Clinical Policy Bulletin:
Breath Tests of Airway Inflammation: Exhaled Nitric Oxide and Exhaled Breath Condensate pH

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Policy

Aetna considers measurement of exhaled nitric oxide experimental and investigational for the assessment of the following indications because of insufficient evidence of its effectiveness (not an all-inclusive list):

- Asthma
- Chronic cough due to non-asthmatic eosinophilic bronchitis
- Lung cancer
- Pulmonary diseases (e.g., chronic obstructive pulmonary disease, pulmonary tuberculosis, sino-nasal disease) Sickle cell airway disease

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Background

Exhaled Nitric Oxide

A number of studies have investigated the relationship between exhaled nitric oxide (NO) and airway inflammation in asthma. These studies have found that levels of exhaled NO are elevated in people with asthma who are not taking inhaled glucocorticosteroids compared to people without asthma, yet these findings are not specific for asthma. Exhaled NO is thought to reflect eosinophilic airway inflammation in asthma.

Levels of exhaled NO have been suggested as non-invasive markers of airway inflammation in asthma, as an alternative to invasive methods such as examination of spontaneously produced or hypertonic-saline induced sputum for eosinophils and metachromatic cells. Measurement of exhaled NO may prove to be useful in diagnosing asthma, assessing adherence to treatment with inhaled corticosteroids, or in the identification of patients in whom respiratory symptoms are associated with eosinophilic airway inflammation. A number of methods have been developed to measure exhaled NO, including laser spectroscopy and chemiluminescence.

Current studies on exhaled NO in asthma have focused on the technical feasibility of measurement and correlations with pulmonary function and invasive methods of assessing airway inflammation. The evidence for assessment of exhaled NO for other pulmonary conditions is more limited. However, well-designed, long-term studies are needed to evaluate whether the addition of exhaled NO measurements to clinical and lung function assessment results in improved asthma control.

An assessment on a NO measurement system (NIOX) for monitoring response to asthma treatment prepared by the Canadian Coordinating Office for Health Technology Assessment (CCOHTA) (Hailey, 2004) concluded that no information was found on the extent to which the use of this device improves patients’ compliance with medication use or ensures appropriate prescribing. Comparative studies to obtain such measures of efficacy would be desirable. This is in agreement with the observation of Zeidler et al (2004) who stated that exhaled NO may be a useful parameter for monitoring asthmatic inflammation, adjusting therapy, and diagnosing asthma, although prospective longitudinal trials investigating the correlation between exhaled NO and clinical outcomes are necessary to determine its utility.

An assessment by the BlueCross BlueShield Association (BCBSA) Technology Evaluation Center (TEC) (2006) concluded that use of exhaled NO (FENO) levels for monitoring patients with chronic asthma does not meet the TEC criteria. The TEC assessment found that available evidence does not permit the conclusion that use of FENO monitoring to guide treatment decisions in asthma leads to improved outcomes. The assessment stated that the 7 studies that evaluated the predictive ability of FENO, and its potential to provide prognostic information that could influence management decisions, had considerable methodologic limitations and variability in study methodology that precluded synthesis of their results and definitive conclusions. The assessment stated that the 2 randomized, controlled trials included in the review suggest possible benefits for FENO monitoring, but are not sufficient to conclude that outcomes are improved. The assessment explained that each study reported different benefits that have not been reproduced. The assessment noted that differences in the control management strategy raise questions about the optimal management strategy to which FENO monitoring should be compared.
In a randomized-controlled trial, Smith et al (2005) reported a reduction in use of inhaled corticosteroids in asthma patients (n = 46) using FENO measurements to adjust their doses compared to asthma patients (n = 48) using conventional guidelines to adjust their doses. Patients were followed for 12 months. The final mean daily doses of fluticasone were 370 µg for the FENO group versus 641 µg for the control group, a difference of 270 µg per day. The rates of exacerbation were 0.49 episodes per patient in the FENO group and 0.90 in the control group, representing a non-significant reduction of 45.6% in the FENO group. The authors reported no significant differences in other markers of asthma control, use of oral prednisone, pulmonary function, or levels of airway inflammation (sputum eosinophils). Earlier studies, however, have demonstrated that control can be maintained through a standardized approach involving a uniform dose reduction. Smith’s findings may reflect over treatment in the control group rather than more appropriately targeted treatment in the FENO group. It is also unclear what effects multiple medications may have on the relationship between asthma control and FENO measurements.

van Mastrigt et al (2007) stated that there is still much uncertainty about the potential clinical utility of measurement of fractional FENO in infants. There is a need for clinical studies showing the merits and limitations of different methodologies, to standardize measurements of fractional FENO in infants, and to obtain normal reference values for this age group. Turner (2007) stated that over a relatively short time FENO has become recognized as an useful objective tool for diagnosing and monitoring asthma. However, more clinical studies are needed in order for FENO to become fully established in the diagnosis and management of asthma.

Shaw and co-workers (2007) tested the hypothesis that titrating corticosteroid dose using the concentration of FENO in exhaled breath results in fewer asthma exacerbations and more efficient use of corticosteroids, when compared with traditional management. A total of 118 subjects with a primary care diagnosis of asthma were randomized to a single-blind trial of corticosteroid therapy based on either FENO measurements (n = 58) or British Thoracic Society guidelines (n = 60). Participants were assessed monthly for 4 months and then every 2 months for a further 8 months. The primary outcome was the number of severe asthma exacerbations. Analyzes were by intention-to-treat. The estimated mean exacerbation frequency was 0.33 per patient per year (0.69) in the FENO group and 0.42 (0.79) in the control group (mean difference, -0.1 %; 95% confidence interval [CI]: -5.7 % to 4.3 %; p = 0.43). Overall, the FENO group used 11% more inhaled corticosteroid (95% CI: -17% to 42%; p = 0.40), although the final daily dose of inhaled corticosteroid was lower in the FENO group (557 versus 895 microg; mean difference, 338 microg; 95% CI: -640 to -37; p = 0.028). The authors concluded that an asthma treatment strategy based on the measurement of FENO did not result in a large reduction in asthma exacerbations or in the total amount of inhaled corticosteroid therapy used over a 12-month period, when compared with current asthma guidelines.

Travers and associates (2007) attempted to develop reference ranges for FENO and ascertained which factors in health and disease influence FENO levels. Subjects aged between 25 and 75 years were drawn from a random sample of the predominantly white population of Wellington, New Zealand. Exhaled NO was measured using an online NO monitor in accordance with international guidelines. A detailed respiratory questionnaire and pulmonary function tests were performed.
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The geometric mean FENO was 17.9 parts per billion (ppb) with a 90% CI for an individual prediction (reference range) for normal subjects of 7.8 to 41.1 ppb. Sex, atopy, and smoking status significantly affected FENO levels, and several reference ranges were presented adjusting for these factors. Asthma and allergic rhinitis were associated with higher FENO. Measurement of FENO had poor discriminant ability to identify steroid-naive subjects with asthma.

Ramser and colleagues (2008) evaluated FENO as a surrogate for bronchial hyper-responsiveness (BHR) in children with possible reactive airway disease. Exhaled NO was measured using the single-breath method in 169 successive outpatients 11 +/- 5 years of age before lung function testing and subsequent bronchial provocation by exercise (n = 165) and methacholine (n = 134). Baseline forced expiratory volume in 1 second (FEV1) less than 80% of predicted and/or BHR were seen in 59%. Exhaled NO correlated weakly with PD(20) to methacholine (r = -0.24, p < 0.05), but not with the change in FEV1 due to exercise-induced bronchoconstriction (EIB) (r = 0.1, p > 0.05). The negative predictive value of FENO less than 10 ppb for EIB was 94%, but overall accuracy for predicting BHR was low. The authors concluded that measurement of FENO is not a substitute for bronchial provocation in children.

A study by Szefler et al (2008) found that measurement of FENO did not improve asthma outcomes. In a randomized clinical trial, 546 adolescents and young adults with persistent or uncontrolled asthma were assigned to receive computer-protocol-driven care, with or without measurement of FENO. During the 46-week study, patients returned for visits every 6 to 8 weeks, and their medicines were adjusted as required. Mean numbers of days with asthma symptoms during the 2 weeks before each visit was similar in the NO and control groups (1.93 versus 1.89). No differences between groups were noted in secondary outcomes, including measures of pulmonary function and asthma exacerbations.

Guidelines from the National Asthma Education Program (NIH, 2007) state that NO expired gas determination is among several biomarkers that may potentially be used for asthma management in the future. The guidelines note that fractional FENO concentration is one of many biomarkers that have been proposed. "Few studies, however, have validated or 'anchored' assessment of these markers by analyzing their relationship to the rate of adverse events or decline in pulmonary function over time. Further complicating the matter is that the relationship between normalization of a biomarker and normalization of risk of an adverse event may depend on the specific treatment given. What is found true for treatment with an [inhaled corticosteroid] ICS may not be true for treatment with a leukotriene receptor antagonist (LTRA) or an inhaled long-acting beta2-agonist (LABA), or vice versa." The guidelines concluded that, "[i]n the future, assessment of a combination of historical features and of biomarkers may allow accurate estimation of the risk of future adverse events, but it must be kept in mind that laboratory tests only indirectly estimate control of risk. In the end, only symptoms, exacerbations, and quality of life over time are the measures of asthma control."

