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Risk factors associated with frequent exacerbations in asthma

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ABSTRACT

Background: Asthma is a chronic airway inflammatory disease with various degrees of severity. Exacerbations are commonly seen in uncontrolled asthma and their treatment involves oral corticosteroids use with a lot of side effects.
Objective: The aim of the study was to identify easily available predictors for future exacerbations in patients with asthma.
Methods: This is a prospective study on 250 consecutive patients with asthma with a successful sputum induction. Exacerbation rate in the following year was assessed by telephone interview. Logistic regression was used to test the relationship between the binary outcomes (<1 or ≥1 exacerbation, <2 or ≥2 exacerbations) and a set of covariates including demographic, clinical, functional and inflammatory characteristics such as FeNO, sputum and blood cell counts. The results were then applied and validated in a new cohort of 1450 patients.
Results: Sputum and blood eosinophils were able to identify patients presenting ≥1 or ≥2 exacerbations with the same discriminative power (AUC:0.65 and 0.64 respectively). The multiple regression analysis identified that exacerbations in the previous year (OR = 9.3), treatment with high doses ICS (OR = 27.1), blood eosinophils (cells/mm^3, OR = 1.8) and FEV_1/FVC (OR = 0.93) were independent predictors of exacerbations in the year following the visit with an AUC of 0.93 for this model. Frequent exacerbations (≥2) were also predicted by exacerbations in the previous year (OR = 10.5), treatment with high doses ICS (OR = 39.2) and blood eosinophils (OR = 3.5) with an AUC of 0.95 for the model.
Conclusion: Blood and sputum eosinophils have similar predictive value for future exacerbations. Prediction could be improved by combining this information with lung function, ICS dose and history of previous exacerbations.

1. Introduction

Asthma is a chronic airway inflammatory disease with various degrees of severity. Exacerbations are commonly seen in uncontrolled asthma and their treatment includes oral corticosteroids use with a lot of side effects such as diabetes, osteoporosis and peptic ulcers. Price et al. indeed reported that the dose-response relationship for cumulative systemic corticosteroids exposure with most adverse outcomes began at cumulative exposures of 1.0-<2.5 g and for some outcomes at cumulative exposures of only 0.5-<1 g, equivalent to four lifetime SCS courses [1]. We clearly need diagnostic tools to predict future exacerbations. The aim of the study was to identify easily available predictors for future exacerbations in a general population of patients with asthma and in a subpopulation of patients with severe asthma. These biomarkers would be useful to target therapy for this at-risk population. Important studies have shown that targeting sputum eosinophil count <2–3% results in marked reduction in asthma exacerbations [2,3]. It was previously shown that exacerbations are associated with eosinophilic inflammation [2,4] and that sputum eosinophil count increases several weeks before exacerbations [5,6]. Several studies including ours have previously found that sputum eosinophils was a predicting factor of loss of asthma control when stepping down ICS [5,7]. Leuppi also reported that sputum eosinophil percentage >6.3% was a predictor of loss of asthma control after ICS cessation [8]. Previous studies have highlighted that higher number of blood or/and sputum eosinophils [9–11] are associated with higher risk of exacerbations. Moreover, new therapies targeting eosinophils lead to improvement of exacerbation frequencies [12–14].

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Abbreviations list

ACQ	asthma control questionnaire
ACT	asthma control test
AQLQ	asthma quality of life questionnaire
AUC	area under the curve
BEC	blood eosinophil counts
BMI	body mass index
CRP	C reactive protein
FeNO	fraction of exhaled nitric oxide
FEV_1	forced expiratory volume in 1 s
FVC	forced vital capacity
ICS	inhaled corticosteroids
IgE	Immunoglobulin E
IL:	interleukin
OCS	oral corticosteroids
OR	odds ratio
ROC	receiver operating curve
RV	residual volume
sGaw	specific airway conductance

However there are few data in the literature concerning the thresholds for both sputum and blood eosinophilic inflammation to predict frequent exacerbations.

In a previous study, we found that patients with asthma exhibiting eosinophilic inflammation both in the blood and in the sputum were characterized by the highest sputum eosinophil counts, a higher rate of exacerbations, a poorer asthma control, and more frequent nasal polyps and chronic rhinosinusitis than patients with increased eosinophils in only one compartment and non-eosinophilic patients with asthma [9]. Induced sputum is not widely applicable in the management of moderate to severe asthma and other clinical parameters and biomarkers are available in clinical practice. Exhaled nitric oxide (FeNO) is a marker of type 2 inflammation and previous studies have shown that the combination of elevated FeNO and elevated blood eosinophils relates to more exacerbations [10,15,16] and better response to anti-IL-4/13 with regard of reduction of exacerbation frequency [17]. In this study we evaluated, which of the markers collected in the asthma clinic could be predictors for future exacerbations in asthmatics. Special focus is put to biomarkers, blood eosinophils, sputum eosinophils and FeNO, especially.

