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REVIEW

Current evidence and future research needs for FeNO measurement in respiratory diseases

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Summary
Although not yet widely implemented, fraction of exhaled nitric oxide (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 suggests 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 inhaled corticosteroids (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

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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. This article reviews the current evidence, both supportive and critical of FeNO measurement, in the diagnosis and management of asthma and other inflammatory airway diseases. It additionally provides 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. Although some unanswered questions remain, current evidence suggests that FeNO is potentially a valuable tool for improving the personalised management of inflammatory airway diseases.

Contents

Introduction

The majority of patients presenting to primary care physicians with non-specific respiratory symptoms such as wheeze, cough and breathlessness are treated with inhaled corticosteroids based on the presumptive diagnosis of asthma [1]. On detailed assessment, however, many patients lack objective evidence of asthma or inflammatory airway disease [1,2].

The identification of airway obstruction and abnormal airways physiology is the objective of diagnostic tests such as spirometry, reversibility testing, peak flow monitoring, and bronchoprovocation tests, commonly used in the investigation of airway disease [3,4]. However, diagnosis and management of patients with airway diseases based on these physiological parameters alone without assessing underlying inflammation may be inadequate in targeting anti-inflammatory treatment to those who lack confirmatory evidence. This is important as ineffective treatment is costly and may also be associated with adverse effects, while delaying appropriate treatment. Hence, exploring more adequate diagnostic and management strategies is required. In addition to current practice would be the assessment of airway inflammation and corticosteroid responsiveness on an individual basis for a personalized diagnostic and treatment approach. One such approach involves measuring the fraction of exhaled nitric oxide (FeNO), which can be performed easily and in close to real time by utilising chemiluminescence, electrochemical detection or laser spectroscopy devices [5], and which has the potential to identify patients with corticosteroid-responsive, T-helper cell 2 (Th2)-mediated airway inflammation [6]. In conjunction with symptom scores and lung function tests, FeNO measurement could provide a more useful and effective approach for the identification of asthma and other corticosteroid-responsive inflammatory airway conditions.

Nitric oxide synthase (NOS) enzymes, which catalyse the conversion of L-arginine to L-citrulline to generate NO exist in three distinct isoforms: endothelial (eNOS), inducible (iNOS), and neuronal (nNOS) [7]. Recent evidence shows that in atopic asthmatics, the upregulation of iNOS in the

respiratory epithelium via STAT-6 and pro-inflammatory Th2-cytokines interleukin (IL)-4 and IL-13 [6], produces enhanced NO concentrations in exhaled air [8,9]. Exhaled NO, can thus be regarded as a direct biomarker of Th2-mediated mechanisms within the bronchial mucosa, and can provide a direct indication of ongoing Th2-driven inflammation. Further research shows that in patients with Th2-driven airway inflammation, FeNO measurement provides information on potential responsiveness to corticosteroid treatment. FeNO may consequently provide the ability to (i) identify individuals with inflammatory airway diseases who will benefit from existing and future anti-inflammatory treatments, particularly inhaled corticosteroid (ICS) treatment [4,10–12], and (ii) to monitor and manage the treatment of patients with inflammatory airway diseases [13].

We believe that personalised strategies, specifically the application of inflammometry, should be rigorously assessed for widespread use in clinical practice, including analysis of cost-effectiveness. This would allow formal testing of the practical utility of such an integral and personalised approach in daily routine care.

Factors influencing FeNO measurement and interpretation

Individual factors

Certain considerations should be applied when interpreting FeNO values as they are influenced by various factors. A consistent finding in children is that FeNO levels increase with age, most likely due to increases in airway mucosal surface area. Data in adults are inconsistent, with variations in the age ranges of normal subjects included in these studies. Olin et al. showed that individuals aged >64 years had 40% higher FeNO levels than those aged 35–44 years [14]. This finding was corroborated by Gelb et al., who recently showed that the effect of age on FeNO is greater in individuals >60 years [15]. Another study, also conducted in healthy, non-smoking adults with normal spirometry values showed no correlation between age and FeNO, but few older subjects were included in this study. Instead, a significant gender difference was observed; at expiratory flows of 50 mL/s, mean FeNO levels were 11.7 (range 2.6–28.8 ppb) in men and 9.9 (range 1.6–21.5 ppb) in women ($p = 0.01$) [16]. Most studies show a relationship between height and FeNO levels, both in children and adults [14,17]. Recent studies have indicated that a more accurate and generalisable interpretation of FeNO could be derived by taking individual factors into consideration and assessing values based on percent predicted of reference values, or z-scores [18,19]. However, in a recent study including 13,275 participants aged 6–80 years (normal population), prediction equations based on multiple linear regression models justified only 10.3–15.7% of the variation in FeNO levels [20]. Thus, the prediction equation models need to be improved.

