Why use FeNO for asthma diagnosis and routine management?

A NIOX® WHITE PAPER
Summary

This white paper summarizes current evidence supporting the use of fractional exhaled nitric oxide (FeNO) to aid in the initial diagnosis and routine management of asthma. The clinical benefits of measuring and monitoring FeNO in asthma are well recognized and FeNO testing is currently recommended by all key international and national asthma guidelines.

Awareness of a patient’s FeNO score and how it changes with disease progression and treatment helps clinicians optimize management in a cost-effective manner by:

- Identifying patients with airway inflammation, specifically Type 2 airway inflammation, who are likely to respond to corticosteroid therapy,
- Guiding inhaled corticosteroid (ICS) dosing and uncovering non-adherence to treatment,
- Predicting patients who are more likely to experience an asthma exacerbation and reducing exacerbations [1], and
- Assessing suitability for and predicting response to specific asthma biologic agents when such treatments are indicated [2].

Asthma

Asthma is defined as a heterogeneous disease, usually characterized by airway inflammation. A history of respiratory symptoms (such as wheezing, shortness of breath, chest tightness and cough) that vary over time will be present, along with variable expiratory airflow limitation. There is marked heterogeneity in individual patient triggers and responses to therapy and several clinical phenotypes have been identified [3].

The diagnosis of asthma is usually based on identification of a characteristic pattern of symptoms as well as documented airflow limitation with excessive variability. To assess the latter, variability of twice daily peak expiratory flow, bronchodilator reversibility, exercise challenge testing and bronchial challenge testing may be utilized [3]. None of these assessments, however, examine underlying airway inflammation.

Airway inflammation

The inflammatory nature of asthma has been appreciated for decades and this awareness has influenced management strategies and the development of treatment guidelines [4]. More recently, attention has focused on understanding the underlying mechanisms of airway inflammation, so that treatment can be efficiently targeted. Two general asthma subtypes (endotypes, but often referred to as phenotypes), based on the underlying inflammation, have been described:

1. Type 2 or T2 or T2-high: comprised of allergic and non-allergic eosinophilic asthma, a type 2 immune response regulated by T helper 2 (Th2) cells and other Type 2 inflammatory cells (eosinophils, macrophages, basophils, mast cells) secreting interleukin-4 (IL-4), IL- 5, and IL-13, typically responsive to corticosteroids.
2. Non-type 2 or non-T2 or T2-low: non-eosinophilic non-atopic asthma, may be neutrophilic or paucigranulotytic (neither eosinophils nor neutrophils elevated), much less well defined and often not responsive to conventional asthma therapy [5,6].

The proportion of patients with Type 2 vs. non-type 2 inflammation or asthma varies by cohort and perhaps severity of disease. One early paper suggested that Type 2 asthma constitutes 50% of mild to moderate asthma [7], whereas a more recent study found that almost 84% of patients with severe asthma most likely exhibit an eosinophilic phenotype, with over 92% likely or most likely to have the eosinophilic phenotype. In contrast, only 1.6% of the severe asthma patients had a non-eosinophilic phenotype [8], reflecting the small number (3-4%) of asthma patients who respond poorly to ICS therapy [9]. Regardless of the exact percentages, it is evident that clearer understanding of the
pathophysiologic mechanisms can clarify the phenotype and help guide treatment to the most effective options [4] with the potential to spare exposure to unhelpful ICS and avoid adverse effects [10,11].

However, assessing airway inflammation is not always an easy or simple task. Bronchoscopy with bronchial biopsy or brushing or bronchoalveolar lavage can be quite informative but is highly invasive and not indicated for routine assessment [12]. Sputum induction, usually for analysis of sputum neutrophils and/or eosinophils, requires careful attention to the induction procedures and a high degree of patient cooperation to achieve adequate accuracy and repeatability, close monitoring of pulmonary function for safety reasons and sample processing that has been described as laborious [13,14]. Neither technique affords immediate results and both are most often used for research purposes.

**Biomarkers of airway inflammation**

A biological marker or biomarker is defined as a characteristic that can be objectively measured and evaluated as an indicator of normal biological or pathogenic processes or pharmacological responses to a therapeutic intervention [15].

Given the many disadvantages of bronchoscopic sampling and evaluation (invasiveness, inability to repeat frequently, safety concerns in severe patients, timeliness of results, specialized expertise required, cost, etc.) induced sputum eosinophils is another means to investigate airway inflammation. However, even sputum induction has limitations, as noted in the previous section. Since both are less cumbersome than sputum eosinophils [14], FeNO and peripheral blood eosinophil count have emerged as alternative biomarkers of airway inflammation. Blood eosinophils, FeNO and sputum eosinophils are all are recommended for assessing the severe asthma phenotype and confirming the presence of Type 2 inflammation [2].

