Histol Histopathol (2015) 30: 1447-1454

http://www.hh.um.es

Histology and Histopathology

Cellular and Molecular Biology

Histopathological classification of refractory chronic rhinosinusitis with nasal polips

G. Mortuaire[1,2], X. Leroy[3], I. Gengler[1], D. Chevalier[1], L. Prin[2] and A. Picry[1]

[1]Otorhinolaryngology - Head and Neck Surgery Department, [2]EA 2686- French Eosinophil Network and [3]Pathology Department, University Hospital of Lille, Lille, France

Summary. Objective: To delineate the histopathological characteristics of nasal mucosa in refractory chronic rhinosinusitis with nasal polyps (CRSwNP) in order to demonstrate subtypes of nasal polyps and their potential relation with lower airway comorbidity. Study Design: Clinical- and pathological-based cross-sectional study. Methods: Nasal polyp specimens were prospectively collected from patients with refractory CRSwNP referred to our institution for endoscopic sinus surgery. Oral and topical steroids were stopped 1 month before surgery. The pathological analysis was conducted by 2 independent reviewers with light microscopy on Hematoxylin-Eosin-Saffron stained slides. Each observer fulfilled a standardized protocol with cell count and stromal characterization on the most representative field. Mean grading scores were established. Morphological aspects were compared with the cell distribution and the clinical conditions. Results: Among 36 patients, three subtypes of nasal polyps were depicted: eosinophilic edematous (64%), fibrous (9%) and intermediate with mixed edematous and collagen stromal structure (27%). Basement membrane thickening and seromucous gland hyperplasia were observed in the fibrosis sub-type ($p<0.03$). Eosinophilic mucosal infiltrate was significantly increased ($p=0.026$) in patients with concomitant pulmonary disease (n=21). Nasal polyp distribution was not influenced by asthma, allergy, previous surgery and smoking. Conclusion: Our 3-subtype classification of refractory CRSwNP in Caucasian population shows a predominant edematous structure whatever the clinical conditions may have been. Eosinophilia as a major factor of adaptive immune response in nasal inflammation is a feature of concomitant pulmonary disease. Further studies concerning mucosal remodelling and outcome assessment after sinus surgery are required to evaluate the impact of our classification on a daily basis.

Key words: Nasal polyps, Eosinophil, Histological classification, Asthma

Offprint requests to: Geoffrey Mortuaire, Service d'ORL de chirurgie cervico-faciale, Hôpital Huriez, CHRU Lille, 59000 France. e-mail: geoffrey.mortuaire@chru-lille.fr

DOI: 10.4670/HH-11-632

Introduction

Chronic rhinosinusitis with nasal polyps (CRSwNP) is an inflammatory disease of the nose and paranasal sinuses mucosa with symptoms lasting more than 12 weeks (Fokkens et al., 2012). With a prevalence from 2 to 4% in the general population, CRSwNP has a significant impact on the quality of life and is characterized by a variable response to corticosteroids, with potential recurrences leading to surgery (Klossek et al., 2005; Mortuaire et al., 2010; Fokkens et al., 2012). The impact of allergy is controversial as allergy incidences in CRSwNP and general population are equal (Fokkens et al., 2012). Asthma or lower airway hyperreactivity observed in 42% of cases could delineate a specific group of patients with a more severe disease (Klossek et al., 2005; Fokkens et al., 2012).

Although the pathogenesis of CRSwNP is still poorly understood, a massive eosinophilic mucosal

infiltrate is considered to be one of the histologic hallmarks of this disease in Caucasian population (Bachert et al., 2001). It is still unclear whether a more precise histopathological categorization of nasal polyps, regarding the clinical status of patients, could have implications for treatment and outcomes. Several studies have already been published about the morphological characterization of chronic nasal mucosa inflammation (Berger et al., 2002; Couto et al., 2008; Soler et al., 2009; Payne et al., 2011). A predominant edematous and eosinophilic structure was often described (Couto et al., 2008). However, the results can be misinterpreted as patients with different diseases (CRS with or without polyps, allergic fungal sinusitis) were included (Berger et al., 2002; Payne et al., 2011). In a recent study, histological analyses were performed on nasal polyps collected during sinus surgery in patients regardless of ongoing oral corticosteroid or antibiotic therapies (Soler et al., 2009). At last, those studies did not compare histopathological phenotypes with relevant clinical data like asthma or allergy.

Herein, we described the detailed histopathological findings from a cross-sectional study of patients undergoing endoscopic sinus surgery for refractory CRSwNP, without any previous drug therapy. Using strict observation rules through optic microscopy, we proposed to classify these nasal polyps exhaustively in accordance with morphological criteria encountered in the literature.

