

FeNO and Exercise Testing in Children at Risk of Asthma



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What is already known about this topic? Exercise testing is the gold standard for diagnosing exercise-induced bronchoconstriction in children, but requires considerable cooperation and medical resources. Fraction of exhaled nitric oxide (FeNO) has been proposed as a tool to predict the need for exercise testing.

What does this article add to our knowledge? FeNO and degree of bronchoconstriction after exercise testing are linearly associated traits in children at risk of asthma, but FeNO cannot reliably be used dichotomized in clinical practice to predict the need of exercise testing.

How does this study impact current management guidelines? This study suggests that the clinician should refrain from using FeNO to predict the need for exercise testing in children reporting respiratory symptoms during exercise.

BACKGROUND: Exercise testing is the gold standard for diagnosing exercise-induced bronchoconstriction in children, but requires considerable cooperation and medical resources. Therefore, fraction of exhaled nitric oxide (FeNO) has been proposed as a tool to predict the need for exercise testing.

OBJECTIVE: The objective of this study was to investigate the relationship between FeNO, exercise test results, and a history of respiratory symptoms during exercise in children at risk of asthma.

METHODS: FeNO measurement, exercise testing, and interview about respiratory symptoms during exercise were completed in 224 seven-year-old children from the at-risk Copenhagen Prospective Studies on Asthma in Childhood₂₀₀₀ birth cohort. The

associations between FeNO, exercise test results, and reported respiratory symptoms during exercise were analyzed adjusting for gender, respiratory infections, and inhaled corticosteroid treatment. The associations were also analyzed stratified by asthma and atopic status.

RESULTS: Of the 224 children, 28 (13%) had an established asthma diagnosis and 58 (26%) had a positive exercise test ($\geq 15\%$ drop in forced expiratory volume in 1 second [FEV₁] from baseline). FeNO and bronchial obstruction after exercise were linearly associated with a doubling of FeNO corresponding to a 2.4% drop in FEV₁ (95% confidence interval, 0.8–4.1; $P < .01$). However, a receiver operating characteristic curve analysis showed that the best cutoff of FeNO for predicting exercise test outcome among children who reported respiratory symptoms during exercise was 17 ppb, which only had 74% negative predictive value. There was no association between FeNO and reported respiratory symptoms during exercise (odds ratio = 1.3 [0.8–1.9]; $P = .29$) or reported symptoms during exercise and exercise test results (odds ratio = 1.0 [1.0–1.1]; $P = .12$).

CONCLUSIONS: A history of respiratory symptoms during exercise was not associated with either elevated FeNO or a positive exercise test in children at risk of asthma. FeNO and exercise test results were linearly associated traits, but FeNO could not reliably be used dichotomized to predict the need of exercise testing. © 2017 American Academy of Allergy, Asthma & Immunology (J Allergy Clin Immunol Pract 2018;6:855–62)

Key words: Asthma; Children; Exercise-induced bronchoconstriction; Exercise test; FeNO

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All funding received by COPSAC is listed on www.copsac.com. The Lundbeck Foundation (Grant no R16-A1694); The Ministry of Health (Grant no 903516); Danish Council for Strategic Research (Grant no 0603-00280B); and the Capital Region Research Foundation have provided core support to the COPSAC research center.

Conflicts of interest: K. Bønnelykke has received consultancy fees from ALK Abello; has received research support from the Lundbeck Foundation, Novo Nordisk Foundation, Danish Ministry of Health, and Danish Strategic Research Foundation; has an approved but unpaid patent for Diagnosing asthma and other wheezing disorders by genotyping of CDHR3 variants (Danish patent no. PA 2012 00368). H. Bisgaard has received research support from the Danish Ministry of Health, Lundbeck Foundation, Danish Council for Strategic Research, Danish Advanced Technology Foundation, Danish State Budget; and has received consultancy fees from Chiesi Pharmaceuticals and Boehringer Ingelheim. The rest of the authors declare that they have no relevant conflicts of interest.

Received for publication March 21, 2017; revised October 5, 2017; accepted for publication October 13, 2017.

Available online November 10, 2017.

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2213-2198

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<https://doi.org/10.1016/j.jaip.2017.10.014>

Exercise-induced bronchoconstriction is a common manifestation of childhood asthma,^{1,2} which compromises the child's participation in physical activities and may have a negative influence on the quality of life, cardiovascular health, and psychomotor development.^{3–5} Exercise-induced bronchoconstriction in young children is difficult to diagnose as children often have a poor perception of their respiratory symptoms during exercise or