2. Methods

2.1. Study populations

We have previously conducted a prospective study on 250 patients with asthma (the discovery cohort) seen in our University Asthma Clinic in CHU of Liege, Belgium, between June 30, 2011 and January 12, 2013. This population was recruited to confirm the link between blood eosinophils and asthma control found in a retrospective analysis [9] and to identify predictors of exacerbations. The results were then applied and validated in a new cohort (the validation cohort) of 1450 patients with asthma seen between January 2013 and March 2020, none of these patients included in the discovery cohort. In this population, we performed the same measurements as in the discovery study.

In both populations, patients came from routine practice to University Hospital, Liege, and were recruited by two clinicians involved in asthma. Entry criteria were any patients with asthma age $\geq$18 years who agreed to undergo detailed investigation at the asthma clinic. The visits were not parts of an asthma trial. All the patients who had a successful sputum induction were included in the study.

2.2. Asthma diagnosis

Asthma was diagnosed based on the presence of chronic respiratory symptoms such as cough, breathlessness or dyspnea together with the demonstration of airflow variability. The latter was defined by airway hyperresponsiveness shown by one or more of the following: increase in forced expiratory volume in 1 s (FEV_1) of 12% and 200 mL following inhalation of 400 mg salbutamol; or inhaled concentration of methacholine provoking a 20% fall in FEV_1 of 16 mg/mL. Methacholine challenge was performed according to a standardized methodology as previously described.

2.3. Comorbidities

Subjects were characterized as atopic if they had at least one positive specific IgE test (0.35 kU/L; Phadia, Groot-Bijgaarden, Belgium) for at least one common aeroallergen (cat, dog, house dust mites, grass pollen, tree pollen and a mixture of moulds). Nasal polyps and sinusitis was diagnosed by an ear, nose and throat physician either by endoscopy or sinus computed tomography. Gastro-esophageal reflux was diagnosed either by symptoms of pyrosis at history taking or the presence of esophagitis demonstrated by gastroscopy.

2.4. Exacerbation definition

Exacerbation of asthma are episodes characterized by a progressive increase in symptoms such as shortness of breath, cough, wheezing or chest tightness and progressive decrease in lung function sufficient to require a change in treatment. In our study, we collected data on severe exacerbation defined by the requirement of a course of oral corticoids for $\geq$3 days or hospitalization for a case of asthma worsening [18–21]. Exacerbations in the previous year were collected at the visit to the asthma clinic during which treatment was initiated or adjusted according to asthma control, lung function and inflammatory markers in the sputum and in the blood at the discretion of the clinician. Most of exacerbations were managed at the asthma clinic and recorded in the medical file during the last twelve months but some of them were treated by the general practitioner and reported by the patients during the visit at the asthma clinic. Exacerbation rate in the following year was measured through a telephone call by a nurse given 12 months after the visit to the asthma clinic.

2.5. Statistical analyses

We evaluated the value of FeNO, sputum eosinophils and blood eosinophils taken in different combinations of normal and/or elevated values as predictor of exacerbations. We also conducted a logistic regression analysis to assess the relationship between the binary outcomes ($<$1 or $\geq$1 exacerbation in the year following the visit, $<$2 or $\geq$2 exacerbations in the year following the visit and ACQ$<$1.5 or $\geq$1.5) and a set of covariates, individually or in combination. Covariates included FeNO (log-transformed), age, age of onset, ICS dose, sputum eosinophil counts, sputum neutrophil counts, FEV_1% predicted, FEV_1/FVC, blood eosinophils, fibrinogen, CRP, IgE, gender, BMI, gastro-esophageal reflux, chronic rhinosinusitis, smoking history and exacerbations in the last year. The results were considered to be significant at the 5% critical level ($p < 0.05$). Calculations were done using SAS Version 9.1 (SAS Institute, Cary, North Carolina, USA).

2.6. Ethics

This study was conducted with the approval of the ethics committee of CHU Liege and all patients gave written informed consent.

3. Results

The demographic, functional and inflammatory characteristics of the prospective population were described earlier [9] and were similar to those of the validation study except for sputum eosinophils that were slightly higher in the discovery population (Table 1).

3.1. Prediction of exacerbations

3.1.1. FeNO, sputum eosinophils and blood eosinophils

In this study we first evaluated the ability of well-known type-2 biomarkers for the prediction of exacerbations, using thresholds reported in the literature for normal values. We classified patients according to the level of FeNO, blood eosinophils and sputum eosinophils and looked at the number of exacerbations observed within each category (Table 2). The percentage of patients exhibiting at least one or at least two exacerbations was higher in patients with blood eosinophils ≥360/mm^3, sputum eosinophils ≥3% or FeNO >50 ppb. When combining these biomarkers, the proportion of patients with exacerbations was higher in patients with high blood and sputum eosinophils, whatever the FeNO level (Table 3).

