

Fluctuation phenotyping based on daily fraction of exhaled nitric oxide values in asthmatic children

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Background: Fraction of exhaled nitric oxide (FENO), a marker of airway inflammation, has been proposed to be useful for asthma management, but conclusions are inconsistent. This might be due to the failure of mean statistics to characterize individual variability in FENO values, which is possibly a better indicator of asthma control than single measurements.

Objective: We characterized fractal fluctuations in daily FENO values over time and the relationship between FENO values and symptom scores. We investigated whether these are associated with asthma severity, control, and exacerbation risk.

Methods: Daily FENO values and symptom scores over 192 days in 41 atopic asthmatic children from the Childhood Asthma Respiratory Inflammatory Status Monitoring study were analyzed. Two methods of time-series analysis were used: detrended fluctuation analysis to quantify fractal patterns in fluctuations in daily FENO values (α value) and cross-correlation to quantify the strength of the relationship between daily FENO values and symptom scores. The associations of α values and cross-correlation with markers of asthma severity and control were assessed by means of regression analysis.

Results: Daily fluctuations in FENO values exhibited fractal-type long-range correlations. Those subjects receiving higher doses of inhaled corticosteroids at study entry had a significantly lower α value, corresponding to more random fluctuations in FENO values in those with greater inhaled corticosteroid need. The cross-correlation between FENO values and symptom scores was significantly higher in those subjects who had exacerbations.

Conclusions: Fluctuation in FENO values and their cross-correlation to symptom scores contains information on asthma severity and control. Methods that quantify the complexity of asthma over time might assist in identifying asthmatic subjects with concordance between eosinophilic inflammation and

symptoms and thus increased exacerbation risk. (*J Allergy Clin Immunol* 2011;■■■:■■■-■■■.)

Key words: Exhaled nitric oxide, airway inflammation, asthma control, children, time-series analysis, detrended fluctuation analysis, cross-correlation analysis, fractal

The heterogeneous expressions of asthma make it a complex disease to characterize.¹ Clinically, the goal is to achieve asthma control, which can be facilitated by identifying functional asthma phenotypes and reliable markers of disease, and to design a targeted treatment strategy with a minimal inhaled corticosteroid (ICS) dose. Fraction of exhaled nitric oxide (FENO), a biomarker of airway inflammation, has been shown to reflect asthma control²⁻⁴ and therapy response⁵⁻⁷ and has been proposed to be useful for asthma management.⁸⁻¹¹ Longitudinal randomized controlled trials in which ICSs were titrated based on FENO values showed only limited benefits,^{12,13} but several studies found that longitudinal measurements of FENO in asthmatic subjects are helpful to predict deterioration.^{2,14,15} Furthermore, studies with positive results using the FENO value as a predictor of deterioration concluded that the changes in FENO values were more predictive than isolated FENO measurements,² especially in those with medium- to high-dose ICS treatment.¹⁴ This provides support for studying the day-to-day changes in FENO values^{16,17} for long-term asthma management.

The study of variability in physiological systems over time allows patients to be considered as nonlinear dynamic systems that are constantly adapting to changes in their environment. By using methods from time-series analysis, different aspects of dynamic systems can be quantified. Detrended fluctuation analysis (DFA)¹⁸ is a technique that quantifies the extent to which signal fluctuations are correlated over different time scales, a property consistent with fractal systems. By using DFA, it has been shown that fluctuations in heart-rate dynamics,¹⁸⁻²⁰ tidal breathing,^{21,22} and lung function in adult asthmatic subjects^{23,24} exhibit fractal behavior. Specifically, fractal correlations in fluctuations of peak expiratory flow in adult asthmatic subjects were shown to be related to asthma severity and control²⁴ and could be used to predict the risk of future obstructive events,^{24,25} as well as β_2 -agonist treatment responses.²³ Therefore the quantification of fractal properties in fluctuations in FENO values over long periods of time might provide information on a subject's asthma severity in relation to asthma control and exacerbation risk.