The American Thoracic Society (ATS) guidelines on evaluation of chronic cough in gastro-esophageal reflux disease (GERD) (Irwin, 2006) stated that FENO measurements are not routinely recommended, because they do not appear to be helpful in diagnosing cough due to GERD.
Kowk et al (2009) examined the feasibility of obtaining FENO concentrations in 133 children aged 2 to 18 years, who were treated in the emergency department for acute asthma exacerbation and to examine the association between FENO concentrations and other measures of acute asthma severity. The investigators measured FENO concentrations before and 1 hour after the administration of corticosteroids and at discharge from the emergency department. Outcome measures included pulmonary index score (PIS), hospital admission, and short-term outcomes (e.g., missed days of school). The investigators reported that 68% of the subjects provided adequate breaths for FENO measurement. The investigators found no difference in the median initial FENO concentration among subjects, regardless of the severity of their acute asthma. Most subjects showed no change in their FENO concentrations from the start to the end of treatment. The investigators stated that FENO concentrations were not significantly associated with other short-term outcomes. The investigators concluded that measurement of FENO is difficult for a large proportion of children with acute asthma exacerbation, and that FENO concentration during an asthma exacerbation does not correlate with other measures of acute severity and has limited utility in the emergency department management of acute asthma in children.

de Jongste et al (2009) found no added value of daily FENO monitoring with symptom monitoring versus symptom monitoring only. Children with atopic asthma (n = 151) were randomly assigned to 2 groups: (i) FENO monitoring plus symptom monitoring, or (ii) monitoring of symptoms only. All patients scored asthma symptoms in an electronic diary over 30 weeks; 77 received a portable NO analyzer. Data were transmitted daily to the coordinating centers. Patients were phoned every 3 weeks and their steroid dose was adapted according to FENO and symptoms, or according to symptoms. Children were seen at 3, 12, 21, and 30 weeks for examination and lung function testing. The primary end point was the proportion of symptom-free days in the last 12 study weeks. The investigators reported that both groups showed an increase in symptom-free days, improvement of FEV1 and quality of life, and a reduction in steroid dose. None of the changes from baseline differed between groups.

A randomized clinical study of step-up therapy in children (n = 182) with uncontrolled asthma despite corticosteroids (Lemanske et al, 2010) found that patterns of differential response were not predicted by the fraction of FENO, either dichotomized at the median baseline value or examined as a continuous covariate. The authors stated that more expensive and labor-intensive measures of physiological factors (e.g., methacholine PC20) and biomarkers (e.g., the fraction of ENO) did not have predictive value.

Powell et al (2011) concluded that asthma exacerbations during pregnancy can be significantly reduced with a validated FENO-based treatment algorithm. The investigators reported on a double-blind, parallel-group, controlled trial in two antenatal clinics in Australia. The investigators randomly assigned 220 pregnant, non-smoking women with asthma to treatment adjustment at monthly visits by an algorithm using clinical symptoms (control group) or FENO concentrations (active intervention group). The primary outcome was total asthma exacerbations. The investigators reported that the exacerbation rate was lower in the FENO group than in the control group (0.288 versus 0.615 exacerbations per pregnancy; incidence
rate ratio 0.496, 95% CI 0.325-0.755; p=0.001). In the FENO group, quality of life was improved (score on short form 12 mental summary was 56-9 [95% CI 50.2-59.3] in FENO group versus 54.2 [46.1-57.6] in control group; p=0.037) and neonatal hospitalizations were reduced (eight [8%] versus 18 [17%]; p=0.046). A limitation of this study is that the control group's algorithm differed from current guidelines and that significantly more women in the FENO group were receiving corticosteroids than in the control group.

Using a similar treatment algorithm, Pike et al (2012) concluded that FENO-guided inhaled corticosteroid titration did not reduce corticosteroid usage or exacerbation frequency in children with moderate to severe asthma. The investigators conducted a randomized controlled clinical trial to evaluate whether monitoring FENO can improve outpatient management of children aged 6 to 17 years with moderate to severe asthma. Ninety children were randomized to FENO-driven therapy or to a standard management group where therapy was driven by conventional markers of asthma control. Inhaled corticosteroids or long-acting bronchodilator therapies were altered according to FENO levels in combination with reported symptoms in the FENO group. Subjects were assessed twice monthly for 12 months. Inhaled corticosteroid dose and exacerbation frequency change were compared between groups in an intention to treat analysis. The investigators reported that no difference was found between the two groups in either change in corticosteroid dose or exacerbation frequency. The investigators stated that results were similar in a planned secondary analysis of atopic asthmatics.

Calhoun et al (2012) reported the results of the use of FENO in adults from the Best Adjustment Strategy for Asthma in the Long Term (BASALT) trial, a randomized controlled clinical trial conducted by the Asthma Clinical Research Network at 10 academic medical centers in the United States. The investigators found that, among adults with mild to moderate persistent asthma controlled with low-dose inhaled corticosteroid therapy, the use of either FENO-based or symptom-based adjustment of inhaled corticosteroids was not superior to physician assessment-based adjustment of inhaled corticosteroids in time to treatment failure. The investigators reported on the results of a randomized, parallel, 3-group, placebo-controlled, multiply-blinded trial of 342 adults with mild to moderate asthma controlled by low-dose inhaled corticosteroid therapy who were assigned to physician assessment-based adjustment, FENO-based adjustment, and symptom-based adjustment. For physician assessment-based adjustment and FENO-based adjustment, the dose of inhaled corticosteroids was adjusted every 6 weeks; for symptom-based adjustment, inhaled corticosteroids were taken with each albuterol rescue use. The primary outcome was time to treatment failure. The investigators reported that there were no significant differences in time to treatment failure. The 9-month Kaplan-Meier failure rates were 22% (97.5% CI, 14%-33%; 24 events) for physician assessment-based adjustment, 20% (97.5% CI, 13%-30%; 21 events) for FENO-based adjustment, and 15% (97.5% CI, 9%-25%; 16 events) for symptom-based adjustment. The hazard ratio for physician assessment-based adjustment versus FENO-based adjustment was 1.2 (97.5% CI, 0.6-2.3). The hazard ratio for physician assessment-based adjustment versus symptom-based adjustment was 1.6 (97.5% CI, 0.8-3.3).
An editorial accompanying the BASALT trial (O’Connor and Reibman, 2012) concluded that dose adjustment based on exhaled nitric oxide measurements has not been shown to improve outcomes in routine asthma management. The editorialist commented that the result of the BASALT trial is consistent with prior evidence that routine exhaled nitric oxide monitoring is not warranted for managing most patients with asthma. The editorialist noted that the recent ATS practice guideline recommends the use of exhaled nitric oxide measurement “in monitoring airway inflammation in patients with asthma (strong recommendation, low quality of evidence),” but that, in light of the BASALT findings, it is difficult to justify additional health care expenditures for routinely monitoring exhaled nitric oxide in adults with mild to moderate asthma. The editorialist noted that there may be a role for exhaled nitric oxide measurement, however, when the diagnosis of asthma is not clear or for specific patient subgroups, but that further research is needed to identify the clinical scenarios in which exhaled nitric oxide measurement may improve clinical outcomes.

Lester et al (2012) reported on the use of a comprehensive asthma management program by an urban community health center. The program included serial FeNO measurements among several program components. Other components of the program were: use of asthma management guidelines; use of a team approach to asthma management; use of a standardized tool for screening for asthma risk factors, symptoms, and level of asthma control; use of a workflow algorithm and chart form incorporated into an electronic health record to collect data and track clinical measures for an asthma registry; use of asthma health educators to assist patients in setting asthma self-management goals and educate them in asthma self-management; use of community resources (visiting nurses, a state-funded pest control program, and durable medical equipment vendors for products such as aerochambers and nebulizers); and regular followup, with frequency based upon asthma severity. The authors reported that 95.8 percent of patients enrolled in the program had an asthma severity assessment, 95.4 percent of persistent asthmatics were on anti-inflammatory medications, 68.2 percent of asthma patients have documented asthma self-management goals, and 5.2 percent of asthma patients have a self-reported visit to the emergency room in the six months preceding their most recent visit. Because the program includes multiple components, the contribution of FeNO measurements to these outcomes cannot be determined.

Guidelines from the British Thoracic Society and the Scottish Intercollegiate Guidelines Network (2008) stated that "more research needs to be done before recommendations can be made for the use of exhaled nitric oxide concentration". Updated guidelines from the Scottish Intercollegiate Guidelines Network (2009) stated that more experience with FENO and more information on the long-term response to corticosteroid in patients who do not have a raised FENO is needed before this approach can be recommended to identify patients who are going to respond to corticosteroid therapy. Updated guidelines from the British Thoracic Society and the Scottish Intercollegiate Guidelines Network (2012) stated that studies in children have shown that routine serial measurements of exhaled nitric oxide do not provide additional benefit when added to a symptom based management strategy. The guidelines (2012) stated that a better understanding of the
natural variability of biomarkers independent of asthma is required and studies are needed to establish whether subgroups of patients can be identified in which biomarker guided management is effective.