External factors

Cigarette smoking has consistently been shown to reduce FeNO levels, and the magnitude of the reduction seems to depend on the daily cigarette consumption [21,22]. However, FeNO is still raised in smokers with asthma, compared to smokers without asthma, and it has been shown that FeNO can differentiate asthma from non-asthma with asthma-like symptoms equally well in smokers as in never-smokers [23]. In contrast, FeNO increases following consumption of nitrate rich food, for example green-leaved vegetables such as lettuce and spinach [6,24]. ATS/ERS guidelines recommend performing FeNO measurements before spirometric manoeuvres [25]. However, while some studies show a marginal reduction in FeNO levels in children [26], others show no effect in adults [27,28]. In addition to individual determinants, other factors including allergen exposure, rhinovirus infections, physical exercise, and air pollution influence FeNO [6]. IgE sensitisation and subsequent allergen exposure has been reported to increase FeNO levels in asthmatic individuals [29–33]. Rhinovirus infections induce increases in FeNO levels as a result of upregulated iNOS expression in airway epithelium [34]. Discrepant effects of exercise on FeNO have been reported; some studies show up to a 10% decrease in FeNO following exercise in healthy and asthmatic subjects [35,36], while others show no effect [37,38]. Air pollution due to increased ozone levels appears to increase FeNO levels particularly in asthmatics, possibly due to increased iNOS expression in airway epithelium in an AP-1- and STAT-1-dependent mechanism [6].

Most external factors reported to influence FeNO have only small and clinically nonsignificant effects. However, three major confounders can be distinguished; cigarette smoking, virus infections and certain food intake. These confounders are summarized in Table 1 with suggestions on how to deal with them in clinical practice.

It should also be noted that absolute FeNO values vary depending on the device used. A study by Boot et al. showed that a chemiluminescence device and an electrochemical device, from two different manufacturers, could

Table 1 Clinically important confounding factors for FeNO measurements, their approximate effect size and advice on how to manage these in clinical practice.

Factor	Effect size	Measure
Cigarette smoking	Reduction of 30–60%, dependent on daily cigarette consumption	Use intraindividual changes, for example after introduction of anti-inflammatory therapy
Rhinovirus infections	Increase of 50–150%	Repeat measurement after at least 14 days
Intake of nitrate-containing food	Increase of up to 40–60%, with peak 1–2 h after intake	Ask patients to refrain from a meal consisting primarily of green-leaved vegetables on the day of assessment, or at least record such intake

not be used interchangeably because the chemiluminecence device produced slightly lower values [39]. To maintain accuracy in interpretation and comparison of FeNO values, it is essential that the same device is used during research and in general clinical practice, as calibration procedures may differ between devices [5].

Clinical applications of FeNO

Diagnosing and assessing ICS-responsive inflammatory airway disease

Asthma is a chronic inflammatory airway disease associated with airway hyperresponsiveness (AHR) [40,41]. While the majority of asthma is associated with eosinophilic inflammation, not all patients exhibit this feature [42]. Various studies have shown that in individuals with asthma, increased FeNO levels are associated with eosinophilia in blood, sputum, bronchoalveolar lavage (BAL) fluid and airway mucosa [3,13,43–45]. FeNO is potentially a valuable aid in asthma diagnosis; in a study comparing FeNO and sputum cell counts against serial peak flow recordings and spirometry in children and adults, the sensitivity of spirometry was lower (47%) than that of either FeNO (88%) or sputum eosinophils (86%). FeNO and sputum eosinophils additionally exhibited a specificity of 92% as compared with 73% for spirometry [46].

FeNO has long been regarded as a surrogate marker of eosinophilic airway inflammation. Recent studies, however, indicate that FeNO is more representative of a Th2-driven local inflammation, specifically of the bronchial mucosa, rather than general eosinophilic inflammation, as measured by blood or induced sputum. For example, FeNO levels correlate better with bronchial eosinophils than with sputum eosinophils [6,47,48]. The disconnect between FeNO and eosinophilic inflammation has been highlighted by studies with monoclonal antibodies (mAb) against IL-5 and IL-13, which show that treatment with mepolizumab, an anti-IL-5 mAb, significantly reduces blood and sputum eosinophils without affecting FeNO levels [48], while treatment with lebrikizumab, an anti-IL-13 mAb, significantly reduces FeNO levels without reducing blood eosinophils [49].

