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Association of Inflammation Mediator in Mucosal and Tissue of Chronic Rhinosinusitis with Recurrent Nasal Polyp

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Abstract

BACKGROUND: Chronic rhinosinusitis with polyps (CRSwNP) have a high risk of recurrence and patients often experience repeated surgery. There are several types of inflammatory patterns in CRSwNP, such as Th2 inflammation (eosinophilic) and Th1/Th17 inflammation (neutrophilic).

AIM: This study aims to determine the expression of IL-5, IL-8, IL-17A and TGF-β in recurrent CRSwNP using the most convenient and non-invasive examination tool such as brushing the mucosal polyp and find out its correlation with polyp tissues.

MATERIAL AND METHODS: A cross-sectional comparative study was carried out on 15 samples of mucosal brushing and polyp tissue. Expressions of IL-5, IL-8, IL-17A and TGF-β on mucosa were measured using the Enzyme-Linked Immunosorbent Assay (ELISA) examination and tissues using Immunohistochemical (IHC) examination.

RESULT: The result showed that Only IL-5 has a significant relationship between mucosa and tissue with moderate positive correlation ($p < 0.05$; $r = 0.527$).

CONCLUSION: This study concluded that mucosa brushing could be used as a simple and non-invasive examination to observe the expression of IL-5 in recurrent CRSwNP. IL-5 is one of the cytokines that mark the Th2 (eosinophilic) inflammatory pattern where eosinophilic polyps are closely related to recurrence.

Introduction

Chronic rhinosinusitis with polyps (CRS) is a chronic inflammatory disease of the sinuses and paranasal sinuses that last for more than 12 weeks, CRS can be accompanied by polyps (CRSwNP) or without polyps (CRSsNP) [1], [2]. This disease could reduce the quality of life of patients and cause an economic burden because of the high cost of treatment, especially because of the high rate of recurrence, causing patients to experience repeated surgery [3], [4], [5]. In the United States, chronic rhinosinusitis accounts for almost 16% of adults per year, which consumes health funds of around 5.8 billion US/year [6]. The prevalence of CRS in Indonesia is quite high. Based on data from the ENT polyclinic at M. Djamil Central Public Hospital, Padang from October 2011 to September 2012, 106 new cases of CRS were found in which 87 cases were CRSWNP without polyps and 19 cases of CRSwNP.

There have been many theories and research, the exact aetiology of nasal polyps and the causes of recurrence is still unknown [6]. The role of several mediators (cytokines) such as interleukin, growth factors and chemokines in the inflammatory process had been widely investigated. In addition, the

effect of a combination of cytokines, which serves as a signal to the cell to provide response and growth factors, produced by T lymphocytes, fibroblasts, epithelial cells such as the Granulocyte Macrophage Colony Stimulating Factor (GM-CSF), Interleukin-3 (IL-3) and Interleukin-5 (IL-5) plays an important role in the formation of polyps [7].

Several recent studies have reported that there were differences in CRS immunoprofile between Caucasian and Asian races. The American and European populations have Th2 inflammatory pattern (eosinophilic inflammation) whereas in the Asian population have Th1/Th17 inflammatory pattern (neutrophilic inflammation [3], [8], [9].

The differences in histological and inflammatory patterns between CRSwNP in the Caucasians and Asians and the high recurrence rate of CRSwNP require further study to find out a comfortable and non-invasive examination tool that can detect polyps early. We hope the polyp can be diagnosed early without having to go through a polypectomy that makes the CRSwNP patient uncomfortable and spend more cost for treatment, such as brushing the mucosa of the polyp. Based on that, we will study the correlation of cytokine expression (IL-5, IL-8, IL-17A, and TGF-β1) in CRSwNP recurrence between mucosa (brushing) and tissue (biopsy) of nasal polyps.