Puckett and George (2008) stated that the airway NO flux and alveolar NO concentration can be elevated in adults and children with asthma and have been correlated with markers of airway inflammation and airflow obstruction in cross-sectional studies. They noted that longitudinal studies that specifically address the clinical potential of partitioning FENO for diagnosis, managing therapy, and predicting exacerbation are needed.

In a review on FENO in the diagnosis and management of asthma, Rodway and colleagues (2009) stated that further evidence of the clinical utility of FENO in asthma management is needed. The authors stated that,"[r]egardless of the rapid, convenient, and noninvasive nature of this test, additional well-designed, long-term longitudinal studies are necessary to fully evaluate the clinical utility of eNO in asthma management".

In a review on the utility of FENO in the diagnosis and management of asthma, Majid and Kao (2010) noted that "FeNO shows promise as a tool in the diagnosis and treatment of asthma. However, further studies are needed to address outstanding questions about its exact role in guiding asthma management".

Guidelines from the Global Initiative for Asthma (GINA, 2012) state that levels of FENO and carbon monoxide have been suggested as non-invasive markers of airway inflammation in asthma. The guidelines state that levels of FENO are elevated in people with asthma (who are not taking inhaled glucocorticosteroids) compared to people without asthma, yet these findings are not specific for asthma. The guidelines note the FENO has not been evaluated prospectively as an aid in asthma diagnosis, but these measurements are being evaluated for potential use in determining optimal treatment, "although it has been shown that the use of FeNO as a measure of asthma control does not improve control or enable reduction in dose of inhaled glucocorticosteroid." The Global Initiative for Asthma was launched in conjunction with the National Heart Lung and Blood Institute and the World Health Organization.

A Cochrane review evaluated clinical studies of the efficacy of tailoring asthma interventions based on FENO in comparison to clinical symptoms (with or without spirometry/peak flow) for asthma related outcomes in children and adults (Petsky et al, 2009). The assessment found that tailoring the dose of inhaled corticosteroids based on FENO in comparison to clinical symptoms was carried out in different ways in the 6 randomized controlled clinical studies that were included in the review, and the results showed only modest differences at best and potentially higher doses of inhaled corticosteroids in children. The authors of the Cochrane review concluded that the role of utilizing FENO to tailor the dose of inhaled corticosteroids is currently uncertain and can not be routinely recommended for clinical practice. In this Cochrane review, Petsky et al (2009) evaluated the effectiveness of tailoring asthma interventions based on FENO in comparison to clinical symptoms (with or without spirometry/peak flow) for asthma related outcomes in children and adults. These investigators searched the Cochrane Airways Group Specialised Register of Trials, the Cochrane Central Register of Controlled Trials (CENTRAL), MEDLINE, EMBASE and reference lists of articles.
The last search was completed in February 2009. All randomized controlled comparisons of adjustment of asthma therapy based on FENO compared to traditional methods (primarily clinical symptoms and spirometry/peak flow) were selected for analysis. Results of searches were reviewed against pre-determined criteria for inclusion. Relevant studies were independently selected in duplicate. Two authors independently assessed trial quality and extracted data. Two studies have been added for this update, which included 6 (2 adults and 4 children/adolescent) studies; these studies differed in a variety of ways including definition of asthma exacerbations, FENO cut-off levels, the way in which FENO was used to adjust therapy and duration of study. Of 1,053 randomized subjects, 1,010 completed the trials. In the meta-analysis, there was no significant difference between groups for the primary outcome of asthma exacerbations or for other outcomes (clinical symptoms, FENO level and spirometry). In post-hoc analysis, a significant reduction in mean final daily dose inhaled corticosteroid per adult was found in the group where treatment was based on FENO in comparison to clinical symptoms, (mean difference of -450 mcg; 95 % CI: -677 to -223 mcg budesonide equivalent per day). However, the total amount of inhaled corticosteroid used in one of the adult studies was 11 % greater in the FENO arm. In contrast, in the pediatric studies, there was a significant increase in inhaled corticosteroid dose in the FENO strategy arm (mean difference of 140 mcg; 95 % CI: 29 to 251 mcg budesonide equivalent/day). The authors concluded that tailoring the dose of inhaled corticosteroids based on FENO in comparison to clinical symptoms was carried out in different ways in the 6 studies and found only modest benefit at best and potentially higher doses of inhaled corticosteroids in children. The authors stated that the role of utilizing FENO to tailor the dose of inhaled corticosteroids can not be routinely recommended for clinical practice at this stage and remains uncertain.

Donohue and Jain (2013) evaluated the evidence for FeNO as a predictor of corticosteroid responsive airway inflammation. The authors conducted a meta-analysis of three adult studies comparing asthma exacerbation rates with FeNO-based versus clinically based asthma management algorithms, including one study that was not included in the Cochrane metaanalysis. The authors reported that the results indicate that the rate of asthma exacerbations was significantly reduced in favor of FeNO-based asthma management (mean treatment difference - 0.27; 95% CI [0.42,0.12] as was the relative rate of asthma exacerbations (relative rate - 0.57; 95% CI [0.41, 0.80]).

An ATS ad hoc committee was organized by the committee chairman to devise “interpretive strategies” for the different potential uses and applications of FENO (Dweik et al, 2011). The committee identified a number of potential uses of FENO, which were graded by the committee members on the strength of the recommendation and strength of the evidence. Several potential uses of FENO were identified, each of which were related to the use of FENO to identify steroid responsiveness. However, there is a lack of reliable evidence demonstrating that clinical outcomes are improved by using FENO to identify steroid responsiveness and direct patient care in persons with asthma. One of the recommendations, identification of eosinophilic airway inflammation, was judged by the Committee to be supported by “strong consensus” and “moderate quality evidence.” The report stated that the importance of identifying eosinophilic airway inflammation rests on its correlation with steroid responsiveness: “the finding that FENO correlates with
eosinophilic inflammation suggests its use as indirect indicator not only of eosinophilic inflammation, but more importantly of the potential for steroid responsiveness." However, the evidence cited to support the recommendation found widely varying correlations between FENO and other recognized measures of eosinophilic airway inflammation (biopsy, sputum, and bronchiolar lavage). Another potential use recommended by the Committee, the use of FENO to support the diagnosis of asthma in situations in which objective evidence is needed, was a "weak recommendation" based upon "moderate quality evidence." The report explained that "the importance of FENO lies in its potential to identify steroid responsiveness, rather than the exact clinical diagnosis." Other potential uses of FENO identified by the Committee were judged to have "low quality evidence", including use of FENO to determine the likelihood of steroid responsiveness in individuals with chronic respiratory symptoms possibly due to airway inflammation, and use of FENO in monitoring airway inflammation in patients with asthma. Also, the summary of this ATS position paper (Dweik et al, 2011) stated that "the use of exhaled nitric oxide levels (FENO) in COPD and pulmonary hypertension and the use of nasal NO in diagnosis and monitoring of other respiratory disorders (e.g., allergic rhinitis, sinusitis, nasal polyposis, CF) are potentially of interest, but more research is needed before we know how clinically useful these tests can be for these disorders".

A systematic evidence review of the literature on the usefulness of exhaled nitric oxide in childhood asthma conducted by the Andalusian Agency for Health Technology Assessment (AETSA) (García Estepa, et al., 2011) identified four clinical trials, one systematic evidence review and two health technology assessment reports that met inclusion criteria. The review found that the selected trials, with the exception of one, had some methodological weaknesses, as did the health technology assessment reports. However, the systematic evidence review, despite several limitations, was of high methodological quality. The AETSA review found that the selected studies do not provide significant correlations between FENO levels and clinically relevant outcomes such as optimal therapy, reduction of inhaled corticosteroid doses or more appropriate drug combinations, reduced exacerbations, or decrease in symptoms. Furthermore, in the variety of secondary outcomes in each study, significant differences were detected only in some of them, from which it might be concluded that the usefulness of FENO levels for the control of childhood asthma has not been demonstrated. The authors of the systematic evidence review concluded that the clinical validity of using FENO levels to control childhood asthma has not been conclusively established. The report concluded that, according to available evidence, the use of the determination of FENO levels does not improve important outcomes in asthma such as: reduction of symptoms and prevention of crisis or exacerbations, improved lung function and reduction or better management of inhaled corticosteroid treatment, compared to the usual practice, based on symptoms with or without spirometry. The authors stated that the studies analyzed did not demonstrate the clinical usefulness of determining FENO levels in the control of childhood asthma.

Jartti et al (2012) noted that FENO has gained interest as a non-invasive tool to measure airway inflammation in asthma since it reflects allergic inflammation. The authors stated that recent controlled clinical studies have, however, questioned its role in the management of asthma in children. To assess the clinical value of FENO in pediatric asthma management, the authors performed a meta-analysis on
the controlled studies of childhood asthma management guided by repeated FENO measurements, and relevant publications on the confounders of FENO were reviewed. The authors concluded that the data suggested that utilizing FENO to tailor the dose of inhaled corticosteroids in children can not be recommended for routine clinical practice since there is a danger of excessive inhaled corticosteroid doses in children without meaningful changes in clinical outcomes. Many disease and non-disease related factors (most importantly atopy, height/age and infection) affect FENO levels that can easily confound the interpretation.

