OCCASIONAL REVIEW

Exhaled nitric oxide measurements: clinical application and interpretation

D R Taylor, M W Pijnenburg, A D Smith, J C De Jongste

Thorax 2006;**61**:817–827. doi: 10.1136/thx.2005.056093

The use of exhaled nitric oxide measurements (F_ENO) in clinical practice is now coming of age. There are a number of theoretical and practical factors which have brought this about. Firstly, F_ENO is a good surrogate marker for eosinophilic airway inflammation. High F_ENO levels may be used to distinguish eosinophilic from non-eosinophilic pathologies. This information complements conventional pulmonary function testing in the assessment of patients with non-specific respiratory symptoms. Secondly, eosinophilic airway inflammation is steroid responsive. There are now sufficient data to justify the claim that F_ENO measurements may be used successfully to identify and monitor steroid response as well as steroid requirements in the diagnosis and management of airways disease. F_ENO measurements are also helpful in identifying patients who do/do not require ongoing treatment with inhaled steroids. Thirdly, portable nitric oxide analysers are now available, making routine testing a practical possibility. However, a number of issues still need to be resolved, including the diagnostic role of F_ENO in preschool children and the use of reference values versus individual F_ENO profiles in managing patients with difficult or severe asthma.

See end of article for authors' affiliations

Correspondence to: Professor D R Taylor, Department of Medical and Surgical Sciences, Dunedin School of Medicine, University of Otago, P O Box 913, Dunedin, New Zealand; robin.taylor@stonebow.otago.ac.nz

Received 22 November 2005
Accepted 7 February 2006

It is now 12 years since it was first reported that exhaled nitric oxide (NO) levels are increased in bronchial asthma.[1] This discovery followed a period of intense interest in the biology of NO during the late 1980s.[2] The numerous roles of NO in respiratory pathophysiology have been extensively reviewed.[3] NO is an endogenous messenger with a diverse range of effects including non-adrenergic, non-cholinergic neurotransmission, vascular and non-vascular smooth muscle relaxation.[3]

There is contradictory evidence regarding the exact function of NO in lung disease. In pathological situations NO is a pro-inflammatory mediator with immunomodulatory effects.[3] This appears to predispose to the development of airway hyperresponsiveness (AHR), although this is not a consistent finding.[3 4] On the other hand, under physiological conditions NO acts as a weak mediator of smooth muscle relaxation and protects against AHR.[5] In exhaled air NO appears to originate in the airway epithelium.[6] Although raised levels may occur with a number of airway or lung diseases,[7] the most important context in which the measurement of NO is clinically useful is that of allergic airways disease.

It is against this background that measuring the fraction of NO in exhaled air (F_ENO) has emerged as a potentially important clinical tool. F_ENO can be measured easily using a range of commercially available analysers, and smaller less costly devices are now becoming available. This opens the possibility that F_ENO measurements might be used routinely in the assessment of airway disease. This is a significant advance. To date, assessing airway physiology—that is, changes in airway calibre and/or bronchodilator or bronchoconstrictor responsiveness—has been the principal means of providing supportive evidence for the diagnosis of airways disease and assessing severity. Although pulmonary function tests will always remain important, they are one step removed from the issue of interest—that is, airway inflammation. Thus, F_ENO measurements provide a complementary and, in some instances, a more relevant perspective.

In this paper we will address key issues of importance to both adult and paediatric respiratory clinicians who are contemplating using F_ENO measurements in day to day practice.

RATIONALE FOR THE USE AND INTERPRETATION OF F_ENO MEASUREMENTS

A number of lines of evidence converge to provide the rationale for using F_ENO measurements in the assessment and management of respiratory disease. There are two key points: (1) there is a highly significant relationship between F_ENO and eosinophilic airway inflammation, and (2) there is an equally important relationship between eosinophilic airway inflammation and steroid responsiveness. The evidence is summarised as follows:

- F_ENO measurements are highly correlated with eosinophilic airway inflammation.
- Eosinophilic airway inflammation is associated with a positive response to steroid treatment.
- Raised F_ENO levels predict steroid responsiveness in patients with non-specific respiratory symptoms.

Abbreviations: AHR, airway hyperresponsiveness; CF, cystic fibrosis; COPD, chronic obstructive pulmonary disease; F_ENO, exhaled nitric oxide; FEV_1, forced expiratory volume in 1 second; ICS, inhaled corticosteroids; NO, nitric oxide; PCD, primary ciliary dyskinesia

- The use of inhaled corticosteroid (ICS) treatment in asthma results in a fall in F_ENO, and there is a dose-dependent relationship between ICS and F_ENO.

F_ENO measurements are highly correlated with eosinophilic airway inflammation

Atopic asthma is characterised by an inflammatory infiltrate in the airways, with a predominance of mast cells and eosinophils.[8] Studies confirm that F_ENO measurements correlate well with airway eosinophilia in induced sputum,[9 10] biopsy material,[11–13] and bronchoalveolar lavage fluid.[14] In one study a significant relationship between F_ENO and blood eosinophils was also reported.[15] A similar relationship has been described between F_ENO and sputum eosinophilic cationic protein[16] in patients with asthma.

Importantly, two studies have shown that the relationship between F_ENO levels and airway eosinophilia is independent of the clinical diagnosis. It has been reported in patients with chronic obstructive pulmonary disease (COPD).[17] In the study by Brightling *et al*[11] patients who did not fulfil criteria for the diagnosis of asthma but who had eosinophilic bronchitis had raised F_ENO levels. In atopic patients with allergic rhinitis but no asthma, F_ENO levels are also raised.[18–20] Similarly, in atopic asthmatic subjects in remission for many years, but who nevertheless have eosinophilic airway inflammation in bronchial biopsies, F_ENO levels are increased.[13] All of these data form the basis on which F_ENO measurements are considered reliable as a non-invasive marker of eosinophilic airway inflammation.

Eosinophilic airway inflammation is associated with a positive response to steroid treatment

Treatment with corticosteroids results in a reduction in airway eosinophilia in asthma and a simultaneous improvement in clinical parameters.[21 22] In contrast, in asthma which is not characterised by eosinophilia (at least in sputum), the response to steroids is likely to be poor.[23 24] These findings also apply in patients with fixed airflow obstruction in whom neither the history nor physiological measurements permit easy discrimination between asthma and COPD. In such patients, a positive outcome with a trial of steroid treatment is associated with the presence of sputum eosinophilia.[25 26] Thus, assessing the character of airway inflammation (eosinophilia) appears to be important in the initial management of patients with chronic respiratory symptoms in order to identify those who are more likely to benefit from treatment with steroids.

Raised F_ENO levels predict steroid responsiveness in patients with non-specific respiratory symptoms

Little *et al*[27] have shown that the clinical benefit of increased steroid treatment in patients with asthma is greatest in patients with raised F_ENO levels. This has been taken a step further by Smith *et al*[28] who evaluated the predictive accuracy of F_ENO measurements (as a surrogate for airway eosinophilia) in adult patients with undiagnosed respiratory symptoms. In that study, the positive and negative predictive values for a range of outcomes following a trial of inhaled fluticasone were superior for F_ENO as a predictor than spirometry, bronchodilator response, and measurements of AHR. Importantly, this study identified an optimum cut point for steroid response at an F_ENO of 47 ppb (fig 1). This outcome was largely independent of the final diagnosis. A similar result has been reported by Szefler *et al*[29] who showed that children with high F_ENO values are more likely to respond to ICS than children with lower F_ENO values.

ICS treatment in asthma results in a fall in F_ENO with a dose-dependent relationship between ICS and F_ENO

A number of studies have shown that ICS treatment results in a fall in F_ENO levels in patients with mild asthma.[30–32] Both the magnitude and the time interval over which the reduction occurs are dose-dependent[33 34] and the response is

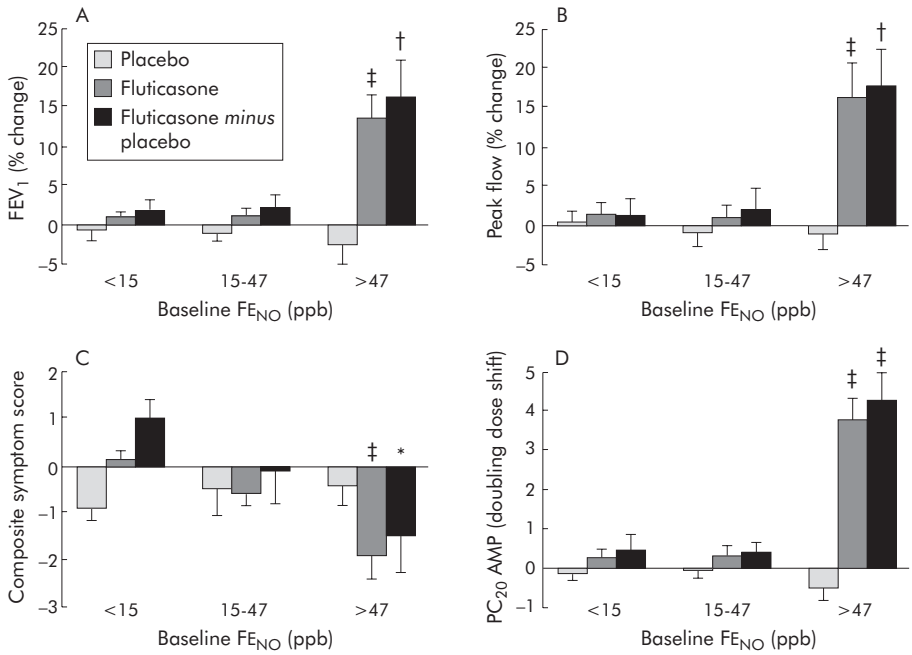

Figure 1 Steroid responsiveness in relation to F_ENO measurements in patients with non-specific chronic respiratory symptoms. Mean (SE) changes from baseline in (A) forced expiratory volume in 1 second (FEV_1), (B) morning peak flow over last 7 days of treatment, (C) composite symptom score, and (D) provocative concentration of adenosine monophosphate causing a 20% fall in FEV_1 (PC_{20}AMP) following treatment with inhaled fluticasone 500 μg/day (minus change with placebo), stratified by baseline F_ENO expressed as tertiles. Comparisons between tertiles were performed using one way analysis of variance with linear contrasts to identify any trend across the three tertiles; *p<0.05; †p<0.01; ‡p<0.001. Reproduced from Smith *et al*[28] with permission of the publishers.

reproducible.[35] $F_{E}NO$ levels tend to plateau at higher doses of ICS.[36] In addition, there are highly significant correlations between the *changes* in $F_{E}NO$ and *changes* in induced sputum eosinophils with ICS therapy.[37]

Taken together, these data provide foundational evidence that $F_{E}NO$ measurements have a potentially important role in evaluating and treating patients with airways disease. Firstly, $F_{E}NO$ may be used as a surrogate marker for airway diseases characterised by eosinophilia such as atopic asthma, cough variant asthma, and eosinophilic bronchitis. Secondly, because of the close relationship between steroid responsiveness and airway eosinophilia (in contrast to other histological phenotypes), $F_{E}NO$ measurements have a role in predicting and monitoring the response to ICS treatment.

