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ORIGINAL ARTICLE

Correlation between cytokine levels in nasal fluid and eosinophil counts in nasal polyp tissue in asthmatic and non-asthmatic patients

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Abstract
Background/Aims: Concentrations of mediators in nasal secretions could reflect the inflammatory status of the nasal mucosa and evolution of sinus disease. So, the aim of our study was to evaluate local immune reaction by measuring crucial Th1, Th2 and inflammatory cytokines in nasal fluid samples of patients with nasal polyps (NP), and to correlate them to clinical, radiological findings and to the degree of eosinophil infiltration of polyp tissue. Therefore, in our study we compared the cytokine levels in nasal fluid of asthmatic and non-asthmatic patients with nasal polyposis, the eosinophil counts in NP tissues of these patients, and we correlated cytokine levels with eosinophil counts in NP tissue specimens.
Material and methods: Thirty patients with nasal polyposis (NP) (15 asthmatic and 15 non-asthmatic) were included in this prospective study. Nasal secretion samples were collected from nasal cavities of all subjects. The levels of 11 cytokines (TNF-α, TNF-β, IL-1β, IL-2, IL-4, IL-5, IL-6, IL-8, IL-10, IL-12, and IFN-γ) were measured using commercial flow cytometric kit. Eosinophils were counted in haematoxylin-and-eosin-stained NP sections.
Results: The concentrations of Th2 cytokines IL-5, IL-6, IL-10, and Th1 cytokine IFN-γ were significantly higher in patients with NP and asthma compared with non-asthmatic subjects. A positive correlation was found between IL-6 and TNF-α levels in nasal fluid and eosinophil counts in polyp tissue in non-asthmatic subjects. In asthmatic NP patients, we found positive correlation between level of IL-6 and eosinophil counts and negative correlation between IFN-γ level and number of eosinophils in NP tissue specimens.
Conclusion: Our results showed that these patients with similar clinical findings had significantly different mediator profiles in their nasal secretions, implying clear differences in pathogenesis of their NP.

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Introduction

Asthma is a chronic disease characterised by intermittent obstruction and inflammatory changes of the airways, and bronchial hyperresponsiveness.[1] Seven percent of asthma patients have nasal polyps.[2] Nasal polyps (NP) originate from the paranasal sinuses, most often from the anterior ethmoid complex and from there they can descend between the middle turbinate and the lateral nasal wall into the nasal cavity causing symptoms such as nasal obstruction, anosmia, sneezing, rhinorrhea, and itching.[2] Similar typical findings can be found in microscopic examination of NP when compared with the bronchial mucosa in patients with asthma. In both tissues there is epithelial damage, goblet cell hyperplasia, thickening of basement membrane, accumulation of extracellular matrix, fibrosis and eosinophil-dominated inflammation.[3] Eosinophil accumulation is a hallmark of NP. Activated eosinophils release a wide range of cytotoxic proteins and transformed growth factors that cause injury of epithelium, thickening of epithelial basement membrane, stromal fibrosis and angiogenesis, and glandular hyperplasia.[3] The link between these two diseases is still made plausible by observation that the nasal polyp eosinophilic inflammation is significantly higher in NP patients with concomitant asthma when compared with non-asthmatic NP patients.[3]

NP is a multifactorial disease with several aetiological factors. It has been suggested that an ineffective local Th1-based immune response in these patients is associated with increased Th2-cytokine-based activity, which contributes to a chronic infection as well as to an increased presence of eosinophils, which then lead to further polyp formation.[4] It has been further proposed that the weakened Th1 response in these patients may be secondary to the down-regulation of some specific toll-like receptors involved in the innate immune response.[4] Different mutations which alter toll-like receptor function have revealed the significance of these receptors in susceptibility to infection and their involvement in the pathogenesis of a large number of non-infective inflammatory disorders.[5]

Nasal secretions represent a first-line defence medium, in which the leukocyte compartment probably acts as an efficient part of the defence mechanism along with the mucociliary transport system and the biochemical properties of the mucus.[6] To characterise inflammatory changes of the upper respiratory mucosa, cellular secretory products in nasal secretions may be determined.[7] Nasal secretions contain minute amounts of cytokines, potent biological factors involved in the regulation of inflammation and immune defence, and other inflammatory mediators expressed by various epithelial and non-epithelial cells.[8] Because cytokines play a dominant role in the pathophysiology of airway disease, the cytokine profile in nasal secretions may help to recognise mechanisms underlying NP associated with bronchial asthma.