Petsky et al (2012) evaluated the efficacy of tailoring asthma interventions based on inflammatory markers (sputum analysis and FENO) in comparison with clinical symptoms (with or without spirometry/peak flow) for asthma-related outcomes in children and adults. Cochrane Airways Group Specialised Register of Trials, the Cochrane Central Register of Controlled Trials (CENTRAL), MEDLINE, EMBASE and reference lists of articles were searched. The last searches were in February 2009. All randomized controlled comparisons of adjustment of asthma treatment based on sputum analysis or FENO compared with traditional methods (primarily clinical symptoms and spirometry/peak flow) were selected. Results of searches were reviewed against pre-determined criteria for inclusion. Relevant studies were selected, assessed and data extracted independently by at least 2 people. The trial authors were contacted for further information. Data were analysed as "intervention received" and sensitivity analyses performed. Six (2 adults and 4 children/adolescent) studies utilising FENO and 3 adult studies utilizing sputum eosinophils were included. These studies had a degree of clinical heterogeneity including definition of asthma exacerbations, duration of study and variations in cut-off levels for percentage of sputum eosinophils and FENO to alter management in each study. Adults who had treatment adjusted according to sputum eosinophils had a reduced number of exacerbations compared with the control group (52 versus 77 patients with greater than or equal to 1 exacerbation in the study period; p = 0.0006). There was no significant difference in exacerbations between groups for FENO compared with controls. The daily dose of inhaled corticosteroids at the end of the study was decreased in adults whose treatment was based on FENO in comparison with the control group (mean difference -450.03 μg, 95% CI: -676.73 to -223.34; p < 0.0001). However, children who had treatment adjusted according to FENO had an increase in their mean daily dose of inhaled corticosteroids (mean difference 140.18 μg, 95% CI: 28.94 to 251.42; p = 0.014). It was concluded that tailoring of asthma treatment based on sputum eosinophils is effective in decreasing asthma exacerbations. However, tailoring of asthma treatment based on FENO levels has not been shown to be effective in improving asthma outcomes in children and adults. The authors concluded that, at present, there is insufficient justification to advocate the routine use of either sputum analysis (due to technical expertise required) or FENO in everyday clinical practice.

Malby Schoos et al (2012) stated that elevated FENO and bronchial hyper-responsiveness are used as surrogate markers of asthma. These traits may be continuous in the population. The objective of this study was to investigate whether FENO and bronchial responsiveness are associated in both children with and children without a history of asthma symptoms. A total of 196 children (6-year old) with no asthma symptoms, intermittent asthma symptoms, and persistent asthma were randomly included from the Copenhagen Prospective Study on Asthma in Childhood prospective clinical birth cohort of mothers with asthma. Bronchial
Responsiveness was assessed as the relative change in specific airway resistance after cold dry air hyperventilation. FENO measurements were performed prior to the hyperventilation test. The association between FENO and bronchial responsiveness was assessed by generalized linear models. Bronchial responsiveness and FENO exhibited a significant and linear association in the population. A doubling of FENO corresponded to an 8.4% (95% CI: 3.7% to 13.1%; p = 0.0006) increase in airway resistance after challenge testing and remained significant after adjustment for sex, allergic rhinitis, current asthma, inhaled corticosteroid treatment, and upper respiratory tract infections prior to testing. Stratified analyses showed similar associations in children with and without asthma. The authors concluded that FENO and bronchial responsiveness are associated and continuous traits in young children regardless of asthma symptoms, suggesting a continuous subclinical to clinical process underlying asthma. The authors stated that these findings also suggested caution against the use of these surrogate markers for a dichotomized approach to asthma diagnosis.

Gelb and colleagues (2012) noted that the up-regulation of NO by inflammatory cytokines and mediators in central and peripheral airway sites can be easily monitored in exhaled air FENO. It is now possible to estimate the predominant airway site of increased FENO, i.e., large versus peripheral airway/alveoli, and its potential pathologic and physiologic role in obstructive lung disease. In asthma, 6 double-blind, randomized, controlled algorithm trials have reported only equivocal benefits of add-on measurements of FENO to usual clinical guideline management including spirometry. Significant design issues, as emphasized by Gibson, may exist. However, meta-analysis of these 6 studies (Petsky et al, 2012) concluded that routine serial measurements of FENO for clinical asthma management does not appear warranted. In COPD including chronic bronchitis and emphysema, despite significant expiratory airflow limitation, when clinically stable as well as during exacerbation, FENO, \( \text{j(NO)} \) and C(ANO) may all be normal or increased. Furthermore, the role of add-on monitoring of exhaled NO to GOLD management guidelines is less clear because of the absence of conclusive double-blind, randomized, control trial studies concerning potential clinical benefits in the management of COPD.

Guidelines from the Canadian Thoracic Society (Lougheed et al, 2012) concluded: "there is still insufficient evidence to recommend the use of FeNO to tailor the dose of ICS compared with titrating ICS dose based on clinical symptoms alone. As such, the routine use of FeNO measurements as a guide to tailor the dose of ICS in asthma cannot be endorsed for clinical practice at this time."

A consultation document from the National Institute for Health and Clinical Excellence (2013) states that fractional exhaled nitric oxide (FeNO) testing is recommended as an option to help with diagnosing asthma and as an option to support asthma management in people who are symptomatic despite using inhaled corticosteroids.

Guidelines on work-related asthma from the European Respiratory Society (Bauer et al, 2012) state that "in the clinical setting, a finding of a normal exhaled nitric oxide fraction cannot be used to exclude occupational asthma." The guideline explained that exhaled nitric oxide is not sufficiently sensitive to be used to exclude occupational asthma suggested by history. Guidelines from the British
Occupational Health Research Foundation (Nicholson, et al., 2010) concluded that the measurement of exhaled nitric oxide "has not been fully validated as an effective diagnostic test for occupational asthma." The guidelines explain that exhaled nitric oxide is increased in other inflammatory lung disorders, and levels are lower in persons who smoke and in those using inhaled corticosteroids.

An UpToDate review on “Exhaled nitric oxide analysis” (Deykin and Massaro, 2013) states that “In the future, measurement of exhaled nitric oxide (eNO), a simple, noninvasive test, could be used to aid in the diagnosis of asthma and potentially monitor the inflammatory status of the asthmatic airway during treatment with antiinflammatory medications. Although it is possible that therapeutic agents could be titrated to the lowest doses that suppress elevated levels of eNO, the role of eNO in the ongoing care of patients with established asthma is uncertain. Moreover, due to the expense of the chemiluminescence NO analyzer, measurement of eNO is largely restricted to research hospitals and large medical centers at the present time”.

See and Christiani (2013) stated that elevated FENO reflects airway inflammation, but few studies have established its normal values. This study aimed to establish the normal values and thresholds for the clinical interpretation of FENO in the U.S. general population. A total of 13,275 subjects aged 6 to 80 years sampled for the National Health and Nutrition Examination Survey (NHANES) 2007-2010 underwent interviews, physical examination, and FENO analysis at 50 ml/s using an online chemiluminescence device according to ATS/European Respiratory Society (ERS) guidelines. After excluding subjects with self-reported asthma and subjects with wheeze in the prior 12 months, prediction equations for the natural logarithm (ln) of FENO were constructed using age, sex, ethnicity, height, body mass index (BMI), active/passive smoke exposure, and hay fever episodes as covariates. The 5th to 95th percentile values of FENO were 3.5 to 36.5 ppb for children less than 12 years of age and 3.5 to 39 ppb for subjects 12 to 80 years of age. Using multiple linear regression, prediction equations explained only 10.3 % to 15.7 % of the variation in the general population. In the general population, 39 % to 45 % had ln(FENO) levels greater than 2 standard deviations of the predicted means. When applied to the general population inclusive of subjects who reported asthma but who did not have attacks within the past year, nearly identical results were obtained. The authors concluded that assuming 95 % of the healthy U.S. general population had no clinically significant airway inflammation as assessed by FENO, values exceeding the 95th percentiles indicated abnormality and a high-risk of airway inflammation. A large variation of normal FENO values existed in the general population, which was poorly predicted by multiple linear regression models.

Columbo et al (2013) studied the role of serial measurements of FENO in elderly subjects with asthma. A total of 30 stable asthematics 65 years old and older were followed for 1 year with evaluations at baseline and every 3 months. These researchers looked for associations between FENO and subjects' demographics, co-morbidities, asthma treatment, spirometric values and asthma control test (ACT) scores. Fractional exhaled nitric oxide was not elevated in these subjects throughout the study period (mean less than 30 ppb); FENO significantly increased and FEV1 % decreased between first and last study visit, while ACT scores and steroid dose remained unchanged. No significant correlation was found between
FENO and FEV1/FVC, other spirometric values, inhaled steroid dose or ACT scores at any time point. No associations of FENO were found with age, sex, BMI, atopic status, disease duration, presence of rhinitis or GERD, or other medications used. Moderate asthma exacerbations did not consistently cause an increase of FENO. The authors concluded that in stable elderly asthmatic patients, FENO was not elevated and did not correlate with subjects' demographics, co-morbidities, treatment, symptoms or spirometric values. They stated that routine measurements of FENO may not be clinically valuable in elderly asthmatics.

