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Eosinophilic COPD — a distinct phenotype of the disease

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Abstract

Chronic obstructive pulmonary disease (COPD) has been traditionally associated with neutrophilic inflammation of the bronchi. Studies from the early 1990s demonstrated that eosinophils may also migrate into the lower airways of patients with COPD and their increased numbers can be noticed during exacerbations as well as stable disease. Eosinophilic phenotype of COPD is characterized by several unique features, *i.e.* a specific pattern of airway inflammation and distinct clinical course or susceptibility to corticosteroid treatment. In this paper, we present an up-to-date review of the literature on clinical characteristics of eosinophilic COPD, as well as the role of eosinophils as a biomarker-guided therapy in COPD.

Key words: COPD, eosinophils, biomarkers

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Introduction

Chronic Obstructive Pulmonary Disease (COPD) is one of the most frequent chronic diseases in adults and one of the main causes of morbidity and mortality. Data on the worldwide prevalence of COPD has been provided by the Burden of Obstructive Lung Disease (BOLD) study. Based on these results, the prevalence of COPD at Global Initiative for Chronic Obstructive Lung Disease (GOLD) stage 2 or higher was estimated at 10.1% overall, with a number of regional differences [1]. In Poland, the Małopolska Region, the estimated population prevalence of COPD was 22.1%, whereas 10.9% of the subjects had COPD GOLD stage ≥ 2 [2].

Cigarette smoke exposure is a well-recognized risk factor in the development of COPD. Cigarette smoke leads to activation of pro-inflammatory cascades resulting in lung injury. It is generally accepted that the neutrophilic inflammation observed in the lungs of COPD patients is intrinsically linked to the tissue destruction and alveolar airspace enlargement, leading to disease

progression [3]. However, a subset of patients with COPD develops eosinophilic inflammation in the airways that is reflected also in increased blood eosinophil number. A growing body of evidence suggests that eosinophilic COPD is a distinct phenotype of the disease. Eosinophilic COPD could be labelled as a part of asthma-COPD overlap (ACO), a condition sharing pathophysiological and clinical features of both asthma and COPD. However, a recent study demonstrated that eosinophilic COPD patients have distinct characteristics compared to COPD patients with a history of asthma, *i.e.* they are characterized by little evidence of allergies and less exacerbations, but more pronounced eosinophilic inflammation [4]. Here, we present a review of the literature on clinical characteristics of eosinophilic COPD, as well as the role of eosinophils in a biomarker-guided therapy in COPD.

Eosinophilic inflammation in COPD

Although COPD has been considered a condition characterized by neutrophilic airway in-

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flammation, as early as in the 1990s, the concept of eosinophilic inflammation arose. In 1994, Saetta *et al.* [5] showed that patients with chronic bronchitis at exacerbation had significantly more eosinophils in sputum and bronchial biopsies than subjects examined at stable disease. Further studies revealed that the influx of eosinophils into the airways is rather associated with viral exacerbations, whereas during bacterial infections a decrease in blood and sputum eosinophils occurs [6, 7].

Evidence implicating eosinophils in airway inflammation in stable COPD was presented by Balzano *et al.* [8] who compared sputum cell composition and eosinophil cationic protein (ECP) levels between clinically stable COPD subjects, healthy smokers, mild asthmatics and healthy subjects. The authors showed that sputum eosinophil percentage was increased in patients with COPD, as compared with healthy controls. Sputum ECP levels were significantly higher in both COPD and asthma than in the other two groups. In patients with COPD and asymptomatic smokers, considered as a whole, eosinophil percentage and ECP concentrations correlated negatively with FEV₁ % predicted and FEV₁/FVC ratio.

A complex clinical characteristics of COPD subjects with evidence of eosinophilic inflammation was provided by the ECLIPSE study (Evaluation of COPD Longitudinally to Identify Predictive Surrogate End-points) [9]. The data is valuable not only due to the size of the study group, but also the 3-year follow-up period which provided an opportunity to assess stability of eosinophilic COPD phenotype. Blood samples for eosinophil counts were obtained from 1483 subjects at a baseline and then yearly over the observation period. 37.4% of the subjects had eosinophil counts persistently $\geq 2\%$ at all visits, 13.6% had eosinophil counts persistently $\leq 2\%$ at all visits, and 49% subjects had variable eosinophil counts. COPD subjects with eosinophil counts persistently $\geq 2\%$ were slightly older, had a greater proportion of males and fewer current smokers than other COPD groups. They were also characterized by a higher FEV₁ % predicted and fat-free mass index, lower St. George's Respiratory Questionnaire (SGRQ), modified Medical Research Council (mMRC) scores, and BODE (body mass index, airflow obstruction, dyspnea, exercise capacity) index.