Material and Methods

Sample

Samples obtained from CRSwNP patients who visited the Ear, Nose and Throat (ENT) clinic in the Public Central Hospital Dr M Djamil Padang and several hospitals in West Sumatera on August 2016 until September 2018. A total of 15 CRSwNP patients met the inclusion and exclusion criteria in this study. Research approval was requested from the respondent before the operation. Samples were taken from CRSwNP patients who did not use anti-allergic drugs during the wash out period before brushing (chlorpheniramine for 3 days, cetirizine, fexofenadine, loratadine for 5 days each and corticosteroids for 2 weeks) and aged between 18 and 55 years.

Sampling

Brushing performed on nasal polyps mucosa with the nasoendoscopy in a circular motion by using a modified gynecologic cytology brush. Before brushing the polyp, a cotton tampon containing lidocaine and adrenaline installed with a ratio of 4:1 for 10 minutes on the nasal cavity. Brushing was done on the mucosa of the polyp in a circular motion ten times clockwise. Samples obtained from brushing were inserted into a sterile bottle containing PBS liquid and immediately taken to the Biomedical laboratory in the Faculty of Medicine, Andalas University and stored at a temperature of -80°C.

Retrieval of nasal polyp tissue is performed during surgical removal of the polyp by FESS (Functional Endoscopy Sinus Surgery). Polyp tissue samples when FESS performed were put into neutral formalin liquid and immediately taken to the Anatomy Pathology Laboratory of the Andalas University Medical School to make paraffin blocks.

ELISA

In this research, human IL-5, IL-17 and TGF-β1 were used from R&D and human IL-8 from BT lab for examines nasal mucous polyps.

IHC

Immunohistochemical staining techniques using the Labeled Streptavidin-Biotin Complex (LSAB) method were carried out by manual procedure. The results in the form of preparations were measured and calculated by looking under a microscope to assess the expression of IL-5, IL-8, IL-17, and TGF-β1. In this study, IL-5 (GeneTex) antibodies were used as polyclonal IgG from rabbits. Positive values are the results of assessments of the brown intensity of epithelium and stroma seen in the light microscope.

Statistical Method

We use SPSS program version 17.0.0.0. The analysis aimed to determine the correlation of IL-5, IL-8, IL-17A, and TGF-β1 between tissue (IHC) and mucosa (ELISA) in recurrent CRSwNP.

Results

Research had been conducted on 30 samples from 15 CRSwNP patients who underwent FESS surgery for the second time. This study aims to determine the correlation of the expression of inflammation mediators between tissue and mucosa in recurrent CRSwNP. The clinical characteristics of CRSwNP patients can be seen in the table below.

Table 1: Characteristics of respondents based on gender and age

Characteristics	Recurrent CRSWNP (n = 15)
Sex	
Male	12 (80%)
Female	3 (20%)
Age	41.40 ± 10.23

In Table 1, the percentage of males was higher than that of women; there were 12 male respondents and 3 female respondents with a ratio of 4:1. The average age in the recurring CRSwNP group was 41.40 ± 10.23.

Expression of IL-5, IL-8, IL-17, and TGF-β1 between mucosa and recurrent polyp tissue

The mean expression of IL-5 on the mucosa of recurrent CRSwNP was 2.75 (± 2.02) while the tissue was 78.80 (± 15.01).

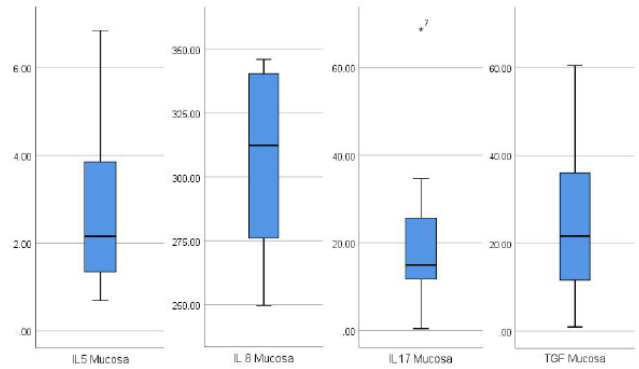

Figure 1: Expression of IL-5, IL-8, IL-17, and TGF-β1 mucosa with ELISA (pg/dl) in recurrent CRSwNP showed in the box plot graphs

