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REVIEW

Biomarkers Guided Treatment Strategies in Adult Patients with Asthma: Ready for the Clinical Field?

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Abstract Asthma is a chronic inflammatory airways disorder mainly characterized by heterogeneity. In the more severe forms, a discordance often exists between symptoms and inflammation. Difficulty in managing asthma derives partly from the multiple phenotypes existing and our inability to recognize them. The use of non-invasive, with main representative the fraction of exhaled nitric oxide, or semi-invasive techniques such as induced sputum are effective tools that can help us to guide asthma treatment. In the latest years, several serum biomarkers related to asthmatic inflammation have been used for the better recognition of asthma sub-phenotypes to achieve optimization of therapy and disease outcome. In patients with mild–moderate asthma, as well as patients with more severe asthma, the use of blood eosinophils revealed an acceptable accuracy for the prediction of airway eosinophilia indicating that in future studies may facilitate both individualized treatment and management of asthma. None of the above techniques have been incorporated in clinical practice although sputum eosinophils can be used in patients with severe asthma particularly in specialized centers with great experience. Of great interest are blood eosinophils since current data support their role either as tool for treatment selections or/and as a biomarker of airway eosinophilia.

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Abbreviations

ACQ	Asthma control questionnaire
FeNO	Fraction of exhaled nitric oxide
ICS	Inhaled corticosteroids
LABA	Long-acting beta two-agonists
NOS	Nitric oxide synthase

Introduction

"Asthma is a heterogeneous disease, usually characterized by chronic airway inflammation. It is defined by the history of respiratory symptoms such as wheeze, shortness of breath, chest tightness and cough that vary over time and in intensity, together with variable expiratory airflow limitation" (Bateman et al. 2008). Considering the definition of asthma, it is important to emphasize that the heterogeneity is mainly attributed to the underlying airway inflammation, the presence of clinical syndromes and finally airflow limitation and/or obstruction. Such complexity has the potential to explain in a substantial extent why, despite the use of available therapies, a significant percentage of asthma patients still cannot control their symptoms (Agache 2013; Martin et al. 2007; Szefler et al. 2002). There is a clear need to therapeutically address the complexity of asthma, beyond a symptom-based approach utilizing beta agonists and corticosteroids to a more tailored approach of individualized management that correlates therapeutic choices to specific underlying inflammatory processes (Szefler et al. 2002). This need is particularly evident and urgent in more severe, less-

controlled forms of the disease where there is a clear discordance between symptoms and inflammation. This discordance is mainly associated with the presence or/and absence of eosinophilic inflammation (Haldar et al. 2008). The heterogeneity of asthma is the main reason why the treatment of the disease is so challenging, since a symptom-led approach would be effective only for patients with mild to moderate atopic and mostly early onset asthma in which it seems to be a concordance between inflammation and symptoms. Regarding this discordance, clinicians need easy non-invasive and affordable biomarkers for the evaluation of asthmatic inflammation which will help them to assess asthma control and to provide the correct and most effective therapeutic intervention.

A biomarker has been defined as a characteristic that is objectively measured and evaluated as an indicator of normal biological processes, pathogenic processes or pharmacologic responses to a therapeutic intervention (Biomarkers Definitions Working Group 2001). The term biomarker historically refers to analytes in biological samples. The ideal biomarker that could be used in patients with asthma, should: (1) be clearly associated to the pathophysiological events of the disease (2) be sensitive and disease-specific, (3) provide information on the disease process and prognosis, (4) be reliable and reproducible in a routine clinical setting, (5) be inexpensive, (6) be able to identify responses to the therapeutic interventions, (7) have little or no diurnal variation, (8) have high positive and negative predictive values and finally (9) have a simple and acceptable to patients sampling method.