Abbreviations used*COPSAC- Copenhagen Prospective Studies on Asthma in Childhood**FeNO- Fraction of exhaled nitric oxide**FEV₁- Forced expiratory volume in 1 second**OR- Odds ratio**ppb- Parts per billion**ROC- Receiver operating characteristics**SPT- Skin prick tests**TROLS- Troublesome lung symptoms*

adjust their physical behavior according to their bronchoconstriction threshold. Thus, there is a poor correlation between the child-reported and parent-reported respiratory symptoms during exercise and clinically diagnosed exercise-induced bronchoconstriction.^{6,7}

Standardized exercise testing is the gold standard for diagnosing exercise-induced bronchoconstriction in children,^{8,9} but the test is time consuming and requires significant cooperation of the child and medical resources. Therefore, there is a need for simple screening tests to identify children where exercise testing may be required, and measuring fraction of exhaled nitric oxide (FeNO) may serve such purpose. In a population of children diagnosed with asthma, a positive exercise test on a treadmill could be excluded with a probability of 90% (negative predictive value) at a FeNO cutoff level <12 parts per billion (ppb) and <20 ppb in children on versus not on inhaled corticosteroid treatment, respectively.¹⁰ However, most studies have focused on populations of children diagnosed with asthma, whereas there is a need to study the clinical application of FeNO for screening children at risk of asthma.

The purpose of this study was therefore to explore the association between FeNO, exercise test results, and a history of respiratory symptoms during exercise in 7-year-old children with and without a diagnosis of asthma participating in the at-risk Copenhagen Prospective Studies on Asthma in Childhood₂₀₀₀ (COPSAC₂₀₀₀) birth cohort born to mothers with asthma.

METHODS**Study design**

Participants comprised children of COPSAC₂₀₀₀: a single-center, prospective clinical birth cohort study of 411 children born to mothers with a history of asthma. The children were enrolled between August 1998 and December 2001 at 1 month of age and were closely monitored by scheduled visits to the COPSAC clinical research unit every 6 months till the age of 7. The recruitment and baseline characteristics of the participants have previously been described in detail.¹¹⁻¹⁴

Ethics

The Copenhagen Ethics Committee (KF 01-289/96) and the Danish Data Protection Agency (2008-41-2434) approved the study, and written and oral informed consent was obtained from both parents at enrollment.

Exercise testing

An exercise test was performed at the 7-year follow-up visit of the cohort by trained research personnel according to the official guidelines from the American Thoracic Society (ATS).¹⁵ The child ran on a motorized treadmill for 6 minutes with a heart rate of

175-185 beats/minute for at least 5 minutes while inspiring dry air through a mouthpiece wearing a nose clip to avoid nose breathing. Forced expiratory volume in 1 second (FEV₁) was measured at baseline and 1, 3, 5, 10, and 15 minutes after exercise with a Jaeger Powerunit spirometer (Viasys Healthcare, Höchberg, Germany) assuring compliance using a volume-anchored incentive. The exercise test outcome was analyzed continuously as a postexercise test percentage drop in FEV₁, and a positive test was defined as a postexercise decrease in FEV₁ ≥ 15% from baseline measured up to 15 minutes after the exercise test.

FeNO measurement

FeNO was measured before the exercise test by an online technique using a NIOX Flex (Aerocrine, Solna, Sweden) in accordance with international guidelines.¹⁶ The child inhaled to total lung capacity and immediately exhaled at a constant flow of 50 mL/s until a FeNO plateau of ≥2 s could be identified. An exhalation lasted at least 4 s maintaining the expiratory pressure at 5 to 20 cm H₂O to close the velum. An exhalation flow-driven animated computer program guided the child. Two to three repeated measurements where the FeNO level agreed within 5% were completed using the mean FeNO value as the test outcome.

History of respiratory symptoms during exercise

A COPSAC pediatrician interviewed the child and parents about the history of respiratory symptoms during exercise according to the ATS criteria including a history of chest tightness, cough, wheezing, or dyspnea during and/or after fierce physical activity.⁹ If the child was suspected of respiratory symptoms during exercise based on the history, an inhaled short acting β_2 -agonist was prescribed as needed.

Asthma diagnosis

Asthma was diagnosed by the COPSAC pediatricians according to a previously validated quantitative symptom algorithm¹⁷ based on daily diary recordings verifying a history of persistent troublesome lung symptoms (TROLS) (5 episodes of at least 3 consecutive days over 6 months or 1 episode lasting 4 consecutive weeks of TROLS), a requirement for intermittent use of inhaled β_2 -agonist, response to a 3-month course of inhaled corticosteroids, and relapse when stopping treatment.¹³

Atopic status

IgE. Serum specific IgE levels were determined using a screening method (ImmunoCAP, Thermo Fisher Scientific, Uppsala, Sweden), and levels ≥0.35 kU_A/L were considered indicative of sensitization.¹⁸

Skin prick tests (SPT). SPT were performed using standard allergen extracts (ALK Abello USA, Hørsholm, Denmark) and a positive reaction was defined as a diameter ≥3 mm larger than the negative control. We analyzed atopic status by sIgE and SPT data separately using the most common food and inhalant allergens as previously described in detail.¹⁹