In figure 1, IL-5 was the only cytokine that had a significant correlation with moderate positive correlation; it can be concluded that the enhancement of expression of IL-5 in the mucosa accompanied by an increase of the expression of IL-5 in the tissue ($p < 0.05$; $r = 0.527$). The mean of IL-8 in the mucosa was 304.35 (± 34.86) while in the tissue was 78.33% (± 18.79), but there was no significant correlation found ($p > 0.05$).

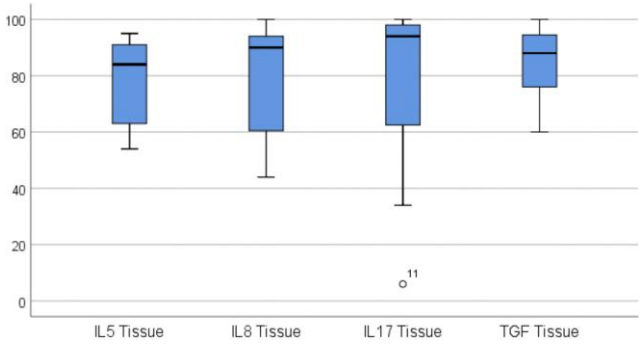

Figure 2: The box plot of expression of IL-5, IL-8, IL-17, and TGF-β1 tissue with IHC (per 100 cells) in recurrent CRSwNP tissue

Like IL-8, IL-17A also did not have a significant correlation, the mean mucosa was 20.13 (± 16.78), and the tissue was 77.13% (± 29.64). The mean of TGF-β1 in the recurrent mucosal CRSwNP was obtained at 24.51 (317.03) while the tissue was 85.33% (± 12.38) with a non-significant correlation ($p > 0.05$).

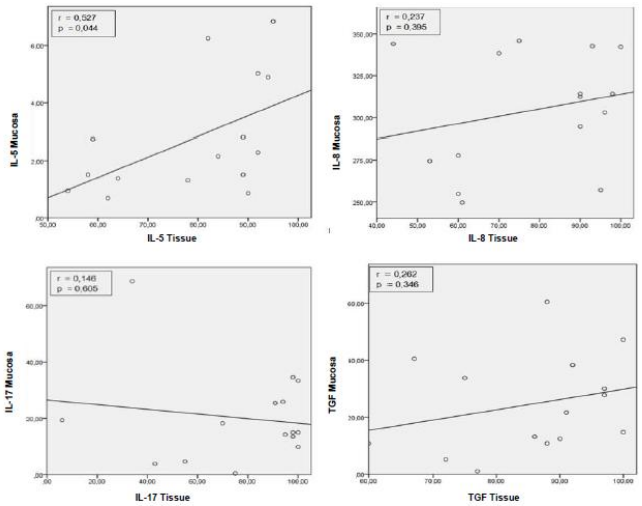

Figure 3: Correlation of expression of IL-5, IL-8, IL-17, and TGF-β1 between mucosa and tissue in CRSwNP recurrent polyps

Of all the cytokines examined, only IL-5 had a significant correlation with a moderate positive correlation ($p < 0.05$; $r = 0.527$).