Consequently, an important attribute of FeNO is its ability to potentially predict the response to ICS therapy in asthma and other inflammatory airway conditions [12,50–53]. Research suggests that subjects (especially patients with asthma) with elevated baseline FeNO levels are more likely to respond to ICS [12] and, in most cases, show a rapid reduction in FeNO levels upon initiation of ICS treatment [54]. In contrast, those with baseline FeNO levels in predefined normal ranges are less likely to respond to ICS [12]. A relatively small study by Smith et al. investigated the utility of FeNO in predicting an ICS response in patients aged 12–75 years with persistent, previously undiagnosed respiratory symptoms [12]. Regardless of the final diagnosis, patients in the highest FeNO tertile (>47 ppb) had significantly greater responses to inhaled fluticasone (increase in FEV_1, increase in mean morning peak flows, improved symptoms and reduction in AHR to adenosine monophosphate [AMP]) than those in the mid (15–47 ppb) or low (<15 ppb) FeNO tertiles. However, less than half of patients in the mid-tertile were later diagnosed with asthma, as compared with almost 90% in the high tertile, which probably explains the low degree of ICS responsiveness in the subjects with intermediate FeNO levels [12]. Hahn et al. demonstrated that in subjects aged ≥18 years with uncontrolled chronic cough, those with elevated FeNO levels (≥35 ppb) had a higher likelihood of responding positively to ICS therapy than those with lower FeNO levels (<35 ppb) [50]. In patients with COPD, some of whom show an eosinophilic rather than the usual neutrophilic inflammatory pattern [55,56], pre-ICS FeNO levels have been shown to correlate positively with short-term improvement in FEV_1 to oral and inhaled corticosteroids [52,57]. Conflicting evidence was reported by others, e.g. Klaassen et al. reported that symptoms, but not FeNO levels, predicted a positive response to ICS therapy in children with recurrent wheeze who were later diagnosed with asthma [58]. Notably however, in this study, FeNO was measured using an offline (tidal breathing) method, rather than the recommended online measurement. Prieto et al. reported that while a significant proportion of patients (aged 18–70 years) with chronic cough responded well to ICS, at a baseline cut-off of 20 ppb, FeNO was not useful in predicting this response [59].

Management of ICS-responsive inflammatory airway disease

Inhaled corticosteroids are recommended for long-term control of persistent asthma in both children and adults due to their ability to target the underlying airway inflammation and to reduce the risk of asthma exacerbations [40,60,61]. The relationship between clinical outcomes and ICS dose is variable in asthma, and some patients may require high ICS doses to achieve acceptable levels of disease control. However, higher ICS doses increase the risk of adverse effects such as oral candidiasis, dysphonia, hoarseness, cataracts, and growth retardation in children [62]. Optimum ICS dosing is important to promote patient safety, whilst maintaining adequate asthma control and minimising exacerbations.

Traditionally, ICS dose titration is based on assessment of patient exacerbation history, symptoms and standard lung function tests. A number of studies have investigated the value of FeNO in the management of asthmatic patients, particularly in predicting the risk of exacerbations, in dose titration, and in assessing compliance to ICS. Dose titration studies have been inconsistent, with some studies reporting benefits and others not. Smith et al. randomly allocated 97 asthmatic patients requiring ICS to treatment adjustment based on FeNO measurements or conventional guidelines. As compared with the control group, FeNO-guided therapy resulted in a significant reduction in the mean ICS dose in the active group (641 vs 370 μg; $p = 0.003$), accompanied by a non-significant trend towards reduced exacerbation rates (0.49 vs 0.90) [63]. Powell et al. used a FeNO-based treatment algorithm to optimise ICS dosing in non-smoking, pregnant asthmatic women. Patients were randomly assigned to ICS adjustment using either clinical symptoms (control group) or FeNO levels (active group). ICS doses were increased at FeNO concentrations >29 ppb and reduced at <16 ppb. The mean maintenance daily ICS dose and exacerbation rates were