Hanania et al (2013) assessed the potential of FENO, peripheral blood eosinophil count, and serum periostin as biomarkers of Th2 inflammation and predictors of treatment effects of omalizumab. The EXTRA omalizumab study enrolled patients (aged 12 to 75 years) with uncontrolled severe persistent allergic asthma. Analyses were performed evaluating treatment effects in relation to FENO, blood eosinophils, and serum periostin at baseline. Patients were divided into low- and high-biomarker subgroups. Treatment effects were evaluated as number of protocol-defined asthma exacerbations during the 48-week treatment period (primary endpoint). A total of 850 patients were enrolled. Data were available from 394 (46.4 %), 797 (93.8 %), and 534 (62.8 %) patients for FENO, blood eosinophils, and serum periostin, respectively. After 48 weeks of omalizumab, reductions in protocol-defined exacerbations were greater in high versus low subgroups for all 3 biomarkers: FENO, 53 % (95 % CI: 37 to 70; p = 0.001) versus 16 % (95 % CI: -32 to 46; p = 0.45); eosinophils, 32 % (95 % CI: 11 to 48; p = 0.005) versus 9 % (95 % CI: -24 to 34; p = 0.54); and periostin, 30 % (95 % CI: -2 to 51; p = 0.07) versus 3 % (95 % CI: -43 to 32; p = 0.94). The authors concluded that the difference in exacerbation frequency between omalizumab and placebo was greatest in the 3 high-biomarker subgroups, probably associated with the greater risk for exacerbations in high subgroups. Moreover, they stated that additional studies are needed to explore the value of these biomarkers in clinical practice.

In order to evaluate the clinical usefulness of FENO assessment for monitoring asthma during pregnancy, Nittner-Marszalska et al (2013) monitored 72 pregnant asthmatics who underwent monthly investigations including: the level of asthma control according to Global Initiative for Asthma (GINA), the occurrence of exacerbations, Asthma Control Test (ACT), as well as FENO and spirometry measurements. In 50 women, during all visits, asthma was well-controlled. In the remaining 22 women, asthma was periodically uncontrolled. FENO measured at the beginning of the study did not show significant correlation with retrospectively evaluated asthma severity (r=0.07; p=0.97). An analysis of data collected during all 254 visits showed that FENO correlated significantly but weakly with ACT scores (r=0.25; p=0.0004) and FEV1 (r=0.21; p=0.0014). FENO at consecutive visits in women with well-controlled asthma (N=50) showed large variability expressed by median coefficient of variation (CV)=32.0 % (Min 2.4 %, Max 121.9 %). This concerned both: atopic and nonatopic groups (35.5 %; and 26.7 %, respectively). Large FENO variability (35.5 %) was also found in a subgroup of women (N=11) with ACT=25 constantly throughout the study. FENO measured at visits when women temporarily lost control of asthma (N=22; 38 visits), showed an increasing tendency (64.2ppb; 9.5ppb-188.3ppb), but did not differ significantly (p=0.13) from measurements taken at visits during which asthma was well-controlled (27.6ppb; 6.2ppb-103.4ppb). The authors observed that the comparison of FENO in
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consecutive months of pregnancy in women who had well-controlled asthma did not show significant differences in FENO values during the time of observation. The authors concluded that assessment of asthma during pregnancy by means of monitoring FENO is of limited practical value due to this parameter's considerable intrasubject variability, regardless of the degree of asthma control.

Piersman et al (2013) reported that FeNO measurements in childhood asthma management did not improve the proportion of symptom-free days, but did result in fewer asthma exacerbations associated with an increased leukotriene receptor antagonist use and an augmentation of the inhaled corticosteroid doses. The authors investigated the potential yield of incorporating FeNO measurements in childhood allergic asthma management. Ninety-nine children with persistent allergic asthma were included in this multicentre, single-blind, randomized controlled trial. Treatment was based on the Global Initiative for Asthma (GINA) guidelines. In the FeNO group, asthma management was also guided by FeNO measurements. Health outcomes were evaluated over a 52-week timeframe. Results: Fewer asthma exacerbations were registered in the FeNO group. 24% of the children in the FeNO group experienced one or more exacerbations per year, compared with 48% in the clinical group (P = 0.017). The proportion of symptom-free days did not differ between groups. In the FeNO group, more months of leukotriene receptor antagonist use (median [interquartile range]) were observed: 12 (9–12) months, compared with 9 (3–12) months in the clinical group (P = 0.019). The evolution of inhaled corticosteroid doses between visits 1 and 5 (median change [interquartile range]) showed a significant increase of -100 micrograms (0, -400) in the FeNO group and a change of 0 mg (+200, -80) in the clinical group (p = 0.016).

Syk et al (2013) reported on the results of a study where 187 patients with asthma and who were nonsmokers (age range of 18 to 64 years) with perennial allergy and who were on regular inhaled corticosteroid treatment were recruited at 17 primary health care centers, randomly assigned to 2 groups and followed up for 1 year. For the controls (n = 88), FENO measurement was blinded to both operator and patient, and anti-inflammatory treatment was adjusted according to usual care. In the active group (n = 93), treatment was adjusted according to FENO. Patients in both groups were not allowed to use long-acting beta agonists. Questionnaires on asthma-related quality of life (Mini Asthma Quality of Life Questionnaire) and asthma control (Asthma Control Questionnaire) were completed, and asthma events were noted. The Asthma Control Questionnaire score change over 1 year improved statistically significantly more in the FENO-guided group (0.17 [interquartile range (IQR), 0.67 to 0.17] versus 0 [IQR, 0.33 to 0.50]; p < 0.045), although of questionable clinical significance. The Mini Asthma Quality of Life Questionnaire score did not improve significantly more in the FENO-guided group (0.23 [IQR, 0.07 to 0.73] versus 0.07 [IQR, 0.20 to 0.80]; p < 0.197). The change in Asthma Control Questionnaire was clinically important in subpopulations with poor control at baseline (p < 0.03). Furthermore, the exacerbation rate (exacerbations/patient/y) was reduced by almost 50% in the FENO-guided group (0.22 [CI: 0.14 to 0.34] versus 0.41 [CI: 0.29 to 0.58]; p < 0.024). Mean overall inhaled corticosteroid use was similar in both groups (p < 0.95). Limitations of the study include that the control group was provided "usual care" and not assigned to best available guideline supported care, whereas patients assigned to the active treatment group was managed by protocol. Treatments deviated from current guideline supported care in that long-acting beta-agonists were not allowed to be
used. Other limitations of the study included the lack of participant personnel
blinding, incomplete outcome data and selective reporting.

McCormack et al (2013) noted that little is known about use of FENO to predict
asthma exacerbations among high-risk, urban, minority populations receiving usual
care. A total of 138 children with persistent asthma were enrolled in a prospective
observational cohort study and skin tested at baseline (wheal greater than or equal
to 3 mm = +SPT). Fractional exhaled nitric oxide levels, lung function, and asthma-
related health care use were assessed at baseline and every 3 months thereafter
for 1 year. Relationships between FENO and health care utilization in the
subsequent 3 months were examined. Final models accounted for repeated
outcome measures and were adjusted for age, gender and lung function. The
mean age was 11 years (range of 5 to 17), and most were male (57 %), African
American (91 %), and atopic (90 %). At baseline, FENO was (median IQR: 31.5
ppb [16 to 61]) and FEV1/FVC was (mean +/- SD: 80.7 +/- 9.6 %). There were 237
acute asthma-related health care visits, 105 unscheduled doctor (UD) visits, 125
ED visits, and 7 hospitalizations during the follow-up period. Fractional exhaled
nitric oxide was not a significant predictor of acute visits, ED visits, UD visits, or
hospitalization in either unadjusted or adjusted analyses. Use of recommended cut-
-off points did not improve the predictive value of FENO (positive-predictive value
[PPV]: 0.6 to 32.8 %), nor did application of the guideline-based algorithm to
assess change over time. The authors concluded that FENO may not be a
clinically useful predictor of health care use for asthma exacerbations in urban
minority children with asthma.