There is little knowledge about the mechanism leading to the development of eosinophil infiltration in the airways of some COPD subjects, as well as differences in systemic and airway

inflammation pattern between eosinophilic and non-eosinophilic COPD. In a recent study, Kolsum *et al.* [10] assessed the nature of airway inflammation in patients with COPD based on blood eosinophil numbers. The study subjects were classified according to blood eosinophil counts as "eosinophil low" or "eosinophil high" (< 150 cells/ μL or > 250 cells/ μL respectively). "Eosinophil high" patients had also higher eosinophil numbers in sputum, bronchoalveolar lavage (BAL) and bronchial submucosa. This was accompanied by higher sputum interleukin(IL)-5 and haptoglobin levels, as well as CCL20 and CCL24 concentrations in BAL. "Eosinophil high" subjects were also characterized by more pronounced airway remodelling as indicated by increased reticular basement thickness and tenascin thickness, as well as increased BAL metalloproteinase-7 and -9 concentrations. In addition, our group reported that IL-33, one of so-called "epithelium derived cytokines" extensively examined in allergic asthma in the past few years, might be involved in the pathogenesis of eosinophilic COPD. IL-33 is an alarmingly critical for eosinophil differentiation, maturation, activation and survival, produced by epithelial cells in response to danger signals [11]. Our preliminary data demonstrated that serum and sputum IL-33 concentrations are significantly elevated in COPD patients with sputum eosinophil counts $> 3\%$ versus patients without airway eosinophilia [12]. Moreover, IL-33 serum and sputum levels correlated positively with sputum eosinophil percentage. Hence, IL-33 may be implicated in the development of eosinophilic phenotype of COPD. It is plausible that other cells and cytokines released by epithelial cells and/or airway smooth muscles may play a role in the development of airway eosinophilia in a similar mechanism to that observed in eosinophilic asthma (Fig. 1).

Several studies assessed fractional exhaled nitric oxide (FeNO) as a surrogate marker of eosinophilic airway inflammation in COPD. According to the available data, FeNO correlated positively with sputum but not blood eosinophils at exacerbation and in lesser extent at recovery [13, 14]. In addition, FeNO levels determined at hospital admission may predict the overall response to corticosteroid treatment in COPD patients with acute exacerbations [10]. However, no difference in FeNO levels between high and low eosinophil counts patients with stable COPD was detected [10]. Thus, FeNO may reflect eosinophil accumulation in the airways during exacerbation and therefore may help guide therapeutic decisions.