Materials and methods

Subjects

A prospective study was conducted including 36 patients with refractory CRSwNP. The diagnosis was established according to the EPOS criteria (Fokkens et al., 2012). The patients were addressed in our department for persistent symptoms despite optimal medical treatment (at least 3 courses of 7-day oral corticosteroid and maximum dose of topical nasal corticosteroid spray). Age, gender and history of prior sinus surgery were obtained from clinical record. Concomitant asthma or lower airway hyperreactivity (diagnosed respectively by spirometric measurement of forced expired volume in one second/vital capacity ratio or by metacholine bronchial challenge test), allergy (diagnosed by an allergy focused history in conjunction with skin prick testing) and history of acetylsalicylic acid (ASA) intolerance were systematically assessed. No case of smoker chronic obstructive pulmonary disease was recorded. Adult patients (≥18 years old) were enrolled after maximum medical management failure and surgery indication. The burden of sinonasal symptoms was measured by visual analogical scale (VAS) with 0=no symptom. Patients with immunodeficiency, autoimmune disease and/or cystic fibrosis were excluded. A procedure of endoscopic sinus surgery was scheduled for each patient.

To improve the homogeneity of the population, any kind of medical therapy (oral and topical nasal corticosteroid), except nasal douching, was stopped at least one month before surgery.

All patients provided written informed consent before participation. This study was part of an institutional protocol on eosinophilic syndromes for which the sample collection was approved by local ethics review board (registration number: 2009-A00314-53). The protocol was also agreed according to institutional requirements for routine histopathological studies. The day before surgery, a computed tomography (CT) scan was performed to evaluate sinus opacifications with the Lund-Mackay score (maximum total score: 24) (Lund and Mackay, 1993). The nasal polyp size was endoscopically measured using a 30-degree rigid nasal fiberoptic scope according to the Lildholdt grading system (0-absence of nasal polyps, 1-nasal polyps in middle meatus only, 2-nasal polyps beyond the middle meatus but not reaching the inferior edge of the inferior turbinate, 3-nasal polyps completely obstructing the nose) (Lidholdt et al., 1995).

Tissue preparation and staining

Nasal polyps were obtained at the beginning of the surgical procedure. Through endoscopic view, one nasal polyp was gently removed from the middle meatus for each patient and fixed in 4% paraformaldehyde overnight at 4°C. Standard tissue dehydration in graded alcohol solutions and embedment in paraffin blocks were undergone with Shandon® Excelsior ES Tissue Processor (Thermo Fisher Scientific, Waltham, MA). Thereafter, blocks were cut into 3-μm thick sections with RM2155® rotary microtome (Leica Microsystems, Wetzlar, Germany) and put on Superfrost® Plus Micro Slides (VWR International, Radnor, PA). Subsequently, samples were deparaffinized, cleared in xylene and rehydrated through graded series of alcohol. Hematoxylin-Eosin-Saffron (HES) staining was obtained with the Tissue-Tek® Prisma®/Film® Automated Slide Stainer and Coverslipper (Sakura Finetechnical Co, Tokyo, Japan).

Histologic scoring

A microscopic review was performed by two trained authors (G.M. and A.P.) using a binocular Leica DM4000B® microscope (Leica Microsystems, Wetzlar, Germany). Panels of 4 slides from sections of the distal part of the nasal polyp were randomly selected for subsequent observations. The pathologic review was done in a blinded fashion in regard to all clinical data. Two slides were assigned to each observer. The different items evaluated were chosen according to the criteria used in pathologic practice and in the literature to assess nasal mucosa (Table 1). At 400× power, cell infiltrates were measured on a field of 250 μm×250 μm, yielding an area of approximately 0.13 mm^2. Each observer used

a cell-counter to define the mean proportions of cells population by high-power field (percentage of cell types for a count of 100 cells) on the most representative field of the 2 assigned slides. Correlations between the two observers' counts were checked before setting an average cell count for each patient. Epithelial and stromal structures were assessed through 100× to 400× magnifications. Specific grading scales were used for each item (Table 1). In case of grading discrepancy between the two observers, an agreement was set by a third certified author (X.L.). Maximal basement membrane (BM) thickening was measured with the most optimal transverse mucosal histologic orientation and recorded as <5μm, 5-15 μm and >15μm with a graduated reticle mounted within one of the eyepiece objectives.

The mean percentage difference of the two major cell components (eosinophil and T/plasma cell) and the mean grading score difference in terms of stromal structure for edema and fibrosis were used in a matrix approach. The polyp was considered as eosinophilic if a positive difference between eosinophil and T/plasma cell counts was observed.

Statistical analyses

Data were input into Microsoft Excel® and a statistical analysis was performed with SPSS® v 15.0 statistical software (SPSS Inc., Chicago, IL, USA). Means, standard deviations (SD) or ranges were reported for descriptive statistics. The Chi-squared test was used for comparison of demographic data. The non-parametric Kruskal-Wallis test was used to compare the means of non-paired samples. If a statistically significant difference among the groups was found, a post hoc analysis was followed by means of Mann-Whitney test. Correlation analyses for cell counts were conducted with the Pearson test (using *rho* as correlation coefficient). A two-tailed p-value ≤0.05 was considered statistically significant.