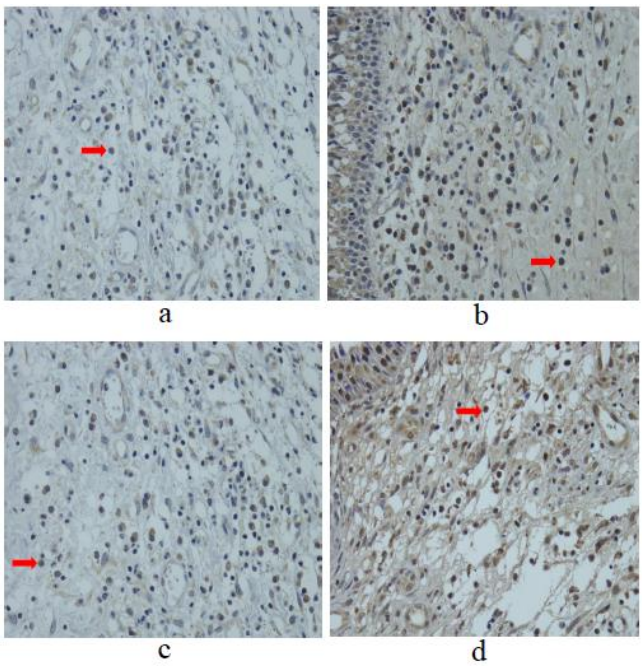

Figure 4: Description of cell expression that produces cytokines in recurrent CRSwNP tissue with 40 X 10 enlargement: a) description of IL-5 expression; b) description of IL-8 expression; c) description of IL-17A expression; d) description of TGF-β1 expression. Red arrows indicate cells that contain positive cytokines

Discussion

Interleukin-5 in the mucosa and recurrent tissue CRSWNP has a significant relationship ($p < 0.05$) with a moderate positive correlation ($r = 0.527$), we can conclude that the enhancement of expression

of IL-5 in the mucosa accompanied by an increase of the expression of IL-5 in the tissue. Interleukin-5 is a cytokine produced by T helper 2 (Th2) and ILC2 cells, which is important in the differentiation, maturation, toxicity, activation and survival of eosinophil [10], [11]. Interleukin-5, together with CCL11 (eotaxin) mobilises eosinophils into the tissues. Interleukin-5 stimulates eosinophils to increase its defence and cytotoxicity. Eosinophil infiltration and granulation encourage to release the toxic products such as major basic protein, eosinophil cationic protein, an eosinophil-derived neurotoxin and eosinophil peroxidase which contribute to airway epithelial damage, mucus hypersecretion of goblet cells, and ciliary movement damage [12]. In a study conducted by Bachert, in immunohistochemical examination 70% of cells containing IL-5 were eosinophils, and there was a strong relationship between IL-5 and Eosinophilic Cationic Protein (ECP) which is a marker of eosinophil activation [10].

Many studies have reported that eosinophilic type CRSwNP has a worse recurrence and prognosis rate. Matsuwaki compared the neutrophilic type with eosinophilic type in 2008. Matsuwaki reported that patients with eosinophilic mucosa showed a high incidence of recurrence in 5 years, where the positive predictive value for recurrence was 85.7% [13]. Nakayama also reported that the presence of eosinophilic mucosa is often associated with disease severity and recurrence of nasal polyps [14]. Ikeda in 2013 compared eosinophilic CRSWNP and neutrophilic CRSWNP types, found that eosinophilic type of CRSWNP had a poor prognosis of the disease and higher recurrence, especially eosinophilic CRSWNP with aspirin intolerance [15]. Similar was also reported by Tecimer in 2014 in which the study reported a higher recurrence rate in eosinophilic nasal polyps (55%) compared with neutrophilic nasal polyps (42.8%) [16]. Van Zele reported that the type of Th2 inflammation and eosinophilic inflammation are the main risk factors for recurrence and the severity of the disease [3]. A study conducted by Lou in 2015 in the Chinese population, also reported that the proportion of tissue eosinophilia over 27% of total tissue cells could predict recurrence in nasal polyps with a sensitivity of 87.4% and specificity of 97.1% [17].

Histologically, Gevaert reported in 2004 that eosinophils were localised in blood vessels, borders, and just below the mucosal epithelium where mast cells were generally granulated and were found increased in the nasal polyp stroma. The enhancement of plasma cells and lymphocytes are also found on cellular levels [18]. The positive correlation in this study proved that in CRSwNP recurrence, the examination of IL-5 by brushing technique on the mucosa could be assessed with IL-5 on the tissue so that the techniques can be used to look expression of IL-5 in early developmental of CRSwNP and can predict the possibility of recurrence.