A trial by Petsky, et al. (2014) found that taking atopy into account when using
FeNO to tailor asthma medications is likely beneficial in reducing the number of
children with severe exacerbations at the expense of increased inhaled
corticosteroid use. However, the investigators concluded that this strategy is
unlikely beneficial for improving asthma control. The investigators stated that
controversy remains of the benefits of use of FeNO for routine asthma
management, and the appropriate FeNO cut-off remains elusive. The investigators
noted that there are four published randomized controlled trials that have evaluated
the benefit of an FeNO-based strategy in adjusting medications for asthma in
children, and none of these had shown any significant benefit of using FeNO
compared to controls with respect to exacerbations, but none of the studies used
exacerbation as their primary end-point. The authors noted that adult-based studies
used exacerbations as their primary endpoint, but none showed any benefit in
reducing asthma exacerbations. The investigators conducted the first clinical trial to
evaluated the use of FeNO cut-offs adjusted for atopy to improve asthma
outcomes. In a randomized controlled clinical trial conducted at two centers, the
investigators assessed whether a treatment strategy based on FeNO levels,
adjusted for atopy, reduces asthma exacerbations compared with the symptoms-
based management (controls). Children with asthma from hospital clinics of two
hospitals were randomly allocated to receive an a-priori determined treatment
hierarchy based on symptoms or FeNO levels. There was a 2-week run-in period
and they were then reviewed 10 times over 12-months. The primary outcome was
the number of children with exacerbations over 12-months. Sixty-three children
were randomized (FeNO = 31, controls = 32); 55 (86%) completed the study. The
investigators reported that, although they did achieve their planned sample size,
significantly fewer children in the FeNO group (6 of 27) had an asthma
exacerbation compared to controls (15 of 28), p = 0.021; number to treat to prevent one child from having any exacerbation in 12 months was 4 (95% CI 3-24). There was a trend toward fewer exacerbations in the FeNO group (0.39 per person-year) compared to controls (0.78 per person-year that did not reach statistical significance. When the groups were subanalyzed into those with 2 or more exacerbations per year, they found no significant difference between the groups. At the end of the 12-month study, the total cumulative dose of inhaled corticosteroid per child-year was significantly higher in the FeNO group compared to the control group. The investigators found no difference between groups for any secondary outcomes (quality of life, symptoms, FEV1). The final daily inhaled corticosteroids (ICS) dose was significantly (p = 0.037) higher in the FeNO group (median 400 μg, IQR 250-600) compared to the controls (200, IQR 100-400). The authors concluded that a larger study is required to confirm or refute their findings.

Van Beek et al (2011) evaluated the potential usefulness of measuring FENO as a screening tool for pulmonary tuberculosis (TB). These researchers compared 90 consecutive smear-positive, culture-confirmed TB patients presenting at a referral hospital with office workers (no X-ray confirming TB) at a hospital (n = 52) and at a construction firm (n = 84). Exhaled NO levels were analysed using a validated hand-held analyser. Exhaled NO levels among TB patients (median 15 parts per billion [ppb], inter-quartile range [IQR] 10 to 20) were equal to those among construction firm workers (15 ppb, IQR 12-19, p = 0.517) but higher than those among hospital workers (8.5 ppb, IQR 5 to 12.5, p < 0.001). Taking the hospital workers as the comparison group, best performance as a diagnostic tool was at a cut-off of 10 ppb, with sensitivity 78 % (95 %CI: 68 to 86) and specificity 62 % (95 % CI: 47 to 75). Test characteristics could be optimized to 84 % versus 67 % by excluding individuals who had recently smoked or consumed alcohol. The authors concluded that while FENO measurement has limited value in the direct diagnosis of pulmonary TB, it may be worth developing and evaluating as a cost-effective replacement of chest X-ray in screening algorithms of pulmonary TB where X-ray is not available.

Phillips et al (2011) reviewed the data available on the sino-nasal application of nasal NO measurement, particularly its use as a diagnostic, prognostic, or treatment effect indicator. EMBASE 1980 to February 10, 2010; Medline 1950 to February 10, 2010; Cochrane Collaboration database; NHS Evidence Health Information Resources database were searched using a search strategy designed to include manuscripts relevant both to NO measurement and sinus or nasal problems. A title search was performed on these manuscripts to select those relevant to clinical or basic science aspects of NO measurement. A subsequent abstract search selected those manuscripts concerning the application of NO measurement to sino-nasal problems. The manuscripts selected were subject to a full-text review to extract data sets of nasal NO readings for different patient groups. Initially, 1,088 manuscripts were selected. A title search found 335 manuscripts of basic scientific or clinical interest. An abstract search found 35 manuscripts directly relating to NO measurement in sino-nasal disease. Full-text analysis produced 20 studies with extractable data on nasal NO levels in clearly defined patient groups. Studies did not show sufficient homogeneity to enable substantial meta-analysis of aggregated data. The authors stated that the literature concerning nasal NO is marked by many theories concerning its role in the nose. However, clinical studies show a wide range of measurement methods, the
presence of various confounding factors, and heterogeneity of study populations. Although both the presence of nasal polyps and opening of the sinuses surgically seem to have an effect on nasal NO levels, there is no evidence in any population that low nasal NO causes harmful effects. They concluded that current evidence shows that nasal NO is not a clinically useful measure for sino-nasal disease.

Yoon and Sin (2011) stated that chronic obstructive pulmonary disease (COPD) is a leading cause of morbidity and mortality in the world. None of the current treatments, except for smoking cessation and supplemental domiciliary oxygen for hypoxemic patients, can modify its natural course or alter survival. The pipeline for new compounds is not very promising owing to repeated failures, and many large pharmaceutical companies have abandoned COPD drug discovery altogether. One major barrier to new drug discovery is the lack of modifiable biomarkers that can be used as surrogates of clinical outcomes such as exacerbation and mortality. The only accepted marker in COPD is FEV1. However, by definition, COPD is a non-reversible or poorly reversible condition with respect to FEV1. Thus, very few drugs except for bronchodilators have been able to address this endpoint. Of many candidate molecules, sputum neutrophil counts, FENO and proline-glycine-proline (PGP) and N-α-PGP, which are breakdown products of collagen, are promising lung-based biomarkers. However, their clinical utility has not been validated in large clinical trials. Promising blood biomarkers include surfactant protein D, and pulmonary- and activation-regulated chemokine (PARC/CCL-18). However, the clinical data have been inconsistent. Non-specific inflammatory biomarkers such as C-reactive protein and interleukin-6 lack specificity for COPD and thus are of limited clinical usefulness.

In a pilot study, Donohue, et al. (2014) identified increases in FeNO in a subset of patients with COPD. The investigators conducted a single-visit, outpatient study in 200 patients aged 40 years and older with COPD. All patients underwent spirometry and FeNO testing. COPD severity was classified according to the Global initiative for chronic Obstructive Lung Disease (GOLD) 2010 guidelines. Patients who participated in the study had a mean age of 63.9 ± 11.3 years and a mean smoking history of 46 ± 29 pack years. Patients had a mean forced expiratory volume in 1 second percent predicted of 53.9% ± 22.1%. The percentage of patients classified with COPD severity Stage I, II, III, and IV was 13%, 40%, 39%, and 8%, respectively. In addition, according to current procedural terminology codes, 32% of patients were classified as mixed COPD/asthma, 26% as COPD/emphysema, and 42% as all other codes. The mean FeNO level for all patients was 15.3 ± 17.2 parts per billion (ppb). Overall, 89% of patients had a FeNO <25 ppb, 8% had a FeNO 25-50 ppb, and 3% had a FeNO >50 ppb. The percentages of patients with FeNO in the intermediate or high ranges of FeNO were greatest among patients with mixed COPD/asthma (intermediate, 11.5%; high, 6.6%) compared with COPD/emphysema (intermediate, 8%; high, 0) and all other codes (intermediate, 6.3%; high, 1.3%).

In a review on "Biomarkers in chronic obstructive pulmonary disease", Rosenberg and Kalhan (2012) concluded that "So far, no single biomarker in COPD warrants wide acceptance emphasizing the need for future investigation of biomarkers in large-scale longitudinal studies".
See and Christiani (2013) stated that elevated FENO reflects airway inflammation, but few studies have established its normal values. This study aimed to establish the normal values and thresholds for the clinical interpretation of FENO in the U.S. general population. A total of 13,275 subjects aged 6 to 80 years sampled for the National Health and Nutrition Examination Survey (NHANES) 2007-2010 underwent interviews, physical examination, and FENO analysis at 50 ml/s using an online chemiluminescence device according to ATS/European Respiratory Society (ERS) guidelines. After excluding subjects with self-reported asthma and subjects with wheeze in the prior 12 months, prediction equations for the natural logarithm (ln) of FENO were constructed using age, sex, ethnicity, height, body mass index (BMI), active/passive smoke exposure, and hay fever episodes as co-variates. The 5th to 95th percentile values of FENO were 3.5 to 36.5 ppb for children less than 12 years of age and 3.5 to 39 ppb for subjects 12 to 80 years of age. Using multiple linear regression, prediction equations explained only 10.3 % to 15.7 % of the variation in the general population. In the general population, 39 % to 45 % had ln(FENO) levels greater than 2 standard deviations of the predicted means. When applied to the general population inclusive of subjects who reported asthma but who did not have attacks within the past year, nearly identical results were obtained. The authors concluded that assuming 95 % of the healthy U.S. general population had no clinically significant airway inflammation as assessed by FENO, values exceeding the 95th percentiles indicated abnormality and a high-risk of airway inflammation. A large variation of normal FENO values existed in the general population, which was poorly predicted by multiple linear regression models.

