Relevance of Th2 markers in the assessment and therapeutic management of severe allergic asthma: a real life perspective

Running title: Asthma severity markers are disputable

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Abstract

Background: Although blood eosinophils are currently recognized as the main clinical marker of Th2 inflammation, their relevance in identifying asthma severity is still matter of debate.

Methods: Our retrospective real-life study on severe asthmatics included in the NEONet Italian database, aimed at investigating the relevance of blood eosinophil count and FeNO in severe asthma clinical assessment and their role as a potential predictor of responsiveness to anti-IgE therapy. As cut-off values 300 blood eosinophils/mm3 and 30 ppm for FeNO were chosen.

Results: Overall 132 adult patients were evaluated. No significant differences could be observed between the high and low basal eosinophils groups, in terms of demographic data, total IgE, lung function, Patient Reported Outcomes (PROs), nasal comorbidities. The patients with FeNO $\geq$ 30 ppb showed a worse ACT score, and a lower AQLQ score in comparison with the FeNO< 30 ppb ones. In the high FeNO subgroup, more frequent hospital admissions and a higher number of lost working days in the last year were registered. A combined score including both eosinophils and FeNO did not to improve the accuracy of the single parameters. In the high eosinophil subgroup the proportion of responders to omalizumab treatment was greater and ~~significantly~~ increased at every follow-up time point.

Conclusions: According to our findings blood eosinophils do not represent a univocal marker of asthma severity, whilst a higher FeNO level is associated with more frequent hospital admissions and lost working days. Blood eosinophils seem to act as predictor of treatment responsiveness to omalizumab.

Key words: Severe Asthma, Eosinophils, Omalizumab, Biomarker, Th2 Inflammation, Asthma Network.

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Resumen

Antecedentes: Aunque los eosinófilos en la sangre actualmente son reconocidos como el principal marcador clínico de la inflamación Th2, su relevancia en la identificación de la gravedad del asma sigue siendo un tema de debate.

Métodos: Nuestro estudio retrospectivo de la vida real sobre asmáticos graves, incluido en la base de datos italiana de NEONet, tuvo como objetivo investigar la relevancia del recuento de eosinófilos en sangre y el FeNO en la evaluación clínica del asma grave y su función como posible factor predictivo de la capacidad de respuesta al tratamiento con anti-IgE. Como valores de corte se eligieron 300 eosinófilos / mm3 en sangre y 30 ppm para FeNO.

Resultados: En total se evaluaron 132 pacientes adultos. No se pudieron observar diferencias significativas entre los grupos de eosinófilos basales altos y bajos, en términos de datos demográficos, IgE total, función pulmonar, resultados informados por el paciente (PRO) o comorbilidades nasales. Los pacientes con $\geq$ FeNO 30 ppb mostraron una puntuación de ACT peor y una puntuación AQLQ más baja en comparación con los de FeNO <30 ppb. En el subgrupo de FeNO alto, se registraron ingresos hospitalarios con más frecuencia y un mayor número de días de trabajo perdidos en el último año. Una puntuación combinada que incluye tanto a los eosinófilos como el FeNO no mejoró la precisión de los parámetros individuales. En el subgrupo de eosinófilos altos, la proporción de pacientes que respondieron al tratamiento con omalizumab fue mayor y aumentó significativamente en cada punto de tiempo de seguimiento.

Conclusiones: De acuerdo con nuestros hallazgos, los eosinófilos en sangre no representan un marcador unívoco de la gravedad del asma, mientras que un nivel más alto de FeNO se asocia con más ingresos hospitalarios y más días de trabajo perdidos. Los eosinófilos de la sangre parecen actuar como predictores de la respuesta del tratamiento al omalizumab.