Recent investigations have advocated the need to focus on the multidimensional nature of asthma phenotypes and their temporal patterns in the assessment of asthma control.^{1,26} It has been shown that the concordance between FENO values and symptom scores

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Abbreviations used

CHARISM: Childhood Asthma Respiratory Inflammatory Status Monitoring

DFA: Detrended fluctuation analysis

FENO: Fraction of exhaled nitric oxide

GINA: Global Initiative for Asthma

ICS: Inhaled corticosteroid

might identify a phenotype of adult asthma that has a greater exacerbation risk, with possible therapeutic implications.²⁶ One method to quantify this relationship between FENO values and symptom scores is cross-correlation, which calculates the correlation between 2 fluctuating signals as a function of a time lag applied to one of them. This type of analysis can reveal the extent to which different expressions of asthma change together.

In this study we investigated fluctuations in FENO values and symptom scores over 30 weeks using DFA and cross-correlation and examined their association with clinical status and asthma severity and control in atopic asthmatic children who participated in a previous study.¹² We hypothesized that information contained within fluctuations in FENO values over time and their relationship to symptoms might be an additional marker to describe individual asthma severity and control.

METHODS

Study population and design

We retrospectively analyzed data from the Childhood Asthma Respiratory Inflammatory Status Monitoring (CHARISM) study,¹² a prospective, open-label, randomized, multicenter, parallel-group study in Italy and The Netherlands. Asthma was monitored daily over 30 weeks in 151 children aged 6 to 18 years with mild-to-moderate atopic asthma. The aim of the original study was to investigate whether ICS doses are better titrated based on FENO values and symptom scores rather than symptom scores only. We analyzed data from the FENO arm of the trial, in which ICS doses were adjusted every 3 weeks based on FENO values and symptom scores assessed daily in 77 randomly assigned children. Before analysis, we defined a technical inclusion criterion of less than 10% of FENO data missing out of the first 192 days of the study (<19 days missing) to minimize errors in the DFA.²² Missing days were filled in with values of the previous day, as detailed previously.²⁴

Assessments

FENO measurements. Daily FENO telemonitoring was performed with a portable airway inflammation monitor (NIOX MINO; Aerocrine, Solna, Sweden) with a constant flow of 50 mL/s, according to European Respiratory Society/American Thoracic Society criteria.²⁷ Measurements were transferred daily to the study center with a palm-top computer.

Clinical status. Subjects recorded daily symptom scores, ICS use, rescue bronchodilator use, and adverse events in a palm-top electronic diary (PalmOne Tungsten W TrialMax; CRF, Inc, Helsinki, Finland). Similar to the Santanello score,²⁸ sleep disturbance and symptom scores of cough, wheeze, and shortness of breath were scored from 0 to 3 based on increasing severity (0, no symptoms; 1, occasional symptoms; 2, symptoms most of the day; and 3, very bad asthma with inability to perform normal activities). Asthma control was estimated weekly and then averaged over the observation period according to the Global Initiative for Asthma (GINA) guidelines,^{29,30} in which symptoms, bronchodilator use, exacerbations, and lung function contributed to an asthma control rating of controlled (code = 0), partly controlled (code = 1), and uncontrolled (code = 2). FEV₁ and reversibility were measured at clinical visits 5 times during the study. The lung function results were applied to all weeks after the measurement until the next measurement. Weeks were

excluded if there were not at least 6 of 7 days of symptom data in the electronic diary. Symptoms, medications, and asthma control were summarized over the entire study period, at baseline (asthma activity in the first 3 weeks of the study), and at the end of the study (asthma activity in the last 12 weeks).

Exacerbations. In accordance with the 2009 American Thoracic Society/European Respiratory Society statement,³¹ severe exacerbations were defined as 1 or more days of oral prednisone prescription. Moderate exacerbations were defined as 2 or more days with a total symptom score of 3 or greater above the average total symptom score of the preceding 2 weeks or 1 or more days with a symptom score of greater than 5 above the preceding 2-week average and increased rescue bronchodilator use.

Comorbidities. All subjects had a positive RAST class of 2 or higher or a positive skin prick test response for at least 1 airborne allergen. Comorbidities were recorded at each clinic visit and entered into the electronic diary by the study nurses. Reports of cold, pharyngitis, fever, flu, otitis, bronchitis, and pneumonia during the study period were classified as "infectious comorbidities." Allergic rhinitis, conjunctivitis, and eczema were classified as "allergic comorbidities."