Statistical analysis

Log₂-transformation of FeNO values was performed before analysis to obtain normal distribution of the data. The associations between FeNO, postexercise test percentage drop in FEV₁, and reported symptoms during exercise were investigated using logistic regression and general linear models with FeNO and percentage drop in FEV₁ as continuous explanatory variables and reported symptoms during exercise as a dichotomous outcome. Results are

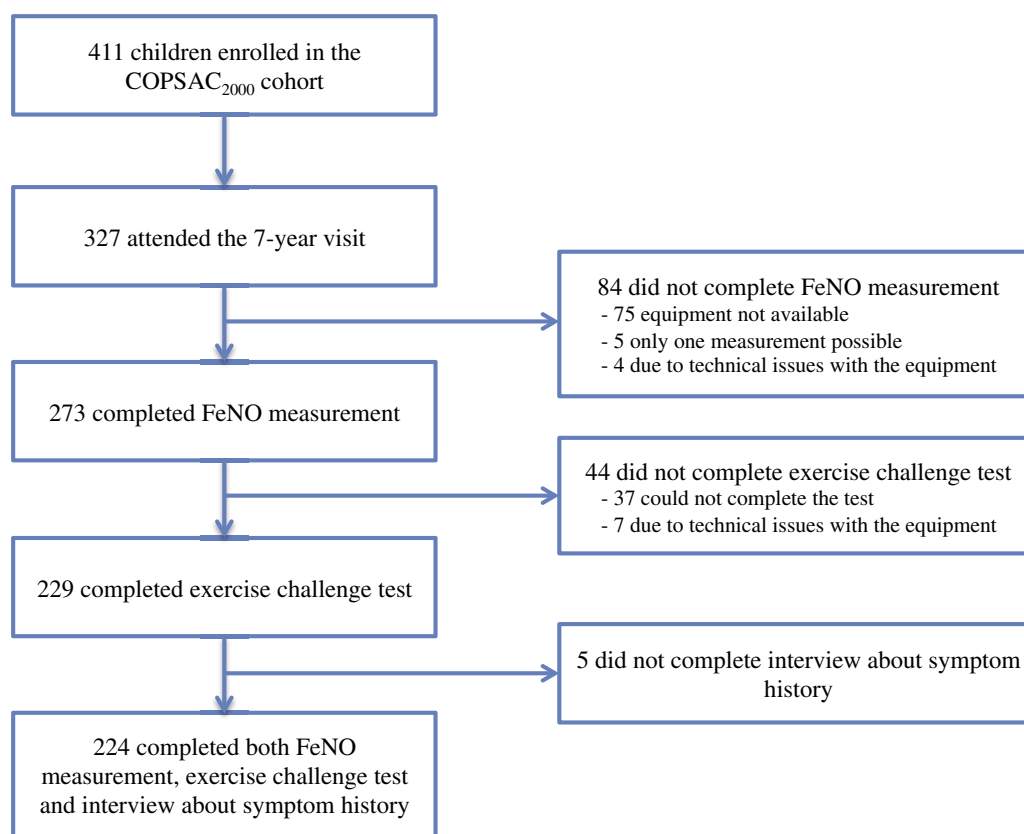


FIGURE 1. Study group flowchart. *COPSAC*, Copenhagen Prospective Studies on Asthma in Childhood; *FeNO*, fraction of exhaled nitric oxide.

reported crude and adjusted for gender, upper respiratory tract infections 7 days before FeNO measurement (yes/no), and any inhaled corticosteroid treatment within 28 days before FeNO measurement (yes/no).²⁰ The associations were further explored by stratifying the study group as children with and without asthma and by atopic status.

Thereafter, a ROC curve analysis was conducted to estimate the best cutoff value of FeNO for predicting a positive exercise test within the group of children with a history of respiratory symptoms during exercise.

Results of the logistic regression analyses are presented as OR with 95% confidence intervals (CIs) and results of the general linear models as β -coefficients with 95% CIs. A *P* value $\leq .05$ was considered significant. All analyses were performed using SAS, version 9.4 for Windows (SAS Institute, Cary, NC).