The correlation of FeNO and sputum eosinophils has been examined in both children and adults with asthma [16-20]. In a study of steroid-naïve school-age children and those treated with controller medications, FeNO at >19 parts per billion (ppb) had high sensitivity, specificity and positive and negative predictive values for sputum eosinophils >3% (the generally regarded cutoff for Type 2 inflammation). The authors also noted a significantly higher diagnostic yield over the standard approach of spirometry-based asthma diagnosis when FeNO was measured and considered [18]. In a study of adults with varying severities of asthma, a FeNO cut point of 42 ppb was a good differentiator between eosinophilic and non-eosinophilic airway inflammation [19].

Blood eosinophil count has performed marginally better than FeNO in a few studies and the two biomarkers appear to be only weakly correlated with each other in most studies. This suggests
that FeNO and blood eosinophils relate to different, but perhaps slightly overlapping, inflammatory pathways. Therefore, each should be regarded as distinct indicators of different aspects of Type 2 airway inflammation, providing complementary information [19,21-23].

**Mechanism of FeNO in asthma**

Nitric oxide (NO) is present in all organ systems and has a variety of functions. Its discovery as a cardiovascular signalling molecule won the Nobel Prize in Physiology or Medicine in 1998. While the exact role of NO in the lung remains unclear, it is generally regarded as marker for upregulated airway inflammation [24].

NO in exhaled breath, FeNO, is derived from airway epithelial cells via NO synthase enzymes (NOS). Most patients with asthma express high levels of inducible nitric oxide synthase (iNOS) and, thus, high FeNO levels [24]. IL-4 and IL-13, cytokines expressed as a result of activation of antigen-specific Th2 cells following allergen exposure, act through signal transducer and activator of transcription 6 (STAT6) to induce gene transcription that upregulates iNOS in airway epithelial cells, which in turn produce the NO that is released in expired breath [4,5,22].

Not long after the discovery of the elevated FeNO in patients with asthma, it was found that corticosteroid treatment, both oral and inhaled administration, results in a significant reduction of FeNO [24,1]. This effect has been verified in numerous studies. In hindsight, a reduction of FeNO would not be unexpected as corticosteroids act by suppressing a wide range of inflammatory pathways.

These two observations, the increase of FeNO seen during airway inflammation and asthma, and the reduction of FeNO with successful anti-inflammatory therapy, form the foundation of the utility of measuring and monitoring FeNO in patients with asthma.

**Use of FeNO in asthma**

*Identification of Type 2 inflammation and responsiveness to steroids*

Measuring FeNO can aid in the management of asthma by helping to identify patients with Type 2 airway inflammation and, therefore, those who will respond favorably to corticosteroid therapy. Based on the mechanism of FeNO production, it should be clear that an elevation of FeNO is indicative of underlying Type 2 airway inflammation and, thus asthma. Indeed, an analysis of 43 studies with a total of 13,747 patients, showed a high FeNO increases the likelihood of asthma by 2.8 to 7.0 times, depending on the FeNO level [25].
FeNO measurement compares well with other diagnostic procedures for asthma, including induced sputum eosinophils, spirometry, and bronchial challenge testing [26-28]. FeNO was found to have performed well to the extent that authors of a review speculated whether FeNO might render bronchoprovocation testing superfluous [29]. The 2021 European Respiratory Society (ERS) Clinical Practice Guidelines for the Diagnosis of Asthma in Children Aged 5-16 Years and 2022 ERS Guidelines for the Diagnosis of Asthma in Adults both include FeNO as an objective test recommended as part of the diagnostic work-up of asthma [30,31].

A large study in primary care investigated FeNO for predicting response to ICS treatment in difficult to manage patients with undiagnosed, non-specific respiratory symptoms. A higher baseline FeNO was associated with an increased likelihood of a positive response to ICS, determined using the Asthma Control Questionnaire (ACQ7, a 7-item questionnaire on asthma control), symptoms (cough), spirometry (forced expiratory volume in one second, FEV1) and a global evaluation of treatment effectiveness. The baseline FeNO value was a better predictor of clinical improvement than peripheral blood eosinophils, spirometry and clinical opinion [32].

Other studies have affirmed that FeNO predicts response to ICS [11,33-36]. Some have also shown that FeNO is an even better predictor of corticosteroid responsiveness compared to spirometry, bronchodilator response, peak flow variation, and bronchial hyperresponsiveness [34-36]. A FeNO value of >47 ppb was found to have the best predictive value [36].