DIAGNOSING AIRWAYS DISEASE

Establishing a diagnosis is the first step in clinical management but, for diseases of the airways, a diagnostic label has its limitations. The term ''chronic obstructive airways disease'' (COPD) encompasses a spectrum of overlapping pathologies and the phenotype is a mixed one, especially in relation to treatment. The same is true for bronchial asthma, which is increasingly acknowledged to be heterogeneous,[38 39] particularly if it is severe.[40 41] Against this background, and given the specificity of $F_{E}NO$ measurements as a marker for eosinophilic airway inflammation, it is not surprising that $F_{E}NO$ offers advantages as well as limitations as a ''test for asthma''.

Asthma

In adults, $F_{E}NO$ measurements are helpful in discriminating asthma from non-asthma.[42] It is best to reserve the test for investigating chronic symptoms (of 6 weeks duration or longer) because viral illness may give rise to a false positive result.[43 44] In the study by Dupont *et al*,[45] among 240 non-smoking steroid naïve individuals of whom 160 (67%) fulfilled the criteria for the diagnosis of asthma, $F_{E}NO$ levels were highly predictive of asthma with a sensitivity and specificity of 85% and 90%, respectively. In the study by Smith *et al*,[46] similar sensitivity (88%) and specificity (79%) were obtained in 47 patients of whom 17 had asthma. Predictive values were almost identical to those obtained using induced sputum cell counts. A striking feature in that study was the poor performance of almost all the ''conventional'' diagnostic tests against which $F_{E}NO$ measurements were compared. This reflects the fact that, in unselected patients, most will have mild disease with normal lung function. In this setting $F_{E}NO$ measurements may therefore be more relevant than traditional lung function tests. Interestingly, the *combination* of a raised $F_{E}NO$ ($>$33 ppb) and abnormal spirometry (FEV_1 $<$80% predicted) provides even greater sensitivity (94%) and specificity (93%) for the diagnosis of asthma.[46 47]

It is important to remember that patients may fulfil conventional clinical criteria for the diagnosis of asthma and yet $F_{E}NO$ levels will be normal, especially in non-atopic subjects. Normal values do not exclude the diagnosis of asthma. Measuring AHR may reveal a positive clinically relevant result. Thus, $F_{E}NO$ measurements complement AHR rather being a substitute for it, both in population surveys[48] and in patients with asthma.[49] This highlights further the fact that the asthma phenotype is heterogeneous, and that $F_{E}NO$ measurements provide a perspective on only one aspect of the ''asthma syndrome''.

Non-specific respiratory symptoms

$F_{E}NO$ measurements have a wider role in assessing patients with *undiagnosed* chronic respiratory symptoms. There is a broad differential diagnosis in such patients depending on age. It includes eosinophilic bronchitis, cough variant asthma, post-viral bronchial hyperresponsiveness, postnasal drip and other ENT problems, gastro-oesophageal reflux disease, vocal cord dysfunction, primary hyperventilation syndrome, and COPD. In children, recurrent wheezy bronchitis, cystic fibrosis, congenital abnormalities of the airways or lungs, and primary ciliary dyskinesia also need to be considered.

$F_{E}NO$ measurements may also permit the clinician to anticipate treatment responses. For eosinophilic bronchitis and cough variant asthma, which are characterised by eosinophilic airway inflammation and increased $F_{E}NO$ levels, a positive response to a trial of steroid treatment is likely.[11 50] On the other hand, for other diagnoses—for example, vocal cord dysfunction presenting as ''asthma'' which clinicians often treat empirically with steroids with little meaningful benefit[51]—it is just as helpful to have a low normal $F_{E}NO$ level indicating a condition which is *not* characterised by eosinophilic airway inflammation and, in turn, is less likely to respond to steroids.

Preschool children

Given that spirometry and sputum induction cannot easily be performed in preschool children, a non-invasive measurement of airway inflammation is potentially very useful. As the single breath technique for measuring $F_{E}NO$ in this age group is not suitable in preschool children, several alternatives have been developed, varying from modifications of the standard online technique to offline tidal breathing methods without flow control.[52–61] In general, these techniques (which were reviewed by an ERS/ATS Task Force) are less sensitive in discriminating between asthmatic and non-asthmatic subjects.[62 63]

Evidence as to the overall diagnostic usefulness of $F_{E}NO$ measurements in young children is mixed. In an unselected population of preschool children too young to perform spirometric tests, $F_{E}NO$ performed poorly in distinguishing between asthma and non-asthma. Differences in $F_{E}NO$ values between atopic children, children with doctor diagnosed asthma, and healthy children were less pronounced than in older subjects.[64] However, when used in selected children, the performance characteristics are somewhat improved.[65]

In the differential diagnosis of non-specific respiratory symptoms, the same issues are encountered. Baraldi *et al* studied a group of 13 young children with recurrent wheeze and compared their $F_{E}NO$ values with those of nine healthy controls and six children with a first episode of wheezing.[53] Exhaled air was collected offline in a bag during tidal breathing without flow control. During an acute episode, $F_{E}NO$ was significantly higher in those with recurrent wheeze than in controls, while in children with their first episode of wheezing $F_{E}NO$ levels did not differ from normal children. These data are in keeping with those of Ratjen *et al*[60] who measured peak $F_{E}NO$ values online in mixed exhaled air (from mouth and nose).

A test that might allow better targeting of anti-inflammatory treatment, particularly in preschool children, would be very helpful. $F_{E}NO$ is a promising tool in this regard. Treating infants and young children with recurrent wheeze and increased $F_{E}NO$ levels with corticosteroids reduced $F_{E}NO$ to normal or near normal values.[53 66] Also, montelukast reduced $F_{E}NO$ values in young children with early onset asthma.[67 68]

Influence of atopy

Several epidemiological studies have confirmed that $F_{E}NO$ levels are raised in atopic subjects, whether or not they have significant lower respiratory tract symptoms.[18 20 64 69–72] There is also a strong correlation between $F_{E}NO$ levels and total as well as antigen specific IgE.[71 72] This overall picture may be explained by the fact that even asymptomatic atopic patients

may have mild airway inflammation,[13 73] giving rise to increased $F_{E}NO$ levels.

It has been suggested that the usefulness of $F_{E}NO$ measurements may be limited only to atopic subjects, but we disagree with this view. Firstly, not all atopic individuals are identified using skin prick testing—that is, the label ''non-atopic'' may be falsely negative. Secondly, the presence of a low/normal $F_{E}NO$ level in patients with chronic respiratory symptoms may be equally helpful in pointing the clinician *away* from the diagnosis of an atopic condition. In practice, when raised $F_{E}NO$ levels are encountered in atopic subjects, additional investigations or treatment should be based on a history of significant symptoms. There is little evidence at present to support intervention in asymptomatic individuals.

Chronic obstructive pulmonary disease (COPD)

$F_{E}NO$ levels are inconsistent in patients with COPD. This may be due to the confounding effect of current smoking or it may reflect the heterogeneity of underlying airway inflammation. Some studies report no significant change in $F_{E}NO$ levels compared with controls,[74 75] while others report that levels are increased.[76] More recent evidence suggests that measuring alveolar rather than airway $F_{E}NO$ may yield more important information,[77] but at present this is technically demanding and beyond the scope of routine laboratory testing.

In older patients (>45 years) with *fixed* airflow obstruction, physiological tests alone are unhelpful in distinguishing those with asthma who would otherwise be classified as having COPD. Fabbri *et al*[17] have shown that patients with historical evidence of asthma have eosinophilic airway inflammation in association with raised $F_{E}NO$ levels. Earlier, Papi *et al*[78] reported that increased sputum eosinophils and $F_{E}NO$ levels occur in COPD patients with greater degrees of bronchodilator reversibility.

Perhaps the most important question is not whether the diagnostic label is accurate, but whether the response to anti-inflammatory treatment can be predicted. The data provided by Brightling *et al*,[26] in which induced sputum eosinophil counts were used as the predictor, are encouraging. In that study 22 of 67 patients with COPD whose induced sputum eosinophil count was in the uppermost tertile (>4.5%) had significant symptomatic as well as physiological improvements with oral prednisone. Using $F_{E}NO$ measurements, Zietkowski *et al*[76] showed that the increase in post-bronchodilator FEV_1 after 2 months of open label treatment with inhaled budesonide 800 µg/day was strongly correlated ($r = 0.73$, $p = 0.0003$) with baseline $F_{E}NO$ levels in 19 ex-smoking patients with COPD. However, statistically significant correlations are not the same as predictive accuracy. There is a need for more work to be done to establish the exact role of $F_{E}NO$ measurements in assessing COPD. This will require larger randomised controlled studies.