Table 1. Overview of histologic inflammatory markers.

Inflammatory markers	Measurement
Cellular markers (× 400 magnification)	
Eosinophils	Percentage (%) for 100 cells count
T and plasma cells	Percentage (%) for 100 cells count
Macrophages	Percentage (%) for 100 cells count
Neutrophils	Percentage (%) for 100 cells count
Mast cells	Percentage (%) for 100 cells count
Basophils	Percentage (%) for 100 cells count
Mucosal structure (× 100 and × 400 magnifications)	
Epithelial erosion	Grade 0,1,2,3 a
Epithelial metaplasia	Grade 0,1,2,3 a
Epithelial hyperplasia	Grade 0,1,2,3 a
Globet cell hyperplasia	Yes/No
Basement membrane thickening	<5μm, 5-15 μm, >15 μm
Eosinophil infiltrate*	Yes/No
Lamina propria structure (× 400 magnification)	
Stromal edema	Grade 0,1,2,3 b
Fibrosis	Grade 0,1,2,3 b
Seromucous gland abundance	Grade 0,1,2,3 b
Seromucous gland hyperplasia	Yes/No

The analyses were performed on HES stained sections by 2 independent observers. [a] 0=not present, 1=focal (mild), 2=patchy (moderate), 3=extensive (marked). [b] 0=not present, 1=mild, 2=moderate, 3=marked. * presence of eosinophils within the epithelial layer.

Results

From December 2013 to May 2014, 36 consecutive patients were included in our study (Table 2). As a major factor of disease outcome (Klossek et al., 2005; Fokkens et al., 2012), the respiratory status was used to delineate 2 groups of patients (patients with asthma or lower airway hyperreactivity disease (n=21)). Patients with or without lower airway disease were comparable for age, gender, duration of CRSwNP before surgery in our institution and tobacco use. In terms of disease severity, mean VAS, polyp score and CT scan opacification staging were not different in both patient groups.

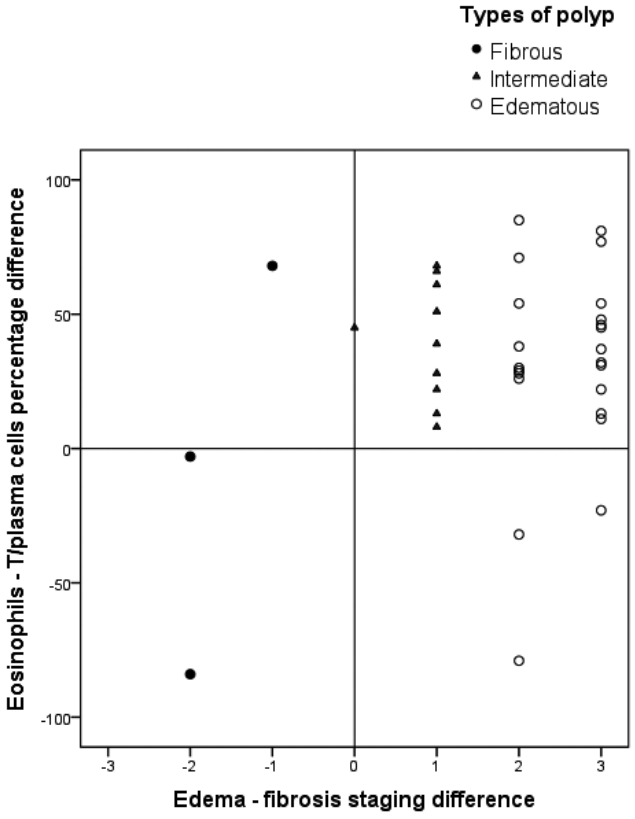

Fig. 1. Nasal polyps classification according to eosinophil infiltrate and stromal framework. The eosinophil infiltrate and the relative edema/fibrosis grading difference were simultaneously assessed. Three types of nasal polyp were observed: edematous (n=23), intermediate (n=10) and fibrous (n=3).

However, allergy was more frequent in patients with lower airway disease (p=0.027). Prior surgery was observed in 33% of these patients. This rate did not reach a significant statistical level.

Pearson correlation test was used to evaluate inter-observer cell count variability. Rho correlation coefficient ranged from 0.73 to 0.93 for the different cell subtypes (p≤0.001). Eosinophilic infiltrate was mainly observed in our population (mean percentage of 62%, SD: 20%). The mean percentages of T and plasma cells, macrophages and other cells were respectively 31%, 4.8% and 2.6%. The presence of eosinophils within the epithelial layer, often described in CRSwNP, was not associated with a more substantial eosinophilic infiltrate in the stroma.