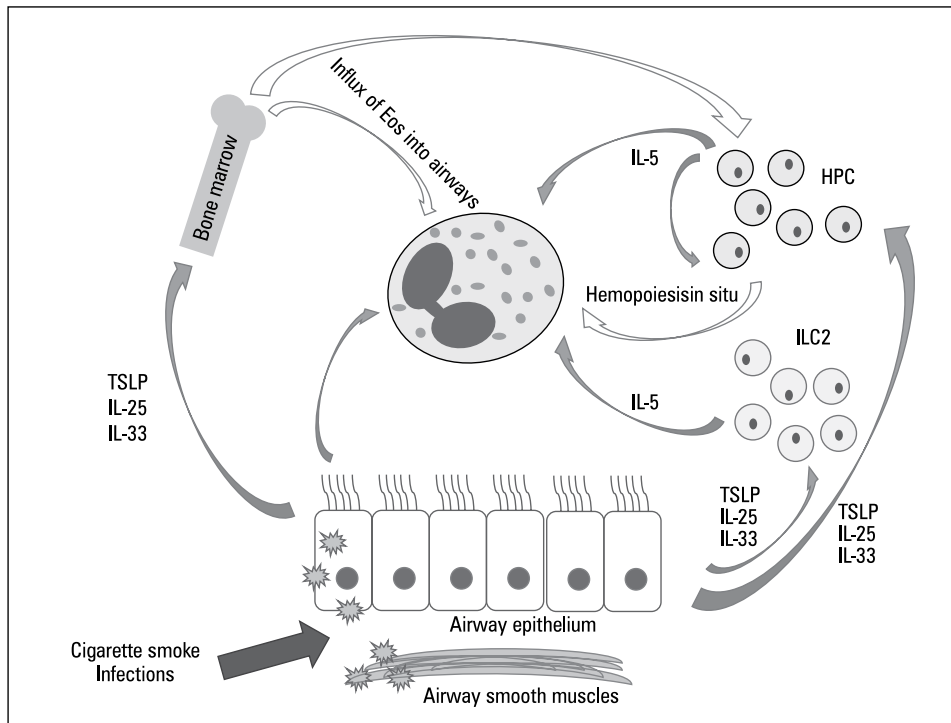


Figure 1. Some of possible mechanisms leading to the development of eosinophilic inflammation in COPD. Airway epithelium produces thymic stromal lymphopietin (TSLP), interleukin (IL)-25 and IL-33 in response to danger signals, e.g. cigarette smoke and infections. Airway smooth muscles also produce IL-33 and TSLP when exposed to cigarette smoke. The three cytokines are directly involved in eosinophil (Eos) activation and migration into the airways. They also activate innate lymphoid cells type 2 (ILC2), an abundant source of IL-5. In addition, they increase lung homing of hemopoietic progenitor cells (HPC), stimulate HPC to produce IL-5 and orchestrate so called “hemopoiesis *in situ*”, i.e. transformation of HPC into mature eosinophils in the airways

Eosinophilia and risk and clinical course of COPD exacerbations

The role of eosinophils as a biomarker predicting risk of COPD exacerbations and response to treatment has been of considerable interest. In a large study including 7225 patients with COPD defined from Copenhagen General Population Study, Vedel-Krogh *et al.* [15] examined an association between baseline blood eosinophil counts and risk of exacerbations, defined as moderate (short course treatment with systemic corticosteroids) or severe (hospitalization). COPD was defined basing on spirometry as FEV_1/FVC ratio < 70%. The authors also assessed exacerbation risk in a subgroup of 203 individuals with “clinical COPD”, described as participants with a smoking history of at least 10 pack-years, FEV_1 less than 70% of predicted value, and at least one moderate or severe exacerbation in the year before a baseline. Among all participants with COPD, blood eosinophils above 0.34×10^9 cells/l were associated with 1.76-fold increase in incidence rate for severe exacerbations and 1.15-fold for moderate exacerbations. Corresponding values in

those with “clinical COPD” were 3.21 and 1.69. In contrast, using a cut point of 2% for blood eosinophils, the risk of exacerbations was increased for severe exacerbations only among individuals with “clinical COPD”. These results suggest that elevated blood eosinophil absolute number is a better predictor of exacerbations than percentage values, and the narrower COPD definition, the stronger association between exacerbation rate and eosinophils can be noticed.

In a metaanalysis of three clinical trials, Bafadhel *et al.* [16] evaluated effectiveness of treatment COPD exacerbations with oral corticosteroids in respect of blood eosinophilia. The primary outcome was the rate of treatment failures following treatment of an exacerbation, defined as retreatment, hospitalization or death within 90 days of randomization. The subjects were grouped according to treatment allocation (prednisolone or non-prednisolone) and blood eosinophil count (< 2% or \geq 2%) at the time of exacerbation. The treatment failure rate was 66% in patients with a blood eosinophil count \geq 2% who did not receive prednisolone and 11% in those who did. In patients with a blood eosinophil

count < 2%, there was no difference in treatment failure rates with and without prednisolone (26% vs 20%). The findings were not modified by severity of the exacerbation, baseline exacerbation lung function, age or smoking history. The authors concluded that a eosinophil-directed corticosteroid treatment strategy using the peripheral blood eosinophil count measured at the onset of exacerbation is a promising approach to maximize benefit and minimize harm in COPD.

A recent study on a group of patients hospitalized due to COPD exacerbation demonstrated that blood eosinophil level ≥ 200 cells/ μ L and/or $\geq 2\%$ of the total white blood cell count on admission was associated with 3.59-fold increase in the risk of 12-month COPD-related readmission, 2.32-fold increase in the risk of 12-month all-cause readmission, and 2.74-higher chance of a shorter time to first COPD-related readmission but not with the length of stay [17]. The study also showed that readmission rate increased proportionally to eosinophil cell count. On the contrary, in another study using similar criteria for blood eosinophilia, the mean length of stay was significantly shorter in patients with eosinophilic than with non-eosinophilic exacerbations (5.0 days vs 6.5 days), whereas readmissions rates were similar in both groups [18].