Table 1 Respiratory and non-respiratory conditions in which $F_{E}NO$ measurements may have a role in diagnosis

Increased $F_{E}NO$	Variable changes in $F_{E}NO$ reported	Decreased $F_{E}NO$
Asthma[1 79]	Bronchiectasis[91–93]	Cystic fibrosis[91 108–110]
Late asthmatic response[80 81]	COPD[17 75 78 94–102]	Primary ciliary dyskinesia[111 112]
Allergic rhinitis[19]	Fibrosing alveolitis[103]	Pulmonary hypertension[113]
Viral infections[43 44 82]	Sarcoidosis[104]	HIV infection[114]
Hepatopulmonary syndrome[83]	Systemic sclerosis[105–107]	ARDS[115]
Liver cirrhosis[84 85]		
Acute/chronic rejection of lung transplant including bronchiolitis obliterans[86–90]		

OTHER DISEASES IN WHICH $F_{E}NO$ MAY HAVE A ROLE

Besides the common airways diseases, $F_{E}NO$ measurements may have a role in the assessment of several other respiratory and non-respiratory conditions (table 1).

Cystic fibrosis (CF)

In patients with CF, $F_{E}NO$ measurements have not been found to be clinically helpful. Values are usually normal or low.[108–110] There are several possible explanations. Firstly, there is decreased expression of nitric oxide synthase (NOS-2) in patients with CF.[116 117] Secondly, increased levels of nitrite are found in the breath condensate of patients with CF, suggesting trapping and metabolism of NO in secretions and mucus in CF airways.[118 119]

Primary ciliary dyskinesia (PCD)

$F_{E}NO$ levels are significantly lower in patients with PCD than in healthy individuals, although with some overlap.[111 120 121] Moreover, nasal NO (nNO) is *extremely* low in patients with PCD of all ages, and discriminates fully between affected and unaffected individuals. Measurement of nNO is likely to become the screening tool of choice.[122] The diagnostic sensitivities and specificities of nNO for PCD range from 89% to 100% and from 97% to 100%, respectively. Low $F_{E}NO$ and nNO levels may also be found in subjects with non-PCD bronchiectasis and sinus disease.[112 121]

Again, there are several possible explanations. Firstly, there may be decreased NOS activity. Administration of L-arginine as a substrate for NO increases nasal and exhaled NO formation in PCD, although not to normal values.[123 124] This favours decreased NOS activity as a mechanism. Secondly, mucus may impair the diffusion of NO from the sinuses to the nasal cavity or from epithelial cells to the airway lumen, or may alter NO elimination.[125] However, even in young infants with PCD, nNO is low, favouring the first explanation.[126]

NO is probably involved in stimulating ciliary motility.[127] Nasal NO may also play a role in non-specific host defences, including direct toxic effects on micro-organisms.[128] Reduced endogenous NO production and damage to NO producing cells may therefore contribute to recurrent airway infections.

Lung transplantation

$F_{E}NO$ levels are increased in post-transplant patients with unstable lung function.[87 88] More recent studies have investigated whether sequential $F_{E}NO$ measurements can identify patients with progressive bronchiolitis obliterans syndrome (BOS).[89 90] In a study by Brugiere *et al*,[90] mean $F_{E}NO$ levels were twice as high in patients with progressive BOS than in those with or without BOS whose lung function remained stable over 14 months. Verleden *et al*[89] evaluated the performance characteristics of $F_{E}NO$ measurements over 2 years and obtained sensitivity, specificity, positive and negative predictive values for BOS of 92%, 84%, 80%, and 94%, respectively. The cut point used was 15 ppb (at an expiratory flow rate of 200 ml/s). This was equivalent to the upper limit of the 95% confidence interval for mean $F_{E}NO$ levels in stable transplant patients. Interestingly, increased $F_{E}NO$ levels preceded the changes in lung function by approximately 9 months. Although promising, the exact role of $F_{E}NO$ measurements in post-transplant monitoring is not yet established.

$F_{E}NO$ MEASUREMENTS IN THE MANAGEMENT OF CHRONIC ASTHMA

Two important questions have emerged regarding $F_{E}NO$ measurements in the ongoing management of asthma:

- Does F_{ENO} have prognostic significance?
- Can F_{ENO} be used to guide treatment decisions relating to anti-inflammatory treatment?

Predicting exacerbations

Asthma is characterised by relapses and remissions, with deterioration in control provoked by a number of triggers as well as due to poor compliance with anti-inflammatory therapy. There is a perceived need for an objective measurement which might provide warning of impending deterioration or the need to change treatment. Peak flow measurements have been used to fulfil this role, but with limited success because changes in peak flow largely coincide with deteriorating symptoms rather than precede them.

Overall, the prognostic value of F_{ENO} measurements to predict deteriorating asthma appears limited. In a small study involving a steroid reduction protocol, Jatakanon *et al*[129] reported that changes in sputum eosinophils were superior to F_{ENO} measurements in predicting loss of control. In a study by Jones *et al*[37] measurements of AHR to hypertonic saline, sputum eosinophils, and F_{ENO} measurements all ranked similarly as predictors of control in 78 asthma patients following ICS withdrawal. Sensitivities ranged from 21% (eosinophils >4%) to 65% (F_{ENO} >10 ppb at a flow rate of 250 ml/s), although positive predictive values were in the range of 80–88%. Interestingly, the measurement of *changes* in these measurements was only marginally better than using single measurements. An increase in F_{ENO} of 60% was deemed to be optimum, but this was only 50% sensitive with a positive predictive value of 83%. These studies used a steroid withdrawal protocol to mimic a clinical exacerbation and are not necessarily ideal. In a much smaller study Harkin *et al*[130] reported that, in routine practice, increased levels of F_{ENO} predicted an exacerbation within the following 2 weeks. It may be that, with the advent of portable monitoring, daily F_{ENO} measurements may prove to be beneficial in anticipating deteriorating asthma. However, as yet no data are available.

Predicting the outcome of ICS withdrawal in stable asthma

A relevant question is whether markers of airway inflammation can be used to predict the successful reduction or withdrawal of ICS treatment. In studies by Leuppi *et al*[131] and Deykin *et al*,[132] while sputum eosinophil counts (>0.8%) were highly predictive of subsequent loss of asthma control over periods of 6 months and 16 weeks respectively, no prognostic significance could be derived from F_{ENO} measurements. In the first study,[131] baseline rather than sequential F_{ENO} values were used in the calculations. In the second, the number of patients in whom F_{ENO} values were obtained was limited, making valid comparisons difficult.[132] In the study by Zacharasiewicz *et al*[133] the negative predictive value of sputum eosinophils (at a cut point of 0%) was 100%—that is, treatment reduction/withdrawal was 100% successful (during the subsequent 8 weeks) when sputum eosinophilia was absent. A negative predictive value of 92% was obtained for F_{ENO} at a cut point of 22 ppb or less. Focusing on F_{ENO}, Pijnenburg *et al*[134] reported that, following steroid withdrawal in currently asymptomatic children, F_{ENO} levels 2 and 4 weeks later were highly predictive of relapse during the subsequent 24 weeks of follow up, with a cut point for F_{ENO} of 49 ppb providing best predictive accuracy—that is, F_{ENO} levels above this threshold predicted likely asthma relapse.

Taken together, we can conclude that sputum eosinophil counts (>1%) probably offer superior prognostic accuracy when evaluating whether or not patients require ongoing ICS treatment. Furthermore, in circumstances where induced sputum cannot be obtained (in the majority of centres and in young children), a high F_{ENO} level (>50 ppb) is likely to predict asthma relapse and a low F_{ENO} level (<20 ppb in children, <25 ppb in adults) is likely to predict asthma stability if measured at least 4 weeks after ICS treatment is reduced/withdrawn in a currently asymptomatic patient. The outcome in those with an intermediate result (F_{ENO} 20–50 ppb) is less certain.

Adjustment of ICS dose

Several studies have recently explored whether ''inflammometry'' can be used to optimise the dose of ICS treatment. Using induced sputum eosinophil counts, Green *et al*[24] showed that a management strategy which prescribed a stepwise reduction in ICS dose if sputum eosinophils were <1% (or an increase in dose if >3%) reduced asthma exacerbations to 32% of the rate observed in the control group. In another randomised study Jayaram *et al*[133] have shown that, when ICS treatment is adjusted to maintain sputum eosinophils below 2%, the risk of *eosinophilic* exacerbations was reduced significantly (by 49%), with the number requiring intervention with prednisone reduced by two thirds.[135] Interestingly, in that study the benefits of the ''inflammometry'' strategy occurred predominantly in patients with moderate or severe asthma.

The underlying rationale for each of these studies is both plausible and desirable—that is, anti-inflammatory treatment should be adjusted to ensure minimum airway inflammation. Currently, clinicians respond to uncontrolled symptoms or impaired lung function assuming that this results from uncontrolled airway inflammation. But the correlation between airway inflammation and either symptoms[13 136] or lung function is weak,[15 137] so the use of these end points to guide treatment can only be regarded as second best. It is far more rational that the ICS dose should be

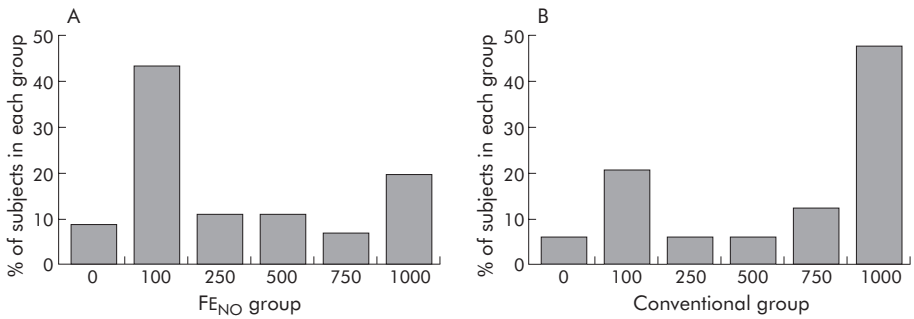

Figure 2 Profile of dose distribution for mean inhaled fluticasone requirements over 12 months in 48 patients (conventional group) whose ICS dose was adjusted using a priori guidelines and 46 patients in whom the ICS dose was adjusted on the basis of F_{ENO} measurements (F_{ENO} group; cut point equivalent to 35 ppb). There was a highly significant difference between the two groups (p = 0.008).[138]

adjusted to take account of the principal action of ICS treatment—namely, control of airway inflammation.