Palabras clave: Asma Grave, Eosinófilos, Omalizumab, Biomarcador, Inflamación Th2, Red De Asma

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Introduction

According to recent studies, eosinophils play a crucial role in asthma pathogenesis and clinical management. Actually, they characterize the Type 2 asthma phenotypes, including early-onset atopic asthma as well as late-onset asthma with nasal polyps [1]. Furthermore, high eosinophil blood count is predictive of increased eosinophil inflammation in the sputum [2,3]. On clinical ground the eosinophil blood count has been recently identified as a reliable biomarker to select patients eligible to biological treatments [4-11]. Fractional exhaled nitric oxide (FeNO) is considered a surrogate of airways eosinophilic inflammation, and it has been described as a good marker of eosinophilic bronchial inflammation and, at a less extent, of blood eosinophilia [12]. In day-to-day asthma management there is an increasing need for biological markers related to the severity of the disease, or able to predict it [13]. However, in order to be widely used, such biomarkers have to be feasible, specific, not time consuming as well as not expensive. In the present real life study, carried out in a population of patients with severe allergic asthma according to the ERS/ATS criteria [14] and selected for omalizumab treatment, we investigated the relevance of well-known TH2-inflammation clinical biomarkers (blood eosinophil count and FeNO) in the frame of severe asthma and their correlation with clinical and functional parameters at baseline. Furthermore the role of baseline eosinophils level as a potential predictor of responsiveness (6, 12 and 18 months follow-up evaluation) to the treatment was explored.

Material and Methods

A retrospective analysis of the North East Omalizumab Network (NEONet) database was carried out. A detailed description of the Network in terms of aims and methods is provided elsewhere [15]. In brief, NEONet is a non-profit initiative involving 19 Allergy and Respiratory Referral Centres

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for Severe Asthma located in the North- East region of Italy and approved by the local ethics committee. NEONet aims at providing real word evidence data, by collecting homogeneous clinical information from adult patients affected by severe allergic asthma and undergoing omalizumab treatment in a real-life setting in order to produce some new insights concerning the current unmet needs (e.g. impact of omalizumab treatment on lung function and on asthma comorbidities, long-term follow-up of treated patients, adherence, non-responders profile, optimal treatment duration). The participating clinicians, once obtained informed consent from the patients, enter anonymous coded data into a shared limited-access web platform. For the present study clinical data, lung function, eosinophil blood count and FeNO levels registered at the enrolment visit were analysed. The sensitization profile was also assessed by dosing blood total and specific IgE. In order to cluster patients with higher eosinophilic inflammation 300 eosinophils/mm3 and 30 ppm for FeNO were chosen as cut-off values [16]. Omalizumab doses and treatment schedule were established according to AIFA (Agenzia Italiana del Farmaco – Italia Drug Regulatory Agency) criteria [17]. Blood eosinophil count was monitored at 6, 12 and 18 months follow-up visits and matched with the evaluation of treatment responsiveness, assessed at the same time points. Treatment responsiveness evaluation relied on GETE (Global Evaluation of Treatment Efficacy) Questionnaire [18]. GETE is a five-point scale, including 5 possible outcomes: excellent (complete control of asthma), good (marked improvement), moderate (discernible, but limited improvement), poor (no appreciable change) and worsening. According to the rating of symptoms control the patients were calissified as ‘responder’ (GETE: excellent’/‘good’) or ‘not responder’ (GETE: ‘moderate’ / ‘poor’).

Statistical analysis

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Results are expressed as mean and standard deviation if variables are continuous and as a percentage if variables are categorical. The Shapiro-Wilk test was used to test the normality for continuous variables. The two-sample t-test or the Wilcoxon (Mann-Whitney) rank-sum test was used to compare the mean of continuous variables while the Anova analysis or Kruskal-Wallis rank test was used when the mean comparison regarded more than two independent groups. A p-value < 0.05 was to be considered statistically significant. Analysis were performed using STATA version 15 (StataCorp, College Station, TX, USA).

Results

The population sample included 151 adult patients. Nineteen subjects where excluded, as an oral steroid treatment was ongoing at the time of the enrolment and eosinophil count assessment. Overall 132 patients were analysed. As previously mentioned, all the included patients had been selected for omalizumab treatment, and they were receiving a GINA step 5 treatment. All the patients were on regular treatment with a combination of ICS (mean dose: 1080.5 +/-487.3 mg of fluticasone propionate equivalents) plus long-acting β-agonists (LABA). Furthermore, in 41.7% of patients the pharmacological treatment also included a leukotriene receptor antagonist and in 39.4% a long-acting muscarinic antagonist. Demographic data are summarized in Table 1.