The aims of this prospective study were: (1) to compare the cytokine levels in nasal fluid of asthmatic and non-asthmatic patients with NP; (2) to compare the counts of eosinophils in NP tissue specimens of asthmatic and non-asthmatic subjects; and (3) to correlate cytokine levels with eosinophil counts in NP tissue specimens.

Materials and methods

Human subjects

Thirty NP patients (15 asthmatic and 15 non-asthmatic) were included in this prospective-analytic study. All the patients had necessity for functional endoscopic sinus surgery (FESS). Written informed consent was obtained from all subjects. The study was approved by the Ethics Committee of the Military Medical Academy, Belgrade, Serbia and in accordance with the Helsinki Declaration and the World Health Association. The diagnosis of NP was based on each patient's medical history and on the results of nasal endoscopy and computed tomography (CT). Fifteen patients had diagnosis of mild persistent bronchial asthma. Diagnosis of asthma was made at the time of inclusion in the study according to the Global Initiative on Asthma (GINA).[9] The assessment of the severity of asthma was done by a pulmonologist based on the patient's medical history, clinical data and on pulmonary function testing, including forced expiratory volume in one second (FEV1) and methacholine provocation test (Mch PD20). Only patients with polyps associated with mild bronchial asthma, without aspirin sensitivity, were included in the study. The diagnosis of aspirin-induced asthma was done by a positive bronchial aspirin-provocation-test. The other exclusion criteria were the presence of antrochoanal and sphenochoanal polyps, cystic fibrosis, and primary ciliary dyskinesia. All subjects included in this investigation did not have bronchial or respiratory tract infection and none of the subjects were treated with oral and topical corticosteroids, antibiotics and antihistamines for at least three weeks before the enrolment. Skin prick tests were performed on all patients for sensitivity to 18 common allergens. A test result was considered positive when at least one of the wheals had a diameter 3 mm higher than that in the negative control. Subjects were considered allergic if they had a serum IgE level > 100 IU/mL.

Clinical score

The presence of nasal symptoms associated with NP (obstruction, anosmia, sneezing, rhinorrhea, and itching) on the day of the enrolment in the study was scored according to Tsicopoulos et al.[10] from 0 to 3: 0 for no symptoms; 1 for mild symptoms; 2 for moderate symptoms; and 3 for severe symptoms, so that the maximal global nasal symptom score is 15.

Endoscopic physical findings were scored according to Lildholdt et al.[11] The degree of nasal polyps is classified in relation to fixed anatomical landmarks in four steps: 0 = ''no polyposis'', 1 = ''mild polyposis (small polyps not reaching the upper edge of the inferior turbinate)'', 2 = ''moderate polyposis (medium-sized polyps reaching between the upper and lower edges of the inferior turbinate)'', 3 = ''severe polyposis (large polyps reaching below the lower edge of the inferior turbinate)''. The maximal endoscopic score is 6, bilaterally.

Findings on CT scans were graded according to the Lund-Mackay score.[12] The mucosal abnormalities were graded as 0 (no abnormality), 1 (partial opacification), or 2 (total opacification) of the frontal, maxillary, anterior ethmoid,

posterior ethmoid and sphenoid sinus, bilaterally. The ostiomeatal complexes were scored bilaterally as 0 (not occluded) or 2 (occluded). The maximal CT grading score is 24.