Biomarkers have been evaluated in different samples obtained from asthma patients, including blood, urine, bronchoscopic samples, sputum, and exhaled air. Biomarkers evaluated by invasive techniques, including bronchial biopsies and bronchoalveolar lavage, are limited by the fact that they are associated with possible adverse events, especially in patients with severe asthma and the difficulty to be repeated consecutively in the same patient (Bergeron et al. 2007). Biomarkers evaluated in induced sputum and in exhaled air represent an attractive option that presents several advantages but simultaneously many arising issues need to be resolved (Loukides et al. 2011). There is still an unmet need in the field of biomarkers in asthma, since only few of them have been properly validated and more information is needed on both repeatability and reproducibility as well as to their association to disease progression. Despite the above limitations every single biomarker used in order to characterize a phenotype or/and endotype of asthma, must provide adequate information regarding its standardization procedure, its reproducibility and its reference values. The above issue is considered crucial since many biomarkers failed to provide accuracy in regard to the above procedures. In asthma, biomarkers can help not only in the monitoring of inflammation and asthma control, but also in the selection of the optimal therapeutic intervention, and optimization of treatment. Furthermore, many of the latest biological therapeutic options of asthma (such as omalizumab and mepolizumab) are based in the measurement of commonly used biomarkers (serum IgE levels and blood eosinophils) showing the way for the design of more personalized therapy for asthmatic patients.

So far, there is very little success in establishing and validating highly specific biomarkers. In this review, we will focus on the use of biomarkers in the selection of the optimal treatment in patients with asthma. Furthermore, we provide some data which are mainly related to biomarkers which have been used for the better recognition of asthma sub-phenotypes to achieve optimization of therapy and disease outcome.

Biomarkers for Guided Treatment Strategies in Asthma

Induced Sputum

Eosinophils play a central role in asthmatic inflammation. Induced sputum has been used as a tool for identifying and clarifying lower airway inflammation. Induced sputum is considered to be a safe and well validated method as normal ranges for sputum cell counts from a relatively large adult population have been published (Belda et al. 2000). Based on sputum cell differentials asthma can be categorized into four subgroups: eosinophilic asthma (>3 % eosinophils), neutrophilic asthma (>60 % neutrophils), mixed granulocytic asthma (>3 % eosinophils and >60 % neutrophils) and paucigranulocytic asthma (<3 % eosinophils and <60 % neutrophils) (Simpson et al. 2006). In regard to cut-off value for defining sputum eosinophilia different definitions exist (Bakakos et al. 2011). Some authors would define it as ≥2 % while others prefer ≥3 %. Regarding sputum neutrophilia some reports consider it as an epiphenomenon of the actual use of ICS particularly the high doses (Bakakos et al. 2011). Persistent eosinophilia in asthmatic patients has been suggested as a predictor of poor asthma control, augmented asthma severity and worsened lung function regardless of treatment (Wenzel et al. 1999). However, the value of sputum eosinophil counts in monitoring asthma severity remains a matter of debate (Louis et al. 2000; Pin et al. 1992). Berry et al. (Berry et al. 2007) have reported that the presence of sputum eosinophilia in asthmatic patients is clearly associated with good response to inhaled corticosteroids (ICS). In contrast, patients with predominance of sputum neutrophilia may not respond to an increase in treatment with ICS (Green et al. 2002a).

Some studies have shown that sputum eosinophilia correlates with the degree of airflow obstruction and clinical symptoms, although others failed to show a significant association between sputum eosinophilia, asthma severity, bronchial hyper-responsiveness, and quality of life (Bartoli et al. 2004; Jayaram et al. 2000). Possible mechanisms for the persistence of sputum eosinophilia despite treatment with ICS are inadequate treatment dosing, overactive and intense inflammation glucocorticoid resistance or reduced glucocorticoid receptor binding affinity (Hamid et al. 1997, 1999).

There is a proposed therapeutic strategy mainly based on sputum cell counts. This strategy stratifies patients into three groups: the first one includes patients with sputum eosinophilia and the recommended approach involves titration of steroid dose to achieve a normal sputum eosinophilic count as well as investigation of the cause of eosinophilia. The second one includes patients with sputum neutrophilia in which we usually reduce the dose of steroids, and initiate alternative treatments like macrolides which are mainly targeting neutrophilic inflammation. Finally, the last one is the one where we can reduce or/and keep the dose of steroids while simultaneously we can consider other causes of airway obstruction (Hargreave and Nair 2011).

