Eur Respir J 2002; 20: 40s–43s
DOI: 10.1183/09031936.02.00004002
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European Respiratory Journal
ISSN 0904-1850

REPORT OF WORKING GROUP 5

Clinical applications of assessment of airway inflammation using induced sputum

Leader of the Working Group: I.D. Pavord*

Members of the Working Group: P.J. Sterk[#], F.E. Hargreave[¶], J.C. Kips[+], M.D. Inman[¶], R. Louis[§], M.M.M. Pizzichini[ƒ], E.H. Bel**, I. Pin[##], D.C. Grootendorst[##], K. Parameswaran[++], R. Djukanović[§§]

Airway diseases account for a significant proportion of the respiratory physician's workload. Traditionally, conditions are classified according to clinical symptoms and abnormalities of function, although the major categories of airway disease (asthma, chronic cough and chronic obstructive pulmonary disease (COPD)) show considerable overlap in terms of both clinical picture and response to anti-inflammatory treatment with corticosteroids. All of the above conditions are associated with airway inflammation, although to date there has been little interest in its routine measurement in clinical practice. The recent development of simple, safe and valid noninvasive techniques for the assessment of airway inflammation has increased interest in such an approach [1, 2]. A number of techniques are available, ranging from measurement of exhaled nitric oxide levels to performing differential cell counts and assessment of mediator concentrations in induced sputum. Relatively little is known about the relationship between these different markers, although it is likely that they measure different aspects of the inflammatory response [3]. A clearer understanding is desirable since some techniques are more suited to routine clinical use than others. Induced sputum cell and mediator measurements are particularly well validated [1], and normal ranges from a relatively large adult population have been published [4].

The present article summarises clinical studies pointing to the usefulness of induced sputum analysis in the following conditions: 1) asthma; 2) cough; 3) COPD; and 4) other respiratory conditions.

Asthma

Asthma is commonly associated with sputum eosinophilia. Up to 80% of corticosteroid-naive subjects [1, 2] and >50% of corticosteroid-treated subjects [5] with currently symptomatic asthma have a sputum eosinophil count that is outside the normal range. Subjects with severe acute asthma usually exhibit marked sputum eosinophilia, although predominant neutrophilia has been noted in some studies in which subjects taking oral corticosteroids at the time of the exacerbation are not excluded [6]. The validity of a high sputum eosinophil count for the identification of asthma (defined as consistent symptoms with objective evidence of abnormal variable airflow obstruction) is better than peak expiratory flow variability, defined as the mean percentage amplitude, and the acute bronchodilator response; it approaches the sensitivity and specificity of measurement of airway responsiveness [7]. There is at best a weak relationship between the severity of asthma as defined by lung function, airway responsiveness or symptoms and the sputum eosinophil count [5, 8–10].

The short-term response to inhaled corticosteroids differs markedly according to the sputum eosinophil count, with little evidence of improvement in symptoms and airway responsiveness in subjects with a baseline sputum eosinophil count of <3% [11]. These findings suggest that measuring the underlying airway inflammation might provide a better guide as to the need for corticosteroid treatment than assessment of functional abnormality. More work is needed to define the relationship between airway inflammation, symptoms, the underlying functional defect and the response to corticosteroids in subjects with asthma. The underlying pathophysiology of noneosinophilic asthma needs to be better understood, and studies are required to determine whether inhaled corticosteroids have any longer-term benefits in this group. Finally, there is a pressing need for a study examining whether asthma management aimed at normalising the sputum

*Institute for Lung Health, Dept of Respiratory Medicine and Thoracic Surgery, Glenfield Hospital, Leicester, UK. [#]Leiden University Medical Centre, Leiden, the Netherlands. [¶]Firestone Institute for Respiratory Health, McMaster University, Hamilton, Ontario, Canada. [+]University Hospital Ghent, Dept of Medical Genetics, Ghent, Belgium. [§]Dept of Pneumology, Hospital of Sart Tilman, Liege, Belgium. [ƒ]Asthma Research Centre, Dept of Internal Medicine, Federal University of Santa Catarina, Florianópolis, Santa Catarina, Brazil. **Dept of Pulmonology, Leiden University Medical Centre, Leiden, the Netherlands. [##]Dept of Paediatrics, CHU of Grenoble, Grenoble, France. [¶¶]Dutch Asthma Centre, Davos, Switzerland. [++]FRCAU, St. Joseph's Hospital, Hamilton, Ontario, Canada. [§§]Southampton University General Hospital, Southampton, UK.