Sampling of nasal fluid and cytokine determination

Nasal fluid samples were collected from nasal cavities of all 30 subjects (15 patients with NP, and 15 patients with NP and asthma) a few days before endoscopic sinus surgery using modified absorption technique with cotton wool sticks (length 10 millimetres, diameter 4 millimetres), which were inserted into the nasal cavity posterior to the mucocutaneous junction for 60 seconds, as previously described.[13] All of the samples were put in a 2 mL Eppendorf tube containing 1 mL of transfer medium (phosphate-buffered saline with gentamicin 50 μg/mL, penicillin G 340 U/mL, fungizone 500 μg/mL) for 30 minutes because of diffusion of cytokines into the medium and then stored at 4 °C for a maximum of 2 h until processed. Nasal fluid was centrifuged at 1000 *g* for 10 minutes to separate the cellular components. After centrifugation, supernatants were portioned and stored at -70 °C until cytokine determination. The levels of eleven cytokines (TNF-α, TNF-β, IL-1β, IL-2, IL-4, IL-5, IL-6, IL-8, IL-10, IL-12, and IFN-γ) were measured in all of the 30 samples using commercial flow cytometric kit (Flow Cytomix, Bender MedSystems, USA) on the flow cytofluorimeter (Beckman Coulter XL-MCL, USA), which was connected with BMS Flow Cytomix Pro 2.2 Software, according to the manufacturer's instructions. The sensitivity, lower and upper limits of detection were as follows: from 22 to 17,000 pg/mL for TNF-α; from 32 to 17,000 pg/mL for TNF-β; from 17 to 11,000 pg/mL for IL-1β; from 28 to 11,000 pg/mL for IL-2; from 20 to 11,000 pg/mL for IL-4; from 30 to 12,000 pg/mL for IL-5, from 21 to 11,000 pg/mL for IL-6; from 13 to 10,500 pg/mL for IL-8; from 20 to 11,000 pg/mL for IL-10; from 15.1 to 2,300 pg/mL for IL-12; from 8 to 11,000 pg/mL for IFN-γ. The examination was performed twice in the same sample, and the concentrations obtained were then averaged. Reproducibility within the assay was evaluated in independent experiments, as follows for cytokine determination in the same sample: coefficient of variation for IL-1β was 5%; for IL-2 was 6%; for IL-4 was 7%; for IL-5 was 3%; for IL-6 was 2%; for IL-8 was 2%; for IL-10 was 2%; for TNF-α was 8%; for TNF-β was 8%; for IL-12 was 5%; and for IFN-γ was 2%. According to the producer's declaration (Bender MedSystems, USA, Flow Cytomix BMS810FF), overall intra assay coefficient of variation should not exceed 10%.

Tissue preparation and histological examination

All the patients were operated endoscopically (FESS), under general anaesthesia. Nasal polyps located in the middle meatus were surgically removed. For histological examination, polyp specimens were fixed in 10% formaldehyde, embedded in paraffin, cut with the microtome into 5-μm sections and stained with haematoxylin and eosin. Histological examination was performed using digital optical microscope (Nikon Coolscope), assisted with computerised picture analysis system. This system was programmed by ImageJ (Java-based image processing program).[14] Once the glass slide was set, Bright field images could be viewed on the monitor. Eosinophils were counted at x 400 magnification. The visual field was oriented along the whole length of the epithelium basement membrane. To yield the mean number of eosinophils per high power field (HPF), 10 randomly chosen HPFs of a single section were examined. The eosinophils in each section were counted, and the average number of eosinophils per HPF was calculated.

Statistical analysis

Data were expressed as mean ± standard deviation (± SD). Statistical comparisons of the results were performed using the non-parametric Mann-Whitney U test, and Chi square-test. Correlations between different parameters were made by the Pearson's correlation test. A p value less than 0.05 was considered to be statistically significant. For statistical analysis, we used Stat Plus 2007 programme as a computerised analysis system.

Results

There were 11 male and four female patients in the NP group (mean age 42.8 ± 13.71 years) and 10 male and five female patients in the NP with asthma group (mean age 46.47 ± 15.25 years). Five patients in the NP group and eight patients in the NP with asthma group were atopic. This finding showed a higher percentage of subjects with allergy in the NP with asthma group than in the NP group ($p < 0.01$; Chi square-test).

The groups did not significantly differ according to sex and age. Comparing the two main groups (NP with asthma and NP without asthma), we did not find any significant difference according to the global nasal symptom score, endoscopic score, and Lund-Mackay score (Table 1). We also did not find significant differences in the levels of TNF-α; TNF-β; IL-1β; IL-2; IL-4; IL-8; and IL-12 in the nasal secretions. The concentrations of IL-10; IL-6; IL-5; and IFN-γ in nasal fluid were significantly higher in patients with NP and asthma, compared with patients with NP without asthma (77.07 ± 67.06 pg/mL *vs* 31.4 ± 52.67 pg/mL, $p < 0.05$; 291 ± 243.01 pg/ml *vs*

Table 1 Patient characteristics.