The results of the above mentioned reports support the notion that peripheral blood eosinophil count can be a biomarker in COPD and a valuable predictor of the response to the treatment of exacerbations.

Exacerbation prevention in eosinophilic COPD

As mentioned above, COPD patients with high blood eosinophil counts are at increased risk of exacerbations, especially severe ones [15]. One must remember that exacerbation in a COPD patient is a major event leading to accelerated lung function decline. Moreover, frequent exacerbations increase mortality risk and 50% of COPD patients will die within 5 years since severe aggravation [19]. Therefore, exacerbation prevention is essential in COPD management.

Studies proved that ICS treatment reduces risk of COPD exacerbations. Early trials using sputum eosinophil count-guided COPD therapy demonstrated that a management strategy aiming to minimize eosinophilic airway inflammation by doubling the dose of ICS in patients with sputum eosinophils > 3%, is associated with a significant reduction, by 62%, in the frequency of severe, but not mild or moderate, COPD exacerbations

compared with patients treated according to traditional guidelines [20]. Subsequent studies reported that sputum eosinophils highly correlated with blood eosinophil count and therefore rendered sputum-guided therapy pointless [21]. Indeed, large clinical trials proved that blood eosinophilia is a useful biomarker of the effect of ICS treatment on COPD exacerbations. A post-hoc analysis of data from two replicate, randomized, double-blind trials of 12-month duration, in which vilanterol once daily was compared with vilanterol plus 50 μ g, 100 μ g, or 200 μ g fluticasone furoate in patients with moderate-to-severe COPD and a history of one or more exacerbation in the previous year [22]. Across all doses of inhaled corticosteroids, fluticasone furoate and vilanterol significantly reduced exacerbations by 29% compared with vilanterol alone (mean 0.91 vs 1.28 exacerbations per patient per year) in patients with eosinophil counts $\geq 2\%$, and non-significantly by 10% (0.79 vs 0.89) in patients with eosinophil count $\leq 2\%$. Reduction in exacerbations increased along with the percentage of blood eosinophils reaching 42% in patients with eosinophil count $\geq 6\%$. Recently, a post-hoc analysis of the FORWARD study, a randomized, double-blind, parallel group trial that compared 48 weeks of treatment with extrafine beclomethasone dipropionate plus formoterol fumarate (BDP/FF), 100/6 mg, two inhalations twice a day, versus FF 12 mg, one inhalation twice a day, in patients with severe COPD and a history of exacerbations, demonstrated that the higher blood eosinophil count, the greater reduction in exacerbations, reaching 46% in patients with blood eosinophil number more than 279.8 cells/ μ l [23]. Noteworthy, withdrawal of ICS in patients with severe to very severe COPD and blood eosinophil counts $\geq 4\%$ or ≥ 300 cells/ μ L before the ICS treatment was introduced, have led to a higher risk of exacerbations compared to a group of patients continuing ICS treatment [24].

Unfortunately, although inhaled steroid treatment can reduce exacerbation rate in COPD patients with high blood eosinophils count, it is associated with increased risk of pneumonia. This phenomenon was first reported in the TORCH (TOWards a Revolution in COPD Health) study, among COPD patients receiving fluticasone propionate alone or in combination with salmeterol, and later confirmed by other studies [25]. Further evidence pointed out that the greatest risk of pneumonia can be seen in patients treated with a daily fluticasone-equivalent dose > 1000 μ g [26]. A subsequent Cochrane

Database systematic review demonstrated that not only fluticasone but also budesonide, delivered alone or in combination with a long-acting β_2 -agonist (LABA), is associated with increased risk of serious adverse pneumonia events [27]. A 2-fold increase in pneumonia incidence was also observed during treatment with beclomethasone/formoterol [28]. In addition, a post-hoc analysis of ten clinical trials demonstrated that COPD subjects with blood eosinophil counts $\geq 2\%$ are slightly less likely to have pneumonia when not treated with inhaled corticosteroids (3.8% of patients with $< 2\%$ of blood eosinophil counts versus 2.4% with $\geq 2\%$ of blood eosinophils) [29]. In patients treated with inhaled corticosteroids, pneumonia events occurred in 4.5% versus 3.9% in those not treated with corticosteroids.