Three sub-types of nasal polyps were defined using the matrix approach (Fig. 1). We noticed a prominent group of patients (n=23) (64%) with eosinophilic edematous nasal polyps (called "edematous"). A second group (n=10) (27%) was characterized with eosinophilic infiltrate, concomitant edema and collagen structure (called "intermediate"). A third group was less prevalent (n=3) and showed a predominant fibrous structure with variable cell infiltrates (called "fibrous"). Edematous nasal polyps were characterized by broad intercellular spaces and eosinophilic gathering underneath the epithelial layer (Fig. 2A). Fibrous nasal polyps showed diffuse cell infiltrate with numerous seromucous glands and ductal structure in the stroma (Fig. 2B). Intermediate nasal polyps could be described as edematous polyps with a significant fibrous component (Fig. 2C). In 3 cases of edematous nasal polyps, we noticed a major T/plasma cell infiltrate. One of those 3 patients was previously followed for thrombocytopenia. No specific past history was noticed for the 2 other patients. To corroborate our classification, we compared the percentage of each cell between the 3 types of nasal polyp (Fig. 3). Eosinophils were significantly enhanced in edematous and intermediate nasal polyps versus

Table 2. Demographic and clinical characteristics of the population.

	Asthma or airway hyper-reactivity	W/o lower airway disease	p-value
Population (n=36) (pts)	21	15	ns
Mean age (years)	50	48.8	ns
Sex ratio	15M/6F	13M/2F	ns
Duration of CRSwNP (years)	9.4	9.2	ns
Prior surgery (%)	33	6.6	ns
Allergy (%)	62	26.6	0.049*
ASA intolerance (pts)	3	-	-
Tobacco use (n=6) (pts)	2	4	ns
Mean polyp size score (/3)	2.47	2.46	ns
Mean CT scan score (/24)	18.2	17.7	ns
Mean VAS score (/10)	8.1	7.2	ns

Apart from allergy (p=0.049), the 2 groups of patients (with or without lower airway disease) were comparable in terms of demographic characteristics and disease severity. (*Chi-squared test).