	Nasal polyposis	Nasal polyposis + asthma
Patients	15	15
Age	42.8 ± 13.71	46.47 ± 15.25
Male/Female ratio	11/4	10/5
FEV1	102.07 ± 3.32	94.4 ± 5.18
MchPD20(μg)	1662.27 ± 59.26	503.8 ± 103.36
Allergic	5	8
Non-allergic	10	7
Nasal symptom score	10.6 ± 1.92	11.47 ± 2.26
Nasal endoscopic score	5.2 ± 1.01	5.07 ± 1.03
Lund-Mackay score	17.13 ± 2.59	18.4 ± 2.53

All results are expressed as means ± SD; FEV1 = forced expiratory volume in 1 second; Mch PD20(μg) = amount of methacholine in micrograms.

82.27 ± 97.53 pg/mL, p < 0.01; 618.8 ± 585.17 pg/mL *vs* 270.4 ± 723.27 pg/mL, p < 0.01; 88.4 ± 67.27 pg/mL *vs* 37.73 ± 37.07 pg/mL, p < 0.05, respectively) (Table 2). The eosinophil counts in NP tissue specimens were higher in the NP with asthma group than in the NP group (35.06 ± 23.23 *vs* 26.4 ± 22.87), but these differences were not statistically significant (Figure 1 a, b). In asthmatic patients' polyps, the subepithelial loose connective tissue and epithelial layer were infiltrated by numerous eosinophils (Figure 2).