3.1.2. New thresholds for the prediction of exacerbations

Looking at cut-offs discriminating between normal and abnormal values of type-2 biomarkers is probably not the best tool for the prediction of exacerbations. Here we provide the best cut-offs to predict exacerbations in a large population of patients with asthma.

In the discovery study (n = 250), sputum and blood eosinophils were able to discriminate between patients presenting at least one exacerbation from those without exacerbation in the previous year with a best cut-off of 7.2% for sputum eosinophils (Sensitivity: 54%, specificity: 76%, AUC: 0.65, p = 0.004) and 360/mm^3 for blood eosinophils (sensitivity 54%, specificity: 78%, AUC:0.64, p = 0.025). There was no significant difference in the ability of sputum or blood eosinophils to predict exacerbations. When sputum and blood eosinophils were taken together for the prediction of at least one exacerbation in the following year, the prediction was not improved as compared to sputum eosinophil or blood eosinophils alone (AUC:0.67, p = 0.016, Fig. 1). FeNO was however not able to discriminate between exacerbators and non-exacerbators (p = 0,17, AUC: 0.52).

We classified patients according to these new thresholds for blood eosinophils ≥ or <360/mm^3 and sputum eosinophils ≥ or <7.2% for the prediction of the risk of exacerbations. We found that patients with asthma exhibiting an increase in sputum or in blood and sputum eosinophils had a higher rate of exacerbations than patients with blood eosinophils below 360/mm^3 and sputum eosinophils <7.2% (Fig. 2). Patients had fewer exacerbations in the year following the visit, after treatment was adjusted or initiated according to inflammatory characteristics.

In the validation cohort (n = 1450), we classified patients according to the thresholds of interest for the prediction of the risk of exacerbations. We found that patients with asthma having increased blood and sputum eosinophils had a higher rate of exacerbations than patients with blood eosinophils below 360/mm^3 and sputum eosinophils <7.2% (Fig. 3).

Table 1
Demographic, clinical and inflammatory characteristics of the validation population. Data are presented as median (interquartile range), mean±standard deviation, % or n (%).

	Discovery cohort	Validation cohort
N	250	1450
Gender (M/F)	99/151	607/843
Age, yrs	50 (36–65)	52 (38–62)
Height, cm	168 ± 9	168 ± 10
Weight, kg	73 ± 17	76 ± 16
BMI	27 ± 6	27 ± 5
Atopy (%)	59%	64%
Current smokers (%) (pack-yr)	22% (25 (2–60))	19% (18 [8–34])
Ex-smokers (%) (pack-yr)	15% (17 (0.5–63))	17% (15 [7–30])
FEV$_1$, %predicted	82 ± 21	84 ± 21
FEV$_1$/FVC, %	71 ± 15	74 ± 10
FeNO	25 (8–58)	22 (12–42)
Blood eosinophils,/mm^3	188 (89–407)	173 (99–309)
Blood eosinophils, %	2,4 (1,2–4,2)	2,5 (1,3-4)
Sputum eosinophils, %	2,8 (0,9–12)	1,4 (0,2–7,5)
Sputum neutrophils, %	49 (25,9–78)	62 (37,9-80,5)

Table 2
Proportions of exacerbations according to FeNO levels, blood eosinophils and sputum eosinophils in the discovery study (N = 250) and validation study (N = 1450).

	≥1 exacerbation		≥2 exacerbations	
	Discovery	Validation	Discovery	Validation
	N = 250	N = 1450	N = 250	N = 1450
FeNO				
<25 ppb	21%	30%	20%	17%
25–50 ppb	24%	30%	14%	19%
≥50 ppb	39%	36%	45%	23%
Blood eosinophils				
<360/mm^3	20%	26%	19%	14%
≥360/mm^3	39%	45%	44%	31%
Sputum eosinophils				
<3%	18%	27%	17%	14%
≥3%	34%	38%	35%	25%

Table 3
Percentage of patients exhibiting low asthma control, more than one exacerbation in the year following the visit or more than two exacerbations in the year following the visit according to the baseline value of FeNO, blood eosinophils and sputum eosinophils (n = 1450).