As shown in Table 2, at baseline no significant differences could be observed between the high and low basal eosinophils groups, in terms of demographic data, total IgE, lung function assessment, Patient Reported Outcomes (PROs). Concerning the nasal comorbidities, rhinitis was slightly more prevalent in the high eosinophil group than in the low one (86.8% vs 69.6%, p: 0.075), whilst nasal polyposis did not reproduce the same trend, being quite uniformly distributed

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in the two groups. Also, the average number of lost working days in the last year is higher in the high eosinophils group when compared with low eosinophils one.

Table 3 summarizes the baseline clinical and functional features of patients when dividing the study population according to the FeNO values, at the cut-off of 30 ppb. Significant differences between the two subgroups could be observed in terms of Body Mass Index (BMI), which is higher in the low FeNO group; in the last, the proportion of patients undertaking oral steroids was significantly lower. The patients with FeNO $\geq$ 30 ppb showed a lower Asthma Control Test (ACT) score, and a lower Asthma Quality of Life Questionnaire (AQLQ) score in comparison with the FeNO< 30 ppb ones. Besides Patient Reported Outcomes, asthma control was worse in the high FeNO subgroup, characterized by more frequent hospital admissions and a higher number of lost working days in the last year.

As shown in Table 4, a combined score including both eosinophils and FeNO seems not to improve the accuracy of the single parameters in discriminating the clinical severity of the disease.

Concerning treatment responsiveness, although without statistical significance, the proportion of responders according to the GETE questionnaire was higher in the high eosinophil subgroup whilst in the low eosinophils subgroup the number of non-responders was prevalent (Figure 1). Furthermore, within the high eosinophil subgroup the proportion of responders increased at every follow-up time point, in comparison with the previous one. On the opposite, in the low eosinophils subgroup a similar trend was not so evident.

Discussion

Our real-life study, including severe asthmatic patients selected for omalizumab treatment, highlighted a poor association between the level of peripheral blood eosinophil count and clinical

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parameters such as lung function, FeNO values and patient reported outcomes. Also, blood eosinophils seemed not relevant in detecting a specific clinical-antropometric patient profile in terms of demographic data, total IgE and nasal comorbidities. FeNO, when higher than 30 ppb, was associated with poorer asthma control defined by ACT, AQLQ score, hospital admissions and number of lost working days in the last year. On the opposite, omalizumab treatment seemed more likely to be effective in patients with higher eosinophilic inflammation (>=300 eosinophils/mm3). FeNO and blood eosinophils cut-offs have been identified according to what has been suggested by the analysis of NHLBI Severe Asthma Research Program big database [16]. However different cut-offs have been investigated (blood eosinophils: 150, 400 eosinophils/mm3; FeNO: 25, 50 ppb – data not shown) without any significant difference in comparison with the described thresholds.

Although up to now eosinophils are recognized as the main clinical marker of Th2 inflammation in respiratory diseases, their relevance in identifying asthma severity is still matter of debate [16,19-21]. When looking at the literature, a substantial lack of correlation between blood eosinophil count and asthma severity has been reported by some authors [16,22,23]. Similarly to our results, this finding seems to be independent of the asthma assessment criterion, including Global Initiative for Asthma parameters, lung function assessment or PROs.

Blood eosinophilia has been identified in the literature as a risk factor for asthma exacerbations, independently of symptoms control [20,21]. In our study we did not observe the same correlation, in fact looking at the variables related to asthma exacerbations, including Emergency Room admissions, Hospital admissions, unscheduled visits and lost working days, no statistically significant differences could be identified between the high and low eosinophils subgroups (Table 2). When considering hospital admission rate for asthma exacerbations, although it can be

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considered a hallmark of asthma control more than of asthma severity, its relationship with blood eosinophil count is controversial as well [16,20,21,24,25]. A number of reasons may account for those divergent findings. Physiologically, eosinophils are interested by a high intra and inter individual variability [26]. Furthermore, especially in the frame of real-life studies, the impact of oral steroids or other drugs influencing blood eosinophils levels cannot be completely ruled out. Our analysis excluded the patients undertaking OCS at the time of basal blood eosinophilia and clinical assessment, in order to increase the population sample homogeneity. For the same purpose we verified the previous use of OCS, which we found homogenously distributed among the high and low eosinophils groups.