Time-series analysis

DFA. The DFA was used to quantify the extent to which fractal-type correlations were present in the FENO signal with a single correlation exponent, the α value.¹⁸ The details of the method have been previously published.²⁴ Briefly, to calculate the α value, a FENO time series of day-by-day FENO measurements was integrated and divided into nonoverlapping time windows of size n . For each window, the fluctuation function $F(n)$ was calculated by taking the root mean square values of the detrended signal.³² This process was repeated for increasing n values and plotted on a log-log plot, in which a linear relationship implies that $F(n)$ follows a power law functional form as follows:

$$F(n) = A n^{\alpha},$$

where A is the amplitude of the power law fluctuation function and α is the correlation exponent, which indicates the extent of long-range correlation in the original signal.

One can think of the presence of correlations in a signal as a form of memory because fluctuations at any time point are related to those at previous points in time. An uncorrelated random signal, such as white noise, has an α value of 0.5. As the α value increases to greater than 0.5, fluctuations have stronger long-range correlations; that is, there is a stronger relationship between values at different short- or long-range time scales (eg, days or weeks).

We chose to analyze the first 192 days of data, which was a compromise between the data length requirements of the DFA calculation²² and inclusion of subjects with sufficient data points. A modified DFA algorithm³² was used to account for long-term linear trends present in the FENO time series.

Cross-correlation of FENO values and symptom scores

Cross-correlation measures the degree to which 2 signals are linearly correlated when one of these signals is lagged in time. Similar to a regression coefficient, the strength and direction of this relationship is quantified by a correlation coefficient: a positive coefficient indicates that as one variable increases, so does the other, and a negative coefficient indicates an inverse relationship between the variables. In this study the FENO value was the reference signal, and symptoms were shifted in time. Thus when a maximum positive cross-correlation is seen at a lag of 2, the strongest relationship between FENO values at present is with symptoms measured 2 days later. We calculated the cross-correlation between daily FENO values and the daily sum of symptom scores, in which symptoms were lagged by FENO values within ± 7 days to allow for the possibility that increases in symptom scores could precede or follow increases in FENO values.

Statistical analysis

Associations between parameters of clinical asthma control status (exposures) and α values or cross-correlation of symptom scores/FENO values (outcomes) were examined separately by using multiple linear regression models adjusted for age, height, and sex. Associations between occurrence of

TABLE I. Characteristics of the study population

	All included subjects (n = 41)	Asthma exacerbations: severe (n = 5) and moderate (n = 10)	No asthma exacerbation (n = 26)	P value*
Male sex, no. (%)	20 (49)	4 (27)	16 (62)	.03
Age (y)	10.9 ± 2.4	10.9 ± 2.4	10.8 ± 2.4	.92
Weight (kg)	39.6 ± 12.1	40.6 ± 12.5	39.1 ± 12.1	.53
Height (cm)	146.3 ± 15.5	146.2 ± 14.7	146.4 ± 16.2	.90
Medication, no. (%)				
Antihistamine	19 (46)	7 (47)	12 (47)	.98
Montelukast	11 (27)	5 (33)	6 (23)	.48
Nasal steroid	15 (37)	5 (33)	10 (38)	.75
Long-acting β -agonist	30 (73)	11 (73)	19 (73)	.46
Mean ICS dose ($\mu\text{g} \cdot \text{day}^{-1}$), median‡	309 (90-1080)	469 (90-1080)	308 (89-1160)	.77†
Mean bronchodilator use (puffs/day), median‡	0.88 (0.16-3.29)	1.32 (0.34-5.05)	0.59 (0.11-3.23)	.02†
Geometric mean FENO value (ppb)	19.8 ± 8.6	18.8 ± 6.4	20.4 ± 9.6	.79
Mean FEV ₁ (% predicted)§	97.6 ± 11.3	96.0 ± 9.8	98.6 ± 12.1	.49
Mean reversibility of FEV ₁ (% predicted)	6.2 ± 6.7	6.9 ± 5.5	5.9 ± 7.3	.65
Mean asthma control¶	0.83 ± 0.46	1.12 ± 0.38	0.66 ± 0.42	.0005
Symptom-free days, median‡	88 (2-176)	75 (2-166)	106 (1-182)	.19†
Mean daily sum symptom score, median‡	0.87 (0.15-3.34)	1.32 (0.29-5.33)	0.62 (0.10-3.27)	.02†
Comorbidities, no. (%)				
Infectious comorbidities	25 (61)	11 (73)	14 (54)	.22
Allergic comorbidities	13 (32)	5 (33)	8 (31)	.87
α Value	1.03 ± 0.13	1.05 ± 0.13	1.02 ± 0.14	.51
Cross-correlation	0.15 ± 0.21	0.27 ± 0.18	0.09 ± 0.21	.01

Mean (SD) values are provided unless otherwise indicated.