Correspondence: I.D. Pavord, Institute for Lung Health, Dept of Respiratory Medicine and Thoracic Surgery, Glenfield Hospital, Groby Road, Leicester, LE3 9QP, UK. Fax: 44 116 2367768. E-mail: ian.pavord@uhl-tr.nhs.uk

Received: April 4 2002; Accepted: April 16 2002

eosinophil count results in better patient outcomes than a standard approach where the aim of treatment is to suppress symptoms.

Categorising subjects according to the presence of sputum eosinophilia might be particularly useful in asthmatics who are symptomatic despite treatment with inhaled corticosteroids since the additional treatment options (long acting β_2-agonists, higher-dose inhaled corticosteroids, leukotriene antagonists and theophylline) differ in their effects on eosinophilic airway inflammation [12–15]. This is an important group (50% of asthmatics in the UK are receiving low or moderate doses of inhaled corticosteroids and >50% have sufficient symptoms to warrant increased treatment); it is therefore necessary to understand better why some patients exhibit persistent eosinophilic airway inflammation despite corticosteroid treatment and whether this group do particularly well with additional anti-inflammatory treatment.

A number of investigators have examined the practical use of assessment of induced sputum markers of airway inflammation in occupational asthma [16–18]. In general, occupational asthma is associated with similar sputum characteristics to nonoccupational asthma, and occupational challenges are associated with an increase in sputum eosinophilia [16, 17] in much the same way as allergen challenge is. There is some evidence that sputum eosinophil counts increase during workplace exposures in subjects with occupational asthma [17]. Further work is required to determine whether incorporation of assessment of airway inflammation into a standard protocol consisting of assessment of peak expiratory flow ands airway responsiveness at work and away from work provides additional diagnostically useful information.

Cough

Chronic cough is associated with predominant sputum neutrophilia [19], but up to 40% of subjects with cough have a sputum eosinophil count of >3% [20, 21]. Half of them show no functional evidence of asthma but suffer from eosinophilic bronchitis [20, 21]. Assessment of airway inflammation is the only way of identifying these subjects and is, therefore, an important addition to the chronic cough investigation algorithm [20]. Further work is required to determine the sputum cell characteristics of cough associated with gastro-oesophageal reflux, rhinitis and viral infections, although a recent bronchoscopic study suggests that no particular patterns will emerge [22]. Lipid-laden macrophages are noted more commonly in induced sputum from patients with oropharyngeal reflux than controls, suggesting that this might be a useful noninvasive marker of oropharyngeal reflux with gastric aspiration [23]. Patients with cough and sputum eosinophilia exhibit an objective response to corticosteroid treatment that occurs in parallel with a treatment-associated fall in the sputum eosinophil count [20, 24]. In contrast, patients without sputum eosinophilia do not respond [25].

Chronic obstructive pulmonary disease

In COPD, the sputum neutrophil count is usually raised, and the absolute or differential neutrophil count is related to reduced forced expiratory volume in one second (FEV1) and the increased rate of decline in FEV1, suggesting that neutrophilic airway inflammation is functionally important [26, 27]. Up to 40% of subjects with COPD have a sputum eosinophil count of >3% [28, 29]. These subjects are in other respects identical to subjects without sputum eosinophilia (*i.e.* no more likely to have features suggesting asthma). There is increasing evidence that the presence of sputum eosinophilia predicts an objective response to corticosteroid treatment in COPD [28, 29]. In one study, the response to a 2-week course of oral prednisolone increased as the baseline sputum eosinophil count increased and was associated with a marked treatment-induced fall in the sputum eosinophil count but no change in sputum markers of neutrophilic inflammation [29]. This suggests that eosinophilic airway inflammation is functionally important in some subjects with COPD, and that the effects of corticosteroids are due to modification of this aspect of the complex airway inflammation. Further work is required to determine whether reduction in exacerbation frequency and decreased rate of decline in FEV1 with long-term corticosteroid treatment is confined to subjects with sputum eosinophilia. Potentially, sputum assessment could be used as a screening test before deciding on long-term corticosteroid treatment in COPD.