Some data suggest that long-acting anti-muscarinic agent (LAMA)/LABA combination treatment might be a new option to decrease exacerbation risk without increasing risk of pneumonia associated with ICS therapy, even in patients with eosinophilic COPD. Indeed, results of the FLAME study, comparing indacaterol/glycopyrronium 110/50 μg with salmeterol/fluticasone combination 50/500 μg in patients with ≥ 1 exacerbation in the preceding year demonstrated that once-daily long-acting β_2 -agonist/long-acting muscarinic antagonist provides superior or similar benefits over twice-daily long-acting β_2 -agonist/inhaled corticosteroid regardless of blood eosinophil levels in patients with COPD [30]. However, this finding needs to be further confirmed.

Biologics in the treatment of eosinophilic COPD

Many monoclonal antibodies have been developed for treatment of eosinophilic asthma and some of them are assumed to have therapeutic potential in the treatment of eosinophilic COPD as well.

A phase 2 trial tested the safety and efficacy of benralizumab, an anti-IL-5 receptor antibody, in eosinophilic COPD defined as sputum eosinophil counts $> 3\%$. In this study, benralizumab did not reduce the exacerbation rate nor modified lung function. However, a subgroup analysis revealed a trend toward an improvement in FEV₁ and exacerbations in patients with a baseline blood eosinophil number greater than 200 cells/ μl and treated with benralizumab. Recently, results of a pilot study on the treatment of eosinophilic COPD (sputum eosinophil counts $\geq 3\%$) with mepolizumab have been published [31]. Six-month treatment with mepolizumab significantly reduced

sputum and blood eosinophil counts, an effect not seen in placebo treated subjects. However, anti-IL-5 treatment did not affect lung function, exacerbation rates, and quality of life scores, suggesting that eosinophils may not play a relevant pathobiological role in COPD. The main limitation of the study was a small number of participants, *i.e.* 8 patients receiving active treatment and 9 subjects in placebo group. Therefore, drawing a final conclusion based on such limited data would be premature. Results of a just completed large clinical trial evaluating efficacy and safety of mepolizumab in severe COPD patients with frequent exacerbations will provide substantially more information on the subject [32].

Thus, additional studies are warranted to evaluate the efficacy of anti-eosinophil therapy and its effects on clinical parameters such as exacerbation rate in COPD. Apart from anti-IL-5 therapies, biologics neutralizing other cytokines critical for eosinophil differentiation, maturation, activation and survival may be an interesting target. Potential molecules of interest include “epithelium derived cytokines”, *i.e.* IL-25, IL-33 and TSLP, traditionally associated with Th2-like conditions. Results of our above mentioned study demonstrating overexpression of IL-33 in COPD suggest that there are still some potential targets for biological therapies that may improve treatment outcomes of COPD, in particular eosinophilic phenotype of the disease.

Summary

A growing body of evidence suggests that eosinophilic COPD is a separate phenotype of the disease with distinct clinical features. Simple laboratory test such as blood differential count may identify patients with eosinophilic inflammation. It is important to do so as subjects with blood and/or airway eosinophilia are more likely to suffer from exacerbations, yet better respond to treatment with corticosteroids. However, COPD subjects treated with inhaled corticosteroids are at increased risk of pneumonia. New treatment approach with biologic agents is expected to be unencumbered by such a side effect. Nonetheless, therapies targeting IL-5 pathway have been investigated showing no clear benefit so far in regard of exacerbation risk reduction and improvement in other clinical parameters. More studies are warranted to confirm if neutralizing other cytokines crucial for eosinophil differentiation and activation may be beneficial in treatment of patients with eosinophilic COPD.

Conflict of interest

The authors declare no conflict of interest.

References:

- Buist AS, McBurnie MA, Vollmer WM, et al. BOLD Collaborative Research Group. International variation in the prevalence of COPD (the BOLD Study): a population-based prevalence study. *Lancet*. 2007; 370(9589): 741–750, doi: [10.1016/S0140-6736\(07\)61377-4](https://doi.org/10.1016/S0140-6736(07)61377-4), indexed in Pubmed: [17765523](https://pubmed.ncbi.nlm.nih.gov/17765523/).
- Nizankowska-Mogilnicka E, Mejza F, Buist AS, et al. Prevalence of COPD and tobacco smoking in Malopolska region—results from the BOLD study in Poland. *Pol Arch Med Wewn*. 2007; 117(9): 402–410, indexed in Pubmed: [18062562](https://pubmed.ncbi.nlm.nih.gov/18062562/).
- Huang G, Xu XC, Zhou JS, et al. Neutrophilic Inflammation in the Immune Responses of Chronic Obstructive Pulmonary Disease: Lessons from Animal Models. *J Immunol Res*. 2017; 2017: 7915975, doi: [10.1155/2017/7915975](https://doi.org/10.1155/2017/7915975), indexed in Pubmed: [28536707](https://pubmed.ncbi.nlm.nih.gov/28536707/).
- Kolsum U, Ravi A, Hitchen P, et al. Clinical characteristics of eosinophilic COPD versus COPD patients with a history of asthma. *Respir Res*. 2017; 18(1): 73, doi: [10.1186/s12931-017-0559-0](https://doi.org/10.1186/s12931-017-0559-0), indexed in Pubmed: [28446172](https://pubmed.ncbi.nlm.nih.gov/28446172/).
- Saetta M, Di Stefano A, Maestrelli P, et al. Airway eosinophilia in chronic bronchitis during exacerbations. *Am J Respir Crit Care Med*. 1994; 150(6 Pt 1): 1646–1652, doi: [10.1164/ajrccm.150.6.7952628](https://doi.org/10.1164/ajrccm.150.6.7952628), indexed in Pubmed: [7952628](https://pubmed.ncbi.nlm.nih.gov/7952628/).
- Rohde G, Gevaert P, Holtappels G, et al. Soluble interleukin-5 receptor alpha is increased in acute exacerbation of chronic obstructive pulmonary disease. *Int Arch Allergy Immunol*. 2004; 135(1): 54–61, doi: [10.1159/000080043](https://doi.org/10.1159/000080043), indexed in Pubmed: [15286446](https://pubmed.ncbi.nlm.nih.gov/15286446/).
- Kolsum U, Donaldson GC, Singh R, et al. Blood and sputum eosinophils in COPD; relationship with bacterial load. *Respir Res*. 2017; 18(1): 88, doi: [10.1186/s12931-017-0570-5](https://doi.org/10.1186/s12931-017-0570-5), indexed in Pubmed: [28482840](https://pubmed.ncbi.nlm.nih.gov/28482840/).
- Balzano G, Stefanelli F, Iorio C, et al. Eosinophilic inflammation in stable chronic obstructive pulmonary disease. Relationship with neutrophils and airway function. *Am J Respir Crit Care Med*. 1999; 160(5 Pt 1): 1486–1492, doi: [10.1164/ajrccm.160.5.9810105](https://doi.org/10.1164/ajrccm.160.5.9810105), indexed in Pubmed: [10556110](https://pubmed.ncbi.nlm.nih.gov/10556110/).
- Kolsum U, Ravi A, Hitchen P, et al. ECLIPSE investigators. Eosinophilic inflammation in COPD: prevalence and clinical characteristics. *Eur Respir J*. 2014; 44(6): 1697–1700, doi: [10.1183/09031936.00162414](https://doi.org/10.1183/09031936.00162414), indexed in Pubmed: [25323230](https://pubmed.ncbi.nlm.nih.gov/25323230/).
- Kolsum U, Damera G, Pham TH, et al. Pulmonary inflammation in patients with chronic obstructive pulmonary disease with higher blood eosinophil counts. *J Allergy Clin Immunol*. 2017; 140(4): 1181–1184.e7, doi: [10.1016/j.jaci.2017.04.027](https://doi.org/10.1016/j.jaci.2017.04.027), indexed in Pubmed: [28506852](https://pubmed.ncbi.nlm.nih.gov/28506852/).
- Mitchell PD, O'Byrne PM. Biologics and the lung: TSLP and other epithelial cell-derived cytokines in asthma. *Pharmacol Ther*. 2017; 169: 104–112, doi: [10.1016/j.pharmthera.2016.06.009](https://doi.org/10.1016/j.pharmthera.2016.06.009), indexed in Pubmed: [27365223](https://pubmed.ncbi.nlm.nih.gov/27365223/).