The mean expression of IL-8 on the mucosal CRSwNP recurrent is 304.35 (± 34.86) and in the tissues 78.33% (± 18.79) but there was no significant relationship between tissue and mucosa ($p > 0.05$). Interleukin-8 is an inflammatory cytokine that has strong neutrophil chemotaxis activity and can induce degranulation, respiratory burst, adherence, deformation, Ca2 + mobilisation and increased regulation of CD11b/CD18 neutrophils. The initial phase of interleukin-8 will be secreted mainly from glandular cells and epithelial cells. These cells tend to secrete IL-8 to attract neutrophils out of the mucosa to the luminal side, and neutrophils that have migrated can also secrete IL-8 resulting in positive feedback [19]. Besides neutrophils, IL-8 is also a chemokine that is very important for eosinophils in all types of CRSwNP and nasal polyps [20]. In the Asian population, CRSWNP polyps occur largely influenced by infectious factors where at the onset of CRSwNP polyps the amount of IL-8 is needed for mobilisation of inflammatory cells such as neutrophils and eosinophils to the nasal polyp CRS site [21], [22]. In recurrent CRSwNP, the role of IL-5 is more dominant where IL-5 plays a role in the differentiation, maturation and survival of eosinophils, which are important in the growth and recurrence of polyps.

In this study, IL-17 expression in the mucosa was 20.13 (± 16.78), while in the tissue was 77.13% (± 29.64). Interleukin-17 is produced by cells induced by proinflammatory cytokines (such as IL-1, IL-6 and TNF-α), chemokines (CXCL1, CXCL2, CXCL5 and CXCL8), and adhesive molecules (ICAM-1 and VCAM-1) [24]. The main source of IL-17 in nasal polyps is lymphocytes. Interleukin-17 can modulate the life of neutrophils in nasal polyps that are not cystic fibrosis. In the study of Derycke et al., it can be proven that IL-17 can prolong the life of neutrophils in non-fibrous nasal polyps where IL-17 can reduce apoptosis [23]. In this study, IL-17 was expressed primarily by macrophages. IL-17 binds to the IL-17 receptor with the involvement of tumour necrosis factor and receptor factors which activate nuclear factor κB and subsequently modulate proinflammatory cytokines. Jiang et al. explained that interactions between IL-17 and IL-17 RD could contribute to the growth of nasal polyps, such as thickening of lamina basal cells and glandular hyperplasia [24].

In this study, the expression of mucosa TGF-β1 was 24.51 (± 17.03) and in tissues was 85.33% (± 12.38). Transforming Growth Factor Beta1 is a mediator related to tissue remodelling. Transforming growth factor-β1 (TGF-β1) is a chemoattractant for fibroblasts, stimulating fibroblast proliferation, and enhancing collagen laydown by fibroblasts. Of the TGF-isoforms, TGF-β1 synthesised by infiltrating eosinophils may contribute to stromal fibrosis and basement membrane thickening, which are characteristic of nasal polyp [25]. Transforming Growth Factor Beta1 plays an important role in the balance of fibrinolysis and fibrogenesis, which means it affects the extracellular matrix. This imbalance is

due to the low levels of TGF-β1 in nasal polyps and the inhibitory effects on MMP-9 activity through the release of TIMP-1 [26].