Some evidence supports a greater accuracy of sputum eosinophils count in comparison with blood eosinophils as a hallmark of asthma severity [16,23,27,28], and the correlation between the two biomarkers seems to be weak [16,26] though reports are controversial [29,30]. In our work the sputum eosinophils assessment is missing and it could represent a limitation of the study. However, the correlation between blood and sputum eosinophilia is claimed by some authors [29,30], and blood eosinophilia has been recently described as a better predictor of response to eosinophil-targeted biological treatments in comparison with sputum eosinophils [19,31]. According to a recent Cochrane review, tailoring asthma management on sputum eosinophilia needs for more evidence and it cannot be currently recommended, unless included in a multiple – approach evaluation [32]. Furthermore investigation of sputum eosinophilia is time consuming and not widely available, so that it cannot be considered a simple tool for the evaluation of severe asthmatic patients “at a glance”.

FeNo measurement allows a more immediate evaluation, although a number of unrelated factors, including diet and upper airway inflammation, may account for its variability [12]. According to our

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findings, differently from blood eosinophils FeNO at 30 ppb cut-off was able to highlight key differences in the study population, particularly in terms of hospital admissions rate in the last year, ACT, AQLQ and lost working days in the last year. Thus FeNO seemed to be more reliable as a marker of asthma severity and control in comparison with eosinophil count, at least in severe allergic asthma patients. However, the correlation between sputum eosinophilia, blood eosinophilia and FeNO is not supported by univocal evidence [16,33,34]. Also the level of agreement between FeNO levels and clinical parameters, including PROs and lung function, is conflicting in different studies [34-37]. Following the better accuracy of FeNO in defining asthma severity and control in our population, the performance of a composite index including both eosinophils (cut-off: 300 eosinophils/mm3) and FeNO (cut-off 30 ppb) has been investigated in our study. Actually the combined score did not improve the accuracy of the single parameters in discriminating the clinical severity of the disease. Similarly, although aiming at investigating potential predictors of sputum eosinophils, Hastie et al [16] demonstrated that blood eosinophils, FeNO, FEV1%predicted or IgE, alone or included in multiple indexes, did not show a enough accurate predictive value for exacerbations, or healthcare utilization in severe asthmatic patients.

As a secondary outcome of our study, the association between basal blood eosinophils and responsiveness to omalizumab treatment has been investigated. In comparison to patients with blood eosinophils lower than 300 eosinophils/mm3, the high eosinophils group included a greater number of responders since the first follow-up, 6 months later the treatment start and at each time-point. Although a statistical significance could not be observed concerning that trend, the p-value related to the difference between the high and low eosinophils subgroups gradually decreased, suggesting the possibility that a longer follow-up time frame would have shown a statistically significant difference between the proportion of responders in the two subgroups.

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Furthermore the observed trend is clinically relevant, besides the statistical significance. Also, the proportion of responders in both high and low eosinophil groups increased at each time-point, but in the first one the increase was greater. These findings suggest that within the low eosinophils group, treatment responsiveness may be not time-dependent, and continuing the treatment after six month seems not likely to increase the number of responders. The relevance of blood eosinophils in predicting the treatment responsiveness to biologicals, particularly Th2-targeted drugs, has been highlighted by different reports [5-11,38]. As far as Omalizumab is concerned, although blood eosinophils are traditionally considered a marker of positive treatment outcome [5,38], a recent large real-life investigation demonstrated that the anti-IgE drug is effective irrespectively of the baseline eosinophil level [39]. As commented by the authors, the retrospective, real-life design of the study may account for the different results in comparison with randomized, controlled clinical trials, together with the more severe asthma phenotype of patients included in the first. The aforementioned physiological intra and inter individual variability in eosinophil levels may also explain different findings, as well as the potential effect of oral steroids or other drugs influencing blood eosinophils, particularly in the real-life life setting where strict inclusion criteria are not applied. Katz et al [31] demonstrated that in patients with severe asthma undergoing mepolizumab treatment, the exacerbation rate reduction was significantly greater in subjects with blood eosinophils of 150/µl or higher compared with subjects with blood eosinophils under 150/µl. A recent review including the data coming from the mepolizumab clinical development program, confirmed the role of blood eosinophil count as a pharmaco-dynamic and predictive biomarker of treatment response in patients with severe eosinophilic asthma [19]. Similarly, the two randomized clinical trials evaluating reslizumab for poorly controlled asthma demonstrated how crucial is the baseline blood eosinophil level in patient selection [8,9]. Elevated