RESULTS

Baseline

Of the 411 children in the COPSAC₂₀₀₀ cohort, 327 (80%) attended the 7-year visit, and of these children, 224 (69%) completed exercise testing, FeNO measurement, and a structured clinic interview about respiratory symptoms during exercise (see the study group flow chart in Figure 1). The number of children with a positive exercise test (postexercise FEV₁ drop $\geq 15\%$ from baseline) was 58 (26%), 29 (13%) reported

TABLE I. Study group characteristics

Variable, 7 y	Study cohort (N = 224)
Asthma, yes (%)	28 (13)
Reported symptoms, yes (%)	29 (13)
Asthma and reported symptoms, yes (%)	15 (7)
Exercise test	
No. of positive tests (%)	58 (26)
Percentage drop in FEV ₁ , mean (SD)	10.6 (9.4)
FeNO	
Increased FeNO (≥ 17 ppb), yes (%)	18 (8)
FeNO (ppb), mean (SD)	8.7 (7)
Spirometry	
FEV ₁ (L), mean (SD)	1.40 (0.24)
Post- β_2 FEV ₁ (L), mean (SD)	1.48 (0.24)
Before the exercise test and FeNO measurement	
Inhaled corticosteroid treatment*, yes (%)	4 (2)
Respiratory infection, yes (%)	2 (1)
Atopic status	
Positive sIgE 6 y (>0.35 kU _A /L), yes (%)	67 (34)
Positive SPT 6 y (>3 mm), yes (%)	28 (15)
Allergic rhinitis, yes (%)	22 (11)

FeNO, Fraction of exhaled nitric oxide; *FEV₁*, forced expiratory volume in 1 s, SD, standard deviation; *SPT*, skin prick test.

*Within 28 d before visit.

TABLE II. Demographics of the selected cohort and nonenrollees

	N	Study cohort (N = 224)	Excluded (N = 187)	P value
Personal				
Male (%)	411	108 (48)	95 (51)	.60
Asthma, 7 y (%)	336	28 (13)	19 (16)	.36
Socioeconomic factors				
Social circumstances (mean, SD)*	382	0.08 (0.98)	−0.10 (−0.26)	.09
Early life exposure				
Mother smoking (last trimester), yes (%)†	411	25 (11)	38 (20)	.01
Older siblings at birth, None/1/2 (% None)	385	135/60/23 (62)	98/48/21 (59)	.76
Allergic disposition—Mother				
Allergic rhinitis, yes (%)	410	166 (74)	144 (77)	.44
Eczema, yes (%)	411	105 (47)	93 (50)	.56
Allergic disposition—Father				
Asthma, yes (%)	399	42 (19)	26 (14)	.17
Allergic rhinitis, yes (%)	397	77 (36)	53 (29)	.18
Eczema, yes (%)	397	31 (14)	21 (12)	.40

SD, Standard deviation.

*Please see this article's Online Repository at www.jaci-inpractice.org.† ≥ 1 cigarette/wk.**TABLE III.** Association between FeNO and exercise test result

TABLE III: Association between FeNO and exercise test result				
Variables	Crude	P value	Adjusted*	P value
	Estimate [95% CI]		Estimate [95% CI]	
All				
FeNO vs exercise test†	2.42 [0.79; 4.05]	.004	1.99 [0.44; 3.53]	.012
Asthmatic				
FeNO vs exercise test†	3.43 [−1.40; 8.27]	.16	6.08 [0.92; 11.2]	.023
Nonasthmatic				
FeNO vs exercise test†	2.15 [0.40; 3.89]	.016	1.11 [−0.49; 2.70]	.17

CI, Confidence interval; FeNO, fraction of exhaled nitric oxide; FEV₁, forced expiratory volume in 1 s.FeNO is analyzed as a log₂ transformed, continuous value and exercise test results as percentage drop in FEV₁.

*Adjusted for gender, inhaled corticosteroid treatment 28 d before tests, upper respiratory infections 7 d before tests.

†General linear model calculating β -coefficient (estimate).

respiratory symptoms during exercise, and 28 (13%) had asthma. The mean (standard deviation) FeNO level was 8.7 ppb (7) (Table I).

Baseline characteristics including gender, social circumstances (see Table E1 in this article's Online Repository at www.jaci-inpractice.org), early life environmental exposures, and allergic predisposition of the 224 children included in the study and the 187 excluded children are outlined in Table II. The excluded versus included children had a significant higher proportion of mothers who smoked in the last trimester of pregnancy (20% vs 11%, $P = .01$) and a trend of lower social circumstances (mean, −0.10 vs 0.08, $P = .09$), but did not differ with respect to the remaining baseline characteristics.

Association between FeNO and exercise test results

In the entire study population of children with and without an established diagnosis of asthma, FeNO level and exercise test results were linearly associated with a doubling of FeNO corresponding to a postexercise test drop in FEV₁ of 2.4% (95% CI, 0.8–4.1), $P < .01$ (Table III), that is, increasing bronchial reactivity with increasing FeNO before the test. The results remained significant after adjusting for gender, respiratory tract

infections, and inhaled corticosteroid treatment: 2.0% drop in FEV₁ (0.4–3.5), $P = .01$. The FeNO level was associated with exercise test results in both strata of children with or without asthma (Figure 2), although the association was strongest in children with an established asthma diagnosis: 6.1% drop in FEV₁ (0.9–11.2), $P = .02$ (Table III).