Other conditions

Little is known about induced sputum cell features of interstitial diseases, although sarcoidosis and extrinsic allergic alveolitis are associated with airway involvement and so might be usefully investigated using induced sputum [30]. One problem is that induced sputum might not be the best way of investigating lymphocytic airway inflammation since lymphocytes are scarce in induced sputum. There have been intriguing findings of mineralogical particles in induced sputum in occupational lung diseases associated with dust exposure [31] and haemosiderin-laden macrophages in subjects with left ventricular failure [32] which deserve further study. Common colds are associated with predominant neutrophilic airway inflammation in normal subjects and subjects with asthma [33]. Further work is also required in this area. Potentially, induced sputum could be a useful means of identifying different patterns of inflammation associated with different pathogens or differences in host response to the same pathogen.

Key points

1) There is considerable overlap of the sputum inflammatory cell characteristics of the main clinical categories of airway disease. 2) The relationship between eosinophilic airway inflammation and disease

expression is uncertain. 3) Anti-asthma therapies differ in their effects on eosinophilic airway inflammation. 4) Eosinophilic bronchitis is a common and treatable cause of chronic cough. It can only be identified by measuring airway inflammation. 5) Sputum eosinophilia predicts a response to corticosteroid therapy in asthma, cough and COPD. 6) There is no evidence that corticosteroids are helpful in the absence of a sputum eosinophilia.

Outstanding questions

1) Does measuring airway inflammation improve diagnosis and treatment outcome? What is the best technique to use? 2) Can patterns of inflammation that are associated with differences in prognosis or treatment response be identified?