Two randomised controlled trials in which $F_{E}NO$ measurements have been used to guide long term treatment with ICS have recently been published.[32 138] In both studies significant but differing benefits were obtained using an $F_{E}NO$ based algorithm. In a study of 94 adult asthmatic patients by Smith *et al*,[138] a 40% reduction in ICS dose requirements was achieved using $F_{E}NO$ measurements at a single cut point of 35 ppb without any significant difference in the rate of asthma exacerbations (fig 2). In a study by Pijnenburg *et al*[32] comprising 85 children with atopic asthma, the cut point for $F_{E}NO$ levels was similar at 30 ppb; there was no difference in cumulative ICS use between the $F_{E}NO$ and control groups. However, in the $F_{E}NO$ group there was a significant reduction in the severity of AHR, with a concomitant (but, for reasons of study size, non-significant) reduction in exacerbations requiring oral prednisone.

Variation in the dose titration protocols and the study end points may account for the apparent differences in the results. However, overall, these studies provide encouraging evidence regarding the use of $F_{E}NO$ in this clinical setting, although for the time being they must be regarded as indicative rather than conclusive. Firstly, in the study by Smith *et al*[138] a single cut point for $F_{E}NO$ was used to prompt either an increase or a decrease in ICS dose. However, a ''one size fits all'' approach may not be appropriate in regular clinical practice. Furthermore, although single cut points are appropriate in ''back titration'' studies with a high starting dose of ICS, *two* cut points defining *three* management choices (increase, no change or decrease in dose) may be more effective. This concept was included in the algorithm used by Pijnenburg *et al*[32] but, clearly, this is an area which requires further investigation. Secondly, although the cut point for $F_{E}NO$ was similar in both studies, substantially different criteria were used to guide ICS dose adjustment in the control groups. Irrespective of where the cut points are set, these will significantly determine the outcome in any dose adjustment strategy. This may explain why differing yet beneficial outcomes have been achieved in the ''strategy'' groups in the studies reported to date.[24 32 138]

An important and as yet unresolved issue is whether $F_{E}NO$ measurements should be used for both upwards as well as downwards ICS dose titration. Clearly, the withdrawal of unnecessary ICS treatment or reducing excessive doses is an important goal of $F_{E}NO$ monitoring. This was the primary objective in the study by Pijnenburg *et al*.[32] In the study by Smith *et al*[138] ICS dose reduction was a secondary end point but proved to be the most important outcome. Thus, data to date suggest that dose reduction is a fairly achievable objective. However, in patients with persistently high $F_{E}NO$ levels (>50 ppb), it remains to be determined whether *increasing* the dose of ICS further will prove to be successful. It is doubtful whether it is justified if the patient is asymptomatic. However, although abolishing symptoms is a valid objective, it is not the only one. Higher doses of ICS reduce the frequency and impact of asthma exacerbations, and it may be that persistently raised $F_{E}NO$ levels represent the signal to prescribe higher ICS doses with this objective in mind. This is controversial. On the one hand, the effect of ICS dose increments in the higher range on $F_{E}NO$ is often small. Also, it is often not possible to normalise $F_{E}NO$ values in patients with persistently high $F_{E}NO$ levels, even on maximum doses.[139] Equally, the cost-benefit ratio for very high doses of ICS in asthma increases. Thus, until further studies have been completed, it seems prudent to put the emphasis on dose reduction when $F_{E}NO$ levels are low (<25 ppb). Only if asthma is poorly controlled and issues of poor compliance and/or poor inhaler technique have been addressed should high $F_{E}NO$ levels prompt an increase in ICS dose.

MEASURING AND INTERPRETING $F_{E}NO$

Obtaining $F_{E}NO$ measurements using commercially available analysers is, like riding a bicycle, fairly straightforward when you know how. The majority are ''online''—that is, the analysis of exhaled gas is immediate and a result is available within a few minutes. Updated recommendations have recently been published and the reader is strongly advised to refer to them for fuller information.[62 63] Establishing reference values for $F_{E}NO$ measurements has been an evolving issue and remains problematic. Most of the studies conducted up to the year 2000 did not include a standardised method for $F_{E}NO$ measurement. This severely limits their generalisablity. Further, our knowledge of factors which affect $F_{E}NO$ levels in health and disease has expanded steadily. Demographic factors include age (adult versus paediatric) and smoking status.[140–142] There is still some disagreement about whether atopy[143] and sex[144–146] are consistently important.

Normal values and clinically important changes

Against this background, current guidelines do not yet specify ''normal'' values.[63] However, several recent studies have attempted to provide reference ranges for adults[143 145 146] and children.[145] In the study by Olin *et al*[146] the interquartile range for $F_{E}NO$ in healthy adults was 11.9–22.4 ppb. In a study comprising 30 healthy non-atopic adult subjects, the upper limit of normal (mean plus two standard deviations) was 33.1 ppb.[146] In the study by Buchvald *et al*[145] in a population of 405 children, the upper limit of the 95% confidence interval was age dependent, ranging from 15.7 ppb at the age of 4 years to 25.2 ppb for adolescents. The reason for this age dependency is unknown, but may be related to increasing airway surface area with age, age dependent induction of NOS secondary to recurrent immunological stimulation, or the progressive reduction over time of a constant exhalation flow rate which is relatively high in younger children. For the time being at least, it is reasonable that upper limits of normal for healthy adults and school age children should be set at 33 ppb and 25 ppb, respectively.

The reproducibility of $F_{E}NO$ measurements is excellent with a very high intraclass correlation (>0.9) for repeated within sitting measurements.[146] This translates into a within subject standard deviation for repeated measurements of 2.1 ppb for adults[146] and 1.6 ppb for children[145] (both at a flow rate of 50 ml/s). Similar high degrees of reproducibility have been reported by other authors.[86 147–149]

In clinical practice, as far as repeated measurements are concerned, the population of greatest interest are patients with chronic asthma. In this group the coefficient of variation for within subject between sitting measurements ranges from 10.5%[37] to 26%.[148] There is no ''normal range'' for patients with asthma. However, anecdotally, it would appear that even when asthma is well controlled, non-asthmatic ''normal'' $F_{E}NO$ levels are rarely achieved. Thus, it may be that $F_{E}NO$ levels measured when asthma is stable become the baseline reference point for individual patients against which subsequent measurements are weighed.

Kharitonov *et al*[146] have reported that an absolute change of >4 ppb is significant. Based on the study by Ekroos *et al*,[148] a percentage change of >26% would be deemed statistically significant. But are absolute or percentage changes of >4 ppb or >26% *clinically* important? And, if not, what constitutes a clinically meaningful change? In the study by Jones *et al*[37] the median change in $F_{E}NO$ which occurred between stability and ''loss of control'' after withdrawal of ICS treatment was 16.9 ppb (mean 24.9), but with a very large range (−10 to

Table 2 $F_{E}NO$ levels as an aid to diagnosis of chronic respiratory symptoms

$F_{E}NO$ (ppb)	Range	Eosinophilic airway inflammation	Interpretation (as an aid to diagnosis of chronic respiratory symptoms)	
			Adults	**Children**
5 10 15 20 25	Low (<20 ppb if 12 years or younger; <25 ppb for adults)	Unlikely	Consider: Neutrophilic asthma Anxiety/hyperventilation Vocal cord dysfunction Rhinosinusitis Gastro-oesophageal reflux Cardiac disease	Consider: Wheezy bronchitis Gastro-oesophageal reflux ENT disorders Cystic fibrosis Primary ciliary dyskinesia ($F_{E}NO$ <5 ppb), (check nasal NO) Congenital abnormalities, e.g. airway malacia Other immunodeficiencies
30 35 40 45	Intermediate	Present but mild	Interpretation based on clinical presentation	Interpretation based on clinical presentation
50 55 60 65 Higher levels	High	Significant	Consider: Atopic asthma *if the history is appropriate.* If FEV_1 <80% predicted, diagnosis of asthma is very likely Eosinophilic bronchitis Churg-Strauss syndrome A positive response to a trial of inhaled or oral steroid is likely. In ex-smokers with COPD this may also be true	If combined with any objective evidence of reversible airway obstruction, asthma is very likely and a positive response to a trial of inhaled or oral steroids is likely

Table 3 $F_{E}NO$ levels as an aid in the management of asthma

$F_{E}NO$ (ppb)	Range	Eosinophilic airway inflammation	Interpretation (as an aid in the management of asthma)	
			Adults	**Children**
5 10 15 20 25	Low	Unlikely	If *symptomatic*, review diagnosis Neutrophilic asthma Anxiety/hyperventilation Vocal cord dysfunction Rhinosinusitis Gastro-oesophageal reflux	If *symptomatic*, review diagnosis Consider also: Wheezy bronchitis Cystic fibrosis Congenital abnormalities, e.g. airway malacia Primary ciliary dyskinesia
			If *asymptomatic* and taking ICS: Implies good compliance with treatment. Reduce dose or, in case of low ICS dose, even withdraw ICS altogether	If *asymptomatic* and taking ICS: as for adults
30 35 40	Intermediate	Present but mild	If *symptomatic*, consider: Infection as reason for worsening High levels of allergen exposure Adding in other therapy apart from ICS (e.g. long acting β agonist) Consider ICS dose increase	If *symptomatic* (besides considerations in adults), consider: Possible inadequate ICS treatment (1) check compliance (2) check for poor inhaler technique and consider metered dose inhaler and spacer if patient is currently using a dry powder device
			If *asymptomatic* No change in ICS dose if patient is stable	If *asymptomatic*: as for adults
45 50 55 60 65 Higher levels	High (or rise of 60% or more since previous measurement)	Significant	If *symptomatic*, consider: Inadequate ICS treatment: (1) check compliance (2) check for poor inhaler technique (3) inadequate ICS dose Continuous high level allergen exposure Imminent exacerbation or relapse depending on history of individual patient. More likely if ICS dose is zero Steroid resistance (rare)	If *symptomatic* (besides considerations in adults) consider: Metered dose inhaler and spacer if patient is currently using a dry powder device
			If *asymptomatic* No change in ICS dose if patient is stable	If *asymptomatic*: as for adults

+141 ppb). The predictive accuracy of a change from baseline of 60% or greater was limited. These data suggest that group mean data may not be helpful in determining a clinically relevant change in individual patients. Further work is required to address this issue, particularly as to the changes (absolute and percentage) which might be anticipated in patients experiencing an asthma exacerbation while continuing to take regular controller therapy.