Three major studies have been conducted in order to clarify whether tailoring of asthma treatment based on sputum eosinophils is superior compared to the clinical assessment (Petsky et al. 2012).

The first study was designed to assess whether a management strategy that minimizes eosinophilic inflammation could reduce the rate of moderate to severe exacerbations compared with the standard clinical management (Green et al. 2002b). It was a 12-month study with a run-in period of 2 weeks where patients were maintained in their usual treatment. If patients, during the run-in period were uncontrolled prednisolone 30 mg was added to their usual treatment for a further 2 weeks' period in order to maximize the chance to achieve asthma control. In the clinical management group, treatment decisions were based on symptoms (daytime and nighttime), peak expiratory flow, and use of rescue medication with β2 agonists. In the inflammatory management group decisions were made in accordance with an algorithm based on maintenance of a sputum eosinophil count below 3 % with a minimum dose of anti-inflammatory treatment. If sputum eosinophils were between 1 and 3 % no change in inflammatory treatment was performed. If sputum eosinophils exceeded 3 % anti-inflammatory treatment was increased. If sputum eosinophils were below 1 % then inflammatory treatment was reduced irrespective of whether asthma control was achieved. Simultaneously any decisions for bronchodilator use were based on clinical symptoms, rescue therapy and lung function impairment. In patients unable to produce sputum, fraction of exhaled nitric oxide (FeNO) was performed by selecting values which properly defined the eosinophilic inflammation. The anti-inflammatory strategy resulted in fewer moderate to severe exacerbations compared to the clinical management. Similar results were also observed for the rate of hospitalizations but in a weaker statistical significance level. Clinical and functional parameters did not significantly differ within the two groups. Similar numbers for both groups received long-acting beta two-agonists (LABA) or leukotrienes receptor antagonists. The mean dose of ICS and oral steroids was similar in both groups. In a subgroup analysis, patients with non-eosinophilic inflammation clearly benefit from the sputum management as they decreased the dose of ICS in a greater degree compared with those treated according to clinical management. Based on the above outcomes specific clusters were proposed. The first one consisted of obese female women where no benefit was observed with the sputum strategy. The second cluster was the inflammation predominant where significant alterations in the rate of exacerbations were observed. The final one was the symptom predominant where a lower dose of ICS was used by the initiation of sputum stratification.

A similar in design study was published 4 years later (Jayaram et al. 2006). This was a 2-year study consisting of two phases and an initial pre-study procedure where patients were stratified/dichotomized according to the duration of their symptoms, ICS dose and FEV_1 % predicted. In phase one the main objective was to minimize treatment at a lower level necessary for the maintenance of asthma control. Patients were followed up at 1-month intervals and at the time of any exacerbation. Phase one was followed by phase two in which the main objective was to evaluate the study outcomes with the maintaining treatment for 2 years. The study compared two different strategies. The first was based on symptoms and spirometry while the second one was based on sputum eosinophils with main target to minimize their count below 2 %. Clinical assessment was also used in the second strategy in order to identify asthma control. The primary outcomes were time to first exacerbation and the relative risk reduction for the first exacerbation. Both outcomes were assessed at phase two. Any co-variates (co-morbidities, allergen exposure) which might affect study measurements were continuously assessed by the investigators. The mean time to achieve the maintenance dose was similar in both groups. Interestingly the above dose was not re-adjusted during phase two.

The sputum strategy group had a lower rate of exacerbations and a longer time to first exacerbation. These advantages were achieved with a similar mean cumulative inhaled steroid dose in phase two. The whole relative risk

for fewer exacerbations was approximately 29 % but did not reach statistical significance. The above outcomes were achieved with similar dose of ICS. Further analysis showed that the patients who benefited from sputum strategy were those with moderate to severe asthma and those with pure eosinophilic profile. The above results extended the findings of Green et al. (2002b) to the direction of the underlying inflammatory profile during exacerbations. Overall, the second study showed that the monitoring of sputum cell counts reduced the risk of exacerbations by 49 % and prolonged the period without an exacerbation with no need for more treatment. These benefits were seen in patients with moderate–severe asthma and were mainly attributed to a reduction of eosinophilic exacerbations. There was no effect on non-eosinophilic exacerbations, which were the most common.