References

1. Pizzichini E, Pizzichini MM, Efthimiadis A, *et al.* Indices of airway inflammation in induced sputum: reproducibility and validity of cell and fluid-phase measurements. *Am J Respir Crit Care Med* 1996; 154: 308–317.
2. Pavord ID, Pizzichini MM, Pizzichini E, Hargreave FE. The use of induced sputum to investigate airway inflammation. *Thorax* 1997; 52: 498–501.
3. Magnussen H, Holz O, Sterk PJ, Hargreave FE. Noninvasive methods to measure airway inflammation: future considerations. *Eur Respir J* 2000; 16: 1175–1179.
4. Belda J, Leigh R, Parameswaran K, O'Byrne PM, Sears MR, Hargreave FE. Induced sputum cell counts in healthy adults. *Am J Respir Crit Care Med* 2000; 161: 475–478.
5. Louis R, Lau LC, Bron AO, Roldaan AC, Radermecker M, Djukanovic R. The relationship between airways inflammation and asthma severity. *Am J Respir Crit Care Med* 2000; 161: 9–16.
6. Fahy JV, Kim KW, Liu J, Boushey HA. Prominent neutrophilic inflammation in sputum from subjects with asthma exacerbation. *J Allergy Clin Immunol* 1995; 95: 843–852.
7. Hunter CJ, Brightling CE, Woltmanw G, Wardlar AJ, Pavord ID. A comparison of the validity of different diagnostic tests in adults with asthma. *Chest* 2002; 121: 1051–1057.
8. Iredale MJ, Wanklyn SA, Phillips IP, Krausz T, Ind PW. Non-invasive assessment of bronchial inflammation in asthma: no correlation between eosinophilia of induced sputum and bronchial responsiveness to inhaled hypertonic saline. *Clin Exp Allergy* 1994; 24: 940–945.
9. Green RH, Brightling C, Woltmann G, Parker D, Wardlaw AJ, Pavord ID. Analysis of induced sputum in adults with asthma. Identification of a subgroup with an isolated sputum neutrophilia and a poor response to inhaled corticosteroids. *Thorax* 2002 (in press).
10. Ronchi MC, Piragino C, Rosi E, *et al.* Do sputum eosinophils and ECP relate to the severity of asthma? *Eur Respir J* 1997; 10: 1809–1813.
11. Pavord ID, Brightling CE, Woltmann G, Wardlaw AJ. Non-eosinophilic corticosteroid unresponsive asthma. *Lancet* 1999; 353: 2213–2214.
12. Kips JC, O'Connor BJ, Inman MD, Svensson K, Pauwels RA, O'Byrne PM. A long-term study of the anti-inflammatory effect of low-dose budesonide plus formoterol *versus* high-dose budesonide in asthma. *Am J Respir Crit Care Med* 2000; 161: 996–1001.
13. Turner MO, Johnston PR, Pizzichini E, Pizzichini MM, Hussack PA, Hargreave FE. Anti-inflammatory effects of salmeterol compared with beclomethasone in eosinophilic mild exacerbations of asthma: a randomized, placebo controlled trial. *Can Respir J* 1998; 5: 261–268.
14. Pizzichini E, Leff JA, Reiss TF, *et al.* Montelukast reduces airway eosinophilic inflammation in asthma: a randomized, controlled trial. *Eur Respir J* 1999; 14: 12–18.
15. Louis R, Bettiol J, Cataldo D, Sele J, Henquet M, Radermecker M. Effect of a 4-week treatment with theophylline on sputum eosinophilia and sputum eosinophil chemotactic activity in steroid-naive asthmatics. *Clin Exp Allergy* 2000; 30: 1151–1160.
16. Obata H, Dittrick M, Chan H, Chan-Yeung M. Sputum eosinophils and exhaled nitric oxide during late asthmatic reaction in patients with western red cedar asthma. *Eur Respir J* 1999; 13: 489–495.
17. Lemiere C, Pizzichini MM, Balkissoon R, *et al.* Diagnosing occupational asthma: use of induced sputum. *Eur Respir J* 1999; 13: 482–488.
18. Malo JL. The case for confirming occupational asthma: why, how much, how far? (revisited). *Eur Respir J* 1999; 13: 477–478.
19. Jatakanon A, Lalloo UG, Lim S, Chung KF, Barnes PJ. Increased neutrophils and cytokines, TNF-α and IL-8, in induced sputum of non-asthmatic patients with chronic dry cough. *Thorax* 1999; 54: 234–237.
20. Brightling CE, Ward R, Goh KL, Wardlaw AJ, Pavord ID. Eosinophilic bronchitis is an important cause of chronic cough. *Am J Respir Crit Care Med* 1999; 160: 406–410.
21. Carney IK, Gibson PG, Murree-Allen K, Saltos N, Olson LG, Hensley MJ. A systematic evaluation of mechanisms in chronic cough. *Am J Respir Crit Care Med* 1997; 156: 211–216.
22. McGarvey LP, Forsythe P, Heaney LG, MacMahon J, Ennis M. Bronchoalveolar lavage findings in patients with chronic nonproductive cough. *Eur Respir J* 1999; 13: 59–65.
23. Parameswaran K, Anvari M, Efthimiadis A, Kamada D, Hargreave FE, Allen CJ. Lipid-laden macrophages in induced sputum are a marker of oropharyngeal reflux and possible gastric aspiration. *Eur Respir J* 2000; 16: 1119–1122.
24. Brightling CE, Ward R, Wardlaw AJ, Pavord ID. Airway inflammation, airway responsiveness and cough before and after inhaled budesonide in patients with eosinophilic bronchitis. *Eur Respir J* 2000; 15: 682–686.
25. Pizzichini MM, Pizzichini E, Parameswaran K, *et al.* Nonasthmatic chronic cough: no effect of treatment with an inhaled corticosteroid in patients without sputum eosinophilia. *Can Respir J* 1999; 6: 323–330.
26. Keatings VM, Collins PD, Scott DM, Barnes PJ. Differences in interleukin-8 and tumor necrosis factor-α in induced sputum from patients with chronic obstructive pulmonary disease or asthma. *Am J Respir Crit Care Med* 1996; 153: 530–534.

27. Stanescu D, Sanna A, Veriter C, *et al.* Airways obstruction, chronic expectoration, and rapid decline of FEV1 in smokers are associated with increased levels of sputum neutrophils. *Thorax* 1996; 51: 267–271.
28. 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: 1511–1517.
29. 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: 1480–1485.
30. Fireman E, Lerman Y. Possible future of induced sputum in interstitial lung disease. *Eur Respir J* 2000; 15: 240–242.
31. Fireman E, Greif J, Schwarz Y, *et al.* Assessment of hazardous dust exposure by BAL and induced sputum. *Chest* 1999; 115: 1720–1728.
32. Leigh R, Sharon RF, Efthimiadis A, Hargreave FE, Kitching AD. Diagnosis of left-ventricular dysfunction from induced sputum examination. *Lancet* 1999; 354: 833–834.
33. Pizzichini MM, Pizzichini E, Efthimiadis A, *et al.* Asthma and natural colds: inflammatory indices in induced sputum: a feasibility study. *Am J Respir Crit Care Med* 1998; 158: 1178–1184.