Interpretation

Based on currently available data, we have recently developed two algorithms for interpreting $F_{E}NO$ results in day to day practice—one for diagnostic use and the other for ongoing asthma management. These are shown in tables 2 and 3.

Diagnostic use is fairly straightforward (table 2). For regular monitoring in subjects with chronic asthma (table 3), raised $F_{E}NO$ levels in a *symptomatic* patient indicate uncontrolled eosinophilic airway inflammation. This is most frequently due to poor compliance with anti-inflammatory treatment or poor inhaler technique rather than inadequate ICS dosing. Although poor inhaler technique resulting in inadequate drug deposition is a plausible reason for raised $F_{E}NO$ values, in a study of asthmatic children on a median dose of 800 μg budesonide this could not be proven.[139] Where $F_{E}NO$ levels remain high despite a seemingly adequate inhaled drug regime, it is theoretically possible that overexpression of constitutive steroid resistant NOS may be the explanation. Alternatively, alveolar NO rather than airway NO may be the source[150 151] and, in such circumstances, a better clinical response may be achieved using oral rather than inhaled treatment.[152] This remains controversial. Only rarely does a persistently high $F_{E}NO$ level indicate true steroid resistance.

A low $F_{E}NO$ level implies the absence of eosinophilic airway inflammation and, assuming that the result is not confounded by current tobacco smoking which may reduce $F_{E}NO$ levels by up to 60%, an alternative or additional diagnosis to atopic asthma should be considered if the patient is symptomatic. The more common examples include non-atopic (possibly neutrophilic) asthma, gastro-oesophageal reflux disease, rhinosinusitis with postnasal drip, and left ventricular dysfunction.

The information contained in tables 2 and 3 is intended for guidance only. Future studies may indicate the need for modifications. Also, as indicated previously, patients with different clinical phenotypes may have different baseline values and different ''target'' $F_{E}NO$ levels may be appropriate. This is because even when asthma is stable, $F_{E}NO$ levels may remain high. As a strategy, evidence that ''normalising'' the $F_{E}NO$ results in clinical benefit has not yet been documented. Rather, individualised ''$F_{E}NO$ typing'' and cut points may be required. In some steroid dependent patients with asthma we have found it appropriate to devise a ''sliding scale'' which relates oral prednisone dose requirements to changes in $F_{E}NO$.

CONCLUSIONS

$F_{E}NO$ measurements offer a step forward in the assessment of airways disease. As an ''inflammometer'', $F_{E}NO$ provides the clinician with hitherto unavailable information regarding the nature of underlying airway inflammation, thus complementing conventional physiological testing, including the measurement of AHR. $F_{E}NO$ measurements are easy to perform, reproducible, and technically less demanding than induced sputum analysis. They are unreliable in current smokers and, when used diagnostically, in patients who have been taking inhaled or oral steroids recently.

$F_{E}NO$ results require careful reference to the clinical context. In symptomatic patients, high $F_{E}NO$ levels (>50 ppb) indicate significant airway eosinophilia which is likely to respond to ICS treatment. This appears to be independent of the diagnostic label. Further work is required to confirm how $F_{E}NO$ measurements should be interpreted in patients with probable COPD. Present data provide support for the diagnostic use of $F_{E}NO$ measurements in children with asthma-like symptoms, but in the very young more evidence is required. Whether or not $F_{E}NO$ may be used to predict steroid response or guide ICS dose requirements in young children with recurrent wheeze is still unclear.

In patients with chronic and/or severe asthma, $F_{E}NO$ levels are helpful to determine whether or not eosinophilic airway inflammation is currently active. Both high (>50 ppb) and low (<25 ppb) $F_{E}NO$ levels may be used to predict outcomes in patients with a definite history of asthma currently in remission, and in whom withdrawal of ICS therapy is being undertaken. Again, depending on the level of symptoms, both high and low $F_{E}NO$ levels offer the clinician information which may help to guide ICS dose adjustment decisions. As yet, however, much more work needs to be done before intermediate values based on group mean data can be used with complete confidence in this setting. The advantages of sequential individual data needs further study.

Authors' affiliations

D R Taylor, A D Smith, Department of Medical and Surgical Sciences, Dunedin School of Medicine, University of Otago, P O Box 913, Dunedin, New Zealand

M W Pijnenburg, J C D Jongste, Department of Paediatric Respiratory Medicine, Erasmus MC - Sophia Children's Hospital, University Medical Centre, P O Box 2060, 3000 CB Rotterdam, The Netherlands

Competing interests: Professor Taylor has received funding from Aerocrine, a manufacturer of nitric oxide analysers.

Note: Unless otherwise stated, all $F_{E}NO$ measurements are reported in parts per billion at a flow rate of 50 ml/s. In some instances corrections for flow rate have been made to ensure consistency and permit appropriate interpretation by the reader.

REFERENCES

1 **Alving K**, Weitzberg E, Lundberg JM. Increased amount of nitric oxide in exhaled air of asthmatics. *Eur Respir J* 1993;**6**:1368–70.
2 **Ignarro LJ**. Nitric oxide as a unique signaling molecule in the vascular system: a historical overview. *J Physiol Pharmacol* 2002;**53**:503–14.
3 **Ricciardolo FL**. Multiple roles of nitric oxide in the airways. *Thorax* 2003;**58**:175–82.
4 **Nijkamp FP**, Folkerts G. Nitric oxide and bronchial hyperresponsiveness. *Arch Int Pharmacodyn Ther* 1995;**329**:81–96.
5 **De Sanctis GT**, MacLean JA, Hamada K, *et al*. Contribution of nitric oxide synthases 1, 2, and 3 to airway hyperresponsiveness and inflammation in a murine model of asthma. *J Exp Med* 1999;**189**:1621–30.
6 **Lane C**, Knight D, Burgess S, *et al*. Epithelial inducible nitric oxide synthase activity is the major determinant of nitric oxide concentration in exhaled breath. *Thorax* 2004;**59**:757–60.
7 **Kharitonov SA**, Barnes PJ. Clinical aspects of exhaled nitric oxide. *Eur Respir J* 2000;**16**:781–92.
8 **Djukanovic R**, Roche WR, Wilson JW, *et al*. Mucosal inflammation in asthma. *Am Rev Respir Dis* 1990;**142**:434–57.
9 **Jatakanon A**, Lim S, Kharitonov SA, *et al*. Correlation between exhaled nitric oxide, sputum eosinophils, and methacholine responsiveness in patients with mild asthma. *Thorax* 1998;**53**:91–5.
10 **Berlyne GS**, Parameswaran K, Kamada D, *et al*. A comparison of exhaled nitric oxide and induced sputum as markers of airway inflammation. *J Allergy Clin Immunol* 2000;**106**:638–44.
11 **Brightling CE**, Symon FA, Birring SS, *et al*. Comparison of airway immunopathology of eosinophilic bronchitis and asthma. *Thorax* 2003;**58**:528–32.
12 **Payne DN**, Adcock IM, Wilson NM, *et al*. Relationship between exhaled nitric oxide and mucosal eosinophilic inflammation in children with difficult asthma, after treatment with oral prednisolone. *Am J Respir Crit Care Med* 2001;**164**:1376–81.
13 **van den Toorn LM**, Overbeek SE, de Jongste JC, *et al*. Airway inflammation is present during clinical remission of atopic asthma. *Am J Respir Crit Care Med* 2001;**164**:2107–13.
14 **Warke TJ**, Fitch PS, Brown V, *et al*. Exhaled nitric oxide correlates with airway eosinophils in childhood asthma. *Thorax* 2002;**57**:383–7.
15 **Strunk RC**, Szefler SJ, Phillips BR, *et al*. Relationship of exhaled nitric oxide to clinical and inflammatory markers of persistent asthma in children. *J Allergy Clin Immunol* 2003;**112**:883–92.