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eosinophils represent an essential condition for the efficacy of Benralizumab too in severe uncontrolled asthmatic patients [10,11]. Thus, blood eosinophil count seems to act as a predictive marker of response to eosinophil-targeted biological treatments more than a hallmark of asthma severity.

Some limitations could weaken our findings, including the retrospective design and the lack of investigation about other potential determinants of treatment response or non-response besides the blood eosinophils level. However according to the above mentioned published studies, the role of eosinophils as predictors of treatment response seems not to be significantly affected by other clinical variables.

In conclusion, according to our findings blood eosinophil level is not associated with a specific clinical profile, in terms of demographic data, total IgE, nasal comorbidities lung function and PROs, whilst FeNO, when higher than 30 ppb, associated with poorer asthma control. On the other hand in our study population the proportion of responder to omalizumab was greater among the patients with higher baseline blood eosinophils level since the first six-months follow-up. Although not supported by statistical significance, the described trend may be relevant from a clinical point of view. Furthermore, continuing the treatment after six month did not significantly increase the number of responders, particularly in the low eosinophils group. Although to be confirmed by wider studies, these findings should be taken into consideration in severe asthmatic patients assessment and selection for biological treatments.

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Declaration of interests

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Conflicts of Interest
The authors declare that they have no conflicts of interest

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Table and Figures

Table 1. Demographic data of study population

Age - mean(sd)	46.9(13.3)
Males (%)	44
Current smokers (%)	5.7
Pack years - mean(sd)	9.4(12.2)
Years of smoking - mean(sd)	11.3(9.1)
BMI - mean(sd)	25.5(5.1)
Total IgE - mean(sd)	395.9(403.9)
Sensitization to aeroallergens (%)	100%

BMI: Body Mass Index; sd: standard deviation

Table 2. Comparison of patients' demographic and clinical variables in high and low basal eosinophils subgroups

	Eosinophil count (mm3)		
	<300	**>=300**	**p-value**
DEMOGRAPHIC DATA			
Age - m(sd)	43.7(12.8)	47.1(14.8)	0.16
Gender (%M)	47.8	49.1	0.92
Smoke (%sì)	13.0	1.9	0.10
BMI - m(sd)	25.8(6.4)	25.4(4.5)	0.40
Total IgE - m(sd)	477.0(588.0)	360.6(291.2)	0.19
Perennial sensitizations (%)	91.3	98.1	0.17
History of oral steroids use (%)	52.2	56.6	0.72
LUNG FUNCTION AND PROs			
FEV1% - m(sd)	69.7(18.8)	69.9(17.4)	0.48
FVC% - m(sd)	83.9(13.4)	84.4(15.5)	0.44
Tiffenau - m(sd)	0.7(0.1)	0.7(0.1)	0.25
ACT - m(sd)	14.2(4.3)	14.2(5.6)	0.47
AQLQ - m(sd)	3.7(1.1)	3.7(1.4)	0.47
FeNO - m(sd)	36.3(35.8)	47.8(51.2)	0.16
DIRECT AND INDIRECT COSTS			
Emergency Room admission in the last year - m(sd)	1.1(2.3)	0.9(1.9)	0.35
Hospital Admissions in the last year - m(ds)	0.3(0.8)	0.4(0.7)	0.31
Unscheduled visits - m(ds)	3.2(3.2)	3.5(3.2)	0.35
Lost working days in the last year - m(sd)	13.4(16.8)	24.7(43.0)	0.07
NASAL COMORBIDITIES			
Poliposis (%)	26.1	37.7	0.32
Rhinitis (%)	69.6	86.8	0.07

m: mean; sd: standard deviation; BMI: body mass index; ACT: asthma control test; AQLQ: asthma quality of life questionnaire