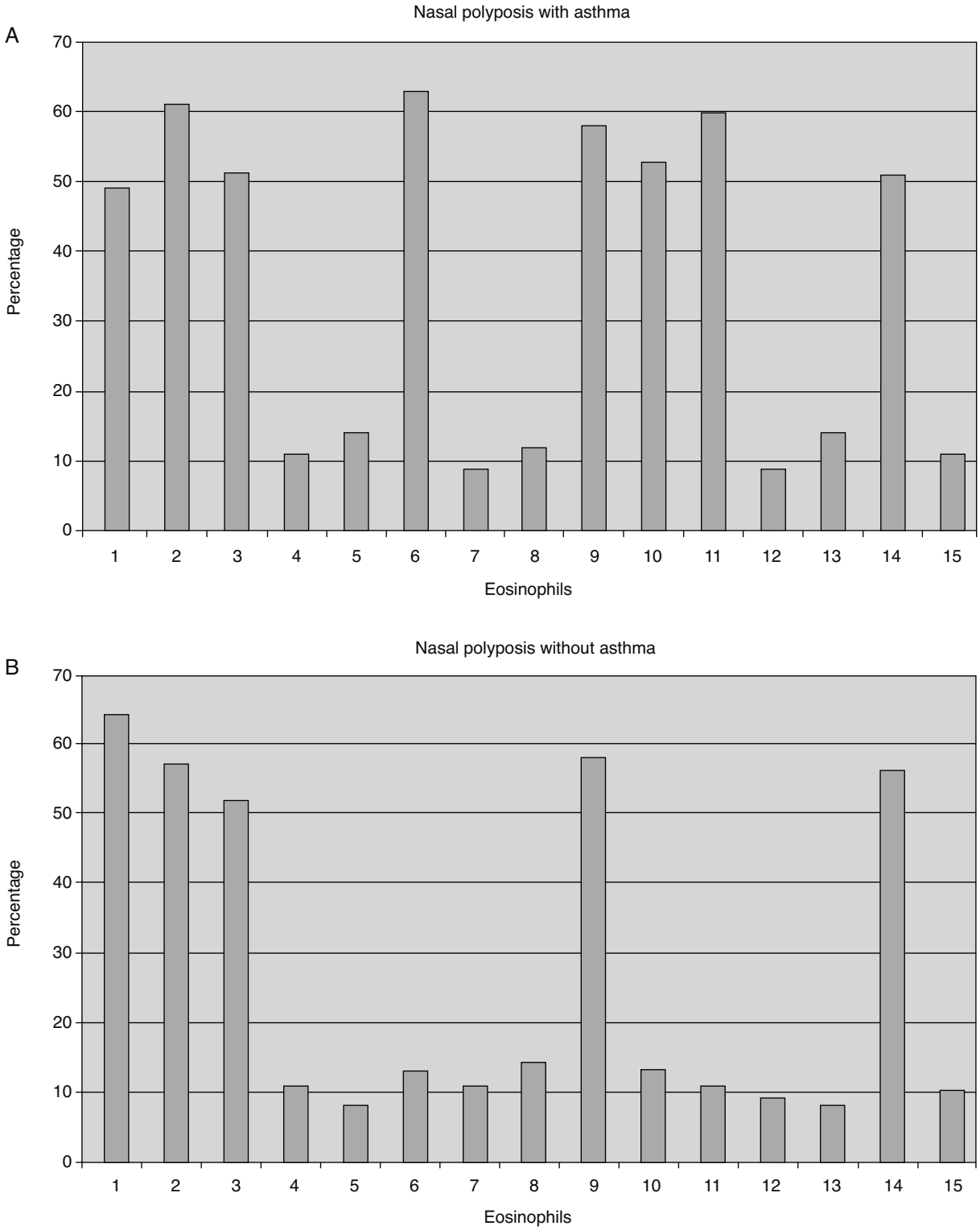

Figure 1 The eosinophil count was higher in the polyp tissue samples from asthmatic patients (a) than in samples from non-asthmatic ones (b), but these differences were not statistically significant.

Table 2 Cytokine levels in nasal secretions.

	IL-12	IFN-γ	IL-2	IL-10	IL-8	IL-6	IL-4	IL-5	IL-1β	TNF-α	TNF-β
Patients with nasal polyps											
mean	6.27	37.73	229.33	31.46	169.82	82.27	908.64	270.45	28.33	25.33	166.33
SD	19.49	37.07	167.36	52.67	278.23	97.53	1045.5	723.27	47.44	24.54	223.27
	IL-12	IFN-γ	IL-2	IL-10	IL-8	IL-6	IL-4	IL-5	IL-1β	TNF-α	TNF-β
Patients with nasal polyps and asthma											
mean	26.6	88.4	353.7	77.07	133.7	291	1352.4	618.8	42.46	46.47	200.93
SD	43.87	67.27	288.6	67.06	100.45	243.01	1317.7	585.18	65.21	42.93	325.78

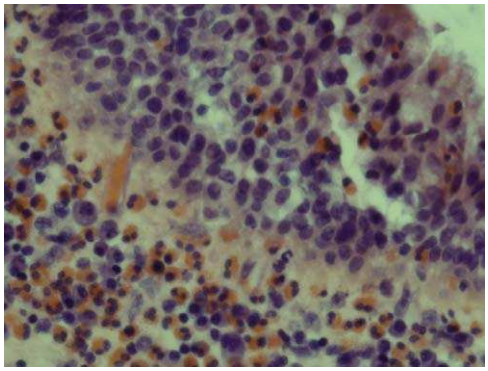

Figure 2 In asthmatic patients' polyps, the epithelium and the subepithelial connective tissue were infiltrated by numerous eosinophils. (Haematoxylin and Eosin staining, x 400 magnification).

We also found significantly higher levels of IFN-γ, IL-6, IL-5 ($p < 0.01$) and TNF-α ($p < 0.05$) in non-allergic patients with NP and asthma compared with non-allergic patients with NP without asthma. In the NP group, we found significantly higher concentrations of IL-6 in nasal fluid ($p < 0.05$) and significantly higher eosinophil counts ($p < 0.01$) in atopic than in non-atopic patients. In the NP with asthma group, the levels of IL-4, IL-6 and IFN-γ, and eosinophil counts in NP tissue specimens were significantly higher in allergic than in non-allergic subjects ($p < 0.05$ for cytokines and $p < 0.01$ for eosinophils).

Our results also showed a positive correlation between IL-6 and TNF-α levels in nasal fluid and eosinophil counts in tissue in the NP group ($r = 0.535$, $p = 0.040$ for IL-6) ($r = 0.567$, $p = 0.028$ for TNF-α). In the NP with asthma group, there was positive correlation between the level of IL-6 and eosinophil counts ($r = 0.546$, $p = 0.035$), and negative correlation between the IFN-γ level and eosinophil counts ($r = -0.614$, $p = 0.015$) (Table 3).

Table 3 Correlation between cytokine levels in nasal fluid and eosinophil counts in nasal polyp tissue.

Cytokines	Eosinophils	
	NP with asthma	NP without asthma
IL-6	$r = 0.546$	$r = 0.535$
	$p = 0.035$	$p = 0.040$
TNF-α	$r = 0.338$	$r = 0.567$
	$p > 0.05$	$p = 0.028$
IFN-γ	$r = -0.614$	$r = -0.242$
	$p = 0.015$	$p > 0.05$

Discussion

Nasal fluid reflects the inflammatory status of the nasal mucosa and the evolution of mucosal disease. However, mechanisms of cytokine release in nasal fluid are not well known. Results published by Ohkubo et al.[15] showed that IL-6 was released to the nasal secretions mainly from the migrating cells and epithelial cells as a result of the antigen provocation, reflex action of methacholine and by direct action of histamine.