significantly lower in the FeNO-guided group than in the control group (ICS dose: $p = 0.043$; exacerbation rates 0.288 vs 0.616, $p = 0.001$) [64]. More recently, a multicentre study performed within primary healthcare by Syk et al. showed improved asthma outcomes without an increase in overall ICS use [65]. Asthmatics were randomised to treatment with ICS and a leukotriene receptor antagonist guided either by FeNO values (active group) or to standard care (control group) and followed for 1 year. In the active group, treatment was stepped up at a FeNO level of $\geq$25 ppb and stepped down at $<$20 ppb. The FeNO-guided group showed significantly improved asthma control (Juniper ACQ score) compared with the control group, and the exacerbation rate was reduced by almost 50%. However, some other studies have failed to significantly show that FeNO-guided treatment strategies provided further benefits in asthma control, when compared with conventional strategies [66–68]. Szefler et al. conducted a randomised, double-blind, parallel-group trial in 546 inner-city adolescents and young adults (aged 12–20 years) with persistent asthma and demonstrated that the addition of FeNO measurements to guideline-based clinical care resulted in significantly higher ICS-doses (118.9 μg/day difference, $p = 0.001$) without clinically important improvements in asthma control. However, FeNO-guided care produced a significant reduction in the risk of requiring at least one prednisone course for asthma exacerbations [67]. Furthermore, post-hoc analyses highlighted that subgroups of asthmatics with obesity, higher blood eosinophil count and greater atopy may benefit from FeNO measurement [67]. de Jongste et al. investigated the effect of daily telemonitoring of asthma symptoms plus-minus FeNO measurements on the management of 151 atopic asthmatic children [66]. Both approaches were associated with improved asthma control and lower ICS use with no statistical difference between study groups. ICS doses were only adjusted every 3 weeks and the authors acknowledge more frequent FeNO-based dose adjustments may have produced better outcomes. In a randomised, single-blind trial by Shaw et al., based either on FeNO measurements or the British Thoracic Society (BTS) guidelines in 118 asthmatic participants [68], a non-significant reduction in asthma exacerbations was achieved together with a significant reduction in final ICS dose in the FeNO-guided group compared with the guidelines-based group. More recently, a study by Calhoun et al. was not able to show a reduced incidence of treatment failure, which was the primary endpoint, by a FeNO-based strategy compared to either a physician-based or a symptom-based strategy [69]. However, the study included primarily patients with mild, well-controlled asthma, which means that very little room was left for further improvement with regard to treatment failures. In a subanalysis by season, the authors showed a significantly lower incidence of treatment failures during the autumn, which is a high-risk season, by the FeNO-based strategy compared to the physician-based strategy. Furthermore, the FeNO strategy provided a significant improvement in daily symptoms as well as methacholine reactivity compared to the physician-guided group.

It is clear that methodological issues and cut-off points used in the different FeNO-guided intervention studies may explain discrepancies between studies. Consequently, a Cochrane review, comparing ICS-adjustments based on either FeNO measurements or clinical symptoms, concluded that FeNO could not be routinely recommended for clinical practice at this time and that further studies were warranted [70]. The primary outcome in this meta-analysis was the proportion of subjects with at least one asthma exacerbation, so the analysis did not account for subjects with multiple exacerbations [4,71]. Further possible analyses include annual exacerbation rates or time to first exacerbation, which has been recommended by an ATS/ERS Task Force on outcomes in asthma clinical trials [72]. Two more recent meta-analyses based on exacerbation rates, have reported that FeNO-guided asthma management was superior to conventional methods [4,71].

FeNO measurement in paediatrics

FeNO may be of particular interest for diagnosing and phenotyping asthma in children with suspected asthma, aiming to achieve optimal treatment and asthma control. Diagnosing asthma in children, particularly preschoolers, may be challenging. Moeller et al. reported that in wheezing children aged 3–47 months, FeNO levels were significantly higher in children with frequent recurring wheeze and a stringent index for the prediction of asthma as compared to children with early recurrent wheeze and a loose index for the prediction of asthma, or children with recurrent cough but no wheeze; thus predicting disease progression [73]. This information may help clinicians identify which children are potential ICS responders [74].