*Assessed by using the Student *t* test.

†Assessed by using the Wilcoxon rank sum test.

‡Expressed as 95% CI.

§Average of 5 FEV₁ tests in weeks 1, 3, 12, 21, and 30.

||Average of 5 reversibility FEV₁ tests.

¶Mean asthma control classified weekly as follows: controlled, 0; partly controlled, 1; or uncontrolled, 2.

exacerbation (exposure) and the α value or cross-correlations (outcome) were examined by using multiple linear regressions adjusted for mean ICS dose. Severe and moderate exacerbations were also examined separately. The potential confounding effect of infectious and allergic comorbidities, as well as medications, on the above associations was investigated. First, univariate associations of each confounder with mean FENO value, α value, and cross-correlation coefficient were investigated with linear regression. Second, significantly associated parameters from these univariate investigations were included as confounders in the above multiple linear regression analysis. In addition, we examined the contribution of treatment decisions on fluctuations in FENO values by calculating the cross-correlation between FENO values and ICS use in 3-week windows. Statistical analyses were performed with Intercooled Stata 10 for Windows (Stata Corp, College Station, Tex).

RESULTS

Forty-one children met the inclusion criterion of less than 10% data missing of daily FENO values and symptom scores of the first 192 days of the study, resulting in a total of 7820 measurements of FENO for analysis. Table I summarizes the subjects' characteristics stratified by severe, moderate, or no asthma exacerbation during the study period. In the whole study group FENO values measured over 192 days displayed a considerable amount of fluctuation. Two examples of a FENO time series over 192 days from subjects with no changes in steroid dose can be seen in Fig 1.

DFA

Fluctuations in FENO values recorded daily over 192 days were correlated on time scales of more than a month (long-range correlation) with a mean long-range correlation exponent ($\alpha = 1.03$ [SD, 0.13]).

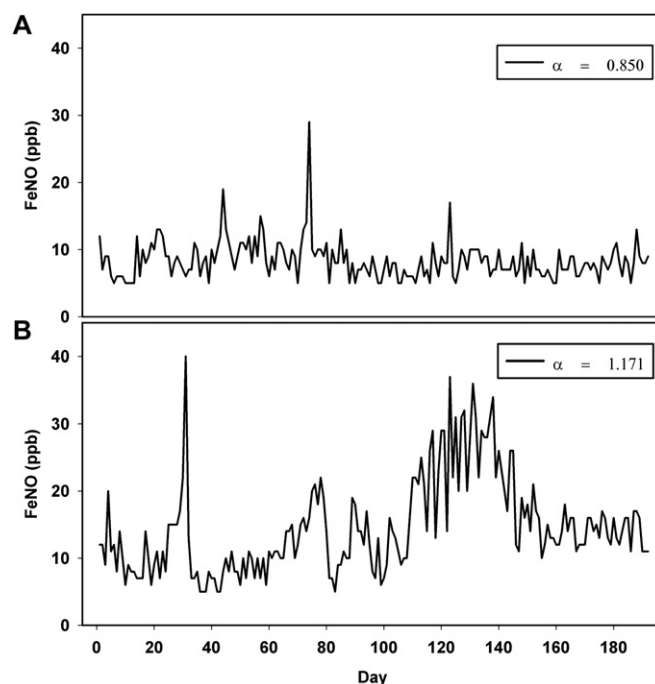


FIG 1. Exhaled nitric oxide values over 192 days from 2 subjects with no changes in ICS dose during the study period. **A**, Subject with a relatively small range of FENO values (5-30 ppb), a constant ICS dose of 1000 $\mu\text{g}/\text{d}$ throughout the study, and weak long-range correlations in FENO values reflected by an α value of 0.850. **B**, Subject with a relatively large range of FENO values (6-40 ppb), a constant ICS dose of 400 $\mu\text{g}/\text{d}$ throughout the study, and stronger long-range correlations reflected by an α value of 1.171.