**Guiding ICS therapy**

Measuring FeNO can aid in the management of asthma by helping to identify patients who may need to step up or can step down their ICS and therefore can be used to optimize corticosteroid treatment. Individualized therapy can help maximize therapeutic benefits while reducing the likelihood of corticosteroid adverse effects.

The benefit of periodically re-assessing airway inflammation using FeNO in chronic asthma was demonstrated in a landmark study by Smith et al. Two groups of patients were evaluated: one used guideline-based care with traditional monitoring (symptoms, spirometry, etc.) and the other included a FeNO-based approach. After 12 months of treatment, the dose of ICS (fluticasone propionate, FP) was significantly lower in the FeNO group compared to traditional monitoring (mean dose 370 vs 641 mcg/day, respectively; p=0.003). Asthma control was also better using FeNO monitoring, with 45.6% fewer exacerbations (not significant) and a greater proportion achieving “total control” of their asthma. Exposure to high doses of ICS was reduced by utilizing the FeNO-based strategy to step down treatment. At the completion of the study 48% of the guideline care group was receiving 1,000 mcg/day of FP compared to just 20% in the FeNO group [37]. These results have been replicated in numerous studies, in both children and adults.

Data from Smith et al. (N Engl J Med 2005) illustrating reduced exacerbations (left) and ICS dose (right) with use of a FeNO-based treatment strategy compared to guideline-based traditional monitoring (control group).
Research has also confirmed that FeNO can predict worsening or loss of asthma control when controller therapy is tapered or stopped. Two studies performed in children being withdrawn from ICS demonstrated that an increase of FeNO two and four weeks after discontinuation of ICS is a predictor of asthma relapse and loss of control [38,39]. A review of available studies reinforced the prognostic value of measuring and monitoring FeNO [40].

In a real-world study conducted in 337 clinical practices in the United States, it was demonstrated that FeNO allows clinicians to assess underlying airway inflammation and alter anti-inflammatory therapy accordingly. Clinical assessment of asthma agreed with the FeNO measurement in only 56% of cases, even less frequently (34%) in patients with high inflammation (FeNO >50 ppb in adults and >35 ppb in children). After becoming aware of their patients’ FeNO values, clinicians often modified treatment. ICS were altered in 90% of cases, with initiation or the dose increased in 66% of patients with high inflammation. Measuring FeNO enabled assessment of underlying inflammation, frequently leading to revision of the treatment plan [41].

Uncovering non-adherence to steroids

Medication non-adherence, specifically with ICS, is a major reason for poor asthma control, asthma-related emergency department visits, inpatient hospitalizations, persistent eosinophilic inflammation and increased oral corticosteroid use [42,43]. Measuring FeNO can aid in the management of asthma by helping to identify patients who are non-adherent with ICS therapy. Recall that the mechanism of NO production in the lung is highly susceptible to corticosteroids, resulting in a reduction of FeNO following treatment.

Children who attended a one-week asthma camp continued their usual medications in an observed manner and FeNO was monitored daily. During the camp, there was a significant decrease of FeNO, that was attributed to improved adherence resulting from the observed ICS administration [44]. Observations such as these highlight the role of FeNO as a surrogate marker of ICS adherence. Since FeNO is elevated in asthma and is responsive to corticosteroid therapy, FeNO is generally lower in adherent vs. non-adherent patients. Reduced FeNO values following confirmed ICS administration could be indicative prior poor adherence [45].

McNicholl et al. introduced the “FeNO Suppression Test” during which FeNO is monitored daily and ICS administration is supervised over seven days [46]. A fall in FeNO during the observation period, a positive test, has proven to be accurate in distinguishing previous ICS non-adherence from refractory asthma [23,45-48] that may require a different treatment strategy [45] and can prevent the unnecessary escalation to biologics [48]. A negative FeNO suppression test, indicating prior good adherence to ICS, correlated with progression to biologic therapy in one investigation of FeNO as a predictive tool. Conversely, a positive test identified patients who could be effectively treated with conventional controller therapy and subsequently discharged from specialist care [49].

Predicting and reducing asthma exacerbations

Measuring FeNO can aid in the management of asthma by helping to reduce the likelihood of asthma exacerbations in patients at risk for future events. While the frequency and severity of exacerbations are variable, the risks for exacerbations are well described and include a history of previous exacerbation, hospitalization, emergency department visit, use or oral corticosteroids and poor adherence to ICS [3]. Elevated FeNO has also been shown to be an indicator of poor asthma control [50].