ROC analysis

We thereafter conducted a ROC curve analysis to explore whether our finding of linear association between FeNO and degree of bronchoconstriction after exercise test had a clinical value, that is, whether the elevated FeNO level can be used for predicting exercise test outcomes. The ROC curve analysis showed an area under the curve of 0.68 (Figure 3) with 17 ppb as the optimal FeNO cutoff value for predicting exercise test outcome among children with a history of respiratory symptoms during exercise (Table E2, available in this article's Online Repository at www.jaci-inpractice.org). This FeNO cutoff resulted in a negative predictive value of 74% and specificity of 89%, which were higher than the positive predictive value of 67% and sensitivity of 40%. This corresponded to a likelihood ratio of a positive test of 3.63, which only indicates a “slight to moderate”

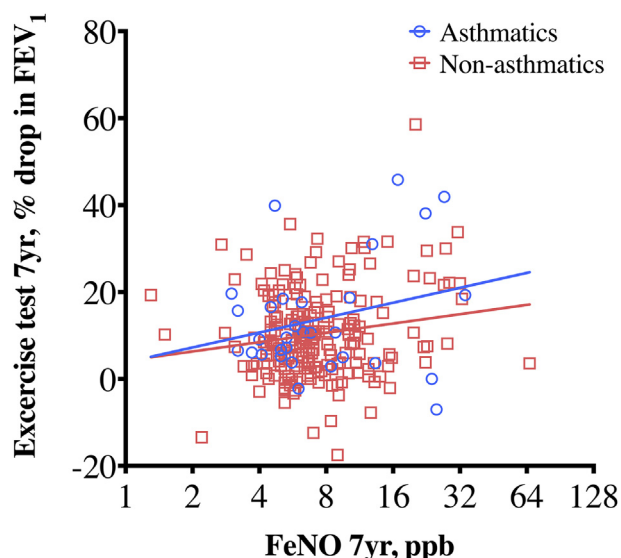


FIGURE 2. FeNO and exercise test results. Scatterplot of FeNO and exercise test results showing the distribution of asthmatic and nonasthmatic children. *FeNO*, Fraction of exhaled nitric oxide; *FEV₁*, forced expiratory volume in 1 second.

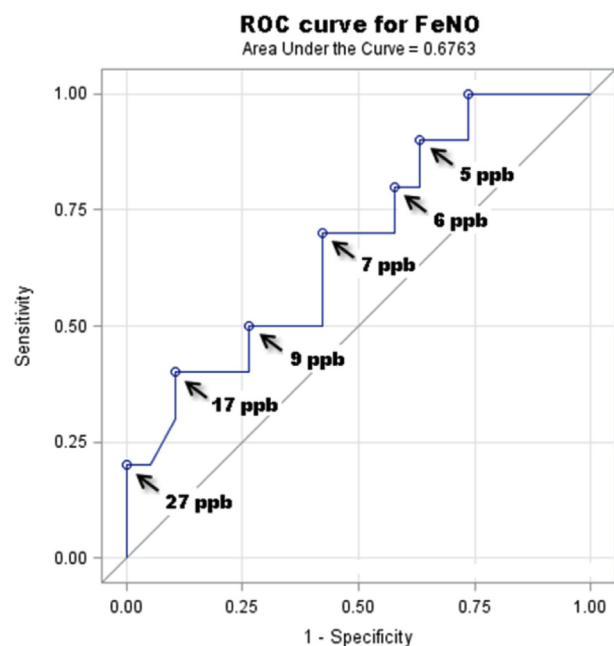


FIGURE 3. ROC curve for FeNO and exercise test at 7 years. The curve shows the predictive value of FeNO at different cutoff levels for predicting a positive exercise test among children with reported symptoms. *FeNO*, Fraction of exhaled nitric oxide; *ROC*, receiver operating characteristic.

(15% to 30%) increase in the likelihood of a positive exercise test when FeNO is above 17 ppb.²¹ Accordingly, the likelihood ratio of a negative test was 0.67, which indicates a “slight to no” (0% to –15%) decrease in the likelihood of a negative exercise test when FeNO is below 17 ppb. The ROC curve analysis also

showed that a FeNO cutoff of 27 ppb resulted in a specificity and positive predictive value of 100% with a negative predictive value of 73% and a sensitivity of 22%.

Elevated FeNO, exercise test outcome, and history of respiratory symptoms during exercise

Finally, we studied the validity of reported respiratory symptoms during exercise by children and their caregivers. We observed no significant association between the FeNO level and report of respiratory symptoms during exercise in the entire study population (adjusted odds ratio, 1.21 [0.74–1.98], $P = .45$), or in either strata of children with or without asthma ($P > .05$) (Table IV). Similarly, we found no association between exercise test results and reported respiratory symptoms during exercise, which may be caused by underreporting, that is, a large proportion of children with positive tests not reporting respiratory symptoms during exercise (83%).