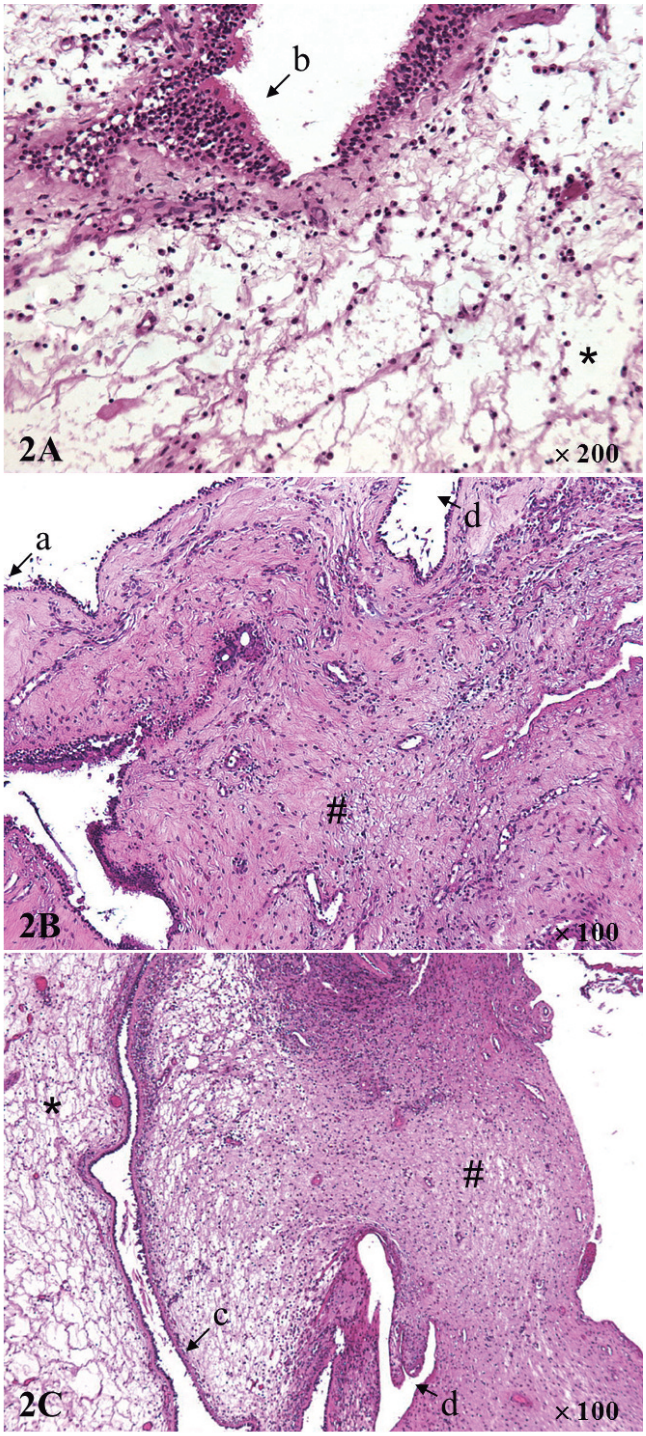

Fig. 2. Microscopical depictions of the nasal polyp classification (×100 and ×200). Edematous nasal polyp was described as a massive stromal eosinophil infiltrate with edema (*) **(A)**. Fibrous nasal polyp was characterized by collagen structure (#) **(B)**. Intermediate nasal polyp was eosinophilic with a mixed composition of the stroma **(C)**. Different features were evenly observed in the 3 types of nasal polyp: complete epithelium desquamation (a) or hyperplasia (b), basement membrane thickening (c), submucosal seromucous gland hyperplasia (d).

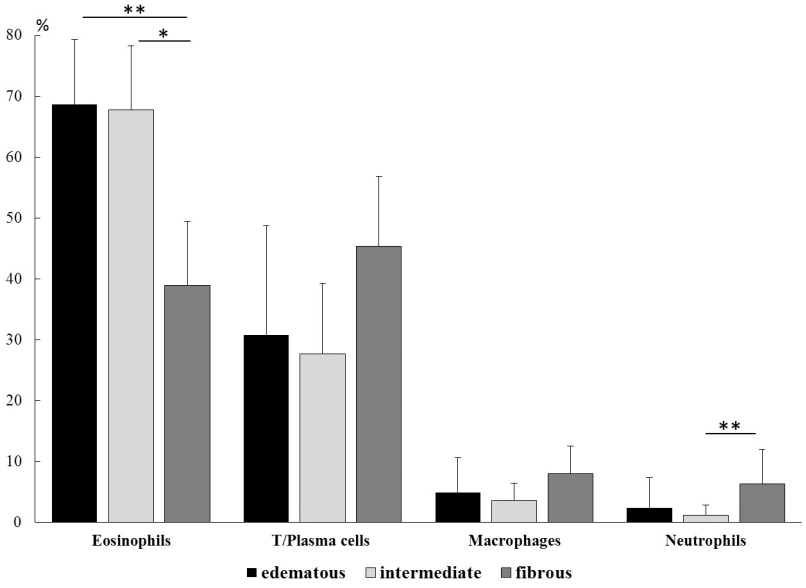

Fig. 3. Cells distribution in the 3-subtype nasal polyp classification. Eosinophils were significantly more present in edematous and intermediate nasal polyps (** p<0.02, *p=0.04). Neutrophils were observed more frequently in fibrous nasal polyps (** p <0.02) (Mann Whitney test). Bar values and error bars represent respectively means and standard errors of the mean (SEM).

4A

Histopathological features	p-value *(Kruskal Wallis test)*
Epithelial erosion	ns
Epithelial hyperplasia	ns
Epithelial metaplasia	ns
Basal membrane thickness	**0,041**
Seromucous glands abundance	**0,002**

4B

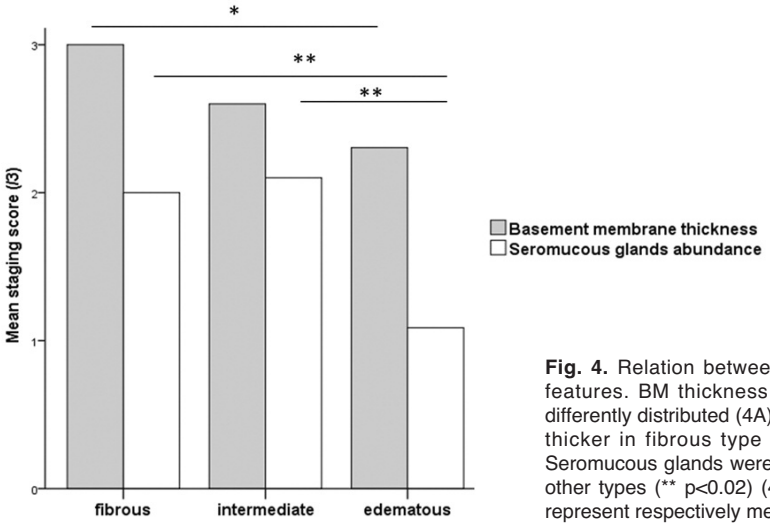

Fig. 4. Relation between nasal polyp classification and histopathological features. BM thickness and seromucous gland abundance scores were differently distributed (4A). Post hoc analysis showed that BM was significantly thicker in fibrous type in comparison with edematous type (* p<0.03). Seromucous glands were scarce in edematous type in comparison with the 2 other types (** p<0.02) (4B) (Mann Whitney test). Bar values and error bars represent respectively means and SEM.

fibrous nasal polyps (respectively p<0.02 and p=0.04). Neutrophils were enhanced in fibrous nasal polyps in comparison with the intermediate sub-type (p<0.02).