As noted previously, numerous studies have demonstrated that FeNO can help optimize ICS therapy and uncover non-adherence, and thereby help avoid asthma exacerbations in patients at risk. Two Cochrane Systematic Reviews examined the efficacy of tailoring asthma treatment based on FeNO; separate meta-analyses were performed for studies conducted in adults and children.
In the adult analysis, seven studies involving 1700 patients, it was found that using FeNO to guide ICS dosing was beneficial in reducing exacerbations. The exacerbation rate was reduced by 40% compared to standard guideline-based care [51]. In the analysis of pediatric studies there were nine clinical trials with over 1400 children. Like adults, there were significantly fewer children with exacerbations with the FeNO-based strategy as compared to traditional clinical monitoring and guideline management [52]. A combined report of 13 studies (five adult, eight pediatric) was subsequently published and reaffirmed that a FeNO-based treatment strategy was significantly better than traditional monitoring in terms of reducing asthma exacerbations [53].

An evidence-based review of measuring FeNO in adults and children was prepared for the United States Department of Health and Human Services Agency for Healthcare Research and Quality (AHRQ). The previously mentioned analysis was conducted on data from 13,747 patients participating in 43 studies. It concluded that the use of asthma management algorithms that incorporate FeNO testing reduce the risk of exacerbations and can possibly reduce the risk of exacerbations requiring treatment with oral corticosteroids [25].

With development of asthma biologic agents and clinical trials conducted with more severe patients, attention has turned to exacerbations and exacerbation rates as outcome measures. Two recently published post-hoc analyses further examined biomarkers and exacerbations in moderate to severe asthma patients participating in the placebo arms in studies of biologic agents. Both found greater exacerbation rates in patients with elevated baseline FeNO, particularly with FeNO ≥50 ppb [54,55]. In one analysis patients with a baseline FeNO of ≥50 ppb had a 1.54 times higher exacerbation rate that patients with FeNO <25 ppb (p=0.0097), and patients with FeNO 25-49 ppb had an exacerbation rate 1.33 times higher than patients with FeNO <25 ppb (p=0.0572) [54]. In the larger analysis, that utilized data pooled from seven clinical trials, patient groups with persistently high FeNO, ≥20 ppb throughout the treatment period, had the greatest exacerbation rates [55]. In a prior study of biomarkers in severe asthma patients, high FeNO was significantly associated (p=0.0008) with more exacerbations. FeNO also demonstrated a stronger correlation with exacerbations than blood eosinophils [56].

**Assessing suitability for and predicting response to biologics**

Measuring FeNO can aid in the management of asthma by helping to identify severe asthma patients who exhibit the Type 2 phenotype and those who will benefit most from treatment with certain biologic agents, when such agents are appropriate. Ruling out non-adherence and assessing the Type 2 phenotype are prerequisites for considering biologic therapy, as recommended by international guidelines [2]. An international expert panel recently stated that FeNO adds “prognostic and predictive value and should be measured in all patients with severe asthma” [57].

Omalizumab binds immunoglobulin E (IgE), an antibody produced in response to specific allergens. By binding free IgE and inhibiting binding of IgE to its receptor, it reduces expression of Type 2 inflammatory mediators (IL-4, IL-5, IL-13, etc.) and eosinophils. In a post-hoc analysis of the biomarkers measured during a clinical trial, a FeNO cut point of 19.5 ppb was determined and evaluated. With FeNO above 19.5 ppb at baseline, omalizumab-treated patients achieved a significantly greater 53% reduction in exacerbations compared to the placebo group [58]. These data comprise the basis for the 20 ppb cutoff, above which a good response is predicted for omalizumab, that is recommended in current guidelines [2].

Dupilumab is a monoclonal antibody to the IL-4 receptor alpha subunit (IL-4Rα). It inhibits signalling of IL-4 and IL-13, both Type 2 inflammatory cytokines. Data from a phase 3 clinical trial demonstrate a greater benefit, improvement of pulmonary function and reduction of exacerbation rate, in patients with a higher baseline FeNO (25-49 and ≥50 ppb) [59]. Based on these data, a FeNO value >25 ppb predicts a good response to dupilumab and is often recommended as one of the criteria
for initiating treatment [2,60]. The role of FeNO with asthma biologics will continue to evolve with the availability of newer biologic agents and newer data. For example, tezepelumab, the mostly recently approved asthma biologic, acts by inhibiting thymic stromal lymphopoetin (TSLP). Like dupilumab, its effectiveness in asthma, as assessed by the reduction of exacerbation rate, is greater in severe uncontrolled patients with higher baseline FeNO values (25-49 and ≥50 ppb) [61].