- Tworek D, Majewski S, Szewczyk K, et al. Serum IL-33 in Stable COPD with Sputum Eosinophilia. *C74 Adv Transl COPD American Thoracic Society*; 2017. p. : A6319–A6319, doi: [doi:10.1164/ajrccm-conference.2017.195.1_MeetingAbstracts.A6319](https://doi.org/10.1164/ajrccm-conference.2017.195.1_MeetingAbstracts.A6319).
- Soter S, Barta I, Antus B. Predicting sputum eosinophilia in exacerbations of COPD using exhaled nitric oxide. *Inflammation*. 2013; 36(5): 1178–1185, doi: [10.1007/s10753-013-9653-8](https://doi.org/10.1007/s10753-013-9653-8), indexed in Pubmed: [23681903](https://pubmed.ncbi.nlm.nih.gov/23681903/).
- Gao J, Zhang M, Zhou L, et al. Correlation between fractional exhaled nitric oxide and sputum eosinophilia in exacerbations of COPD. *Int J Chron Obstruct Pulmon Dis*. 2017; 12: 1287–1293, doi: [10.2147/COPD.S134998](https://doi.org/10.2147/COPD.S134998), indexed in Pubmed: [28490872](https://pubmed.ncbi.nlm.nih.gov/28490872/).
- Vedel-Krogh S, Nielsen SF, Lange P, et al. Blood Eosinophils and Exacerbations in Chronic Obstructive Pulmonary Disease. The Copenhagen General Population Study. *Am J Respir Crit Care Med*. 2016; 193(9): 965–974, doi: [10.1164/rccm.201509-1869OC](https://doi.org/10.1164/rccm.201509-1869OC), indexed in Pubmed: [26641631](https://pubmed.ncbi.nlm.nih.gov/26641631/).
- Bafadhel M, Davies L, Calverley PMA, et al. Blood eosinophil guided prednisolone therapy for exacerbations of COPD: a further analysis. *Eur Respir J*. 2014; 44(3): 789–791, doi: [10.1183/09031936.00062614](https://doi.org/10.1183/09031936.00062614), indexed in Pubmed: [24925917](https://pubmed.ncbi.nlm.nih.gov/24925917/).
- Couillard S, Larivée P, Courteau J, et al. Eosinophils in COPD Exacerbations Are Associated With Increased Readmissions. *Chest*. 2017; 151(2): 366–373, doi: [10.1016/j.chest.2016.10.003](https://doi.org/10.1016/j.chest.2016.10.003), indexed in Pubmed: [27746201](https://pubmed.ncbi.nlm.nih.gov/27746201/).
- Bafadhel M, Greening NJ, Harvey-Dunstan TC, et al. Blood Eosinophils and Outcomes in Severe Hospitalized Exacerbations of COPD. *Chest*. 2016; 150(2): 320–328, doi: [10.1016/j.chest.2016.01.026](https://doi.org/10.1016/j.chest.2016.01.026), indexed in Pubmed: [26851799](https://pubmed.ncbi.nlm.nih.gov/26851799/).
- Soler-Cataluña JJ, Martínez-García MA, Román Sánchez P, et al. Severe acute exacerbations and mortality in patients with chronic obstructive pulmonary disease. *Thorax*. 2005; 60(11): 925–931, doi: [10.1136/thx.2005.040527](https://doi.org/10.1136/thx.2005.040527), indexed in Pubmed: [16055622](https://pubmed.ncbi.nlm.nih.gov/16055622/).
- Siva R, Green RH, Brightling CE, et al. Eosinophilic airway inflammation and exacerbations of COPD: a randomised controlled trial. *Eur Respir J*. 2007; 29(5): 906–913, doi: [10.1183/09031936.00146306](https://doi.org/10.1183/09031936.00146306), indexed in Pubmed: [17301099](https://pubmed.ncbi.nlm.nih.gov/17301099/).
- Negewo NA, McDonald VM, Baines KJ, et al. Peripheral blood eosinophils: a surrogate marker for airway eosinophilia in stable COPD. *Int J Chron Obstruct Pulmon Dis*. 2016; 11: 1495–1504, doi: [10.2147/COPD.S100338](https://doi.org/10.2147/COPD.S100338), indexed in Pubmed: [27445469](https://pubmed.ncbi.nlm.nih.gov/27445469/).
- Pascoe S, Locantore N, Dransfield MT, et al. Blood eosinophil counts, exacerbations, and response to the addition of inhaled fluticasone furoate to vilanterol in patients with chronic obstructive pulmonary disease: a secondary analysis of data from two parallel randomised controlled trials. *Lancet Respir Med*. 2015; 3(6): 435–442, doi: [10.1016/S2213-2600\(15\)00106-X](https://doi.org/10.1016/S2213-2600(15)00106-X), indexed in Pubmed: [25878028](https://pubmed.ncbi.nlm.nih.gov/25878028/).