The third study aimed to determine whether changes in the rate of exacerbations and improvements in asthma control are attributed to a reduction in the number of sputum eosinophils compared to standard clinical assessment (Chlumsky et al. 2006). The study lasted 18 months and used a different cut-off level for eosinophils which was 8 %. This study has shown that the clinical strategy which actually aimed to minimizing symptoms and improving lung function failed to prevent asthma exacerbations. On the other hand, reducing the percentage of sputum eosinophils was more effective in decreasing the frequency of asthma exacerbations particularly in patients with moderate to severe asthma. Furthermore, the anti-inflammatory strategy managed to minimize the dose of ICS.

FeNO

FeNO represents the most widely and successfully used biomarker. Endogenous nitric oxide (NO) is produced from the amino acid L arginine which is metabolized into the amino acid L citrulline catalyzed by the enzyme NO synthase (NOS). The NO synthase has three isoforms. The cellular source of endogenous NO still remains unclear despite the fact that epithelial cells are considered as the most possible source (Guo et al. 2000). It is likely that the NO production is related to the induction of the NOS-2 isoform (inducible NOS) in response to pro-inflammatory cytokines due to the increased transcription mediated by transcriptional factors such as nuclear factor (NF)-κB. NO is produced throughout the respiratory tract, secreted into the lumen of the airways and mixed with alveolar air to provide the final exhaled concentration that is characterized as FeNO. Corticosteroids inhibit the inflammatory induction of NOS-2 in epithelial cells and reduce exhaled NO concentrations (Kharitonov et al. 1994). The role of NO in bronchial mucosa may be closely related to asthmatic inflammation, since it represents a potent chemoattractant of eosinophils that may lead to vasodilatation and plasma leakage (Kharitonov and Barnes 2001).

The measurement of FeNO concentration is a quantitative, noninvasive, simple and safe method of measuring airway inflammation (Dweik et al. 2011). FeNO is known to be increased in patients with bronchial asthma and has been proved useful for the distinction of subjects having asthma from those without asthma with a high degree of discriminatory power (Alving et al. 1993; Dupont et al. 2003; Kharitonov et al. 1994). Atopy significantly influences its levels irrespective of the presence of asthma (Gratziou et al. 1999; Kostikas et al. 2008). Eosinophils in sputum have also been shown to correlate with the levels of FeNO and it has been shown that values over 41 ppb are suggestive of sputum eosinophil count $\geq$3 % with 65 % sensitivity and 79 % specificity (Schleich et al. 2010). Both FeNO and sputum eosinophilia have been shown to have a much greater diagnostic accuracy for the diagnosis of asthma compared with conventional lung function tests (Smith et al. 2004). Finally, the performance of FeNO in predicting asthma control remains controversial since some studies support its positive role while others consider that it has a lower validity compared with sputum eosinophils (Michils et al. 2008, 2009; Papaioannou et al. 2009; Quaedvlieg et al. 2009). Finally, in a recently published study using a pragmatic based design a symptom- plus FeNO driven strategy reduces asthma medication use while sustaining asthma control and quality of life (Honkoop et al. 2015).

The role of FeNO as a tool for guiding treatment strategy has been studied both in adults (Shaw et al. 2007; Smith et al. 2005) and in children (Pijnenburg et al. 2005; Szefler et al. 2008; de Jongste et al. 2009) as well as in asthmatic pregnant women (Powell et al. 2011).