Association between FeNO and exercise test results stratified by atopic status

The FeNO level was similarly associated with exercise test results in both strata of children with or without elevated sIgE; however, the association was stronger in the strata of children without elevated sIgE. The reverse was seen when atopy was defined by SPT results, where the association between FeNO and exercise test results was strongest among the children with a positive SPT (Table E3, available in this article’s Online Repository at www.jaci-inpractice.org).

DISCUSSION

Primary findings

FeNO and exercise test results were linearly associated traits in 7-year-olds at risk of asthma comprising children both with and without an established diagnosis of asthma, suggesting that exercise-induced bronchoconstriction develops as a continuum from health to disease. However, the study also shows that FeNO cannot be used in clinical practice to predict exercise test outcome, as the most optimal cutoff for FeNO only had the potential to exclude a positive exercise test in 74% of children with a history of respiratory symptoms during exercise. Furthermore, the study warrants that children’s report of respiratory symptoms during exercise is unreliable, as we found no association between symptom history and elevated FeNO or symptom history and a positive exercise test.

Strengths and limitations of the study

It is a strength that all exercise tests and FeNO measurements were performed by a small group of trained research personnel according to standard operating procedures following the ATS guidelines, which assured consistency in the objective measures. The clinical interviews about the history of respiratory symptoms during exercise included both the children and their parents, and were performed by the COPSAC research pediatricians strictly adherent to predefined questions with closed response categories, thus avoiding diagnostic heterogeneity. As all mothers have a history of asthma, respiratory symptomatology is well known to the families, which should minimize the risk of recall bias and parental misinterpretation of the symptoms.

It is well known that uncontrolled or steroid naïve asthma and respiratory infections can influence FeNO levels,^{22,23} and hence bias the association analyses. However, the nature of our cohort,

TABLE IV. Association between FeNO and reported symptoms

Variables	Crude	P value	Adjusted*	P value
	OR [95% CI]		aOR [95% CI]	
All				
FeNO vs reported symptoms†	1.25 [0.83; 1.88]	.29	1.21 [0.74; 1.98]	.45
Exercise test vs reported symptoms†	1.03 [0.99; 1.06]	.12	1.04 [0.99; 1.09]	.12
Asthmatic				
FeNO vs reported symptoms†	1.61 [0.77; 3.34]	.20	2.34 [0.88; 6.26]	.09
Exercise test vs reported symptoms†	1.04 [0.97; 1.11]	.24	1.05 [0.98; 1.12]	.20
Nonasthmatic				
FeNO vs reported symptoms†	0.85 [0.43; 1.67]	.64	0.78 [0.37; 1.64]	.51
Exercise test vs reported symptoms†	0.99 [0.94; 1.05]	.78	1.02 [0.96; 1.08]	.58

CI, Confidence interval; FeNO, fraction of exhaled nitric oxide; aOR, adjusted odds ratio.

FeNO is analyzed as a log_e transformed, continuous value.

*Adjusted for gender, inhaled corticosteroid treatment 28 d before tests, upper respiratory infections 7 d before tests.

†Logistic regression calculating odds ratio (estimate).

with close clinical scheduled follow-up and additional acute visits in case of symptoms of asthma or allergy, minimizes the risk of uncontrolled or undiscovered airway diseases. In addition, the relatively low mean FeNO value we found in our cohort²¹ is presumably due to the high number of healthy cohort participants without an asthma diagnosis or allergic symptoms. Furthermore, adjusting the analyses for respiratory infections and inhaled corticosteroid treatment, and stratifying by asthma did not change our results noteworthy, which is an important strength of the study.

A possible limitation of the study is that FEV₁ was followed for only 15 minutes after exercise in accordance with the recommendation by ATS/European Respiratory Society for exercise testing.¹⁵ This may be too short a follow-up time as it has recently been shown that there is a fall in FEV₁ till at least 30 minutes after exercise in adolescents.²⁴

The study investigated children at high risk of having or developing exercise-induced bronchoconstriction, which limits the application of our findings to the general population. The high-risk nature of the cohort could also affect the ROC curve analysis results, as the high prevalence of positive exercise tests in the population automatically leads to a lower negative predictive value, whereas in a normal population the negative predicted value is expected to be higher. However, the high-risk nature of the cohort does not affect our ability to analyze the within subject relationship between FeNO and exercise test results and the relation to history of respiratory symptoms during exercise. Furthermore, it is an important research question to investigate the relationship between markers of airway inflammation (FeNO) and bronchial hyperresponsiveness (exercise challenge test) in children at risk of asthma before the development of symptoms to capture subclinical disease.