Mepolizumab, one of several anti-IL-5 biologics, would not be expected to alter FeNO given its mechanism of acting on the IL-5 pathway (i.e., it does not directly affect IL-4 or IL-13). In fact, some pivotal trials of anti-IL-5 biologics did not include any FeNO measurements. Yet a more recent study generated interesting findings regarding FeNO and mepolizumab. It showed that in severe asthma patients treated with mepolizumab who were experiencing an exacerbation, there were two distinct inflammatory phenotypes. One was non-eosinophilic (low FeNO, high C-reactive protein and high sputum neutrophils) and the other was eosinophilic (high FeNO and high sputum eosinophils). FeNO (≤20 and ≥50 ppb) was the most useful discriminator of the inflammatory phenotype at exacerbation. This information could be used to direct the treatment of asthma exacerbations in patients receiving mepolizumab [62].

FeNO may also be useful for determining when to discontinue biologic therapy, regardless of the mechanism of action or inflammatory pathway affected. Real-word evidence identified a small group of “super-responders” receiving various biologics, including anti-IL-5 therapies, who are candidates for discontinuation. The criteria for stopping included exacerbations, use of oral corticosteroids, asthma control (Asthma Control Test), spirometry and suppression of Type 2 inflammation (FeNO <50 ppb) [63].

**FeNO in asthma guidelines**

Currently, FeNO testing is underutilized in clinical care; however, there is growing global momentum for its support. The 2021 American Thoracic Society (ATS) Clinical Practice Guideline on the Use of FeNO to Guide the Treatment of Asthma recommended “In patients with asthma in whom treatment is being considered, we suggest the use of FENO testing in addition to usual care over usual care alone” and concluded that “Clinicians should consider this recommendation to measure FENO in patients with asthma in whom treatment is being considered based on current best available evidence.” These recommendations were made despite acknowledgement that the overall quality of data is low according to the Grading of Recommendations, Assessment, Development, and Evaluation (GRADE) evidence-to-decision framework [64].
In addition, in just the past few years several other guidelines and recommendations specifically on the use of FeNO in asthma management have been published by national organizations such the Italian Respiratory Society (SIP/IRS) and Italian Society of Allergy, Asthma and Clinical Immunology (SIAAIC) jointly, Japan Respiratory Society (JRS), Asthma Collaborative Group (China), and panels of national experts in Mexico and Scotland [65-69].

Similarly, numerous recent national and international asthma guidelines include guidance regarding FeNO testing. Included are those from the United States (National Asthma Education and Prevention Program Focused Updates), United Kingdom (Scottish Intercollegiate Guidelines Network and British Thoracic Society or SIGN/BTS, and National Institute for Health and Care Excellence, NICE), Spain (Guía Española para el Manejo del Asma, GEMA 5.1), Mexico (Manejo Integral del Asma, MIA), Canada (Canadian Thoracic Society, CTS severe asthma recognition and management), and China (Expert consensus on severe asthma), as well as Europe (ERS diagnosis of asthma in children, ERS diagnosis of asthma in adults), ATS and ERS jointly (severe asthma management) and the Global Initiative for Asthma (GINA, difficult-to-treat and severe asthma) [70-76,30,31,77,2].

Cost-effectiveness of FeNO

Measuring and monitoring FeNO is cost-effective based on the improved outcomes achieved with the modest expenditure. Analyses from around the world have examined the cost-effectiveness of FeNO testing for the diagnosis and management patients with asthma. One early study investigating the addition of FeNO to standard care demonstrated reduced costs and improved accuracy compared to standard diagnostic methods [78]. In another, the accuracy of diagnosis was also improved but with a nominal €12 increase in the per patient cost [79].

Numerous studies have shown that monitoring FeNO, when utilized in the management of asthma, provides a cost savings over standard care alone. Improved outcomes and quality of life have been reported in both children and adults of all severities, even those with severe asthma requiring biologic therapy. Pharmacoeconomic research and modelling of FeNO testing have been conducted in the United States, United Kingdom, Germany, Spain, Sweden and Colombia in private insurer, public and national healthcare systems [78-89]. Given the high cost of exacerbation management (emergency department visits and hospitalizations, reduced quality of life and productivity, oral corticosteroid adverse effects, etc.) and the ability of FeNO monitoring to help reduce exacerbations at limited additional expense, the cost-effectiveness of FeNO testing is not unanticipated.

Conclusions

Current evidence supports measuring and monitoring FeNO to aid in the diagnosis and routine management of asthma. The benefits of FeNO are well recognized and FeNO testing is recommended by a multitude of national and international guidelines. Adding FeNO to the armamentarium of diagnostic and monitoring tools provides a unique, non-invasive and cost-effective measure of airway inflammation that complements pulmonary function and other clinical assessments.
References