The first study was published by Smith et al. (Smith et al. 2005). It was a single-blind, placebo-controlled trial including 97 patients who had been properly treated with ICS. Their daily ICS dose has been titrated in a stepwise mode, on the basis of either FeNO values or an algorithm which was mainly attributed to guidelines promulgated by the Global Initiative for Asthma. The primary outcome of the study was the rate of exacerbations and the secondary outcome was the mean dose of ICS. The study lasted for 12 months and was divided into two phases. The first phase was designed in order, after a proper titration process, to achieve the optimal dose of the ICS. In the control group the dose adjustments were based on pre-specified thresholds in regard to symptoms, use of bronchodilators and lung function. As for the FeNO group, any adjustments were based only on FeNO measurements. A value of 35 ppb was used as a cutoff point in order to increase the dose of ICS. During the second phase which lasted 12 months all subjects continued their treatment at the

optimal dose of phase one. Despite the trend for a lower number of exacerbations in the FeNO group, this did not reach statistical significance. Furthermore, no significant differences were achieved for bronchodilator use, symptoms, night awakenings and use of prednisolone during exacerbations. However, regarding the mean dose of ICS, this was statistically significantly lower in patients treated with the FeNO adjusted strategy. The above outcome was achieved in both study phases. Interestingly, no significant differences were observed in regard to sputum inflammatory cells and particularly eosinophils. However, in a visit where both airway inflammation and asthma control were amplified, significant alterations in both FeNO and sputum eosinophils were observed only in the FeNO group.

Two years later another study including 118 patients with asthma were assigned to be treated either based on a FeNO strategy or using the clinical based British Thoracic society guidelines (Shaw et al. 2007). The primary outcome was the number of severe exacerbations. The study lasted 12 months. Patients were seen on a monthly basis for the first 4 months and then every 2 months until the end of the year. In the control group the dose of ICS was titrated according to Juniper asthma control questionnaire (ACQ) (Juniper et al. 2006). At the FeNO group both values of FeNO and ACQ were used. If FeNO was higher than 26 ppb the dose was increased. If it was less than 16 ppb or less than 26 ppb in two consecutive measurements, then the dose was decreased. Bronchodilator therapy was increased if symptoms were uncontrolled irrespective of FeNO values. Despite the trend for a lower rate of exacerbations in the FeNO group this did not reach statistical significance. The final dose of ICS was lower in the FeNO group despite the fact that the maintenance doses did not differ within groups. No differences were observed in regard to bronchial hyper-responsiveness as assessed by the PC_{20} to methacholine. Interestingly, in participants with amplified eosinophilic inflammation either defined as >3 % of eosinophils in induced sputum or >26 ppb FeNO values the exacerbation rate was significantly lower compared to those who did not fulfill the above pre-determined values.

Finally, in a study where only pregnant women were selected (Powell et al. 2011), 220 pregnant women were allocated in two treatment adjusted management strategies. The first one was based on FeNO plus clinical control while the other one was based on clinical control only. Clinical control was assessed by Juniper ACQ (Juniper et al. 2006). As cut-off level for FeNO the value of 29 ppb was selected. In the initial phase all women already on treatment continued their maintenance dose of ICS while the steroid naïve were started on a low dose of ICS which was equivalent to 100 mcg budesonide. Both strategies also adjusted the addition or/and the increase of a LABA. The FeNO guided management resulted in a significantly lower rate of exacerbations while at the same time there was a delay for the time to first exacerbation. Regarding the other outcomes weak differences but with statistical significant trend were observed for mean ACQ, while no differences were observed in symptom free days. Interestingly the FeNO strategy resulted in a lower dose of ICS but in a higher rate for the use of LABA. Perinatal outcomes involved higher median birthweight, reduction in preterm deliveries and finally reduced neonatal hospitalization and tended to be improved in the FeNO group compared with the control group.