FENO	Blood Eos	Sputum Eos	N	Exacerb ≥1 (%)	Exacerb ≥2 (%)
<50	<360	<3	727	33	19
		≥3	276	38	26
	≥360	<3	62	47	35
		≥3	118	67	55
≥50	<360	<3	31	39	19
		≥3	133	36	27
	≥360	<3	14	64	57
		≥3	89	60	58

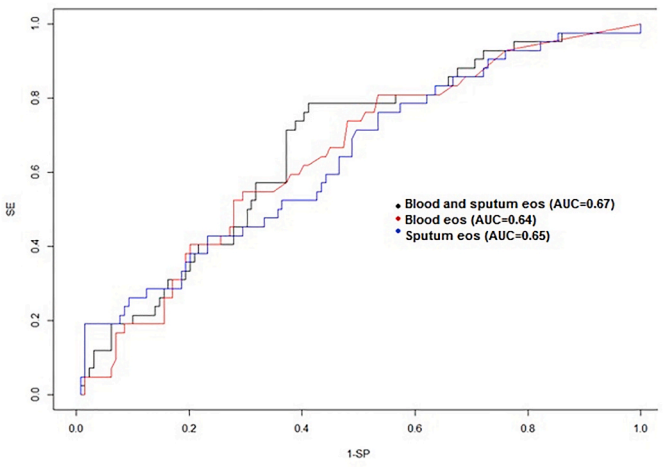

Fig. 1. ROC curves for the prediction of at least one exacerbation in the year following the visit (prospective cohort, n = 250).

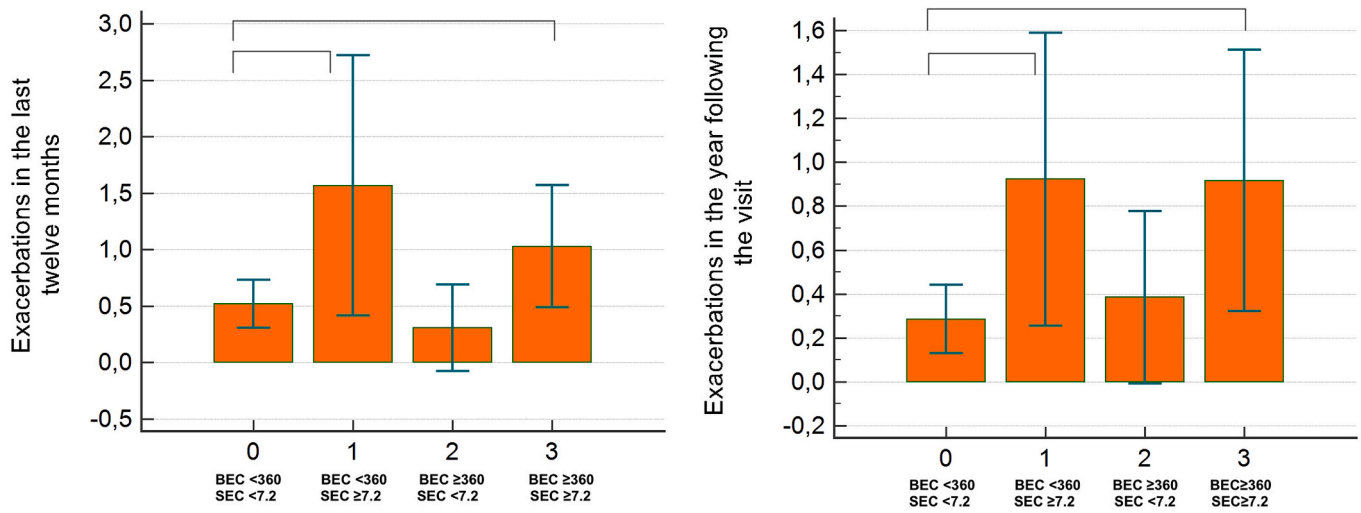

Fig. 2. Exacerbations according to blood and sputum eosinophil counts. Left panel: exacerbation during the 12 months prior to the visit at the asthma clinic. Right panel: exacerbations in the year following the visit. Group 0: Blood eosinophils <360/mm^3 and sputum eosinophil counts <7.2% (n = 145 and 140 respectively). Group 1: Blood eosinophils <360/mm^3 and sputum eosinophils ≥7.2% (n = 51 and 57 respectively), Group 2: blood eosinophils ≥360/mm^3 and sputum eosinophils <7.2% (n = 15 and 18 respectively) and group 4: blood eosinophils ≥360/mm^3 and sputum eosinophils ≥7.2% (n = 39 and 35 respectively). N = 250. Connecting line: $p < 0.05$.

