirometric parameters for evaluating small airway function.²⁸⁻³⁰ Among small airway function parameters, FEF_{50%} showed the highest diagnostic performance (AUC=0.79) and lower variability compared with MMEF and FEF_{75%}. This suggests that FEF_{50%} better captures early airflow limitation in the mid-small airways, where obstruction typically begins in CVA. In addition, FEF_{50%} is less

Table 4 Diagnosis Accuracy for Combined T2 Inflammatory Biomarkers and Small Airway Function in CVA

Parameter Combination	AUC Combined (95% CI)	Biomarker AUC (95% CI)	AUC of FEF _{50%} (95% CI)	Statistical Notes
FeNO+FEF _{50%}	0.82 (0.77–0.87)	FeNO= 0.79 (0.73–0.84)	FEF _{50%} = 0.79 (0.72–0.84)	<i>P</i> = 0.05 (FeNO vs combined model); <i>p</i> < 0.01 (FEF _{50%} vs FeNO alone and the combined model)
sEOS%+FEF _{50%}	0.82 (0.77–0.86)	sEOS%= 0.75 (0.70–0.81)	FEF _{50%} = 0.79 (0.72–0.84)	<i>P</i> = 0.02 (sEOS% vs combined model); <i>p</i> < 0.01 (FEF _{50%} vs sEOS% alone and the combined model)
tlgE+FEF _{50%}	0.79 (0.73–0.84)	tlgE = 0.71 (0.65–0.77)	FEF _{50%} = 0.79 (0.72–0.84)	<i>P</i> = 0.06 (tlgE vs combined model); <i>p</i> < 0.01 (FEF _{50%} vs tlgE alone and the combined model)
PBEC+FEF _{50%}	0.80 (0.74–0.85)	PBEC= 0.66 (0.60–0.73)	FEF _{50%} = 0.79 (0.72–0.84)	<i>P</i> = 0.11 (PBEC vs combined model); <i>p</i> < 0.01 (FEF _{50%} vs PBEC alone and the combined model)

Abbreviations: AUC, area under the curve; CVA, cough-variant asthma; FEF_{50%}, forced expiratory flow at 50% of forced vital capacity; FENO, fractional exhaled nitric oxide; sEOS%, percentage of eosinophil differential cell count in induced sputum; PBEC, peripheral blood eosinophil counts; tlgE, total serum immunoglobulin E; vs, versus.

affected by measurement variability, airway closure, and technical noise than MMEF or FEF_{75%}, making it more stable for clinical application.³¹ Based on these results, FEF_{50%} was selected for inclusion in the combined diagnostic model.

The combined model consistently demonstrated superior diagnostic performance, as reflected by higher AUC values ranging from 0.79 to 0.82 compared to individual predictors. Combining FEF_{50%} with FeNO or sEOS% provided the most accurate diagnosis of CVA, achieving an optimal AUC of 0.82, and substantially improving diagnosis accuracy over any single parameter. These findings are consistent with and extend our previous work, in which we demonstrated the diagnostic value of combining lower airway FeNO with small airway function parameters for CVA diagnosis.³² The present study further strengthens the evidence by incorporating a broader panel of T2 inflammatory biomarkers, including tlgE, sEOS% and PBEC, and by systematically evaluating their combined diagnostic performance. Despite the high diagnostic performance of sEOS% in combination with FEF_{50%}, its clinical applicability is limited due to variability in measurements. sEOS% levels can fluctuate depending on the timing and method of sample collection. In contrast, FeNO has been validated in multiple studies as a reliable and reproducible biomarker for T2 airway inflammation.³³