Another concern is the low sensitivity of 40% that we find in the present study, which is in contrast to other prior studies in older children.^{10,25} This could be due to the lower age, as another study of 3- to 7-year-olds found a similar sensitivity of 41% to 50% depending on the FeNO cutoff level.²⁶

Interpretation

This study included concomitant assessments of FeNO, exercise test, and a physician interview about the history of respiratory symptoms during exercise in a large at-risk population

including children both with and without an established asthma diagnosis. FeNO and exercise test results were linearly associated in the population irrespective of asthma diagnosis, which suggests that the development of exercise-induced bronchoconstriction is a continuum from health to disease. The association was strongest among children with an established asthma diagnosis, who have bronchial hyperresponsiveness secondary to underlying inflammation, and it is plausible that subclinical type 2 inflammation was the driver of the observed association between FeNO and exercise test results among children without an asthma diagnosis. Irrespective of that, in our data there was no obvious clinically applicable FeNO cutoff to predict bronchial obstruction after exercise as the best performance of FeNO with a 17 ppb cutoff resulted in a sensitivity of 40%, specificity of 89%, positive predictive value of 67% and negative predictive value of 74%, meaning that 26% of the children with a positive exercise test would be overlooked using such cutoff. Using a 27 ppb FeNO cutoff resulted in a positive predictive value of 100%, whereas the negative predictive value was 73% with such cutoff.

In line with our findings of poor predictive values, another study reported an optimal FeNO cutoff value of 16 ppb among 126 asthmatic children aged 5 to 18 years treated with inhaled corticosteroids with sensitivity, specificity, positive, and negative predictive values of 83%, 47%, 60%, and 74%, respectively.²⁵ Our own previous study of steroid-naïve asthmatic outpatients aged 7 to 16 years showed better prediction of exercise test outcome with an FeNO cutoff value of 20 ppb resulting in sensitivity, specificity, positive and negative predictive values of 71%, 76%, 53%, and 88%, respectively.¹⁰ However, it is a different approach to investigate selected populations of only asthmatic children versus our approach of investigating an at-risk population, which explores whether FeNO can be used to screen for exercise-induced bronchoconstriction irrespective of the history of respiratory symptoms during exercise.

When stratifying by atopic status, we found conflicting results depending on the definition of atopy. As we have previously shown that there is substantial disagreement between results of SPT and sIgE in young children,²⁷ we analyzed sIgE and SPT results separately. Interestingly, when we defined atopy as a positive SPT, the results were in line with a previous study of 84 wheezy children aged 3 to 7 years that found a stronger predictive value of FeNO among children with a positive SPT.²⁶

However, we also found a significant association among the children with negative SPT, although the association was weaker. In contrast, when defining atopy by elevated sIgE (>0.35 kU_A/L), the estimates were similar among the atopic and nonatopic children, but only significant among the nonatopic children, which could be a power issue as elevated FeNO is associated with type 2 inflammation.

In type 2 asthmatic airway inflammation, the local release of IL-4 and IL-13 upregulates the expression of epithelial inducible nitric oxide synthases that produce nitric oxide from L-arginine.²⁸ These mechanisms are often associated with the presence of eosinophilia as type 2 cells also produce IL-5, and FeNO is therefore proposed as a noninvasive marker of eosinophilic allergic airway inflammation.²⁹

Exercise-induced bronchoconstriction was initially thought to be secondary to underlying inflammation and mediator release from mast cells,³⁰ but is now recognized as a heterogeneous condition with varying underlying pathogenesis. In subjects with asthma, eosinophilic or neutrophilic airway inflammation results in damaged epithelial surfaces and increased airway reactivity after exercise, whereas pathophysiologic changes induced by intense exercising can result in exercise-induced bronchoconstriction in subjects without asthma and underlying inflammation. Hyperventilation through the mouth associated with intense exercise causes the need for humidifying and heating large volumes of air during a short period of time. If the capacity for that is exceeded, breathing poorly conditioned air at high flows may cause an epithelial injury and make the smooth muscle more sensitive to respiratory water loss and the increase in osmolarity of the airway surface liquid, which leads to exercise-induced bronchoconstriction.³¹

The fact that exercise-induced bronchoconstriction can occur in subjects without known asthma and in patients without evident type 2-driven eosinophilic inflammation, both of whom would be expected to have low FeNO levels, may explain our finding that FeNO cannot reliably be used to predict the need for exercise testing.

We found no association between a positive exercise test and a history of respiratory symptoms during exercise, which may be caused by neglect and underreporting. This is in line with a study of 130 asthmatic and nonasthmatic children aged 7 to 19 years showing that half of children who did not report respiratory symptoms during exercise had a positive exercise test.³² Similarly, another study of 43 children with asthma aged 7 to 14 years found that the children's reporting of respiratory symptoms during exercise and the parent's perception of their child's symptoms did not relate to exercise test results.⁷ The lack of association between reported respiratory symptoms during exercise and bronchoconstriction on formal exercise challenge in these and our study may also explain why we did not observe the association between the FeNO level and symptom history.