NP may have a negative impact on lower airway biology, being involved in aggravation of bronchial disease. The mechanisms that connect upper and lower airway dysfunction are: nasal bronchial reflex, mouth breathing caused by nasal obstruction, and pulmonary aspiration of nasal contents.[16] It has been shown in a rabbit model of chronic sinusitis that postnasal drainage of inflammatory mediators may affect lower airway responsiveness.[17] Therefore, one can hypothesise that a local nasal inflammatory stimulus may induce a systemic effect leading to bronchial eosinophilic inflammation.[18]

Th2-type cytokines are thought to regulate inflammatory cell recruitment, activation, survival and the release of tissue-damaging mediators.[19] Hamilos et al.[20] found significantly higher levels of IL-5 in nasal polyp tissue from asthmatic than those from non-asthmatic subjects. The results of our research have also shown significantly higher concentration of Th2 cytokines (IL-5, IL-6 and IL-10) in nasal secretions in patients with NP and asthma than in non-asthmatic NP patients. These findings underline local Th1/Th2 balance as a key factor that governs local inflammation, implying that there is a different mechanism in NP evolution in asthmatic and non-asthmatic patients.

Previous data point to IL-5 as one of the key proteins in the pathomechanism of tissue eosinophilia, enhancing the differentiation, activation, expansion, mobilisation, and in situ survival of eosinophils.[21] It is widely accepted that IL-5 play an important role in the pathogenesis of bronchial asthma where it induces eosinophil mobilisation, B-cell growth and differentiation.[21] The main sources of IL-5 were eosinophils, Th2-lymphocytes and mast cells.[21] Preliminary findings presented by Fan et al.[22] suggest that T-cell-derived

IL-5 and autosecretion of IL-5 from activated eosinophils could be the reasons for the extension and persistence of eosinophil inflammation in NP. Bolard et al.[23] found a positive correlation between IL-5 level and number of eosinophils in nasal fluid. Why did we not find the relationship like that? In our study, cytokine levels in nasal secretions were correlated with the eosinophil counts in nasal polyp tissue. Eosinophils can be released to the nasal fluid not only from the nasal polyps, but can be released from whole inflamed nasal mucosa. In our investigation, the counting of eosinophils was limited only on the nasal polyp tissue. When we analysed all of our patients, we did not find significant correlation of measured IL-5 levels with eosinophil number in NP samples. The group of patients with estimated allergy had a significantly higher number of eosinophils together with significantly elevated IL-5 concentration. Correlation coefficient of IL-5 concentration with eosinophil number in this allergic group of patients with NP was twenty times higher than in the non-allergic group ($r = 0.4$ *vs* $r = 0.02$), although statistical significance was not reached. Our results pointed out that IL-5 levels in nasal secretions could be connected to tissue eosinophil number only in allergic patients.

Unfortunately, we did not enumerate eosinophils and the IL-5 level in the same nasal secretion, nor did we have opportunities to immunehistologically trace IL-5 in the resected polyp tissue, which is a more precise way to establish objective insight into IL-5 function in NP. Furthermore, discrepancies between tissue eosinophil number and IL-5 level in nasal secretion could be due to local Th2 lymphocyte predominance, as they could be one of the major sources of produced IL-5. Due to the findings of significant correlation between IL-6 and eosinophil counts in NP with asthma group, together with elevated IL-5 and IL-10, there is a basis to speculate about Th2 predominance. Certainly, further investigations of local T lymphocyte presence and function are needed as proof.

Our results showed a positive correlation between IL-6 levels in nasal fluid and eosinophil counts in tissue of nasal polyps in both asthmatic and non-asthmatic NP patients. IL-6 is an important proinflammatory Th2 type cytokine involved in the induction of IgE synthesis, as well as in mast cell proliferation and maturation.[19] IL-6 also stimulates fibroblast proliferation and collagen synthesis.[19] Immunohistochemical staining and in situ hybridisation indicated that the main sources of IL-6 are macrophages, T cells, mast cells, and, especially, eosinophils and fibroblasts because of their autocrine activity.[19] Van Zele et al.[24] showed increased colonisation of NP by *Staphylococcus aureus* and presence of specific IgE directed against *Staphylococcus aureus* exotoxins in NP tissue. Rates of colonisation and IgE presence in NP tissue were increased in subjects with NP and comorbid asthma.[24] Hellings et al.[25] demonstrated that nasal application of *Staphylococcus aureus* exotoxin B is capable of aggravating experimental allergic rhinitis and asthma, paralleled with an increase in bronchial and systemic Th2 cytokine levels. Xu et al.[26] also found significantly increased levels of IL-6 in *Staphylococcus aureus* exotoxin B-stimulated nasal polyps.