- Siddiqui SH, Guasconi A, Vestbo J, et al. Blood Eosinophils: A Biomarker of Response to Extrafine Beclomethasone/Formoterol in Chronic Obstructive Pulmonary Disease. *Am J Respir Crit Care Med*. 2015; 192(4): 523–525, doi: [10.1164/rccm.201502-0235LE](https://doi.org/10.1164/rccm.201502-0235LE), indexed in Pubmed: [26051430](https://pubmed.ncbi.nlm.nih.gov/26051430/).
- Watz H, Tetzlaff K, Wouters EFM, et al. Blood eosinophil count and exacerbations in severe chronic obstructive pulmonary disease after withdrawal of inhaled corticosteroids: a post-hoc analysis of the WISDOM trial. *Lancet Respir Med*. 2016; 4(5): 390–398, doi: [10.1016/S2213-2600\(16\)00100-4](https://doi.org/10.1016/S2213-2600(16)00100-4), indexed in Pubmed: [27066739](https://pubmed.ncbi.nlm.nih.gov/27066739/).
- Wedzicha JA, Calverley PMA, Seemungal TA, et al. INSPIRE Investigators. The prevention of chronic obstructive pulmonary disease exacerbations by salmeterol/fluticasone propionate or tiotropium bromide. *Am J Respir Crit Care Med*. 2008; 177(1): 19–26, doi: [10.1164/rccm.200707-973OC](https://doi.org/10.1164/rccm.200707-973OC), indexed in Pubmed: [17916806](https://pubmed.ncbi.nlm.nih.gov/17916806/).
- Ernst P, Gonzalez AV, Brassard P, et al. Inhaled corticosteroid use in chronic obstructive pulmonary disease and the risk of hospitalization for pneumonia. *Am J Respir Crit Care Med*. 2007; 176(2): 162–166, doi: [10.1164/rccm.200611-1630OC](https://doi.org/10.1164/rccm.200611-1630OC), indexed in Pubmed: [17400730](https://pubmed.ncbi.nlm.nih.gov/17400730/).
- Kew KM, Seniukovich A. Inhaled steroids and risk of pneumonia for chronic obstructive pulmonary disease. *Cochrane Database Syst Rev*. 2014(3): CD010115, doi: [10.1002/14651858.CD010115.pub2](https://doi.org/10.1002/14651858.CD010115.pub2), indexed in Pubmed: [24615270](https://pubmed.ncbi.nlm.nih.gov/24615270/).
- Wedzicha JA, Singh D, Vestbo J, et al. FORWARD Investigators. Extrafine beclomethasone/formoterol in severe COPD patients with history of exacerbations. *Respir Med*. 2014; 108(8): 1153–1162, doi: [10.1016/j.rmed.2014.05.013](https://doi.org/10.1016/j.rmed.2014.05.013), indexed in Pubmed: [24953015](https://pubmed.ncbi.nlm.nih.gov/24953015/).
- Pavord ID, Lettis S, Anzueto A, et al. Blood eosinophil count and pneumonia risk in patients with chronic obstructive pulmonary disease: a patient-level meta-analysis. *Lancet Respir Med*. 2016; 4(9): 731–741, doi: [10.1016/S2213-2600\(16\)30148-5](https://doi.org/10.1016/S2213-2600(16)30148-5), indexed in Pubmed: [27460163](https://pubmed.ncbi.nlm.nih.gov/27460163/).
- Holland AE, Spruit MA, Troosters T, et al. An official European Respiratory Society/American Thoracic Society technical standard: field walking tests in chronic respiratory disease. *Eur Respir J*. 2014; 44(6): 1428–1446, doi: [10.1183/09031936.00150314](https://doi.org/10.1183/09031936.00150314), indexed in Pubmed: [25359355](https://pubmed.ncbi.nlm.nih.gov/25359355/).
- Dasgupta A, Kjarsgaard M, Capaldi D, et al. A pilot randomised clinical trial of mepolizumab in COPD with eosinophilic bronchitis. *Eur Respir J*. 2017; 49(3), doi: [10.1183/13993003.02486-2016](https://doi.org/10.1183/13993003.02486-2016), indexed in Pubmed: [28298405](https://pubmed.ncbi.nlm.nih.gov/28298405/).
- Pliskevich D. Efficacy and tolerability of EPs® 7630 in patients with chronic obstructive pulmonary disease (COPD). <http://isrctn.org/>, doi: [10.1186/isrctn01681733](https://doi.org/10.1186/isrctn01681733).