Although monitoring of asthma control in primary care is currently mainly focused on the evaluation of clinical symptoms and lung function parameters, GINA guidelines and ATS FeNO guidelines suggest that airway inflammation could be assessed for optimised treatment strategies [41,75]. However, in patients of all ages, a dissociation between control evaluation tools, such as validated questionnaires, and the level of underlying airway inflammation has been demonstrated [76]. While a reduction in FeNO levels can be indicative of a response to ICS treatment [77], high FeNO levels are indicative of a higher probability of asthma relapse on ICS reduction or withdrawal [77–79]. Based on these data, other studies have investigated the effects of FeNO-guided corticosteroid titration in children with partly conflicting outcomes [66,67,80,81]. Possible explanations for inconsistency in findings have been proposed in the Asthma randomised Treatment Algorithm (ASTRAL) studies report, highlighting design and methodological issues, which may have led to different conclusions between studies as discussed above [82]. In a recent single-blind, randomised, controlled study in 99 paediatric patients with persistent allergic asthma a FeNO-guided measurement strategy failed to improve the proportion of symptom-free days, but was associated with fewer asthma exacerbations, increased LTRA use and augmented ICS doses [83].

Currently, FeNO may be considered as a potentially useful adjunctive tool in clinical paediatric practice, in particular in specialist settings, in order to better characterise airways inflammation in children with wheezing, as a guide to ICS use and to achieve a more comprehensive assessment of disease control.

Treatment adherence

Exhaled nitric oxide may have a role in assessing adherence to ICS therapy, since FeNO responds rapidly and dose-dependently to ICS treatment [84]. This is beneficial because adherence with ICS therapy is a critical prerequisite for asthma control. Beck-Ripp et al. evaluated compliance with inhaled budesonide in children by monitoring FeNO levels following sequential changes in treatment. As opposed to standard lung function tests, there was a significant correlation between FeNO and compliance [85]. Koster et al. reported that increased FeNO levels (>25 ppb) in children prescribed ICS were associated with a reduced adherence (OR = 0.25, 95%CI = 0.15—0.41). The authors suggested that improving parental knowledge of drug characteristics and feedback of FeNO readings could positively influence adherence and thus improve asthma control [86]. Finally, McNicholl et al. showed that when patients with difficult-to-treat asthma treated with budesonide were monitored based on changes in FeNO levels, adherent subjects had a greater reduction in FeNO [87].

Guiding treatment response to drugs other than ICS

In asthma, corticosteroids primarily act on the Th2-mediated cytokine release and subsequent inflammatory response. As ICSs are the standard therapy for patients with allergic airway inflammation, most research has focused on the use of FeNO in tailoring ICS treatment. However, data on its value in determining a response to other treatments including leukotriene-receptor antagonists (LTRAs), and biological drugs including omalizumab (anti-IgE), lebrikizumab (anti-IL-13) and mepolizumab (anti-IL-5), which specifically target the Th2-pathway, are emerging. For example, studies have shown that FeNO can be useful in predicting a response to LTRAs in patients with asthma [88,89]. Omalizumab is indicated for the treatment of patients with inadequately controlled severe persistent allergic asthma despite maximal controller therapy. Hanania et al. evaluated the value of FeNO in predicting exacerbation rates in such patients and showed that those in the high FeNO subgroup had greater reductions in exacerbations by omalizumab treatment as compared with those in the low FeNO subgroup (53% vs 16%) [90]. While the authors suggest that additional studies are required to explore the value of FeNO, this study strongly suggests its value as a predictor of responsiveness to omalizumab treatment [90]. Corren et al. investigated lebrikizumab treatment in patients with uncontrolled asthma. Greater lung function improvements, measured by % change in FEV_1 at 12 weeks, were observed in patients with high pretreatment serum periostin (an IL-13-induced epithelial protein) and FeNO levels (14.0% and 14.2%, respectively) than in those with low periostin and FeNO levels (5.1% and 4.8%, respectively) [49].

Predicting future risk: exacerbations and lung function decline

In children and adults with atopic asthma, Zeiger et al. showed that a FeNO level >300% of expected normal (approximately 35—50 ppb depending on individual factors) predicted both impairment (excessive use of short-acting bronchodilators) and risk (exacerbations with prednisolone courses) in the following year [91]. In an adult population, combined use of FeNO and FEV_1, predicted the risk of an exacerbation. In this study conducted over 18 months, at FeNO levels of ≥28 ppb and FEV_1 ≤76%, clinically stable asthmatics were shown to have an 85% probability of a future exacerbation, while at FeNO levels of ≤28 ppb and FEV_1 >76%, there was no risk of exacerbation [92]. FeNO has also been shown to be useful in predicting loss of asthma control following corticosteroid withdrawal. Jones et al. [93] withdrew ICS therapy from 78 adult patients aged 18—74 years with mild-moderate asthma for a maximum of 6 weeks or until they lost asthma control. In those patients who lost asthma control (77.9%), there was a significantly greater increase in baseline FeNO levels, as compared with patients who maintained control (2.16-fold vs 1.44-fold, respectively; $p = 0.004$). FeNO was additionally associated with a positive predictive value of 80%—90% for predicting and diagnosing loss of control [93]. In another study in children, at a cut-off value of 49 ppb, FeNO exhibited a sensitivity of 71% and a specificity of 93% for predicting asthma relapse (defined as more than one exacerbation per month, or need for beta-agonist treatment 4 days per week for at least 2 weeks, or diurnal peak flow variability of >20% after discontinuation of corticosteroids) [78].