In our study, all the mothers have a history of asthma, which should increase their awareness of the child's respiratory symptoms during exercise and decrease the risk of misinterpretation of the symptoms. In spite of that, it seems that symptoms are still underreported. The observation of a linear association between FeNO and bronchial obstruction after the exercise test in a population of children both with and without an established asthma diagnosis serves as a reminder of the risk of a missing subclinical disease, as underlying inflammation is the most obvious explanation for such association in the strata of children

without an asthma diagnosis. Regrettably, we were unable to find an optimal FeNO cutoff value to exclude the need for exercise testing, which cautions against a dichotomized interpretation of FeNO in this regard. Instead, we suggest using FeNO as a monitoring tool, where variations within the same child over time could be more relevant clinically than single assessments. For that purpose, changes of at least 10 ppb or 20% have been suggested by the ATS guidelines to be a significant increase of FeNO.²¹

CONCLUSIONS

FeNO and bronchial obstruction after the exercise test are linearly associated traits in our at-risk population of 7-year-old children with and without an established asthma diagnosis, suggesting that exercise-induced bronchoconstriction develops as a continuum from health to disease. However, FeNO cannot reliably be used dichotomized in clinical practice to predict the need of exercise testing.

Acknowledgements

We express our deepest gratitude to the children and families of the COPSAC2000 cohort study for all their support and commitment. We acknowledge and appreciate the unique efforts of the COPSAC research team.

The guarantor of the study is HB, from conception and design to conduct of the study and acquisition of data, data analysis, and interpretation of data. A-MMS, CFC, and BLC have written the first draft of the manuscript. All co-authors have provided important intellectual input and contributed considerably to the analyses and interpretation of the data. All authors guarantee that the accuracy and integrity of any part of the work has been appropriately investigated and resolved, and all have approved the final version of the manuscript. The corresponding author had full access to the data and had final responsibility for the decision to submit for publication. No honorarium, grant, or other form of payment was given to any of the authors to produce this manuscript.

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TABLE E1. Baseline table of original information used to derive the measure of social circumstances

Variables	Variable type	COPSAC ₂₀₀₀	
		Categorical: n (%) Numeric: mean (SD)	missing data
Household income	Categorical		29
Below 400,000		120 (31.41)	
400,000-600,000		153 (40.05)	
600,000-800,000		76 (19.90)	
800,000-1000,000		22 (5.76)	
Above 1000,000		11 (2.88)	
Maternal education	Categorical		29
Elementary or college		220 (57.59)	
Medium		104 (27.23)	
University		58 (15.18)	
Maternal age	Numeric	32.11 (4.51)	0

COPSAC₂₀₀₀, Copenhagen Prospective Studies on Asthma in Childhood₂₀₀₀; SD, standard deviation.

Social circumstances are correlated to household income (ratio = 0.73, $P < .001$), to maternal level of education (ratio = 0.68, $P < .001$), and to maternal age (ratio = 0.75, $P < .001$).

TABLE E2. ROC curve analysis for the exercise test

FeNO cutoff (ppb)	Sensitivity (%)	Specificity (%)	Positive predictive value (%)	Negative predictive value (%)	Likelihood ratio of positive test	Likelihood ratio of negative test
27	22	100	100	73	—	0.78
17	40	89	67	74	3.63	0.67
9	50	74	50	74	1.92	0.68

FeNO, Fraction of exhaled nitric oxide; ppb, parts per billion; ROC, receiver operating characteristic.

TABLE E3. Association between FeNO and exercise test result stratified by atopic status

Variables	Crude	P value	Adjusted*	P value
	Estimate [95% CI]		Estimate [95% CI]	
All				
FeNO vs exercise test†	2.42 [0.79; 4.05]	.004	1.99 [0.44; 3.53]	.012
Positive sIgE				
FeNO vs exercise test†	2.66 [−0.17; 5.48]	.06	2.12 [−0.24; 4.48]	.08
Negative sIgE				
FeNO vs exercise test†	3.07 [0.29; 5.85]	.031	3.32 [0.48; 6.16]	.022
Positive SPT				
FeNO vs exercise test†	4.70 [0.44; 8.95]	.032	3.69 [0.05; 7.33]	.047
Negative SPT				
FeNO vs exercise test†	2.19 [−0.27; 4.65]	.08	2.47 [0.27; 4.86]	.044

CI, Confidence interval; FeNO, fraction of exhaled nitric oxide; FEV₁, forced expiratory volume in 1 s; SPT, skin prick test.

FeNO is analyzed as a log₂ transformed, continuous value and exercise test results as percentage drop in FEV₁.

*Adjusted for gender, inhaled corticosteroid treatment 28 d before tests, upper respiratory infections 7 d before tests.

†General linear model calculating β -coefficient (estimate).

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