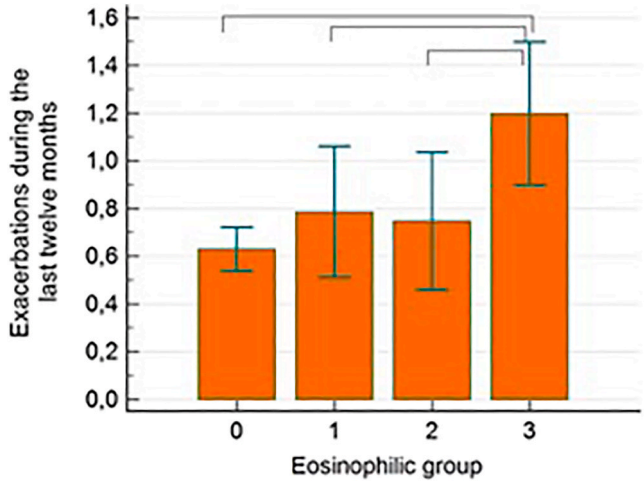

Fig. 3. Exacerbations according to blood and sputum eosinophil counts. Group 0: Blood eosinophils <360/mm^3 and sputum eosinophil counts <7.2% (N = 974). Group 1: Blood eosinophils <360/mm^3 and sputum eosinophils ≥7.2% (N = 188), Group 2: blood eosinophils ≥360/mm^3 and sputum eosinophils <7.2% (N = 118) and group 4: blood eosinophils ≥360/mm^3 and sputum eosinophils ≥7.2% (N = 170). N = 1450. Connecting line: $p < 0.05$.

3.1.3. Predictors of at least one exacerbation identified by the logistic regression analysis

In the discovery cohort, a univariate logistic regression analysis was built to identify predictors of at least one exacerbation in the following year. Sputum eosinophils (AUC = 0.65, OR = 1.6, 95%CI: 1.1–1.9, p = 0.004), FEV_1% predicted (AUC = 0.70, OR = 0.96, 95%CI:0.95–0.98 p < 0.0001), FEV_1/FVC (AUC = 0.73, OR = 0.93, 95%CI:0.89–0.96, p < 0.0001), sGaw (AUC = 0.6, OR 1.31, p < 0.0001), residual volume (AUC = 0.59, OR = 1.29, p = 0.002), blood eosinophils taken in % (AUC = 0.61, OR = 2.1, 95%CI: 1.1–3.9, p = 0.022) and in absolute value (AUC = 0.64, OR = 1.64, 95%CI:1.1–2.5, p = 0.025), total serum IgE levels (AUC: 0.62, OR = 1.33, 95%CI: 1.0–1.7, p = 0.022), high dose of ICS (AUC = 0.79, OR = 34, 95%CI:7.4–155, p < 0.0001), ACQ >1.5 (AUC = 0.63, OR = 3.3, 95%CI:1.5–7.5 p = 0.0036) and exacerbations in the previous year (AUC = 0.72, OR = 18, 95%CI:6.6–51, p < 0.0001) were found to be able to discriminate between patients with at least one exacerbation from those without exacerbation while age (p = 0.23), height (p = 0.91), weight (p = 0.54), BMI (p = 0.60), smoking history (p = 0.75), presence of esophageal reflux (p = 0.27), chronic rhinosinusitis (p = 0.97), sputum neutrophils (p = 0.65), FeNO (p = 0.13), blood neutrophils (p = 0.055), fibrinogen (p = 0.45), CRP (p = 0.11), neutrophil-to-lymphocyte ratio (p = 0.88) and age of onset (p = 0.35) were not significant.

Then we conducted a multivariate logistic regression analysis in order to highlight independent predictors of exacerbations in patients with asthma. We applied a stepwise selection of all non-redundant variables for which p-value was <0.10 in the univariate analysis. The multiple regression analysis identified that exacerbation in the previous year (OR = 9.3, p = 0.0001), treatment with high doses ICS (OR = 27.1, p = 0.0006), blood eosinophils (/mm^3, OR = 1.8, p = 0.044) and FEV_1/FVC (OR = 0.93, p = 0.0031) were independent predictors of exacerbations in the year following the visit. This model gave an AUC of 0.93.

3.1.4. Predictors of at least two exacerbations identified by the logistic regression analysis

We also looked at patients presenting at least 2 exacerbations as compared to those with less than 2 exacerbations per year in the discovery cohort. Sputum and blood eosinophils had comparable accuracy for the discrimination between those populations with a best cut-off of 8.8% (sensitivity: 58%, specificity: 70%, AUC: 0.64, p = 0.023) and 380/mm^3 (sensitivity 54%, specificity: 78%, AUC: 0.65, p = 0.01) respectively.