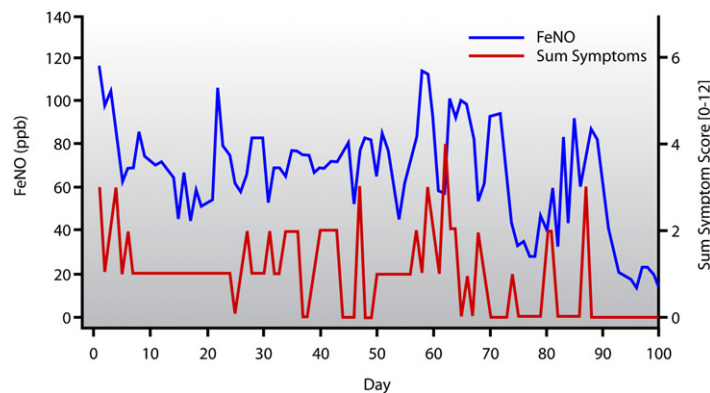


FIG 2. Cross-correlation of daily FeNO values and symptom scores. Day-by-day FeNO values and symptom scores in a subject with a strong concordance between the FeNO values and symptom scores are shown. Here the maximum correlation occurred when increases in FeNO values preceded increases in symptom scores by 2 days (lag 2). Cross-correlation: lag 0, 0.558; lag 1, 0.607; and lag 2, 0.620.

TABLE II. Association of α values and cross-correlation with clinical status

Effect	Multivariable models*					
	Long-range correlation α value†			Cross-correlation‡		
	Coefficient	95% CI	P value	Coefficient	95% CI	P value
Baseline§ mean FeNO value	0.0007	−0.0019 to 0.0034	.58	0.0038	−0.0006 to 0.0082	.09
Baseline symptom score	−0.017	−0.057 to 0.021	.36	0.019	−0.048 to 0.086	.58
Baseline ICS dose¶	−0.013	−0.025 to −0.001	.04	0.002	−0.020 to 0.024	.86
Baseline asthma control	−0.013	−0.095 to 0.069	.76	0.12	−0.02 to 0.25	.09
Mean FeNO value	0.001	−0.004 to 0.006	.74	0.004	−0.004 to 0.012	.35
Mean sum symptom scores	−0.016	−0.053 to 0.021	.38	0.0004	−0.0638 to 0.0647	.99
Mean ICS dose¶	−0.009	−0.021 to 0.004	.15	0.02	0.00 to 0.04	.11
Mean asthma control	0.010	−0.082 to 0.102	.82	0.08	−0.08 to 0.23	.32
Mean FeNO value at study's end#	0.001	−0.004 to 0.006	.76	−0.001	−0.009 to 0.008	.91
Mean symptom score at study's end	−0.017	−0.052 to 0.018	.33	0.018	−0.043 to 0.079	.55
Mean ICS dose at study's end¶	−0.006	−0.018 to 0.006	.32	0.008	−0.013 to 0.029	.44
Mean asthma control at study's end	0.016	−0.067 to 0.098	.70	0.08	−0.06 to 0.22	.23

*Model was adjusted for age, height, and sex.

†Long-range correlations calculated by using DFA adjusted for linear trend.

‡Cross-correlation between FeNO values and symptom scores on the same day (lag 0).

§Clinical status parameters assessed in the first 3 weeks of the study.

||Geometric mean of FeNO values.

¶Calculated as per 100- μ g increase in ICS used.

#Clinical status parameters assessed in the last 12 weeks of the study.

Cross-correlation of FeNO values and symptom scores

More subjects ($n = 10$) had their strongest correlation between FeNO values and symptom scores on the same day (lag 0) than for any other day, whereas the rest of the subjects had maximum correlation when symptom scores were shifted with respect to FeNO values within ± 5 days. To maintain comparability, we used the cross-correlation at lag 0. The cross-correlation coefficient of FeNO values and symptom scores ranged from -0.31 to 0.59 : 23 subjects had a significantly positive relationship between FeNO values and symptom scores ($P < .05$), 4 subjects had a significantly negative relationship, and 14 subjects had a weak relationship (not significant). An example of a subject with a strong positive correlation can be seen in Fig 2. In this subject the strongest cross-correlation occurred when FeNO values preceded symptoms by 2 days (lag 2).