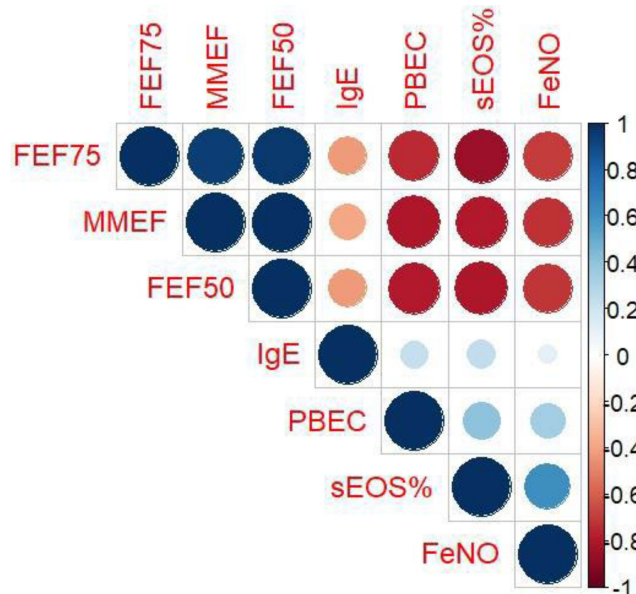


Figure 3 Correlations between T2 inflammatory biomarkers and small airway function for CVA. FEF_{50%}, forced expiratory flow at 50% of forced vital capacity; FEF_{75%}, forced expiratory flow at 75% of forced vital capacity; FENO, fractional exhaled nitric oxide; sEOS%, percentage of eosinophil differential cell count in induced sputum; PBEC, peripheral blood eosinophil counts; tlgE, total serum immunoglobulin E; MMEF, maximal mid-expiratory flow. The color scale represents Pearson correlation coefficients (*r*), ranging from -1 (strong negative correlation, red) to +1 (strong positive correlation, blue). Circle size and color intensity are proportional to the strength of the correlation.

The present study further explored the correlation between T2 inflammatory biomarkers and decreased small airway function. A strong positive correlation was observed among FEF_{75%}, MMEF, and FEF_{50%}, confirming their collective utility in assessing peripheral airflow. Similarly, PBEC and sEOS% demonstrated a very strong positive correlation, indicating that they may be used interchangeably as measures of systemic eosinophilic burden. In contrast, FeNO, PBEC, and sEOS% showed a strong negative correlation with small airway parameters, suggesting that higher levels of these biomarkers are associated with poorer lung function. Additionally, a moderate association has been found between tIgE and reduced lung function.

This study has certain limitations. First, the cough assessment relied on questionnaires prone to recall bias. Measures such as cough frequency monitoring or cough reflex sensitivity testing were not included, potentially missing nuanced insights into cough severity. Second, the success rate for obtaining adequate sputum samples was not explicitly stated, but sputum induction is known to have variable success. This could introduce selection bias and underrepresent patients unable to produce sputum, affecting the accuracy of airway inflammation assessments. Third, factors such as allergen sensitization or comorbidities were not systematically controlled, potentially influencing biomarker levels or lung function parameters.

Future studies incorporating standardized objective cough assessments, improved sputum sampling reporting, and comprehensive clinical phenotyping may help refine diagnostic models for CVA.

Conclusion

In conclusion, this study suggests that T2 inflammatory biomarkers, particularly FeNO and sEOS%, as well as small airway function parameters, provide moderate diagnostic accuracy for CVA when assessed individually. Furthermore, the diagnostic performance was significantly enhanced when FeNO or sEOS% was combined with FEF_{50%}. FeNO has been validated in multiple studies as a reliable and reproducible biomarker, supporting its potential clinical utility. However, the clinical applicability of sputum eosinophils remains limited due to technical complexity and variability in sample processing.

These findings may support earlier clinical assessment and a more individualized approach to CVA management. Nevertheless, given the single-center design, further validation in larger, multicenter studies is required before broad clinical implementation.

Data Sharing Statement

The datasets used and/or analyzed during the current study are available from the corresponding author, Xianghuai Xu, upon reasonable request.

Author Contributions

All authors made a significant contribution to the work reported, whether that is in the conception, study design, execution, acquisition of data, analysis and interpretation, or in all these areas; took part in drafting, revising or critically reviewing the article; gave final approval of the version to be published; have agreed on the journal to which the article has been submitted; and agree to be accountable for all aspects of the work.

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Disclosure

Eloina Margarita Mokuy Ondo Osie, Haodong Bai, and Jianjun Liu are co-first authors for this study. The authors declare that there is no conflict of interest in this work.

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