16 **Mattes J**, Storm van's Gravesande K, Reining U, *et al*. NO in exhaled air is correlated with markers of eosinophilic airway inflammation in corticosteroid-dependent childhood asthma. *Eur Respir J* 1999;**13**:1391–5.
17 **Fabbri LM**, Romagnoli M, Corbetta L, *et al*. Differences in airway inflammation in patients with fixed airflow obstruction due to asthma or chronic obstructive pulmonary disease. *Am J Respir Crit Care Med* 2003;**167**:418–24.
18 **Gratziou C**, Lignos M, Dassiou M, *et al*. Influence of atopy on exhaled nitric oxide in patients with stable asthma and rhinitis. *Eur Respir J* 1999;**14**:897–901.
19 **Henriksen AH**, Sue-Chu M, Lingaas Holmen T, *et al*. Exhaled and nasal NO levels in allergic rhinitis: relation to sensitization, pollen season and bronchial hyperresponsiveness. *Eur Respir J* 1999;**13**:301–6.
20 **Jouaville LF**, Annesi-Maesano I, Nguyen LT, *et al*. Interrelationships among asthma, atopy, rhinitis and exhaled nitric oxide in a population-based sample of children. *Clin Exp Allergy* 2003;**33**:1506–11.
21 **Djukanovic R**, Homeyard S, Gratziou C, *et al*. The effect of treatment with oral corticosteroids on asthma symptoms and airway inflammation. *Am J Respir Crit Care Med* 1997;**155**:826–32.
22 **Lim S**, Jatakanon A, John M, *et al*. Effect of inhaled budesonide on lung function and airway inflammation. Assessment by various inflammatory markers in mild asthma. *Am J Respir Crit Care Med* 1999;**159**:22–30.
23 **Pavord ID**, Brightling CE, Woltmann G, *et al*. Non-eosinophilic corticosteroid unresponsive asthma. *Lancet* 1999;**353**:2213–4.
24 **Green RH**, Brightling CE, Woltmann G, *et al*. Analysis of induced sputum in adults with asthma: identification of subgroup with isolated sputum neutrophilia and poor response to inhaled corticosteroids. *Thorax* 2002;**57**:875–9.
25 **Pizzichini E**, Pizzichini MM, Gibson P, *et al*. Sputum eosinophilia predicts benefit from prednisone in smokers with chronic obstructive bronchitis. *Am J Respir Crit Care Med* 1998;**158**:1511–7.
26 **Brightling CE**, Monteiro W, Ward R, *et al*. Sputum eosinophilia and short-term response to prednisolone in chronic obstructive pulmonary disease: a randomised controlled trial. *Lancet* 2000;**356**:1480–5.
27 **Little SA**, Chalmers GW, MacLeod KJ, *et al*. Non-invasive markers of airway inflammation as predictors of oral steroid responsiveness in asthma. *Thorax* 2000;**55**:232–4.
28 **Smith AD**, Cowan JO, Brassett KP, *et al*. Exhaled nitric oxide: a predictor of steroid response. *Am J Respir Crit Care Med* 2005;**172**:453–9.
29 **Szefler SJ**, Phillips BR, Martinez FD, *et al*. Characterization of within-subject responses to fluticasone and montelukast in childhood asthma. *J Allergy Clin Immunol* 2005;**115**:233–42.
30 **Kharitonov SA**, Yates DH, Barnes PJ. Inhaled glucocorticoids decrease nitric oxide in exhaled air of asthmatic patients. *Am J Respir Crit Care Med* 1996;**153**:454–7.
31 **Beck-Ripp J**, Griese M, Arenz S, *et al*. Changes of exhaled nitric oxide during steroid treatment of childhood asthma. *Eur Respir J* 2002;**19**:1015–9.
32 **Pijnenburg MW**, Bakker EM, Hop WC, *et al*. Titrating steroids on exhaled nitric oxide in children with asthma: a randomized controlled trial. *Am J Respir Crit Care Med* 2005;**172**:831–6.
33 **Kharitonov SA**, Donnelly LE, Montuschi P, *et al*. Dose-dependent onset and cessation of action of inhaled budesonide on exhaled nitric oxide and symptoms in mild asthma. *Thorax* 2002;**57**:889–96.
34 **Jones SL**, Herbison P, Cowan JO, *et al*. Exhaled NO and assessment of anti-inflammatory effects of inhaled steroid: dose-response relationship. *Eur Respir J* 2002;**20**:601–8.
35 **Silkoff PE**, McClean P, Spino M, *et al*. Dose-response relationship and reproducibility of the fall in exhaled nitric oxide after inhaled beclomethasone dipropionate therapy in asthma patients. *Chest* 2001;**119**:1322–8.
36 **Jatakanon A**, Kharitonov S, Lim S, Barnes PJ. Effect of differing doses of inhaled budesonide on markers of airway inflammation in patients with mild asthma. *Thorax* 1999;**54**:108–14.
37 **Jones SL**, Kittelson J, Cowan JO, *et al*. The predictive value of exhaled nitric oxide measurements in assessing changes in asthma control. *Am J Respir Crit Care Med* 2001;**164**:738–43.
38 **Gibson PG**, Henry RL, Thomas P. Noninvasive assessment of airway inflammation in children: induced sputum, exhaled nitric oxide, and breath condensate. *Eur Respir J* 2000;**16**:1008–15.
39 **Wenzel SE**, Schwartz LB, Langmack EL, *et al*. Evidence that severe asthma can be divided pathologically into two inflammatory subtypes with distinct physiologic and clinical characteristics. *Am J Respir Crit Care Med* 1999;**160**:1001–8.
40 **Wenzel SE**. A different disease, many diseases or mild asthma gone bad? Challenges of severe asthma. *Eur Respir J* 2003;**22**:397–8.
41 **Payne DN**, Wilson NM, James A, *et al*. Evidence for different subgroups of difficult asthma in children. *Thorax* 2001;**56**:345–50.
42 **Deykin A**, Massaro AF, Drazen JM, *et al*. Exhaled nitric oxide as a diagnostic test for asthma: online versus offline techniques and effect of flow rate. *Am J Respir Crit Care Med* 2002;**165**:1597–601.
43 **Kharitonov SA**, Yates D, Barnes PJ. Increased nitric oxide in exhaled air of normal human subjects with upper respiratory tract infections. *Eur Respir J* 1995;**8**:295–7.
44 **de Gouw HW**, Grunberg K, Schot R, *et al*. Relationship between exhaled nitric oxide and airway hyperresponsiveness following experimental rhinovirus infection in asthmatic subjects. *Eur Respir J* 1998;**11**:126–32.
45 **Dupont LJ**, Demedts MG, Verleden GM. Prospective evaluation of the validity of exhaled nitric oxide for the diagnosis of asthma. *Chest* 2003;**123**:751–6.
46 **Smith AD**, Cowan JO, Filsell S, *et al*. Diagnosing asthma: comparisons between exhaled nitric oxide measurements and conventional tests. *Am J Respir Crit Care Med* 2004;**169**:473–8.
47 **Smith AD**, Taylor DR. Is exhaled nitric oxide measurement a useful clinical test in asthma? *Curr Opin Allergy Clin Immunol* 2005;**5**:49–56.
48 **Henriksen AH**, Lingaas-Holmen T, Sue-Chu M, *et al*. Combined use of exhaled nitric oxide and airway hyperresponsiveness in characterizing asthma in a large population survey. *Eur Respir J* 2000;**15**:849–55.
49 **Berkman N**, Avital A, Breuer R, *et al*. Exhaled nitric oxide in the diagnosis of asthma: comparison with bronchial provocation tests. *Thorax* 2005;**60**:383–8.
50 **Chatkin JM**, Ansarin K, Silkoff PE, *et al*. Exhaled nitric oxide as a noninvasive assessment of chronic cough. *Am J Respir Crit Care Med* 1999;**159**:1810–3.
51 **Barnes PJ**, Woolcock AJ. Difficult asthma. *Eur Respir J* 1998;**12**:1209–18.
52 **Artlich A**, Jonsson B, Bhiladvala M, *et al*. Single breath analysis of endogenous nitric oxide in the newborn. *Biol Neonate* 2001;**79**:21–6.
53 **Baraldi E**, Dario C, Ongaro R, *et al*. Exhaled nitric oxide concentrations during treatment of wheezing exacerbation in infants and young children. *Am J Respir Crit Care Med* 1999;**159**:1284–8.
54 **Baraldi E**, Scollo M, Zaramella C, *et al*. A simple flow-driven method for online measurement of exhaled NO starting at the age of 4 to 5 years. *Am J Respir Crit Care Med* 2000;**162**:1828–32.
55 **Daniel PF**, Klug B, Valerius NH. Measurement of exhaled nitric oxide in young children during tidal breathing through a facemask. *Pediatr Allergy Immunol* 2005;**16**:248–53.
56 **Buchvald F**, Bisgaard H. FeNO measured at fixed exhalation flow rate during controlled tidal breathing in children from the age of 2 yr. *Am J Respir Crit Care Med* 2001;**163**:699–704.
57 **Jobsis Q**, Schellekens SL, Kroesbergen A, *et al*. Off-line sampling of exhaled air for nitric oxide measurement in children: methodological aspects. *Eur Respir J* 2001;**17**:898–903.
58 **Jobsis Q**, Schellekens SL, Kroesbergen A, *et al*. Sampling of exhaled nitric oxide in children: end-expiratory plateau, balloon and tidal breathing methods compared. *Eur Respir J* 1999;**13**:1406–10.
59 **Jobsis Q**, Raatgeep HC, Hop WC, *et al*. Controlled low flow off line sampling of exhaled nitric oxide in children. *Thorax* 2001;**56**:285–9.
60 **Ratjen F**, Kavuk I, Gartig S, *et al*. Airway nitric oxide in infants with acute wheezy bronchitis. *Pediatr Allergy Immunol* 2000;**11**:230–5.
61 **Wildhaber JH**, Hall GL, Stick SM. Measurements of exhaled nitric oxide with the single-breath technique and positive expiratory pressure in infants. *Am J Respir Crit Care Med* 1999;**159**:74–8.
62 **Baraldi E**, de Jongste JC. Measurement of exhaled nitric oxide in children, 2001. *Eur Respir J* 2002;**20**:223–37.
63 **ATS/ERS**. Recommendations for standardized procedures for the online and offline measurement of exhaled lower respiratory nitric oxide and nasal nitric oxide, 2005. *Am J Respir Crit Care Med* 2005;**171**:912–30.
64 **Brussee JE**, Smit HA, Kerkhof M, *et al*. Exhaled nitric oxide in 4-year-old children: relationship with asthma and atopy. *Eur Respir J* 2005;**25**:455–61.
65 **Malmberg LP**, Pelkonen AS, Haahtela T, *et al*. Exhaled nitric oxide rather than lung function distinguishes preschool children with probable asthma. *Thorax* 2003;**58**:494–9.
66 **Moeller A**, Franklin P, Hall GL, *et al*. Inhaled fluticasone dipropionate decreases levels of nitric oxide in recurrenty wheezy infants. *Pediatr Pulmonol* 2004;**38**:250–5.
67 **Straub DA**, Moeller A, Minocchieri S, *et al*. The effect of montelukast on lung function and exhaled nitric oxide in infants with early childhood asthma. *Eur Respir J* 2005;**25**:289–94.
68 **Straub DA**, Minocchieri S, Moeller A, *et al*. The effect of montelukast on exhaled nitric oxide and lung function in asthmatic children 2 to 5 years old. *Chest* 2005;**127**:509–14.
69 **Frank TL**, Adisesh A, Pickering AC, *et al*. Relationship between exhaled nitric oxide and childhood asthma. *Am J Respir Crit Care Med* 1998;**158**:1032–6.
70 **Moody A**, Fergusson W, Wells A, *et al*. Increased nitric oxide production in the respiratory tract in asymptomatic Pacific Islanders: an association with skin prick reactivity to house dust mite. *J Allergy Clin Immunol* 2000;**105**:895–9.
71 **van Amsterdam JG**, Janssen NA, de Meer G, *et al*. The relationship between exhaled nitric oxide and allergic sensitization in a random sample of school children. *Clin Exp Allergy* 2003;**33**:187–91.
72 **Saito J**, Inoue K, Sugawara A, *et al*. Exhaled nitric oxide as a marker of airway inflammation for an epidemiologic study in schoolchildren. *J Allergy Clin Immunol* 2004;**114**:512–6.
73 **van den Toorn LM**, Prins JB, de Jongste JC, *et al*. Benefit from anti-inflammatory treatment during clinical remission of atopic asthma. *Respir Med* 2005;**99**:779–87.
74 **Clini E**, Bianchi L, Vitacca M, *et al*. Exhaled nitric oxide and exercise in stable COPD patients. *Chest* 2000;**117**:702–7.
75 **Delen FM**, Sippel JM, Osborne ML, *et al*. Increased exhaled nitric oxide in chronic bronchitis: comparison with asthma and COPD. *Chest* 2000;**117**:695–701.
76 **Zietkowski Z**, Kucharewicz I, Bodzenta-Lukaszyk A. The influence of inhaled corticosteroids on exhaled nitric oxide in stable chronic obstructive pulmonary disease. *Respir Med* 2005;**99**:816–24.
77 **Brindicci C**, Ito K, Resta O, *et al*. Exhaled nitric oxide from lung periphery is increased in COPD. *Eur Respir J* 2005;**26**:52–9.
78 **Papi A**, Romagnoli M, Baraldo S, *et al*. Partial reversibility of airflow limitation and increased exhaled NO and sputum eosinophilia in chronic obstructive pulmonary disease. *Am J Respir Crit Care Med* 2000;**162**:1773–7.
79 **Kharitonov SA**, Yates D, Robbins RA, *et al*. Increased nitric oxide in exhaled air of asthmatic patients. *Lancet* 1994;**343**:133–5.
80 **Kharitonov SA**, O'Connor BJ, Evans DJ, *et al*. Allergen-induced late asthmatic reactions are associated with elevation of exhaled nitric oxide. *Am J Respir Crit Care Med* 1995;**151**:1894–9.