The univariate logistic regression analysis applied for the prediction of at least two exacerbations in the following year confirmed that sputum eosinophils (OR = 1.42, p = 0.023), FeNO (OR = 1.59, p = 0.045), FEV_1% pred (OR = 0.97, p = 0.001), FEV_1/FVC (OR = 0.94, p = 0.0025), blood eosinophils (/mm^3, OR = 2.12, p = 0.01), total serum IgE (OR = 1.39, p = 0.031), high dose of ICS (OR = 42.1, p < 0.0001), ACQ (OR = 6.68, p = 0.0028), at least 2 exacerbations in the previous year (OR = 28.1, p < 0.0001) were predictors of at least two exacerbations in the year following the visit while the other parameters were not significant. The only new predictor as compared to prediction of one exacerbation was FeNO value.

The multiple logistic regression analysis found that exacerbations in the previous year (OR = 10.5, p = 0.0009), treatment with high doses ICS (OR = 39.2, p = 0.0005) and blood eosinophils (OR = 3.5, p =

0.024) were independent predictors of at least two exacerbations in the following year. This model gave an AUC of 0.95.

3.2. Construction of Venn diagram with key variables associated with frequent exacerbations

We constructed a Venn diagram to highlight the proportions of patients exhibiting exacerbations according to the best predictors including at least two exacerbations in the previous year, treatment with high dose ICS and blood eosinophils ≥360/mm^3 (Fig. 4).

We looked at clinical and inflammatory characteristics of patients classified according to these predictors in the Venn diagram (Table 4). Patients combining the three predictors had poorer asthma control, higher airway obstruction, higher serum IgE levels, lower conductance and signs of air trapping. Patients with asthma having high blood eosinophil counts and history of exacerbations had similar characteristics as those combining the three predictors while patients with history of exacerbations and high dose of ICS or patients with asthma having as only predictor, a history of exacerbations had poor asthma control and air trapping.

4. Discussion

In a population of 250 patients with asthma recruited prospectively, we have shown that exacerbations in the previous year, treatment with high doses ICS and blood eosinophil counts are independent predictors of exacerbations in patients with asthma. Patients with elevated blood and sputum eosinophils were characterized by higher exacerbation rate, poorer asthma control and distal airway dysfunction. We also provide thresholds for blood and sputum eosinophils that allow discrimination of patients exhibiting exacerbations.

In our study, the best thresholds for blood eosinophils to predict at least one exacerbation in the subsequent year was 360/mm^3 which is very close to the one reported by Price et al. as associated with increased exacerbation risk [22]. Other studies have shown that patients with asthma exhibiting blood eosinophils upper than 400/mm^3 experienced more severe exacerbations [9,11,22,23] and had poorer asthma control [22]. A large recent study showed that the highest the number of blood eosinophils, the highest the exacerbation rate [22]. In our study, we did not find a relationship between blood eosinophil count and asthma control. In the same way, in the DREAM study, blood eosinophil counts appeared to be more strongly associated to exacerbation risk than to measure asthma control [24]. This supports the view that symptoms and exacerbation risk in asthma are to some extent poorly associated and reflect different aspects of the disease [25]. Price et al. confirmed the link between blood eosinophils and frequent exacerbations in another study in which they found a similar OR of 1.48 in patients exhibiting blood eosinophil count >400/mm^3 [26]. Westerhof et al. confirmed that

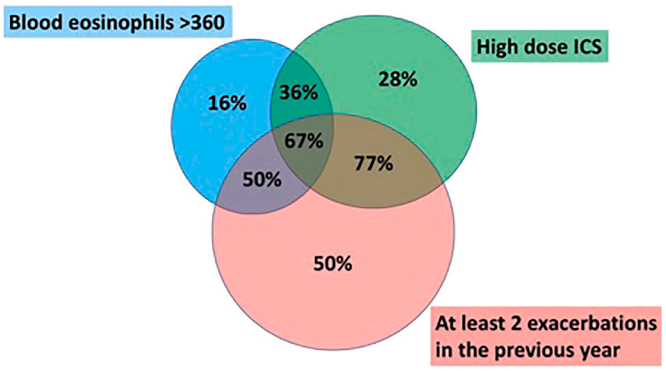

Fig. 4. Venn diagram. Percentage of patients presenting at least one exacerbation in the year following the visit are presented in circles depending on blood eosinophil counts, dose of ICS and exacerbations in the previous year.

blood eosinophils were predictors of exacerbations in never smokers [27] while the analysis of SARP-3 database suggested that blood eosinophils, bronchodilator responsiveness and body mass index were associated with exacerbation frequency [28]. The predictive power of our threshold for blood eosinophils is however limited due to poor sensitivity and will therefore be useful for clinical decision making only if combined with the other cofactors such as exacerbations in the previous year and treatment with high doses ICS that are easily available.