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Hanania et al (2013) assessed the potential of FENO, peripheral blood eosinophil count, and serum periostin as biomarkers of Th2 inflammation and predictors of treatment effects of omalizumab. The EXTRA omalizumab study enrolled patients (aged 12 to 75 years) with uncontrolled severe persistent allergic asthma. Analyses were performed evaluating treatment effects in relation to FENO, blood eosinophils, and serum periostin at baseline. Patients were divided into low- and high-biomarker subgroups. Treatment effects were evaluated as number of
protocol-defined asthma exacerbations during the 48-week treatment period (primary endpoint). A total of 850 patients were enrolled. Data were available from 394 (46.4 %), 797 (93.8 %), and 534 (62.8 %) patients for FENO, blood eosinophils, and serum periostin, respectively. After 48 weeks of omalizumab, reductions in protocol-defined exacerbations were greater in high versus low subgroups for all 3 biomarkers: FENO, 53 % (95 % CI: 37 to 70; p = 0.001) versus 16 % (95 % CI: -32 to 46; p = 0.45); eosinophils, 32 % (95 % CI: 11 to 48; p = 0.005) versus 9 % (95 % CI: -24 to 34; p = 0.54); and periostin, 30 % (95 % CI: -2 to 51; p = 0.07) versus 3 % (95 % CI: -43 to 32; p = 0.94). The authors concluded that the difference in exacerbation frequency between omalizumab and placebo was greatest in the 3 high-biomarker subgroups, probably associated with the greater risk for exacerbations in high subgroups. Moreover, they stated that additional studies are needed to explore the value of these biomarkers in clinical practice.

McCormack et al (2013) noted that little is known about use of FENO to predict asthma exacerbations among high-risk, urban, minority populations receiving usual care. A total of 138 children with persistent asthma were enrolled in a prospective observational cohort study and skin tested at baseline (wheal greater than or equal to 3 mm = +SPT). Fractional exhaled nitric oxide levels, lung function, and asthma-related health care use were assessed at baseline and every 3 months thereafter for 1 year. Relationships between FENO and health care utilization in the subsequent 3 months were examined. Final models accounted for repeated outcome measures and were adjusted for age, gender and lung function. The mean age was 11 years (range of 5 to 17), and most were male (57 %), African American (91 %), and atopic (90 %). At baseline, FENO was (median IQR: 31.5 ppb [16 to 61]) and FEV1/FVC was (mean +/- SD: 80.7 +/- 9.6 %). There were 237 acute asthma-related health care visits, 105 unscheduled doctor (UD) visits, 125 ED visits, and 7 hospitalizations during the follow-up period. Fractional exhaled nitric oxide was not a significant predictor of acute visits, ED visits, UD visits, or hospitalization in either unadjusted or adjusted analyses. Use of recommended cut-off points did not improve the predictive value of FENO (positive-predictive value [PPV]; 0.6 to 32.8 %), nor did application of the guideline-based algorithm to assess change over time. The authors concluded that FENO may not be a clinically useful predictor of health care use for asthma exacerbations in urban minority children with asthma.

An UpToDate review on “Exhaled nitric oxide analysis and applications” (Dweik, 2014) states that “Exhaled nitric oxide has an important role in clinical research and will likely help in expanding our understanding of asthma, such as the factors responsible for asthma exacerbations and the sites and mechanisms of action of medications for asthma. In addition, it is used to characterize the asthma phenotype of subjects in clinical trials. As an example of how FENO levels might improve our understanding of asthma therapy, FENO levels, which are thought to reflect airway inflammation mediated by interleukin (IL)-4 and IL-13, appear to be a good marker for response to inhaled glucocorticoids, lebrikizumab (anti-IL-13), and omalizumab (anti-IgE), consistent with IL-4 and IL-13 mediating the effects of these agents. On the other hand, FENO levels do not correlate with responses to mepolizumab (anti-IL-5), which is thought to affect systemic (rather than local airway) eosinophil production and mobilization. Diseases other than asthma are associated with altered levels of exhaled NO. The role of measuring FENO in
these diseases is not established, although with further study FENO may be useful as a screening test in chronic cough due to non-asthmatic eosinophilic bronchitis .... A few studies have assessed exhaled NO values in interstitial lung disease related to scleroderma and sarcoidosis. In general, the differences in values are within the range of variability of the test. In a study of patients with scleroderma, a higher exhaled NO was noted among patients with ILD compared with those without ILD, while the opposite was found in another study .... Patients with asthma generally have high levels of FENO in their exhaled breath that returns to normal after treatment with glucocorticoids. It is believed that eosinophilic inflammation in the airways stimulates airway epithelial cells to produce NO, thus making FENO a potentially useful marker to monitor airway inflammation in asthma”.

Current guidelines from GINA (2014) stated that “FENO cannot be recommended at present for deciding whether to treat patients with possible asthma with ICS”. The guidelines stated that “treatment guided by FENO has generally not been found to be effective” and that FENO guided treatment cannot be recommended for the general asthma population.

Guidelines from the ATS and the ERS (Chung et al, 2014) concluded that “We suggest that clinicians do not use FeNO to guide therapy in adults or children with severe asthma (conditional recommendation, very low quality evidence)”.

Bjermer et al (2014) stated that although not yet widely implemented, FENO has emerged in recent years as a potentially useful biomarker for the assessment of airway inflammation both in undiagnosed patients with non-specific respiratory symptoms and in those with established airway disease. Research to date essentially suggested that FENO measurement facilitates the identification of patients exhibiting T-helper cell type 2 (Th2)-mediated airway inflammation, and effectively those in whom anti-inflammatory therapy, particularly ICS, is beneficial. In some studies, FENO-guided management of patients with established airway disease is associated with lower exacerbation rates, improvements in adherence to anti-inflammatory therapy, and the ability to predict risk of future exacerbations or decline in lung function. Despite these data, concerns regarding the applicability and utility of FENO in clinical practice still remain. These investigators reviewed the current evidence, both supportive and critical of FENO measurement, in the diagnosis and management of asthma and other inflammatory airway diseases. It additionally provided suggestions regarding the practical application of FENO measurement: how it could be integrated into routine clinical practice, how its utility could be assessed and its true value to both clinicians and patients could be established. The authors concluded that although some unanswered questions remain, current evidence suggested that FENO is potentially a valuable tool for improving the personalized management of inflammatory airway diseases.

Chaudry et al (2014) noted that asthma and airway hyper-responsiveness are reportedly more common in children with sickle cell disease (SCD). In a prospective, single-center study, these researchers determined airway responsiveness, airway inflammation and clinical features of asthma in SCD. A total of 50 SCD children without overt pulmonary vascular disease and 50 controls were included in this study. Exhaled nitric oxide and total serum IgE were measured and spirometry and methacholine challenge were performed. The methacholine dose-response slope (DRS) was calculated. Doctor diagnosis of
asthma was made in 7 (14 \%) SCD versus 12 (24 \%) control subjects (p = 0.203). FENO levels were similar in SCD and controls (p = 0.250), and were higher in those with atopy and an asthma diagnosis (OR 4.33, 95 \% CI: 1.7 to 11.1; p < 0.05). zFEV1 (p = 0.002) and zFEV1/FVC (p = 0.003) but not zFVC (p = 0.098) were lower in SCD versus controls. The methacholine dose-response slope was higher in those with asthma (p = 0.006) but not in SCD versus controls (p = 0.403). The methacholine dose-response slope correlated with FENO and blood eosinophil count in controls but not SCD. In SCD, DRS was higher in those admitted to hospital with respiratory symptoms (n = 27) versus those never admitted (n = 23) (p = 0.046). The methacholine dose-response slope was similar in those with at least 1 acute chest syndrome episode (n = 12) versus those with none (n = 35) (p = 0.247). The authors concluded that SCD children have airflow obstruction despite having minimal evidence of pulmonary vascular disease. Airflow obstruction is not associated with increased methacholine sensitivity or eosinophilic inflammation, at least as judged by FENO. Airflow obstruction in SCD does not appear to be related to childhood eosinophilic asthma, but its pathophysiology remains ill understood.

**Exhaled Breath Condensate pH**

Exhaled breath condensate (EBC) is a non-invasive method for studying the composition of the fluid lining the airway. Researchers have reported abnormalities in EBC concentrations of at least 12 markers in individuals with inflammatory lung disorders. The measurement of EBC pH is one EBC marker that is currently being investigated as a method for assessing asthma and other chronic pulmonary diseases.

Investigators have found that EBC pH values of individuals with respiratory disease (e.g., asthma and other chronic pulmonary diseases) are lower compared with those of healthy controls and that pH levels increase towards control levels after steroid treatment (Kharitonov, 2004; Carpagnano et al, 2004, 2005; Carraro et al, 2005; Effros et al, 2005).

In a study presented at the 2004 Annual Meeting of the American Academy of Allergy, Asthma and Immunology (AAAAI), researchers reported that the pH of EBC in acute asthmatic children was significantly lower than those with stable asthma. Brunetti and colleagues tested 104 asthmatic children with skin prick tests, lung function tests, and EBC pH measurements. The children experiencing asthma exacerbations received steroid and beta-2 agonist treatment for one week before the pH test was repeated. Thirty-four children (34.7 \%) showed evidences of acute asthma and 70 children (67.3 \%) had stable asthma. After treatment, the EBC pH of patients with acute asthma was significantly higher than before therapy.