Biomarkers Obtained from Blood: Targeting Therapeutic Options

In the latest years several serum biomarkers related to asthmatic inflammation have been used for the better recognition of asthma sub-phenotypes to achieve optimization of therapy and disease outcome. Total IgE is the most commonly used serum biomarker and is included in the guidelines for the management of asthma (Reddel et al. 2015), while treatment of asthmatic patients with an anti IgE antibody (omalizumab) resulted to the reduction of several inflammatory markers (Djukanovic et al. 2004). Careful patient selection, proper dosing, and close monitoring can lead to the successful management of a high proportion of persistent severe allergic asthmatics (Holgate et al. 2009). A previous study including patients with allergic asthma, not controlled with high doses of ICS and long-acting β_2-agonists, showed that omalizumab resulted in significantly reduced exacerbations of asthma (Hanania et al. 2011) mainly in patients with increased levels of FeNO, blood eosinophils and serum periostin, suggesting that these biomarkers can be also used to select patients more probable to respond to omalizumab (Hanania et al. 2013). In patients with severe asthma, blood eosinophils have been shown to be a predictor of sputum eosinophilia which as previously mentioned has been an important factor associated with poor asthma control (Wagener et al. 2015; Wenzel et al. 1999). Furthermore, considering the applicability of sputum induction, the fact that it is not so simple, nor patient friendly and it is costly since a specialized lab and qualified/trained staff is needed, then blood could serve as an attractive option for the assessment of eosinophilic profile. In the latest years' research has also focused on interleukin (IL)-5, a cytokine closely related to eosinophilic inflammation since it is responsible for the terminal differentiation and recruitment of human eosinophils. Mepolizumab, is a monoclonal antibody against IL-5 that has been shown to reduce exacerbations (Haldar et al. 2009; Nair et al. 2009) improve symptoms and health

related quality of life (Haldar et al. 2009) in patients with eosinophilic asthma. The important therapeutic role of mepolizumab, in patients with severe eosinophilic asthma, has been also shown in studies reporting reductions in exacerbations, improvement of asthma control and significant sparing effect in oral glucocorticosteroids (Bel et al. 2014; Ortega et al. 2014). In both studies, patients had a blood eosinophil count of at least 150 cells per microliter at screening or at least 300 cells per microliter at some time during the previous year. Thus, blood eosinophils represent an important biomarker for the selection of patients treated with this novel biological agent.

IL-13 is considered as a significant mediator involved in asthmatic inflammation, airway hyper-responsiveness, mucous metaplasia, and activation and proliferation of airway fibroblasts the latest contributing to airway remodeling (Kraft 2011). Lebrikizumab, is an antibody against IL-13 which significantly improved FEV_1 when administered in asthmatic patients expressing increased serum levels of periostin, a surrogate biomarker of Th2 response (Corren et al. 2011). The aforementioned observations suggest that periostin might be a relevant biomarker for the identification of responders to anti-IL-13 therapy.

Table 1 presents a summary of the adults' studies for the use of biomarkers as tools for guided treatment strategy in asthma.

Conclusions

One of the main issues related to asthma management is the absence of consideration of the underlying inflammatory process (Boulet et al. 2015). The above issue is further amplified in the more severe forms of the disease and particularly in patients with a poor correlation between symptoms and inflammatory indices (Haldar et al. 2008). The main arising question is, whether by identifying the cellular features of asthma we would be able to improve disease management. Limited data exist with only three studies to meet the quality criteria for further discussion. We have to focus on persistent sputum eosinophilia as the main hallmark of the cellular component since it represents a cellular profile which is responsive to ICS intervention. On the other hand, no data exist for sputum neutrophilia as a tool to guide an asthma treatment strategy. Sputum eosinophilia is regarded as better indicator of airway inflammation compared to the usual clinical techniques (Hargreave and Nair 2011) and has been associated with positive steroid response. Furthermore, it has been shown to offer better titrating especially in severe asthmatics with lower steroid use and fewer exacerbations. The data from the three studies discussed above provided us with some information which needs to be further analyzed as following: the three studies were designed in a quite similar manner emphasizing the fact that sputum eosinophilic inflammation acted better as a guide to treatment compared to clinical management. We have to clarify three important points. The first one is related to what extent the clinical aspect affected the inflammatory arm of management. We all know that the three sputum studies used two arms for the asthma management. The inflammatory and the clinical one. However, if one considers the study design he will realize that even in the inflammatory arm some decisions mainly related to the use of bronchodilators have been made by the interpretation of both clinical and functional parameters. Considering the synergy and the benefit of adding a LABA to ICS we may speculate that part of the inflammatory benefit may be attributed to the proper management of both clinical and functional parameters (Bateman et al. 2004). The second issue is related to the cut-off value for eosinophils used in the three studies. The cut-off value significantly differed within studies and varied 2–6 % OR 8 %. This may represent a significant bias since titrating the dose of ICS was based on different cut-off values which leads to a reasonable speculation that some of the interventions over- or underestimate the level of eosinophilic inflammation. Interestingly, in the study of Jayaram et al. (2006) a post hoc analysis revealed that any benefit observed in their initial results is mainly applied to those asthmatics with pure eosinophilic profile. Finally, we have to consider carefully the study population in the three studies. Which was the main targeting population? That of mild to moderate asthma or the severe refractory one? Analyzing the baseline characteristics as well as the definition of exacerbations used in each study we have to clarify that Green et al. (2002a) focused on severe asthmatics and used as primary outcome severe exacerbations. Jayaram et al. (2006) did not recruit an homogenous population, meaning that some of their patients were severe asthmatics while other not. They also divided their exacerbations into those treated without and those treated with oral steroids. The Czech study (Chlumsky et al. 2006) was quite similar to that of Green et al. (2002b) mainly recruited severe asthmatics and treated all the exacerbations in a homogenous manner.