Several of the predictors of exacerbation identified in our study have been previously reported as significantly increasing the risk of frequent exacerbations. We found that the history of exacerbations in the previous years was a predictor of frequent exacerbations in the subsequent year. This is in line with what was found in chronic obstructive pulmonary disease. Price et al. found similar results with increased risk observed in patients who received oral corticosteroids in the previous year with an OR between 3.75 (for patients receiving one OCS course) and 25.7 (for patients receiving 3 OCS course [26]. In our study, exacerbations were not only defined by patients taking oral corticosteroids but also patients requiring hospitalization. Other groups have also suggested that asthma exacerbations in the past were key predictors of future exacerbations [29,30].

We found that patients receiving high dose ICS were more prone to exacerbations with an OR of 27.1. Price et al. [26] also found that patients receiving more than 800 μg/d FP equivalent exhibited more exacerbations with an OR of 1.29. The study of Westerhof et al. [27] also found that higher ICS dose was associated with frequent exacerbations. Bateman et al. [31] developed a risk score for asthma exacerbations but the candidate predictors that were selected in that study were clinical and functional parameters. This group included neither inflammatory markers nor the history of exacerbations. They found that patients receiving GINA step 4 treatment, meaning high dose of inhaled corticosteroids, had a 60% higher exacerbation risk.

The multivariable analyses of Price et al. however yielded more than 20 different exacerbation predictors [26]. We did not confirm all the predictors reported in this large study. Some of the predictors were identified in our univariate analysis but not confirmed as independent predictors in the multivariate analysis. The study of Price et al. was not a prospective study. In our prospective study, we confirmed several results of the retrospective study of Price. Moreover, the study of Price et al. did not look at induced sputum inflammatory cells.

In our study, the best threshold for sputum eosinophil counts to predict exacerbations was 7.2%. Studies evaluating the ability of sputum eosinophils to predict the response of patients with severe asthma to biologics in terms of reduction of exacerbations used a cut-off of 3% to select patients [24]. Our study however shows that the best threshold for the prediction of exacerbations is 7.2%. Using this cut-off value would certainly increase the power of the use of sputum eosinophils to select patients in whom a decrease in exacerbations would be obtained with the use of biologicals.

Sputum eosinophils were correlated with exacerbation rate but were not identified as independent predictor in the multivariate analysis probably because they reflect the same process as blood eosinophils. Moreover, in our study, treatment was adapted according to sputum results obtained at the first visit. If sputum eosinophil counts were upper than 3%, the dose of inhaled corticosteroids was increased, thereby decreasing the risk of exacerbations during the following year. When asthma was well controlled and induced sputum eosinophils were lower than 3%, the dose of ICS was reduced. It has been previously shown that targeting sputum eosinophils below 3% resulted in marked reduction in asthma exacerbations [2]. The change in treatment according to sputum results and the consecutive decrease in exacerbations may also play a role in the absence of identification of sputum eosinophils as an independent predictor. It might also be that looking at a severe asthma population would increase the power of sputum inflammation to predict exacerbations. Looking at longitudinal measures, Walsh et al. indeed found that patients with severe eosinophilic asthma phenotype (defined

Table 4
Clinical and inflammatory characteristics of patients according to the presence of absence of predictors of exacerbations. *p < 0.05 as compared to group 1, ¨p < 0.05 compared to group 2, ‘p < 0.05 as compared to group 3, ° p < 0.05 with group 4, ‘p < 0.05 with group 5, ᶜ p < 0.05 with group 6, ᶲp < 0.05 with group 7. BEC: blood eosinophil counts. High ICS: daily dose of inhaled corticosteroids of >1000 mcg beclomethasone equivalent. Exacerb: at least two exacerbations.

	BEC>360	BEC>360 High ICS	High ICS	BEC>360 High ICS Exacerb	BEC>360 Exacerb	High ICS Exacerb	Exacerb	None
	Group 1	Group 2	Group 3	Group 4	Group 5	Group 6	Group 7	Group 8
N	115	121	374	122	12	136	35	470
ACQ	1.71 (0.86–2.71	2.14 (1.14–3)*°	2.14 (1–2.86)°	3.14 (2.43–4)*¨	3.29 (2.5–4)*¨	2.86 (2–3.71)*¨	2.71 (1.75–3.54)*°	1.92 (0.71–2.14)*°‘ᶜᶲ
ACT	16 (13–21)	14 (10–20)*°	16 (11–20)°	10 (8–15)*¨	9 (7–14)*¨	11 (8–14)*¨	12 (9–18)*°	18 (14–22)*°‘ᶜᶲ
AQLQ	5 (4–5.9)	4.5 (3.3–5.7)*	4.3 (3.3–5.7)*	3.5 (2.8–4.3)*¨	3.4 (2.2–3.9)*¨	3.4 (2.7–4.1)*¨	4.1 (3.3–5.1)*ᶜ	5.3 (4.1–6.1)°‘ᶜᶲ
FeNO	37 (19–77)	35 (15–54)	20 (12–38)*	34 (20–60)’	45 (26–133)’	21 (11–40)*°¨	16 (12–25)*°¨	19 (12–33)*°¨
IgE	122 (51–332)	184 (62–401)	113 (39–394)°	187 (69–466)’	290 (62–1202)	136 (50–407)	86 (27–329)°	81 (24–241)*°‘ᶜ
FEV_1 post-BD, %	94 (80–105)	83 (69–100)*	86 (71–97)*	75 (63–89)*¨	68 (56–88)*	82 (61–92)*¨	86 (64–93)*	97 (86–108)*°‘ᶜᶲ
sGaw	0.79 (0.55–0.98)	0.7 (0.55–1.1)	0.69 (0.49–1)	0.49 (0.38–0.78)*¨	0.45 (0.3–0.99)	0.64 (0.43–0.95)*°	0.81 (0.6–1)°	0.87 (0.67–1.1)*°‘ᶜ
RV, %	116 (95–137)	134 (116–161)*	122 (95–149)’	136 (102–157)*	118 (90–159)	134 (101–155)*	134 (108–155)	112 (92–135)°ᶜᶲ