In IL-8, IL-17 and TGF-β1, there was no correlation between the expression of cytokines in the mucosa and tissue in recurrent CRSwNP, because the relationship was not statistically significant ($p > 0.05$). The absence of a significant association with IL-8, IL-17A, TGF-β1 is probably due to the variability of the inflammatory expression in nasal polyps. Stevens reports that from examining 20 cytokines of inflammatory mediators between nasal lavage and biopsy tissue of nasal polyps, only IL-10 had a significant correlation and positive correlation. This finding supports previous observations where there was regional variability in the expression of cytokine inflammatory mediators in sinonasal cavities [27], [28]. Pawankar also explained that the histological characteristics of the mucosa and inflammation cells of the exudate depend on allergic status [29]. Besides of that, in this research, CRSwNP was not differentiated based on the dominant histopathological type of inflammatory cells, eosinophilic CRSwNP and neutrophilic type CRSwNP. Eosinophilic type CRSwNP is CRS with a Th2 inflammatory pattern where IL-5 is one of the signalling cytokines [30]. Neutrophilic type CRSwNP is a polyp which initially influenced by infectious factors and commonly found in Asian ethnicities. In neutrophilic type CRSwNP, the inflammatory pattern that plays a role is Th1 and Th17 characterised by increased expression of IL-8 and IL-17A [31]. Also, in this study, the locus of polyp tissue sampling could not be taken at the same location for each sample.Samplingof tissue polypththe e could not distinguish between the stalk, body (distal, medial and proximal) and peak of the polyp. The difference of sampling location could make different expression due to the discrepancy activity processes in each part of the polyp.

In this study, it was found that only IL-5 has a correlation between mucosa and tissue in recurrent CRSwNP, we can conclude mucosal brushing examination can describe the expression of IL-5 in tissues. Further research is needed to find out the correlation of cytokine between the mucosa and tissue mainly based on the histology type of CRSwNP and to look for an easier and more convenient examination tool to diagnose recurrent CRSwNP especially at the beginning of course of the disease.

References

1. Fokkens WJ, Lund VJ, Mullol J, Bachert C, Baroody AF, Cohen N, et al. European Position Paper on Rhinosinusitis and Nasal Polyps 2012. Rhinology Supplement. 2012:1-298. https://doi.org/10.4193/Rhino50E2 PMid:22469599

2. Gevaert P, Van Bruaene N, Cattaert T, Van Steen K, Van Zele T, Acke F, et al. Mepolizumab, a humanized anti-IL-5 mAb, as a treatment option for severe nasal polyposis. The Journal of allergy and clinical immunology. 2011; 128(5):989-95. https://doi.org/10.1016/j.jaci.2011.07.056 PMid:21958585

3. Van Zele T, Holtappels G, Gevaert P, Bachert C. Differences in initial immunoprofiles between recurrent and nonrecurrent chronic rhinosinusitis with nasal polyps. American journal of rhinology & allergy. 2014; 28(3):192-8. https://doi.org/10.2500/ajra.2014.28.4033 PMid:24980230

4. Tomassen P, Van Zele T, Zhang N, Perez-Novo C, Van Bruaene N, Gevaert P, et al. Pathophysiology of chronic rhinosinusitis. Proceedings of the American Thoracic Society. 2011; 8(1):115-20. https://doi.org/10.1513/pats.201005-036RN PMid:21364229

5. Eloy P, Poirrier AL, De Dorlodot C, Van Zele T, Watelet JB, Bertrand B. Actual concepts in rhinosinusitis: a review of clinical presentations, inflammatory pathways, cytokine profiles, remodeling, and management. Current allergy and asthma reports. 2011; 11(2):146-62. https://doi.org/10.1007/s11882-011-0180-0 PMid:21274665

6. Hsu J, Peters AT. Pathophysiology of chronic rhinosinusitis with nasal polyp. American journal of rhinology & allergy. 2011; 25(5):285-90. https://doi.org/10.2500/ajra.2011.25.3680 PMid:22186239

7. Van Crombruggen K, Zhang N, Gevaert P, Tomassen P, Bachert C. Pathogenesis of chronic rhinosinusitis: inflammation. The Journal of allergy and clinical immunology. 2011; 128(4):728-32. https://doi.org/10.1016/j.jaci.2011.07.049 PMid:21868076

8. Bachert CC, LI. Gevaert,I. Rhinosinusitis and Nasal Polyps. In: Adkinson NF, editor. Middleton's Allergy, Principles and Practice. vol 1. 8th ed. Baltimore: Elsevier Saunder, 2014:686-99.