Despite the positive data of the three above studies, the current recommendations do not support sputum eosinophils as a tool for guiding treatment strategies with a limited exception. In severe asthma guidelines the technique is recommended in specialized centers (Chung et al. 2014). The time-consuming process, the need for trained personnel as well as the non-immediate results constitute the main disadvantages of this procedure Thus, evaluation of eosinophilic airway inflammation with induced sputum

Table 1 Summary of the adults' studies for the use of biomarkers as tools for guided treatment strategy in asthma

Study	Methodology	Subjects	Duration	Primary outcome	Secondary outcomes
Induced sputum					
(Green et al. 2002a)	sputum eosinophils vs BTS management	74	12 months	Patients treated with sputum strategy had fewer severe exacerbations and a lower eosinophil count	Average daily dose of ICS did not differ within groups
(Jayaram et al. 2006)	Sputum eosinophils and clinical control vs clinical control	117	24 months	Patients treated with sputum strategy had fewer severe exacerbations. The above outcome was seen in patients with moderate to severe asthma and refers in eosinophilic exacerbations	Cumulative dose of ICS was similar in both groups
(Chlumsky et al. 2006)	Sputum eosinophils and clinical control as assessed by GINA	55	18 months	Patients treated with sputum strategy had a lower rate of exacerbations	Prolonged time to first exacerbation Improvement in PFTs
FeNO					
(Smith et al. 2005)	FeNO vs GINA control	97	12 months	No significant differences in exacerbations	Lower dose of ICS for FeNO group
(Shaw et al. 2007)	FeNO vs BTS	118	12 months	No significant differences in exacerbations	Total dose for ICS did not differ
(Powell et al. 2011)	FeNO and ACQ vs ACQ	220	22 months before gestation	Significant lower rate of exacerbations	Perinatal outcomes tended to be improved

ACQ Asthma control questionnaire, *FeNO* fraction of exhaled nitric oxide, *ICS* inhaled corticosteroids, *BTS* British thoracic society, *GINA* global initiative for asthma, *PFTs* pulmonary function tests