as sputum eosinophils ≥2% at three time points) had shorter time to first exacerbation and greater risk of exacerbation than patients with non-eosinophilic asthma [32]. In the SARP-3 study [28,33], while the median levels of blood eosinophils did not differ between exacerbators and non-exacerbators, the median value of sputum eosinophils was lower in the group without exacerbation. The measure of type-2 inflammation likely relates to the risk of exacerbation. It seems also clear that eosinophils must be attracted into the airways to induce exacerbations and there is an imperfect correlation between blood eosinophils and sputum eosinophils [9] in patients with asthma. In our study, high doses of ICS were found as predictor of exacerbations and doses of ICS were adapted according to induced sputum eosinophil counts. This might be a reason why blood but not sputum eosinophils were identified as independent predictors in the multivariate analysis. The ongoing therapy has indeed differential response on blood and sputum eosinophils [34]. This emphasizes the need to include more complex biological information than merely eosinophil counts to predict clinical outcome. Other biomarkers in exhaled breath such as VOCs might certainly help to predict the risk of exacerbations as they may reflect local inflammation [35,36].

Other markers present in the sputum might be of interest such as the sputum 6-gene signature providing significant prediction of exacerbations with an AUC of 0.68 [37]. In this study of Fricker [37], they surprisingly did not confirm the ability of blood eosinophils to discriminate between those experiencing at least one exacerbation and those who did not. For the discrimination of patients exhibiting at least 2 exacerbations from others, sputum eosinophil counts (AUC 0.7) and the 6-gene signature (AUC 0.76) provided significant discriminatory capacity in that study. They also found that OCS history could predict future exacerbations frequency. Neutrophil-to-lymphocyte ratio has been previously suggested as a novel predictor of exacerbations in patients with asthma [38]. However, the ability of this ratio when higher than 2.1 was poor to discriminate between exacerbators and non-exacerbators in our study.

Our study has some limitations. Patients with asthma recruited in the discovery and validation cohort were not a true general population of patients with asthma as they were referred by their general practitioner or their pulmonologist to the asthma clinic for their follow-up. Moreover, we did not include children and adolescents in our study. Similar observations were however made in pediatric populations. Hoch et al. indeed found that young patients with asthma having higher blood eosinophil counts taken in percentage or in absolute value, higher treatment step and recent exacerbations were more prone to exacerbate during follow-up [39]. Moreover, Teach et al. found that recent exacerbations and higher blood eosinophil counts were predictors of exacerbations in children with asthma [40].

In conclusion, our study confirms that easily available biomarkers such as the dose of inhaled corticosteroids, the number of exacerbations within the last twelve months and the level of blood eosinophils may help the pulmonologist to identify patients with asthma who are at risk of exacerbations. These predictors may be used to optimize treatment and act as prognostic factors as they reflect exacerbation risk. It is of upmost importance to pay attention to this at risk population in whom regular exacerbations may induce remodeling and irreversible airway obstruction and in whom repeated prescription of systemic corticosteroids may induce irreversible side effects.

Author contribution

Florence Schleich; Conceptualization, Formal analysis, investigation, Writing - original draft, Andrei Malinovschi; Conceptualization, Formal analysis, investigation, Writing - original draft, Anne Chevremont; Database cleaning, Data curation, Laurence Seidel; Statistical analysis, Formal analysis, Renaud Louis; Writing - original draft

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Design of the study: FS, AM, RL. Acquisition of data: FS, AC, RL. Data analysis and interpretation: FS, AM, LS, RL. Drafting the work: FS, AM, RL.

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