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Table 3. Comparison of patients' demographic and clinical variables in high and low basal FeNO subgroups

	FeNO (ppb)		
	<30	**>=30**	**p-value**
DEMOGRAPHIC DATA			
Age - m(sd)	45.1 (15.4)	46.1 (11.5)	0,37
Gender (%M)	45,8	43,8	0,84
Smoke (%sì)	31,3	14,6	0,05
BMI - m(sd)	26.3 (5.9)	24.1 (4.3)	**0,02**
Total IgE - m(sd)	383.3(339.1)	359.0(314.6)	0,36
Perennial sensitizations (%)	95,7	93,8	0,52
History of oral steroids use (%)	33,3	56,3	**0,02**
LUNG FUNCTION, PROs and EOSINOPHILS			
FEV1% - m(sd)	68.0 (15.5)	71.0 (20.5)	0,20
FVC% - m(sd)	83.8 (16.8)	88.9 (17.2)	0,07
Tiffenau - m(sd)	0.7 (0.1)	0.7 (0.1)	0,16
ACT - m(sd)	15.8 (5.9)	13.7 (5.4)	**0,04**
AQLQ - m(sd)	4.1 (1.4)	3.5 (1.2)	**0,04**
Eosinophils - m(sd)	0.87 (0.27)	0.98 (0.22)	0,3803
DIRECT AND INDIRECT COSTS			
Emergency Room admission in the last year - m(sd)	1.2 (2.6)	1.3 (2.2)	0,41
Hospital Admissions in the last year - m(ds)	0.3 (0.9)	0.7 (1.0)	**0,04**
Unscheduled visits - m(ds)	3.4 (3.0)	3.6 (3.2)	0,39
Lost working days in the last year - m(sd)	11.8 (15.8)	26.9 (39.9)	**0,03**
NASAL COMORBIDITIES			
Poliposis (%)	27,1	41,7	0,13
Rhinitis (%)	68,8	75	0,49

m: mean; sd: standard deviation; BMI: body mass index; ACT: asthma control test; AQLQ: asthma quality of life questionnaire

Table 4. Comparison of patients' demographic and clinical variables in different subgroups according to a combined eosinophils/FeNO index

VARIABLE	Eosinophil count (mm3); FeNO (ppb)					
	<300;<30	<300;>=30	>=300;<30	>=300;>=30	p-value	bartlett's
DEMOGRAPHIC DATA						
Age - m(sd)	42.9(13.7)	42.0(15.1)	46.1(16.9)	46.3(10.3)	0,8385	0,213
Gender (%M)	61,5	50	47,8	50	0,88	--
Smoke (%sì)	38,7	33,3	17,4	15	0,35	--
BMI - m(sd)	27.5/7.4)	21.8(4.2)	25.6(4.2)	24.9(5.3)	0,1836	0,12
Total IgE - m(sd)	347.2(415.2)	613.7(475.2)	441.6(338.7)	311.6(244.7)	0,2447	0,132
Perennial sensitizations (%)	92,3	83,3	95,7	100	0,379	--
History of oral steroids use (%)	46,2	50	60,9	55	0,843	--
LUNG FUNCTION and PROs						
FEV1% - m(sd)	63.7(16.7)	74.7(15.9)	73.7(9.4)	66.0(18.8)	0,1563	0,026
FVC% - m(sd)	78.0(12.9)	91.7(8.7)	87.3(11.1)	84.8(15.9)	0,121	0,271
Tiffenau - m(sd)	0.7(0.1)	0.7(0.1)	0.7(0.1)	0.7(0.1)	0,2269	0,349
ACT - m(sd)	14.9(4.4)	13.2(5.3)	14.9(6.0)	14.7(5.9)	0,9256	0,72
AQLQ - m(sd)	3.7(1.2)	3.3(0.6)	4.0(1.4)	3.4(1.4)	0,5051	0,509
DIRECT AND INDIRECT COSTS						
Emergency Room admission in the last year - m(sd)	1.1(2.9)	1.8(1.6)	1.0(2.5)	1.1(1.6)	0,8671	0,088
Hospital Admissions in the last year - m(ds)	0.25(0.62)	0.67(1.2)	0.35(0.71)	0.55(0.89)	0,6257	0,235
Unscheduled visits - m(ds)	3.5(4.0)	3.8(1.5)	3.0(2.1)	4.1(4.4)	0,7562	0,001
Lost working days in the last year - m(sd)	16.8(19.3)	10.0(7.1)	11.7(16.2)	36.6(50.9)	0,0975	<0.001
COMORBIDITIES'						
Poliposis (%yes)	15,4	50	47,8	35	0,238	--
Rhinitis (%yes)	69,2	100	95,7	90	0,078	--