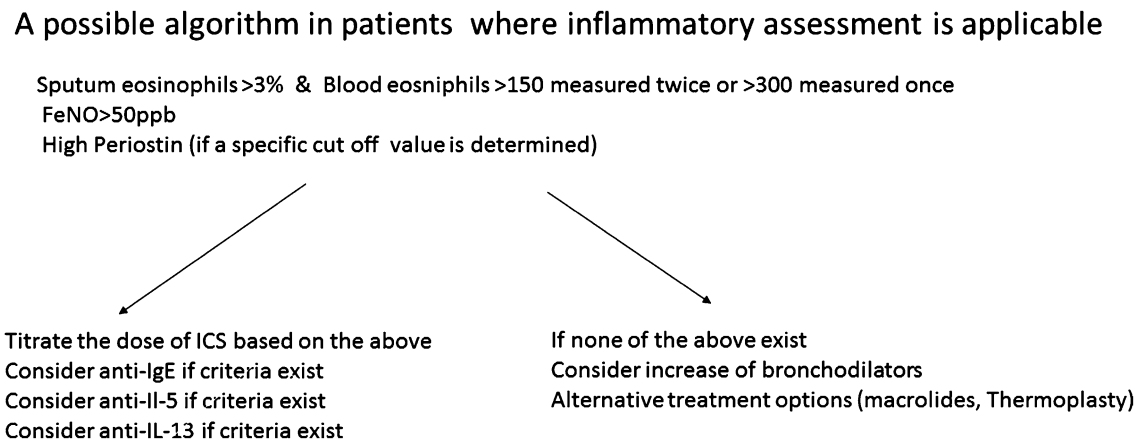

Fig. 1 A possible algorithm in patients where inflammatory assessment is applicable

could be applicable in secondary care but not feasible in primary care.

Regarding the use of FeNO to guide treatment of asthma both studies failed to show an overall benefit in the rate of exacerbations as well as in some secondary outcomes such as asthma control, lung function and use of oral prednisolone during exacerbations. We can consider them as negative studies. However, in the study by Smith et al. (2005) as well as the study by Shaw et al. (2007) a lower dose of ICS was used to maintain control. Interestingly, this was not considered as a benefit. Furthermore, in a subanalysis by Shaw et al. (2007) patients guided by the FeNO strategy and having increased levels of FeNO at baseline showed a clear benefit in regard to exacerbation rate. Interpreting the above result, we may argue that in such a study design we have to focus on patients who despite the use of ICS still have a persistent airway inflammation as assessed by increased levels of FeNO. We should also

consider simultaneously as a limitation that FeNO is still low in the non-pure eosinophilic profile (Tseliou et al. 2010). We strongly believe that the main message for asthma guided treatment strategy by FeNO is coming from pregnancy. It is important to emphasize that the proper asthma medication and the adherence to treatment are major precautions for a good outcome in pregnancy. It is well known that there is an increased risk of pre-term and post birth for fetus malformations among pregnant women with uncontrolled asthma and exacerbations during pregnancy (Murphy and Schatz 2014). So, it is important that pregnant women could keep their asthma controlled and strictly avoid exacerbations, particularly those which require oral steroids since the latest are associated with an increased risk in congenital malformations (Kallen et al. 2013). The study by Powell et al. (2011) clearly showed that the strategy based on FeNO could alter the physical route of asthma during pregnancy since it modulated all the above in a positive direction.

In a pragmatic non blinded trial where the approach was to achieve both clinical and inflammatory control, a strategy consisting of both symptoms and FeNO driven strategy resulted in a lees medication use, better quality of life and better control of the disease. So considering the applicability of FeNO in everyday clinical practice we may speculate that the above approach may provide patients significant benefits either in the clinical direction or/and in the inflammatory one.

Extending all the above data one simple but simultaneously complicating question arises. Which type of asthma requires or benefits from the inflammatory management? Definitely not the mild one since most of the parameters which drive its physical course are significantly affected by the use of ICS. Severe asthma is the ideal phenotype in order the inflammatory management to be applicable in every day clinical practice. But unfortunately many limitations exist at the moment. These are mainly attributed to the lack of a clear inflammatory phenotype, to the low availability, to the time-consuming process of the technique of induced sputum and finally to the absence of an established protocol in order to titrate the dose of ICS when amplified airway inflammation exists. If we overcome these limitations and consider that many endotypes may offer us the opportunity to apply the above techniques, we can then initiate them as attractive options for the future management of the disease. A possible algorithm for the treatment of asthma in patients where inflammatory assessment is applicable is presented in Fig. 1.

Compliance with ethical standards

Conflict of interest None of the authors have any conflicts to disclose.

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