Relationship with clinical status

The long-range correlation exponent α was negatively associated with ICS dose at baseline (Table II) but was not found to be associated with ICS use, symptoms, and asthma control averaged over the whole period or in the last 12 weeks of the study. Furthermore, when stratified by mean asthma control over the study period, there was a significant decrease in the α value with ICS dose only in the uncontrolled children (mean asthma control score >1 over the study period, $n = 14$, 71% of whom had asthma exacerbations) of a 0.03 per 100 μ g/d increase in ICS use (95% CI, -0.05 to -0.01 ; $P = .007$). The strength of cross-correlation between FeNO values and symptom scores was not related to clinical status or ICS dose throughout the study.

Relationship with exacerbations

Of the included subjects, 5 had severe asthma exacerbations, and 10 had moderate exacerbations. Two of those with severe

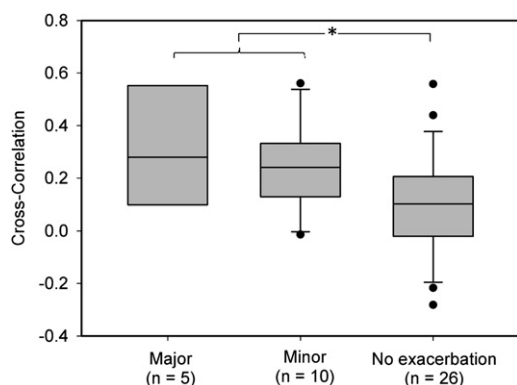


FIG 3. Cross-correlation of daily FENO values and symptom scores in exacerbation groups. Box plots of cross-correlation between symptom scores and FENO values stratified by severe, moderate, or no exacerbation during the study period. Boxes represent 25th and 75th percentiles, with the median line and error bars representing the 10th and 90th percentiles.

exacerbations had more than 1 prednisone course, and 4 of those with moderate exacerbations had more than 1 moderate exacerbation. There was no difference in mean FENO values in the exacerbation groups (Table I). Both α values and cross-correlation were able to distinguish between the exacerbation group (severe and moderate) and the group with no exacerbations. The strength of long-range correlations, the α value, tended to be higher in those children with severe exacerbations, but the differences were not significant. Even when adjusted for mean ICS dose during the study, the strength of cross-correlation between FENO values and symptom scores (Fig 3) was greater in those who had either a severe or moderate exacerbation by 0.17 (95% CI, 0.018-0.30; $P = .030$) compared with those who did not have an exacerbation.

Effect of comorbidities and medications

Children who took antihistamines had a borderline increased geometric mean FENO value of 5.13 ppb (95% CI, -0.10 to 10.37 ppb; $P = .054$) and a stronger cross-correlation of FENO values and symptom scores by 0.13 (95% CI, 0.00 - 0.26 ; $P = .043$) compared with those who did not take antihistamines in a univariate model. These effects disappeared in a multivariable model, whereas the relationship between exacerbations and cross-correlation remained significant. In addition, there was a significant feedback of changes in ICS use on mean FENO values in 71% of the subjects, but this was not related to the long-range correlation exponent α . In a receiver operating curve analysis, the cross-correlation coefficient of FENO values and symptom scores was the best predictor for exacerbations (Fig 4) compared with mean FENO values ($P = .033$), mean symptom scores ($P = .80$), and α values ($P = .16$). A cutoff in cross-correlation above which significant cross-correlation between FENO values and symptom scores was found across all subjects (cross-correlation = 0.134) had a positive predictive value of 54% and a negative predictive value of 84% (sensitivity, 80%; specificity, 61%).

DISCUSSION

In our study of FENO values recorded daily over 192 days in atopic asthmatic children,¹² we found that fluctuations in FENO

values were not random but were correlated over time scales of more than a month (long-range correlation). The long-range correlation exponent α was found to be lower in those subjects requiring higher ICS doses at study entry. Thus fluctuations in FENO values were more random in children requiring higher ICS doses. However, the α value was not related to mean asthma control, as defined by the GINA guidelines. Although mean FENO values were not predictive, the day-by-day cross-correlation of FENO values and symptom scores was significantly stronger in those subjects with severe or moderate exacerbations, even when adjusting for mean ICS dose.