Several researchers, however, have raised concerns regarding the standardization of EBC collection and measurement methods. A recent consensus panel convened by the American Thoracic Society/European Respiratory Society Task Force on EBC (Horvath, 2005) provided general recommendations for both EBC collection and measurement. However, the Task Force stated that more studies are necessary before EBC can be recommended for clinical practice. The following areas for future research were identified: (i) mechanisms and site of EBC particle formation; (ii) determination of dilution markers; (iii) improvement of reproducibility; (iv) longitudinal studies are needed; and (v) determination of the utility of EBC measures in the management of individual patients.
In a review of EBC in chronic obstructive pulmonary disease, Effros et al (2005) stated that EBC pH measurements may not provide accurate estimates of airway pH. Data interpretation is complicated by uncertainty regarding the source of condensate solutes and by variable dilution of respiratory droplets from condensed water vapor, which represents more than 99.9 % of condensate volumes. Furthermore, the Guidelines from the Global Initiative for Asthma (GINA) (2004) stated that neither sputum eosinophilia nor exhaled gases have been evaluated prospectively as an aid in the diagnosis of asthma. The guidelines state that there is a need to develop further noninvasive discriminate measurements of airway inflammation.

Exhaled breath condensate pH is a novel, noninvasive research approach to monitor lung diseases; however, well-designed controlled studies are needed to establish the clinical utility of EBC pH for the assessment of asthma and other chronic pulmonary diseases.

Baraldi and Carraro (2006) stated that EBC is still only a research tool. Ko et al (2007) stated that there is some evidence that certain markers in EBC differ between patients with asthma and controls, and some markers may correlate with asthma severity and lung function, but there are many methodologic pitfalls with EBC assessment that limit its clinical applicability at present. The authors concluded that more studies are needed before this technique can be recommended for clinical use.

Cepelak and Dodig (2007) stated that in spite of many scientific studies involving lung disease patients, methodology for EBC collection and analysis has not yet been realized for daily utilization. Additional studies of the exact origin of condensate constituents and standardization of the overall analytical process, including collection, storage, analysis and result interpretation, are needed. Irrespective of these limitations, further investigation of this sample type is fully justified by the fact that classical specimens used in the management of pulmonary diseases are either obtained by invasive procedures (e.g., induced sputum, biopsy, broncho-alveolar lavage) or can not provide appropriate information (e.g., urine, serum). Analysis of EBC in the future might contribute significantly to the understanding of the physiological and pathophysiological processes in lungs, to early detection, diagnosis and follow-up of disease progression, and to evaluation of therapeutic response.

Guidelines from the National Asthma Education Program (NIH, 2007) stated that many biomarkers have been proposed, including concentration of hydrogen ions and various other metabolites in an exhaled breath condensate, but that few studies have validated these markers. The guidelines stated that these biomarkers may have a role in asthma management in the future.

Chan and colleagues (2009) stated that breath analysis, which includes gaseous phase analysis that measures volatile organic compounds using electronic noses, FENO, and EBC, has been proposed as a non-invasive and simple technique to investigate neoplastic processes in the airways. Exhaled breath condensate can be easily collected by breathing into a cooling system that condenses the water vapor in the breath. It has already been suggested to be a useful method to monitor severity of diseases such as asthma and to act as a surrogate measure of
compliance to medical therapy. Presently, there still remains a relative paucity of lung cancer research involving EBC. However, since EBC is a simple, non-invasive technique that can be easily performed, even in ill patients, it has the potential to be validated for use in screening for the early diagnosis of lung cancer.

Dalaveris and associates (2009) evaluated the levels of vascular endothelial growth factor (VEGF), 8-isoprostanone and tumor necrosis factor (TNF)-alpha in EBC and serum of patients with primary lung cancer prior to the initiation of any treatment, in order to evaluate their possible diagnostic role. Furthermore, associations between VEGF, 8-isoprostanone and TNF-alpha levels in EBC and serum with clinicopathologic factors were examined. These researchers enrolled 30 patients with lung cancer (mean age of 65.2 +/- 10.5 years) and 15 age- and gender-matched healthy smokers as controls. Serum and EBC were collected before any treatment; TNF-alpha, VEGF and 8-isoprostanone levels in EBC and serum were analyzed by an immunoenzymatic method. A statistically significant difference was observed between lung cancer patients and the control group regarding the values of TNF-alpha, both in EBC (52.9 +/- 5.0 pg/ml versus 19.4 +/- 3.9 pg/ml, p < 0.0001) and serum (44.5 +/- 6.3 pg/ml versus 22.2 +/- 4.3 pg/ml, p = 0.035). Moreover, EBC VEGF levels were higher in patients with T3-T4 tumor stage compared to T1-T2 (9.3 +/- 2.8 pg/ml versus 2.3 +/- 0.7 pg/ml, p = 0.047). A statistically significant correlation was also observed between serum and EBC values of VEGF (r = 0.52, p = 0.019). In addition, serum levels of VEGF were higher in lung cancer patients than in controls (369.3 +/- 55.1 pg/ml versus 180.5 +/- 14.7 pg/ml, p = 0.046). Serum VEGF levels were also higher in patients with advanced stage of disease (IIIB-IV) and distant nodal metastasis (N2-N3). No differences were observed in 8-isoprostanone in EBC between lung cancer patients and controls. In contrast, serum 8-isoprostanone levels were higher in lung cancer patients compared to controls (24.9 +/- 3.6 pg/ml versus 12.9 +/- 1.6 pg/ml, p = 0.027) and were higher in patients with advanced disease. All 3 biomarkers presented acceptable reproducibility in the EBC on 2 consecutive days. The authors concluded that TNF-alpha, VEGF and 8-isoprostanone are elevated in the serum of lung cancer patients and increased serum VEGF and 8-isoprostanone levels are related to advanced disease. In EBC, increased TNF-alpha levels were observed in lung cancer patients, whereas increased VEGF levels were observed in advanced T-stage. They stated that further longitudinal studies are needed for the evaluation of the prognostic role of these biomarkers in lung cancer.

Fila and Musil (2010) stated that examination of EBC belongs to experimental methods that are used in many pulmonary diseases and it can take part in the study of their pathophysiology. Many biomarkers of inflammation and oxidative stress were studied in EBC in cystic fibrosis. Examination of pH of EBC is considered to be useful in evaluation of inflammatory acidification of airways, together with evaluation of response to antibiotic treatment of pulmonary exacerbation, due to immediately accessible result. Other important biomarkers include 8-isoprostanone and 3-nitrotyrosine as markers of oxidative stress (both with negative correlation with pulmonary function) and leukotriene B4 as marker of neutrophilic inflammation. Opposite to other pulmonary diseases, hydrogen peroxide does not belong to useful markers of oxidative stress in cystic fibrosis, due to abundant reduced thiols and glutathione peroxidase in sputum of these patients. Attempts to detect bacterial DNA in EBC in cystic fibrosis also failed. In spite of
mentioned progress, examination of EBC remains a research method and it has not been introduced into clinical practice.

Teng et al (2011) examined if EBC hydrogen peroxide \( \text{H}_2\text{O}_2 \) is elevated in people with asthma and if it reflects disease severity and disease control or responds to corticosteroid treatment. Studies were identified by searching PubMed, Embase, Cochrane Database, Cumulative Index to Nursing and Allied Health Literature (CINAHL), and www.controlled-trials.com for relevant reports published before September 2010. Observational studies comparing levels of EBC \( \text{H}_2\text{O}_2 \) between patients with asthma who were non-smokers and healthy subjects were included. Data were independently extracted by 2 investigators and analyzed using Stata 10.0 software. A total of 8 studies (involving 728 participants) were included. EBC \( \text{H}_2\text{O}_2 \) concentrations were significantly higher in patients with asthma who were non-smokers compared with healthy subjects, and higher values of EBC \( \text{H}_2\text{O}_2 \) were observed at each level of asthma, classified either by severity or control level, and the values were negatively correlated with FEV1. In addition, EBC \( \text{H}_2\text{O}_2 \) concentrations were lower in patients with asthma treated with corticosteroids than in patients with asthma not treated with corticosteroids. The authors concluded that \( \text{H}_2\text{O}_2 \) might be a promising biomarker for guiding asthma treatment; however, further investigation is needed to establish its role.

Thomas et al (2013) performed a systematic review to identify studies of EBC markers in childhood asthma. Most of the studies were cross-sectional in design, and the results suggested that simple chemical entities such as hydrogen ions (as \( \text{pH} \)), hydrogen peroxide, and oxides of nitrogen are associated with pediatric allergic asthma and exacerbations. In addition, more complex molecules including leukotrienes, prostaglandins, and cytokines such as the interleukins IL-4 and IL-5 are also elevated in the breath of those with asthma. The authors concluded that EBC has the potential to aid diagnosis, and to evaluate the inflammatory status of asthmatic children. They stated that future studies may be able to refine further how best to collect EBC samples, to interpret them, and the technique has the potential to allow repeated sampling that will allow studies of natural history, pathogenesis and response to treatment to be undertaken.

**CPT Codes / HCPCS Codes / ICD-9 Codes**

**CPT codes not covered for indications listed in the CPB:**

83987

95012

**ICD-9 codes not covered for indications listed in the CPB (not all-inclusive):**

011.0 - 011.6

162.2 - 162.9

470 - 478.19
The above policy is based on the following references:

**Exhaled Nitric Oxide**


70. Deykin A, Massaro AF. Exhaled nitric oxide analysis. UpToDate [online serial]. Waltham, MA: UpToDate; last reviewed June 2013.


86. Dweik RA. Exhaled nitric oxide analysis and applications. UpToDate [serial online]. Waltham, MA: UpToDate; reviewed June 2013.