9. Mahdavinia M, Suh LA, Carter RG, Stevens WW, Norton JE, Kato A, et al. Increased noneosinophilic nasal polyps in chronic rhinosinusitis in US second-generation Asians suggest genetic regulation of eosinophilia. The Journal of allergy and clinical immunology. 2015; 135(2):576-9. https://doi.org/10.1016/j.jaci.2014.08.031 PMid:25312761 PMCid:PMC4323954

10. Bachert C, Zhang N, Holtappels G, De Lobel L, van Cauwenberge P, Liu S, et al. Presence of IL-5 protein and IgE antibodies to staphylococcal enterotoxins in nasal polyps is associated with comorbid asthma. The Journal of allergy and clinical immunology. 2010; 126(5):962-8. https://doi.org/10.1016/j.jaci.2010.07.007 PMid:20810157

11. Tomassen P, Vandeplas G, Van Zele T, Cardell LO, Arebro J, Olze H, et al. Inflammatory endotypes of chronic rhinosinusitis based on cluster analysis of biomarkers. The Journal of allergy and clinical immunology. 2016:1449-56. https://doi.org/10.1016/j.jaci.2015.12.1324 PMid:26949058

12. Chung KF. Targeting the interleukin pathway in the treatment of asthma. The Lancet. 2015; 386(9998):1086-96. https://doi.org/10.1016/S0140-6736(15)00157-9

13. Matsuwaki Y, Ookushi T, Asaka D, Mori E, Nakajima T, Yoshida T, et al. Chronic rhinosinusitis: risk factors for the recurrence of chronic rhinosinusitis based on 5-year follow-up after endoscopic sinus surgery. International archives of allergy and immunology. 2008; 146 Suppl 1:77-81. https://doi.org/10.1159/000126066 PMid:18504412

14. Nakayama T, Yoshikawa M, Asaka D, Okushi T, Matsuwaki Y, Otori N, et al. Mucosal eosinophilia and recurrence of nasal polyps - new classification of chronic rhinosinusitis. Rhinology. 2011; 49(4):392-6.

15. Ikeda K, Shiozawa A, Ono N, Kusunoki T, Hirotsu M, Homma H, et al. Subclassification of chronic rhinosinusitis with nasal polyp based on eosinophil and neutrophil. The Laryngoscope. 2013; 123(11):E1-9. https://doi.org/10.1002/lary.24154 PMid:23670893

16. Tecimer SH, Kasapoglu F, Demir UL, Ozmen OA, Coskun H, Basut O. Correlation between clinical findings and eosinophil/neutrophil ratio in patients with nasal polyps. European Archives of Oto-Rhino-Laryngology. 2014; 272(4):915-21. https://doi.org/10.1007/s00405-014-3174-4 PMid:25007735

17. Lou H, Meng Y, Piao Y, Wang C, Zhang L, Bachert C. Predictive significance of tissue eosinophilia for nasal polyp recurrence in the Chinese population. American journal of rhinology & allergy. 2015; 29(5):350-6. https://doi.org/10.2500/ajra.2015.29.4231 PMid:26219765

18. Gevaert P. Eosinophilic Inflammation in Nasal Polyposis : Regulation of Interleukin-5 and Interleukin 5 Receptor alfa isoformis [Disertation]: University Gent; 2004.