81 **Adisesh LA**, Kharitonov SA, Yates DH, *et al*. Exhaled and nasal nitric oxide is increased in laboratory animal allergy. *Clin Exp Allergy* 1998;**28**:876–80.
82 **Murphy AW**, Platts-Mills TA, Lobo M, *et al*. Respiratory nitric oxide levels in experimental human influenza. *Chest* 1998;**114**:452–6.
83 **Cremona G**, Higenbottam TW, Mayoral V, *et al*. Elevated exhaled nitric oxide in patients with hepatopulmonary syndrome. *Eur Respir J* 1995;**8**:1883–5.
84 **Rolla G**, Brussino L, Colagrande P, *et al*. Exhaled nitric oxide and oxygenation abnormalities in hepatic cirrhosis. *Hepatology* 1997;**26**:842–7.
85 **Sogni P**, Garnier P, Gadano A, *et al*. Endogenous pulmonary nitric oxide production measured from exhaled air is increased in patients with severe cirrhosis. *J Hepatol* 1995;**23**:471–3.
86 **Gabbay E**, Fisher AJ, Small T, *et al*. Exhaled single-breath nitric oxide measurements are reproducible, repeatable and reflect levels of nitric oxide found in the lower airways. *Eur Respir J* 1998;**11**:467–72.
87 **Gabbay E**, Walters EH, Orsida B, *et al*. Post-lung transplant bronchiolitis obliterans syndrome (BOS) is characterized by increased exhaled nitric oxide levels and epithelial inducible nitric oxide synthase. *Am J Respir Crit Care Med* 2000;**162**:2182–7.
88 **Fisher AJ**, Gabbay E, Small T, *et al*. Cross sectional study of exhaled nitric oxide levels following lung transplantation. *Thorax* 1998;**53**:454–8.
89 **Verleden GM**, Dupont LJ, Van Raemdonck DE, *et al*. Accuracy of exhaled nitric oxide measurements for the diagnosis of bronchiolitis obliterans syndrome after lung transplantation. *Transplantation* 2004;**78**:730–3.
90 **Brugiere O**, Thabut G, Mal H, *et al*. Exhaled NO may predict the decline in lung function in bronchiolitis obliterans syndrome. *Eur Respir J* 2005;**25**:813–9.
91 **Ho LP**, Innes JA, Greening AP. Exhaled nitric oxide is not elevated in the inflammatory airways diseases of cystic fibrosis and bronchiectasis. *Eur Respir J* 1998;**12**:1290–4.
92 **Kharitonov SA**, Wells AU, O'Connor BJ, *et al*. Elevated levels of exhaled nitric oxide in bronchiectasis. *Am J Respir Crit Care Med* 1995;**151**:1889–93.
93 **Tsang KW**, Leung R, Fung PC, *et al*. Exhaled and sputum nitric oxide in bronchiectasis: correlation with clinical parameters. *Chest* 2002;**121**:88–94.
94 **Agusti AG**, Villaverde JM, Togores B, *et al*. Serial measurements of exhaled nitric oxide during exacerbations of chronic obstructive pulmonary disease. *Eur Respir J* 1999;**14**:523–8.
95 **Ansarin K**, Chatkin JM, Ferreira IM, *et al*. Exhaled nitric oxide in chronic obstructive pulmonary disease: relationship to pulmonary function. *Eur Respir J* 2001;**17**:934–8.
96 **Clini E**, Bianchi L, Ambrosino N. Exhaled nitric oxide in COPD patients. *Monaldi Arch Chest Dis* 2001;**56**:169–70.
97 **Clini E**, Cremona G, Campana M, *et al*. Production of endogenous nitric oxide in chronic obstructive pulmonary disease and patients with cor pulmonale. Correlates with echo-Doppler assessment. *Am J Respir Crit Care Med* 2000;**162**:446–50.
98 **Corradi M**, Majori M, Cacciani GC, *et al*. Increased exhaled nitric oxide in patients with stable chronic obstructive pulmonary disease. *Thorax* 1999;**54**:572–5.
99 **Kanazawa H**, Shoji S, Yoshikawa T, *et al*. Increased production of endogenous nitric oxide in patients with bronchial asthma and chronic obstructive pulmonary disease. *Clin Exp Allergy* 1998;**28**:1244–50.
100 **Maziak W**, Loukides S, Culpitt S, *et al*. Exhaled nitric oxide in chronic obstructive pulmonary disease. *Am J Respir Crit Care Med* 1998;**157**:998–1002.
101 **Rutgers SR**, van der Mark TW, Coers W, *et al*. Markers of nitric oxide metabolism in sputum and exhaled air are not increased in chronic obstructive pulmonary disease. *Thorax* 1999;**54**:576–80.
102 **Silkoff PE**, Martin D, Pak J, *et al*. Exhaled nitric oxide correlated with induced sputum findings in COPD. *Chest* 2001;**119**:1049–55.
103 **Paredi P**, Kharitonov SA, Loukides S, *et al*. Exhaled nitric oxide is increased in active fibrosing alveolitis. *Chest* 1999;**115**:1352–6.
104 **Moodley YP**, Chetty R, Lalloo UG. Nitric oxide levels in exhaled air and inducible nitric oxide synthase immunolocalization in pulmonary sarcoidosis. *Eur Respir J* 1999;**14**:822–7.
105 **Rolla G**, Colagrande P, Scappaticci E, *et al*. Exhaled nitric oxide in systemic sclerosis: relationships with lung involvement and pulmonary hypertension. *J Rheumatol* 2000;**27**:1693–8.
106 **Moodley YP**, Lalloo UG. Exhaled nitric oxide is elevated in patients with progressive systemic sclerosis without interstitial lung disease. *Chest* 2001;**119**:1449–54.
107 **Kharitonov SA**, Cailes JB, Black CM, *et al*. Decreased nitric oxide in the exhaled air of patients with systemic sclerosis with pulmonary hypertension. *Thorax* 1997;**52**:1051–5.
108 **Grasemann H**, Michler E, Wallot M, *et al*. Decreased concentration of exhaled nitric oxide (NO) in patients with cystic fibrosis. *Pediatr Pulmonol* 1997;**24**:173–7.
109 **Dotsch J**, Demirakca S, Terbrack HG, *et al*. Airway nitric oxide in asthmatic children and patients with cystic fibrosis. *Eur Respir J* 1996;**9**:2537–40.
110 **Elphick HE**, Demoncheaux EA, Ritson S, *et al*. Exhaled nitric oxide is reduced in infants with cystic fibrosis. *Thorax* 2001;**56**:151–2.
111 **Karadag B**, James AJ, Gultekin E, *et al*. Nasal and lower airway level of nitric oxide in children with primary ciliary dyskinesia. *Eur Respir J* 1999;**13**:1402–5.
112 **Wodehouse T**, Kharitonov SA, Mackay IS, *et al*. Nasal nitric oxide measurements for the screening of primary ciliary dyskinesia. *Eur Respir J* 2003;**21**:43–7.
113 **Riley MS**, Porszasz J, Miranda J, *et al*. Exhaled nitric oxide during exercise in primary pulmonary hypertension and pulmonary fibrosis. *Chest* 1997;**111**:44–50.
114 **Loveless MO**, Phillips CR, Giraud GD, *et al*. Decreased exhaled nitric oxide in subjects with HIV infection. *Thorax* 1997;**52**:185–6.
115 **Brett SJ**, Evans TW. Measurement of endogenous nitric oxide in the lungs of patients with the acute respiratory distress syndrome. *Am J Respir Crit Care Med* 1998;**157**:993–7.
116 **Downey D**, Elborn JS. Nitric oxide, iNOS, and inflammation in cystic fibrosis. *J Pathol* 2000;**190**:115–6.
117 **Kelley TJ**, Drumm ML. Inducible nitric oxide synthase expression is reduced in cystic fibrosis murine and human airway epithelial cells. *J Clin Invest* 1998;**102**:1200–7.
118 **Ho LP**, Innes JA, Greening AP. Nitrite levels in breath condensate of patients with cystic fibrosis is elevated in contrast to exhaled nitric oxide. *Thorax* 1998;**53**:680–4.
119 **Grasemann H**, Ioannidis I, Tomkiewicz RP, de Groot H, Rubin BK, Ratjen F. Nitric oxide metabolites in cystic fibrosis lung disease. *Arch Dis Child* 1998;**78**:49–53.
120 **Lundberg JO**, Rinder J, Weitzberg E, *et al*. Nasally exhaled nitric oxide in humans originates mainly in the paranasal sinuses. *Acta Physiol Scand* 1994;**152**:431–2.
121 **Narang I**, Ersu R, Wilson NM, *et al*. Nitric oxide in chronic airway inflammation in children: diagnostic use and pathophysiological significance. *Thorax* 2002;**57**:586–9.
122 **Corbelli R**, Bringolf-Isler B, Amacher A, *et al*. Nasal nitric oxide measurements to screen children for primary ciliary dyskinesia. *Chest* 2004;**126**:1054–9.
123 **Grasemann H**, Gartig SS, Wiesemann HG, *et al*. Effect of L-arginine infusion on airway NO in cystic fibrosis and primary ciliary dyskinesia syndrome. *Eur Respir J* 1999;**13**:114–8.
124 **Loukides S**, Kharitonov S, Wodehouse T, *et al*. Effect of arginine on mucociliary function in primary ciliary dyskinesia. *Lancet* 1998;**352**:371–2.
125 **Csoma Z**, Bush A, Wilson NM, *et al*. Nitric oxide metabolites are not reduced in exhaled breath condensate of patients with primary ciliary dyskinesia. *Chest* 2003;**124**:633–8.
126 **Baraldi E**, Pasquale MF, Cangiotti AM, *et al*. Nasal nitric oxide is low early in life: case study of two infants with primary ciliary dyskinesia. *Eur Respir J* 2004;**24**:881–3.
127 **Jain B**, Rubinstein I, Robbins RA, *et al*. Modulation of airway epithelial cell ciliary beat frequency by nitric oxide. *Biochem Biophys Res Commun* 1993;**191**:83–8.
128 **Sanders SP**, Proud D, Permutt S, *et al*. Role of nasal nitric oxide in the resolution of experimental rhinovirus infection. *J Allergy Clin Immunol* 2004;**113**:697–702.
129 **Jatakanon A**, Lim S, Barnes PJ. Changes in sputum eosinophils predict loss of asthma control. *Am J Respir Crit Care Med* 2000;**161**:64–72.
130 **Harkins MS**, Fiato KL, Iwamoto GK. Exhaled nitric oxide predicts asthma exacerbation. *J Asthma* 2004;**41**:471–6.
131 **Leuppi JD**, Salome CM, Jenkins CR, *et al*. Predictive markers of asthma exacerbation during stepwise dose reduction of inhaled corticosteroids. *Am J Respir Crit Care Med* 2001;**163**:406–12.
132 **Deykin A**, Lazarus SC, Fahy JV, *et al*. Sputum eosinophil counts predict asthma control after discontinuation of inhaled corticosteroids. *J Allergy Clin Immunol* 2005;**115**:720–7.
133 **Zacharasiewicz A**, Wilson N, Lex C, *et al*. Clinical use of noninvasive measurements of airway inflammation in steroid reduction in children. *Am J Respir Crit Care Med* 2005;**171**:1077–82.
134 **Pijnenburg MW**, Hofhuis W, Hop WC, *et al*. Exhaled nitric oxide predicts asthma relapse in children with clinical asthma remission. *Thorax* 2005;**60**:215–8.
135 **Jayaram L**, Pizzichini MM, Cook RJ, *et al*. Determining asthma treatment by monitoring sputum cell counts: effect on exacerbation. *Eur Respir J* 2006;**27**:483–94.
136 **Sont JK**, Han J, van Krieken JM, *et al*. Relationship between the inflammatory infiltrate in bronchial biopsy specimens and clinical severity of asthma in patients treated with inhaled steroids. *Thorax* 1996;**51**:496–502.
137 **Silvestri M**, Sabatini F, Sale R, *et al*. Correlations between exhaled nitric oxide levels, blood eosinophilia, and airway obstruction reversibility in childhood asthma are detectable only in atopic individuals. *Pediatr Pulmonol* 2003;**35**:358–63.
138 **Smith AD**, Cowan JO, Brassett KP, *et al*. Use of exhaled nitric oxide measurements to guide treatment in chronic asthma. *N Engl J Med* 2005;**352**:2163–73.
139 **Pijnenburg MW**, Bakker EM, Lever S, *et al*. High fractional concentration of nitric oxide in exhaled air despite steroid treatment in asthmatic children. *Clin Exp Allergy* 2005;**35**:920–5.
140 **Kharitonov SA**, Robbins RA, Yates D, *et al*. Acute and chronic effects of cigarette smoking on exhaled nitric oxide. *Am J Respir Crit Care Med* 1995;**152**:609–12.
141 **McSharry CP**, McKay IC, Chaudhuri R, *et al*. Short and long-term effects of cigarette smoking independently influence exhaled nitric oxide concentration in asthma. *J Allergy Clin Immunol* 2005;**116**:88–93.
142 **Robbins RA**, Millatmal T, Lassi K, *et al*. Smoking cessation is associated with an increase in exhaled nitric oxide. *Chest* 1997;**112**:313–8.
143 **Olin AC**, Alving K, Toren K. Exhaled nitric oxide: relation to sensitization and respiratory symptoms. *Clin Exp Allergy* 2004;**34**:221–6.
144 **van der Lee I**, van den Bosch JM, Zanen P. Reduction of variability of exhaled nitric oxide in healthy volunteers. *Respir Med* 2002;**96**:1014–20.
145 **Buchvald F**, Baraldi E, Carraro S, *et al*. Measurements of exhaled nitric oxide in healthy subjects age 4 to 17 years. *J Allergy Clin Immunol* 2005;**115**:1130–6.
146 **Kharitonov SA**, Gonio F, Kelly C, *et al*. Reproducibility of exhaled nitric oxide measurements in healthy and asthmatic adults and children. *Eur Respir J* 2003;**21**:433–8.