Long-range correlations

There is accumulating evidence that there is a healthy amount of variability in physiologic systems with a complex but organized structure. The outputs of such systems have been shown to contain long-range fractal correlations, a property that might render them more adaptive to external perturbations. These fractal correlations have been shown to break down with disease, possibly reflecting a reduction in the adaptive ability of the system and prospective critical events. In asthmatic subjects the study of long-range fractal correlations in lung function has been shown to provide valuable information on the complex nature of asthma.^{23,24} We have previously shown that patients with a more random pattern in twice-daily peak expiratory flows had more severe asthma,²⁴ poor treatment response,²³ and higher probability for an obstructive event.²⁴ Our present findings indicate that long-range fractal correlations are also manifested in fluctuations of an inflammatory marker of asthma, FENO.

We found that long-range correlations in fluctuations in daily FENO values were associated with ICS doses but were not related to clinical status and were not predictive of treatment response throughout the study or at the end of the study. This could be due to the constant adaptation of ICSs to FENO values every 3 weeks, as a consequence of the original study design. This association between the α value and mean ICS dose, particularly when stratified by asthma control, is likely to be complex. Although GINA guidelines proposed that asthma severity could best be determined in the untreated patient,³⁰ recent guidelines^{33,34} suggest that asthma severity might be a function of asthma control because it is related to ICS treatment that can mask the underlying disease process. Our data provide evidence that the relationship between ICS dose, severity of asthma, and long-range correlations in FENO values were most obvious in subjects with poorly controlled asthma. This might be due to the amount of the disease process that is not masked by treatment.

The characterizations of long-range correlations in twice-daily peak expiratory flow in asthmatic subjects in combination with variability potentially offer the possibility to predict future obstructive events.^{24,25} In this study we provide the first evidence that fluctuations in daily inflammatory markers also contain long-range correlations. Based on the present findings, it is not possible to draw conclusions on the predictive ability of long-range correlations in FENO values because our data only show a trend for higher α values in those with exacerbations. Future trials of fluctuation analysis of daily FENO values in steroid-naïve asthmatic subjects before and after ICS treatment could help to elucidate the relationship between the severity of the underlying disease process and asthma control in relation to ICS dose.

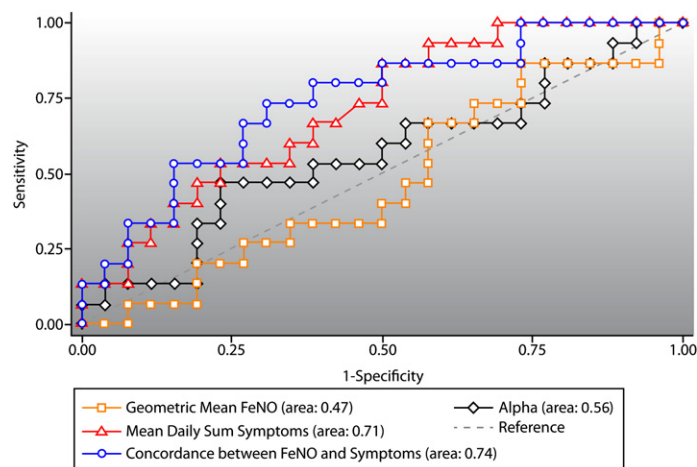


FIG 4. Receiver operating curves for mean FeNO values, mean sum symptom scores, the cross-correlation between FeNO values and symptom scores, and the long-range correlation coefficient α to predict exacerbations. The cross-correlation coefficient of FeNO values and symptom scores was the best predictor for exacerbations compared with mean FeNO values ($P = .033$), mean symptom scores ($P = .80$), and α values ($P = .16$).

Concordance of FeNO values and symptom scores

Although the relationship between FeNO values and symptom scores has been investigated,³⁵⁻³⁷ this is the first study that compares day-by-day FeNO values and symptom scores over such a long period of time by using time-series analysis techniques in asthmatic children. In our study population we found that the majority of subjects had the strongest positive relationship between FeNO values and symptom scores on the same day. Subjects who had severe or moderate exacerbations had a stronger positive cross-correlation between FeNO values and symptom scores, suggesting that concordance of FeNO values and symptom scores is an indicator of increased risk of exacerbation.