The other cell types were evenly distributed or poorly observed.

The different criteria of our histo-morphological review were compared within the 3 subtypes of nasal polyps (Fig. 4A). By grading scale comparison, we showed that BM thickening was significantly increased in fibrous nasal polyps (p<0.03) (Fig. 4B). Seromucous glands were more abundant in fibrous and intermediate nasal polyps (p<0.02) (Fig. 4B). Goblet cell hyperplasia was evenly observed in the 3 subtypes of nasal polyps.

Histological findings were compared with the clinical status of CRSwNP patients. In patients with asthma or airway hyperreactivity, eosinophilic infiltrate was significantly increased (p=0.026). Inversely, the rate of T/plasma cells was increased in patients without concomitant respiratory disease (p=0.043). Noteworthy, the other types of cells were scarce, with no difference in terms of distribution (Fig. 5). The cell composition was not influenced by the other demographic (age, gender) or clinical data (allergy, prior surgery, disease severity or duration, smoking). The 3 types of nasal polyps were equally distributed in patients with or without airway disease. The same results were observed for gender, allergy and smoking. Neither time to surgery in our institution nor past history of sinus surgery modified nasal polyp distribution. Meanwhile, metaplastic changes into squamous epithelium score were higher in patients with pulmonary disease (p=0.02). The impact of ASA intolerance could not be evaluated as only 3 patients were concerned.

Discussion

With current advances in the immuno-phenotyping of tissues, the systematic pathological review of nasal polyps could be considered as basic assessment. Still, published studies about nasal polyp categorization are often blurring as they included different types of patients with different types of medical management before sample collection. In an attempt to give a new insight with clinical perspectives, we focused our study on a homogenous group of patients with refractory disease. CRSwNP was strictly diagnosed according to the EPOS criteria (Fokkens et al., 2012). Topical and/or oral medical therapies were stopped one month before surgery to avoid inflammation dimming. To improve the quality of our samples collection, nasal polyps were gently removed by the same surgeon (G.M.) and histologic analyses were carried out caudally off the polyp attachment. Thereby, we avoided to harvest bony structures and thick adjacent mucosa. An optimal assessment of the polyp framework was achieved with a standardized protocol and a double evaluation by two independent reviewers with no awareness of the clinical data. Small inter-rater variability for cell counting was confirmed by Pearson correlation test. In an attempt to reduce the heterogeneity in the results, means for counting cells and grading scores were established by each observer on the most representative field of 2 slides. Likewise, a third trained observer was called for grading scale measurement in case of disagreement between the first 2 observers.

With this approach, we were able to delineate 3 types of nasal polyps: edematous, fibrous and

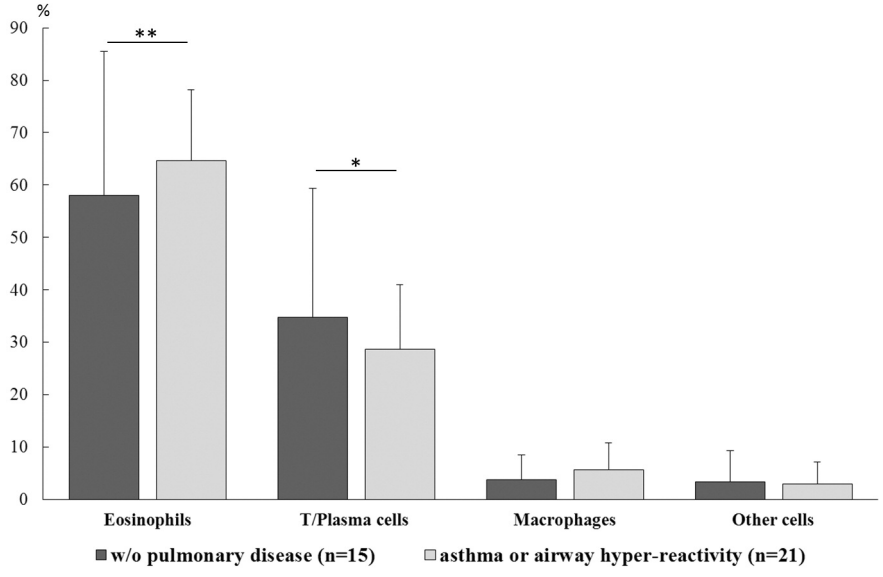

Fig. 5. Stromal cell distribution in nasal polyps according to lower airway status. We showed that eosinophils were more abundant in patients with asthma or lower airway hyper-reactivity (** p=0.026) whereas T and Plasma cells were inversely more frequent in patients without lower airway disease (* p=0.043) (Mann Whitney test). Bar values and error bars represent respectively means and SEM.