m: mean; sd: standard deviation; BMI: body mass index; ACT: asthma control test; AQLQ: asthma quality of life questionnaire

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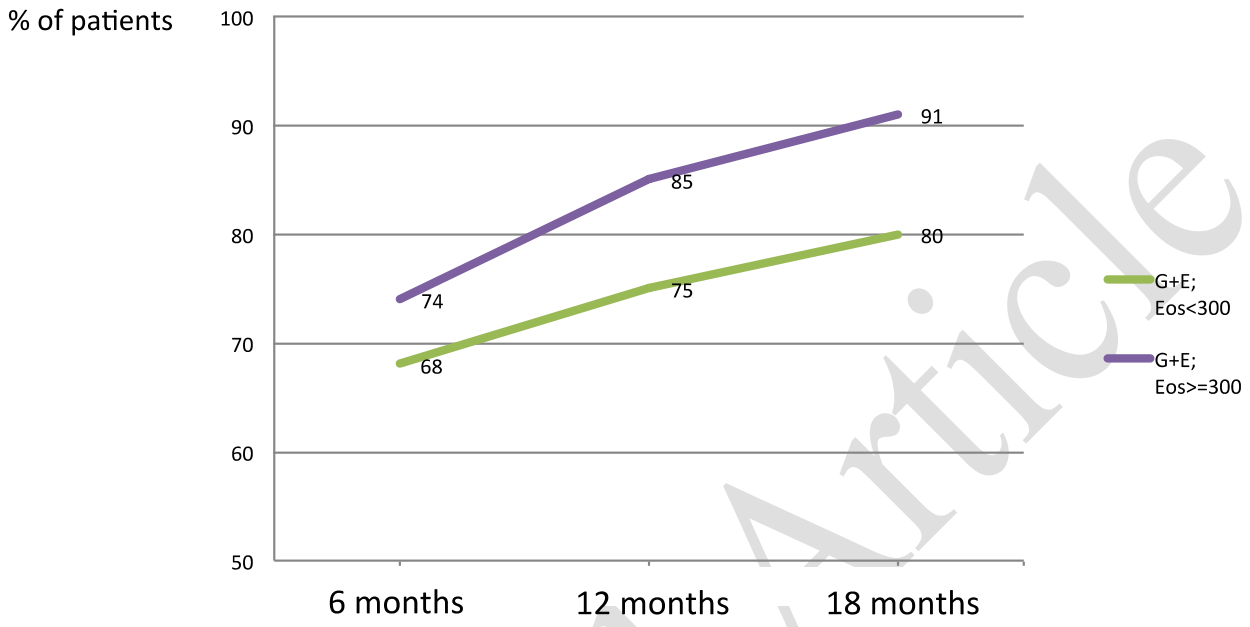
Figure 1. Trend of responders (defined by GETE questionnaire) in high and low basal eosinophils subgroups



	>=300 (n=23)	<300 (n=53)	Difference	CI95% (referred to the difference)	p-value (referred to the difference)
6 months	74%	68%	6%	-16%;28%	0.3003
12 months	85%	75%	10%	-9%;29%	0.1667
18 months	91%	80%	11%	-5%;27%	0.1186

Eos: blood eosinophils basal level; G: good responder; E: excellent responder