FeNO may also predict future lung function decline. [4] Sonnappa et al. investigated the correlation between airway pathology at age 2 and lung function at age 5 (median) in previous severe preschool wheezers by performing biopsies, lung function tests and FeNO measurements. Reticular basement membrane (RBM) thickness and mucosal eosinophilia (but not lung function) measured at age 2 significantly correlated with FeNO measurements at age 5 [94]. Multiple-trigger wheeze in children is associated with abnormal pulmonary function, whereas episodic (viral) wheeze is not [95]. Sonnappa et al. previously demonstrated that despite similar lung function in both groups, multiple-trigger wheezers exhibit significantly higher FeNO levels than episodic wheezers [95]. Van Veen et al. reported that FeNO could predict an accelerated decline in lung function in asthma patients refractory to ICS therapy. FeNO levels of ≥20 ppb (measured at an exhalation flow rate of 100 mL/s) were shown to be associated with an increased decline in FEV_1 compared with FeNO levels of <20 ppb, with an excess decline in lung function of 40.3 mL/year. Patients with a FeNO level of ≥20 ppb had a 57% risk of an accelerated decline in FEV_1 (≥25 mL/year) compared with 30% in patients with a FeNO level of <20 ppb [96].

Proposed framework to guide FeNO use in clinical practice

Clinical scope

The cornerstone of asthma diagnosis is the evaluation of airway dysfunction and airway inflammation, while the aim of asthma management is to achieve control, which according to GINA/BTS guidelines constitutes prevention of symptoms, night-time symptoms/awakening, the need for

rescue medication, exacerbations, limitations of activity and the achievement of normal lung function (FEV_1 and/or PEF >80%) [40,41].

Current evidence suggests that FeNO is useful in: (i) detecting Th2-driven inflammation of the lower airways in conditions like asthma, chronic cough, eosinophilic bronchitis, and sometimes COPD; (ii) predicting a response to ICS and other anti-inflammatory therapy; (iii) continued disease monitoring and follow-up of asthma patients after initial diagnosis using standard procedures. Taking into consideration the previously discussed factors that influence FeNO values (i.e. age, height, gender, smoking, allergen exposure, rhinovirus infections and nitrate intake) we propose a framework to guide treatment decisions that incorporates FeNO measurements into existing GINA/BTS asthma management guidelines. However, further clinical trials, preferably real-world studies, will be required to investigate and validate each of these propositions.

Cut-off values for FeNO

Generic cut-off values are difficult to define due to the effect of the aforementioned individual factors. The 2011 ATS FeNO guidelines suggest that a FeNO level of <25 ppb (<20 ppb in children) provides a strong indication for an unlikely ICS response, while a FeNO level of >50 ppb (>35 ppb in children) provides a strong indication for a likely ICS response. A FeNO level of between 25 and 50 ppb (20–35 ppb in children) should, however, be interpreted cautiously, and with reference to the clinical context, accounting for persistent and/or high allergen exposure as a factor associated with higher FeNO levels, according to these guidelines [75]. However, more recent evidence from a study on 154 steroid-naive adult patients with asthma suggests that subjects with intermediate FeNO levels (25–50 ppb, as defined above) respond to ICS treatment in a similar fashion to patients with high FeNO levels (>50 ppb), whereas patients with a low baseline FeNO value (<25 ppb) respond much less [97]. This is in line with the positive outcomes in the studies by Powell et al. and Syk et al., where ICS treatment was stepped up at FeNO levels of 29 and 25 ppb, respectively, in the FeNO-guided groups [64,65]. Furthermore, Hanania et al. reported that patients with a baseline FeNO level of approximately 20 ppb and above responded significantly more to omalizumab compared to patients below this level [90], and Corren et al. reported that patients with a FeNO level above 21 ppb responded better than patients with lower values [49]. All the outcomes above are supported by the study by Sverrild et al., which showed that a FeNO level of <20 ppb ruled out mannitol reactivity with a sensitivity of 100% and a level of ≥30 ppb ruled in mannitol reactivity with a specificity of 90% in an unselected sample of 180 steroid-naive, non-smoking adolescents and young adults [19]. Moreover, our clinical experience suggests that many patients with intermediate FeNO values, as defined by the ATS guidelines may indeed respond positively to ICS.