This concept of concordance between FeNO values and symptom scores has recently been advocated by Haldar et al²⁶ through phenotype-cluster analysis of an asthmatic cohort. They found that within a group of adult asthmatic patients, there are subjects in whom FeNO values are strongly associated with asthma symptoms, whereas in others this relationship is weak. They suggested a new concept of “concordant” and “discordant” phenotypes of asthma. In our study population of atopic asthmatic children, such a distinction might explain why in some patients FeNO values and symptom scores are dissociated in time. Cross-correlation analysis of biomarkers and symptoms fluctuating over time might be a new way of quantifying such concordance and might help to identify new “fluctuation phenotypes” of asthma. This is particularly important because there is increasing evidence that asthma therapy might benefit from a more individualized and phenotype-specific approach.^{26,38-40} Therefore the temporal concordance between fluctuations in FeNO values and symptom scores, as quantified by using cross-correlation methods, could help to identify an increased risk of asthma instability and exacerbation risk. Moreover, asthmatic subjects in whom FeNO values and symptom scores are discordant might specifically benefit from FeNO monitoring.

In a receiver operating curve analysis we see that the cross-correlation between FeNO values and symptom scores is the best predictor of exacerbations compared with the other predictor variables but only slightly better than symptom scores. Because the definition of exacerbations depends on symptoms, the

usefulness of symptoms as a predictor is limited. Therefore the cross-correlation of FeNO values and symptom scores is attractive as an independent predictor of exacerbations. If we use a cutoff cross-correlation coefficient of 0.134, we find a positive predictive value of 54% and a negative predictive value of 84% (sensitivity, 80%; specificity, 61%). This is similar to the predictive value of FeNO to predict loss of control found in other studies.^{2,41} Because the cutoff value for concordance is derived from this relatively specific dataset, with no information on physiological meaningfulness, the predictive ability should be taken with caution.

Limitations of the study

In human studies over long periods of time, it is impossible to control the external environment. In this study, in which asthma was monitored over 30 weeks, not only were children exposed to various undocumented external factors, such as the school environment, local pollution, and allergens, but also their asthma medication was potentially changed every 3 weeks, depending on FeNO values and symptom scores. Because ICSs have an effect on FeNO values, one could presume that this would have an effect on FeNO value variability and asthma control, as well as contributing to the intrinsic correlations in FeNO values. Although we could show that there was a strong cross-correlation between FeNO values and ICS dose in most subjects, we did not find an effect of this feedback on long-range correlations in FeNO values. We also did not find an effect of the average ICS dose or the number of ICS step changes, implying that the amount of ICS that a subject received did not seem to affect the level of internal correlation. Better insight into FeNO's behavior might be possible in a study in which ICS doses do not depend on FeNO values.

We excluded 34 children because of the inclusion criterion of less than 10% missing data. These children tended to be older and have higher FeNO values. They did not differ in terms of ICS doses at study entry, but because the titration of ICSs based on FeNO values, they tended to have higher ICS doses throughout the study. Whereas the exclusion of this population did not affect our main findings, one should be cautious when extrapolating these results to a more general asthma population.

Clinical implications and future hypotheses

As proposed in a recent review,¹ monitoring multiple disease parameters over time and characterizing their fluctuations might provide new tools to characterize the dynamic and complex nature of asthma better than mean values. In our proof-of-concept study we show that by monitoring fluctuations in symptom scores and FENO values, we can better identify which children are at risk for exacerbations compared with cutoff or mean FENO values. Furthermore, by studying long-range patterns in FENO values, we might be able to better characterize asthma severity and control, which could aid in treatment decisions. We speculate that the application of time-series analysis to clinical parameters could help to define new fluctuation phenotypes of asthma.

Although this dataset was not ideal to answer all our questions on the clinical utility of such methods, future studies with a prospective design might be able to disentangle the effect of ICSs on correlations in FENO values and help to determine which treatment will be most effective for a given patient. Presently, FENO analyzers are still relatively expensive and complex, but with the foreseen development of cheaper simpler devices, daily monitoring of FENO values might become a reality for clinical practice. Such methods are attractive as we move toward patient-initiated personalized medicine.

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Clinical implications: New methods that quantify the complexity of asthma over time might provide additional information on asthma severity and control. The present findings support a concept of asthma phenotyping based on fluctuations in biomarkers.

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