147 **Ekroos H**, Tuominen J, Sovijarvi AR. Exhaled nitric oxide and its long-term variation in healthy non-smoking subjects. *Clin Physiol* 2000;**20**:434–9.
148 **Ekroos H**, Karjalainen J, Sarna S, *et al*. Short-term variability of exhaled nitric oxide in young male patients with mild asthma and in healthy subjects. *Respir Med* 2002;**96**:895–900.
149 **Kissoon N**, Duckworth LJ, Blake KV, *et al*. Exhaled nitric oxide concentrations: online versus offline values in healthy children. *Pediatr Pulmonol* 2002;**33**:283–92.
150 **Silkoff PE**, Sylvester JT, Zamel N, *et al*. Airway nitric oxide diffusion in asthma: Role in pulmonary function and bronchial responsiveness. *Am J Respir Crit Care Med* 2000;**161**:1218–28.
151 **Shin HW**, Rose-Gottron CM, Cooper DM, *et al*. Airway diffusing capacity of nitric oxide and steroid therapy in asthma. *J Appl Physiol* 2004;**96**:65–75.
152 **Berry M**, Hargadon B, Morgan A, *et al*. Alveolar nitric oxide in adults with asthma: evidence of distal lung inflammation in refractory asthma. *Eur Respir J* 2005;**25**:986–91.

LUNG ALERT

Interstitial lung disease and leflunomide use

▲ Suissa S, Hudson M, Ernst P. Leflunomide use and the risk of interstitial lung disease in rheumatoid arthritis. *Arthritis Rheum* 2006;**54**:1435–9

There have been numerous reports of interstitial lung disease associated with the use of the new disease modifying anti-rheumatic drug leflunomide. This epidemiological study examined the risk of developing interstitial lung disease (ILD) in patients on leflunomide.

Data from 62 734 patients with rheumatoid arthritis were examined in a case-control study. The risk of ILD was not higher for patients on leflunomide provided they had no previous methotrexate use or a history of ILD (relative risk (RR) 1.2, 95% confidence interval (CI) 0.4 to 3.1). There was, however, an increased risk of ILD with leflunomide in patients who did have a history of previous methotrexate use or ILD (RR 2.6, 95% CI 1.2 to 5.6).

The use of leflunomide as a disease modifying anti-rheumatic drug is increasing in patients with rheumatoid arthritis and reports of ILD are rising. Respiratory physicians should be aware of the potential for developing ILD.

V Aiyappan
Senior House Office, Huddersfield Royal Infirmary, Huddersfield, UK; aiyapps@yahoo.com

LUNG ALERT

Rhinovirus and severe asthma exacerbations requiring admission to hospital

▲ Venarske D, Busse WW, Griffin MR, *et al*. The relationship of rhinovirus-associated asthma hospitalizations with inhaled corticosteroids and smoking. *J Infect Dis* 2006;**193**:1536–43

Rhinovirus (RV) is the respiratory virus that has been most frequently associated with asthma exacerbations (40–60% using viral culture and molecular techniques in previous studies). This prospective, small, single centre study examined the role of RV in severe asthma exacerbations requiring admission to hospital using reverse transcription polymerase chain reaction detection in nasal wash samples at two different time points: hospital admission and 3 month convalescent follow up visit.

One hundred and one adult patients admitted with acute asthma to the Vanderbilt University Medical Centre, Nashville over a 4 year period were enrolled. Twenty one (21%) were found to be positive for RV at admission. Of these, 12 returned 3 months later for an outpatient convalescence visit; none were RV positive. Of the total 76 patients who returned for the 3 month visit, nasal wash samples were found to be positive for RV in only one. Interestingly, RV positive asthmatics had relatively mild disease and were less likely to have a history of hospitalisation for an asthma exacerbation. Current smoking history and non-use of inhaled corticosteroids (perhaps due to a high number of mild asthmatics) were significantly associated with RV infection.

K Ibrahim
Specialist Registrar, Colchester General Hospital, Essex, UK; topkimo@aol.com