Based on current evidence, it could be proposed that treatment decisions are made using two cut-off levels: A low cut-off range of ≤15–25 ppb, and a high cut-off range of ≥35–50 ppb, with both cut-offs depending on individual factors, for example age (see Table 2). These cutoffs are

Table 2 Summary of the suggested clinical decision-making role of FeNO measurement in the management of patients with suspected or diagnosed asthma.

FeNO levels and inflammation			
FeNO (ppb)	Normal	Elevated	High
Adults	<20–25[a]	20/25–50	>50
Children	<15–20[a]	15/20–35	>35
Th2-driven inflammation	Unlikely	Likely	Significant
Initial assessment in treatment-naïve patients with suspected asthma			
Guide to diagnosis	Consider other diagnosis than asthma	Supports a diagnosis of asthma	Supports a diagnosis of asthma
Guide to treatment decision	The patient will likely not respond to ICS	The patient will likely respond to ICS. A trial of low dose ICS is suggested	The patient will likely respond to ICS. A trial of intermediate dose ICS is suggested
Assessment in treated patients with a confirmed diagnosis of asthma			
Guide to management	Th2-driven inflammation is under control	Check treatment adherence, inhalation technique and allergen exposure	Check treatment adherence, inhalation technique and allergen exposure. Indicates increased risk of exacerbations/disease worsening regardless of clinical history
Guide to treatment change decision	Consider step-down of ICS treatment if the asthma has been controlled for at least 3–6 months	If there is a history of exacerbations, step-up anti-inflammatory treatment	Step-up anti-inflammatory treatment, especially if combined with high blood eosinophil count, or consider fine-particle ICS. May suggest ICS-resistant asthma and need for add-on systemic anti-inflammatory therapy

[a] Exact cut-off dependent on age, height and gender.

suggested to be used differently depending on the clinical situation (see below).

Clinical algorithms guided by FeNO

To aid initial diagnosis/treatment decisions in previously untreated patients with uncertain diagnosis (see also Table 2):

- A FeNO value below the low cut-off (15—25 ppb) could be interpreted as a low likelihood of Th2-driven inflammation and an unlikely response to ICS/anti-inflammatory therapy in a (non-smoking) treatment-naive patient. A FeNO value below this cut-off in a previously undiagnosed patient probably indicates non-Th2-driven inflammation and a diagnosis other than asthma or eosinophilic bronchitis. The diagnosis should be re-evaluated and other (anti-inflammatory) treatment strategies should be investigated.

To guide treatment decisions in diagnosed patients with ongoing anti-inflammatory treatment:

- A FeNO value above the high cut-off (35—50 ppb) could be interpreted as a high degree of Th2-driven inflammation and a high likelihood of asthma diagnosis, with increased risk of worsening of symptoms and exacerbations in asthmatics with ongoing treatment, especially when combined with elevated blood eosinophil count [18]. A level above this cut-off indicates a check-up of treatment adherence including inhalation technique and environmental exposures plus-minus the need for stepping up or change to other anti-inflammatory treatment.

To manage/monitor treatment decisions, the change in FeNO following anti-inflammatory therapy may be more applicable and easier to interpret than an absolute FeNO value:

- A reduction in FeNO from a higher range to a lower range (see Table 2) could be interpreted as a high likelihood of a positive response to the introduction or the step-up of ICS or other anti-inflammatory therapy.

Future directions and conclusions

As highlighted within this publication, there are still several areas that need further investigation to strengthen the clinical and cost benefits of FeNO measurement in the standard diagnosis and management of respiratory diseases.

Some pertinent questions to demonstrate the clinical and cost benefits of FeNO measurements in this area might include:

- Does a low FeNO value preclude the long-term need for ICS treatment in an untreated patient?
- Does FeNO measurement provide better asthma control?
- Why do some studies fail to show a clinical benefit for adjunctive FeNO measurements?
- Which patient groups do most likely benefit from FeNO-based ICS-titration?
- Is there a role for alveolar nitric oxide measurements in standard clinical practice and if so, for which patients?
- What is the clinical and economical yield of FeNO measurements in real-life settings as a tool to facilitate the diagnosis and treatment of inflammatory airway diseases?