A positive correlation between TNF-α level in nasal secretions and eosinophil counts in the NP tissue can be explained by several recently published findings. Eosinophil infiltration is regulated by numerous chemokines and adhesion molecules such as eotaxin, regulated on activation normal T cell expressed and secreted (RANTES), and vascular cell adhesion molecule (VCAM)-1.[27] To infiltrate sites of inflammation, eosinophils leave the bloodstream and pass through the endothelium in four steps, namely rolling, adhesion, transendothelial migration, and chemotaxis.[27] Adhesion molecules, such as VCAM-1, play an important role during adhesion to endothelial cells.[27] Experiments performed by Ohori et al.[27] demonstrated that TNF-α stimulation induces VCAM-1 protein production and mRNA expression in human nasal polyp fibroblasts. Epithelial and immunecompetent cells such as macrophages, mast cells and eosinophils produce TNF-α. These findings suggest that TNF-α increases VCAM-1 production in nasal fibroblasts and activates the transmigration of eosinophils, which induce further production of TNF-α and accelerate the accumulation of eosinophils in nasal polyps. Saji et al.[28] demonstrated that nasal polyp fibroblasts produced RANTES by stimulation with TNF-α and IL-1β. Therefore, results published by Yoshifuku et al.[29] showed that eotaxin secretion from fibroblasts was induced by stimulation with IL-4 and synergistically enhanced by simultaneous stimulation with TNF-α and IL-4.

IFN-γ is a Th1 cytokine which leads via macrophage activation to extensive inflammatory process that also enable the killing of intracellular pathogens.[1] It is an inhibitor of allergic responses through its capacity to inhibit the effects of IL-4 on B-cells and eosinophils.[1] A negative correlation between the concentration of IFN-γ in nasal fluid and eosinophil counts in nasal polyps may be explained by inhibitory effects of IFN-γ on the IL-4-induced selective influx of eosinophils and VCAM-1 expression on microvascular endothelium in NP tissue.[21] We found higher level of IFN-γ in nasal secretions in NP with asthma subjects than in NP patients probably because of higher level of inflammatory reaction in asthmatic patients.

Dhong et al.[30] recently found that eosinophil infiltrations were more prominent in asthmatic NP patients compared to non-asthmatic ones, which is in accordance with our results.

Conclusions

Our objective was to estimate possibilities to study the process and achievable therapies in NP patients at ways that differ than that in ordinary clinical and radiological findings. Therefore, we analysed dominant Th1 mediators (IL-2, IL-12, and IFN-γ), dominant Th2 mediators (IL-4, IL-5, IL-6, and IL-10), one crucial chemokine (IL-8) and inflammation mediators (IL-1β, TNF-α, and TNF-β). Our results showed that the presence of Th2 cytokines in NP is a prominent feature that relates to the increased eosinophilic inflammatory process. Our findings also suggest that upregulation of Th2 cytokines (IL-5, IL-6 and IL-10) is a more significant characteristic of NP in asthmatic than in non-asthmatic subjects. The eosinophil counts were higher in asthmatic patients' polyps compared with non-asthmatic ones, but these differences were not statistically significant. IL-6 and TNF-α levels in nasal fluid correlate with the eosinophil counts in NP tissue in non-asthmatic subjects, whilst in asthmatic patients, only IL-6 levels correlate positively and IFN-γ negatively with eosinophil numbers in tissue. These results showed that these patients with similar clinical findings had

significantly different mediator profiles in their nasal secretions, implying clear differences in pathogenesis of their NP. These findings give basis to move the balance from eosinophils as damage effectors to Th2 lymphocytes, marking them as a crucial target in NP therapy. Also, these results showed that evaluation of local immune reaction mediators in nasal secretion could be an accessible and valuable path in the monitoring of these patients, as well as a sensitive way to estimate new therapies of NP and to study the NP pathogenesis.

Conflict of Interest

The authors have no conflict of interest to declare.

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