intermediate. A major edematous component was originally described in the literature, but with little concern about the influence of previous therapies on the histology of nasal polyps (Tos and Mogensen, 1977; Davidsson and Hellquist, 1993). Our results are in accordance with Couto et al. (2008) study. In 89 patients with sinonasal inflammation, the authors described edematous nasal polyps in 73% of cases and fibro-inflammatory nasal polyps in 18% of cases. Herein we delineate a third type of nasal polyp called intermediate. So far, the intermediate type was not reported in previous CRS histopathological studies. Given that the different histological features of nasal polyps may be found in the same sample, the authors usually preferred to describe the most relevant structure observed (Couto et al., 2008). We chose to take heed of this mixed structure in our classification to reduce subjectivity in the histological assessment. Basement membrane thickening and seromucinous gland abundance were displayed mostly in nasal polyps with a fibrous component. Basal membrane thickening indicates long-lasting inflammation and is considered a marker of airway remodelling (Watelet et al., 2004). Berger et al. (2002) showed substantial hyperplasia of submucosal glandular structure in non-edematous inflammed tissue of 29 patients with CRS. Similar results were observed by Payne et al. (2011). We assume that major remodelling processes observed in patients with fibrotic nasal polyps could lead to a thicker mucosal structure with excretory glandular ducts obstruction and seromucous gland hyperplasia. It is worth noticing that intermediate and fibrous nasal polyps were observed in patients both with and without prior surgery. Thus, previous mucosal resection does not necessarily imply wound healing processes with thicker mucosa. Nasal polyps with stromal atypia described by Couto et al. (2008) were not observed in our patients.

Eosinophilic stromal infiltrate predominantly observed in our patients is typical of CRSwNP in Caucasian population (Fokkens et al., 2012). With a reliable quantification technique of eosinophil counting, Bhattacharyya et al. (2001) showed relatively strong correlations between CT scan stage and tissue eosinophilia in CRS. Although eosinophilia was associated with edema in our nasal polyp classification, neither CT scan score nor nasal polyp size were correlated with eosinophil count (data not shown). As our population was composed of patients with a refractory disease, CT scan and polyp scores were high for all of them (Table 2). This bias could account for the lack of correlation between eosinophilia and the staging systems. Further studies revealed that plasma cells were also abundant in nasal polyps, indicating an elevated production of immunoglobulins (Polzehl et al., 2006). We showed that T cells and plasma cells were the second most common type of cell population in our study. These results corroborate the immunopathological hypothesis of an adaptive immune response to local allergens in nasal polyps. This response leads to immunoglobulins E and A release by B cells, pro-inflammatory cytokine production by T and epithelial cells and eosinophil recruitment (Bachert et al., 2001). In 3 cases of edematous polyp, we observed a prevailing T/plasma cell population without obvious explanation. One of them was followed for a concomitant hematopoietic disorder. The impact of this comorbidity is difficult to explain as the 2 other patients with rich T/plasma cell infiltrate did not show any specific disorder. Intensive immune-regulatory processes counteracting eosinophil inflammation could be involved in those 3 patients.

As concomitant lower airway disease (asthma or airway hyperreactivity), associated with CRSwNP, is a major factor of disease severity and of poor outcomes in steroid resistant patients (Mendelsohn et al., 2011; Zhang et al., 2011), we decided to focus our study on potential relations between the respiratory status of patients and histopathological findings. Like in previous studies, we showed that eosinophil infiltration was more prominent in the asthmatic compared to the non-asthmatic group (Dhong et al., 2005; Soler et al., 2009). Eosinophil mucosal recruitment and survival rely on mucosal release of cytokines (i.e. interleukin-5) involved in the Th2-biaised immune adaptive response (Simon et al., 1997). As blood and nasal interleukin-5 levels are increased in asthmatic patients (Bachert et al., 2010), pronounced mucosal eosinophilic infiltrate could be considered as a marker of potential bronchial disease in CRSwNP patients. Even though we focused our study on patients with refractory CRSwNP and who underwent surgery, this specific relation between airway disease and eosinophilia underlines the impact of this predominant inflammatory process and the need for specific immune therapies for this patient sub-type. Several studies revealed that the accumulation of inflammatory cells could be different, depending on the geographical origin. Zhang et al. (2008) showed in Asian patients a neutrophil predominance with a different T-cell pattern. Our findings did not show major neutrophilic infiltrate in Caucasian population. Distinct disease parameters and therapeutic approaches are needed for different types of CRSwNP inflammations.