Thus far, evidence regarding the value of FeNO measurement for the diagnosis and management of inflammatory airway diseases has not been unequivocally supportive due to a number of perturbing factors: differences in study designs, sample size, methodology, clinical parameters, the application of different FeNO algorithms and devices, and inconsistencies in predefined study endpoints. Despite these factors, when used to assess ICS-responsive disease in conjunction with clinical data and standard lung function tests, current evidence supports the additional value of FeNO measurements. FeNO is capable of providing discriminating information on Th2-driven airway inflammation, in a simple, fast, non-invasive and reproducible manner. FeNO measurement is even simpler than spirometry and may thus easily be implemented even within primary care. To date, no other test possesses these attributes. As such, FeNO has been shown to provide additional useful information for clinical practice to aid diagnosis, predict and tailor responsiveness to ICS and biological therapies, and to assess therapy compliance. In this manner, FeNO may be a useful asset to cost-effective, personalised medicine. FeNO use has been associated with lower exacerbation rates and can assist in the identification of patients with distinct asthma phenotypes, e.g. those at risk of future lung function decline or loss of asthma control during ICS or biological therapy. While ongoing research will provide further evidence that should quell lingering doubts, current evidence suggests that the routine use of FeNO in conjunction with conventional clinical measures and lung function tests could help the diagnosis and management of inflammatory airway disease, particularly asthma.

Approaches directed at improving asthma diagnosis and management could lower healthcare costs as well as improve quality of life in patients with poorly controlled asthma and those unnecessarily prescribed ICS [98]. Some health economic models in Europe suggest the use of FeNO in the management of persistent asthma and potentially in the diagnosis of asthma provides cost savings [98,99]. However, further health economic evaluations and real-world observational studies are required to validate the cost-effectiveness of FeNO measurements in asthma diagnosis and management.

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Conflict of interest

Leif Bjermer has during the last five years given lectures and/or attended advisory board for the following companies: Almirall, AstraZeneca, Airsonette, Andre Pharma-Chiesi, Boehringer, GlaxoSmithKline, Meda, Merck, Mundipharma, Nigard Pharma, Novartis, Pfizer, Takeda/Nycomed, Teva.

Kjell Alving is an associate and minority shareholder of Aerocrine. He has received research funding from Aerocrine and Phadia.

Zuzana Diamant works freelance at a CRO (QPS the Netherlands). She serves at an advisory board of Aerocrine and received consultation fees from HAL Allergy, Mundipharma, 7TM, and Sandoz.

Helgo Magnussen is an associated editor in Respiratory Medicine, a) a member of EFWG group (the meetings were sponsored by Aerocrine) b) an advisor of Aerocrine Germany.

In addition, he is director of the Pulmonary Research Institute received research funding or lecture fees or advisory board fees from the following companies: Almirall, Aerocrine, Astra Zeneca, Boehringer Ingelheim, Chiesi, GSK, Novartis, Nycomed, Revotar, Schering Plough (Merck), Pfizer.

Ian Pavord. In the last 5 years IDP has received speaker's honoraria for speaking at sponsored meetings from Astra Zeneca, Boehringer Ingelheim, Aerocrine and GSK. He has received honoraria for attending advisory panels with; Almirall, Astra Zeneca, Boehringer Ingelheim, GSK, MSD, Schering-Plough, Novartis, Dey, Napp. He has received sponsorship to attend international scientific meetings from Boehringer Ingelheim, GSK, Astra Zeneca and Napp. He is Chief Medical Advisor to Asthma UK, a member of the UK Department of Health Asthma Strategy Group, a member of the BTS SIGN Asthma guideline group and joint editor in chief of Thorax. Neither IDP nor any member of his family has any shares in pharmaceutical companies.

Giorgio Piacentini has during the last five years given lectures and/or attended advisory board for the following companies: Chiesi Farmaceutici, Italchimici, Valeas, GSK, MSD, Neopharmed Gentili, Aerocrine, Sensor Medics Italia.

David Price. Board Membership: Almirall, Astra Zeneca, Boehringer Ingelheim, Chiesi, GlaxoSmithKline, Merck, Mundipharma, Medapharma, Novartis, Napp, Nycomed, Pfizer, Sandoz and Teva.

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