19. Suzuki H, Ikedab K. Mode of Action of Long-Term Low-Dose Macrolide Therapy for Chronic Sinusitis in the Light of Neutrophil Recruitment. Current Drug Targets - Inflammation & Allergy. 2002; 1(1):117-26. https://doi.org/10.2174/1568010023344832

20. Peric A, Vojvodic D, Radulovic V, Vukomanovic-Durdevic B, Miljanovic O. Correlation between cytokine levels in nasal fluid and eosinophil counts in nasal polyp tissue in asthmatic and non-asthmatic patients. Allergologia et immunopathologia. 2011; 39(3):133-9. https://doi.org/10.1016/j.aller.2010.05.001 PMid:21211895

21. Zhang Y, Gevaert E, Lou H, Wang X, Zhang L, Bachert C, et al. Chronic rhinosinusitis in Asia. The Journal of allergy and clinical immunology. 2017; 140(5):1230-9. https://doi.org/10.1016/j.jaci.2017.09.009 PMid:28987810

22. Yoon BN, Choi NG, Lee HS, Cho KS, Roh HJ. Induction of interleukin-8 from nasal epithelial cells during bacterial infection: the role of IL-8 for neutrophil recruitment in chronic rhinosinusitis. Mediators of inflammation. 2010; 2010:1-6. https://doi.org/10.1155/2010/813610 PMid:20634942 PMCid:PMC2904024

23. Avelino MA, Wastowski IJ, Ferri RG, Elias TG, Lima AP, Nunes LM, et al. Interleukin-17A expression in patients presenting with nasal polyposis. Brazilian journal of otorhinolaryngology. 2013; 79(5):616-9. https://doi.org/10.5935/1808-8694.20130110 PMid:24141678

24. Jiang XD, Li GY, Li L, Dong Z, Zhu DD. The characterization of IL-17A expression in patients with chronic rhinosinusitis with nasal polyps. American journal of rhinology & allergy. 2011; 25(5):e171-5. https://doi.org/10.2500/ajra.2011.25.3645 PMid:22186234

25. Lee YM, Kim SS, Kim HA, Suh YJ, Lee SK, Nahm DH, et al. Eosinophil inflammation of nasal polyp tissue: relationships with matrix metalloproteinases, tissue inhibitor of metalloproteinase-1, and transforming growth factor-beta1. Journal of Korean medical science. 2003; 18(1):97-102. https://doi.org/10.3346/jkms.2010.25.1.97

26. Van Bruaene N, Derycke L, Perez-Novo CA, Gevaert P, Holtappels G, De Ruyck N, et al. TGF-beta signaling and collagen deposition in chronic rhinosinusitis. The Journal of allergy and clinical immunology. 2009; 124(2):253-9, 9 e1-2. https://doi.org/10.1016/j.jaci.2009.04.013 PMid:19500825

27. Stevens WW, Lee RJ, Schleimer RP, Cohen NA. Chronic rhinosinusitis pathogenesis. The Journal of allergy and clinical immunology. 2015; 136(6):1442-53. https://doi.org/10.1016/j.jaci.2015.10.009 PMid:26654193 PMCid:PMC4680986

28. Stevens WW, Schleimer RP, Kern RC. Chronic Rhinosinusitis with Nasal Polyps. The journal of allergy and clinical immunology In practice. 2016; 4(4):565-72. https://doi.org/10.1016/j.jaip.2016.04.012 PMid:27393770 PMCid:PMC4939220

29. Pawankar R, Nonaka M, Satoru Masuno a, Kimura S. Nasal Polyposis. Current Concepts on the Pathomechanisms of Chronic Rhinosinusitis and Nasal Polyps. Berlin Heidelberg: Springer, 2010:185-8. https://doi.org/10.1007/978-3-642-11412-0_21

30. Zhang N, Zele TV, Gevaert P, Holtappels G, Cauwenberge PV, Bachert C. Remodelling in Nasal Polyposis is Independent of TH-Cell Polarisation. Journal of Allergy and Clinical Immunology. 2007; 119(1):S141. https://doi.org/10.1016/j.jaci.2006.11.681

31. Derycke L, Zhang N, Holtappels G, Dutre T, Bachert C. IL-17A as a regulator of neutrophil survival in nasal polyp disease of patients with and without cystic fibrosis. Journal of cystic fibrosis : official journal of the European Cystic Fibrosis Society. 2012; 11(3):193-200. https://doi.org/10.1016/j.jcf.2011.11.007 PMid:22178076