Concerning the nasal polyp classification, we did not find a specific histo-pathological pattern for patients with lower airway disease. As described by Berger et al. (2002), asthmatic patients were equally distributed in each nasal polyp subtype. Clinical biases were excluded in our study, as groups with or without lower airway disease were comparable in terms of demographic data, previous surgery or disease severity (Table 2). Allergy per se, more frequently observed in asthmatic patients, has already been described as poorly involved in CRSwNP pathophysiology (Kern, 1993; Fokkens et al., 2012). Like in our study, Soler et al. (2009) showed that allergy was not associated with mucosal eosinophilia. Epithelial metaplasia was higher for asthmatic patients in our analysis. We assume that epithelial damage caused by inflammatory mediators could induce this cell proliferation via epithelial repair processes (Coste et al.,

1996). At last, smoking was not involved in any significant change in the histopathological characteristics of nasal polyps, as previously described (Kule et al., 2014).

Conclusion

With a rigorous histologic methodology and a strict selection of representative refractory CRSwNP patients free from steroid therapy before tissue collection, we have been able to delineate a 3-subtype histopathological classification of nasal polyps. Edematous polyp with eosinophilic infiltrate is the most prevailing pattern in the Caucasian population. No subtype of nasal polyp was predictive of a specific clinical status. Meanwhile, we emphasized that patients with concomitant asthma or lower airway hyperreactivity showed more eosinophils in the stromal compartment, underlining the role of mucosal adaptive immune response in those patients.

Subsequent studies are required to further explore the potential impact of our nasal polyp classification on surgical outcomes in terms of tissue remodelling and symptom relief.

Acknowledgements. The help of Dr Juliette Beaujot and Dr Agnès Wacrenier, Pathology Department, University Hospital of Lille, France, for technical support and study design is gratefully acknowledged.

References

Bachert C., Gevaert P., Holtappels G., Johansson S.G. and Van Cauwenberge P. (2001). Total and specific IgE in nasal polyps is related to local eosinophilic inflammation. J. Allergy Clin. Immunol. 107, 607-614.

Bachert C., Zhang N., Holtappels G., De Lobel L., Van Cauwenberge P., Liu S., Lin P., Bousquet J. and Van Steen K. (2010). Presence of IL-5 protein and IgE antibodies to staphylococcal enterotoxins in nasal polyps is associated with comorbid asthma. J. Allergy Clin. Immunol. 126, 962-968.

Berger G., Kattan A., Bernheim J. and Ophir D. (2002). Polypoid mucosa with eosinophilia and glandular hyperplasia in chronic sinusitis: a histopathological and immunohistochemical study. Laryngoscope 112, 738-745.

Bhattacharyya N., Vyas D.K., Fechner F.P., Gliklich R.E. and Metson R. (2001). Tissue eosinophilia in chronic sinusitis: quantification techniques. Arch. Otolaryngol. Head Neck Surg. 127, 1102-1105.

Coste A. , Rateau J.G., Roudot-Thoraval F., Chapelin C., Gilain L., Poron F., Peynegre R., Bernaudin J.F. and Escudier E. (1996). Increased epithelial cell proliferation in nasal polyps. Arch. Otolaryngol. Head Neck Surg. 122, 432-436.

Couto L.G., Fernades A.M., Brandão D.F., Santi Neto D., Valera F.C. and Anselmo-Lima W.T. (2008). Histological aspects of rhinosinusal polyps. Braz. J. Otorhinolaryngol. 74, 207-212.

Davidsson A. and Hellquist H.B. (1993). The so-called 'allergic' nasal polyp. ORL J. Otorhinolaryngol. Relat. Spec. 55, 30-35.

Dhong H.J., Kim H.Y. and Cho D.Y. (2005). Histopathologic characteristics of chronic sinusitis with bronchial asthma. Acta Otolaryngol. 125, 169-176.

Fokkens W.J., Lund V.J., Mullol J., Bachert C., Alobid I., Baroody F., Cohen N., Cervin A., Douglas R., Gevaert P., Georgalas C., Goossens H., Harvey R., Hellings P., Hopkins C., Jones N., Joos G., Kalogjera L., Kern B., Kowalski M., Price D., Riechelmann H., Schlosser R., Senior B., Thomas M., Toskala E., Voegels R., Wang de Y. and Wormald P.J. (2012). EPOS 2012: European position paper on rhinosinusitis and nasal polyps 2012. A summary for otorhinolaryngologists. Rhinology. 50, 1-12.

Kern R.(1993). Allergy: a constant factor in the etiology of so-called mucous nasal polyps. J. Allergy 4, 483.

Klossek J.M., Neukirch F., Pribil C., Jankowski R., Serrano E., Chanal I. and El Hasnaoui A. (2005). Prevalence of nasal polyposis in France: A cross sectional, case-control study. Allergy 60, 233-237.

Kule Z.G., Habesoglu T.E., Somay A., Deveci H.S., Kule M. and Gursel A.O. (2014). Histopathological characteristics of nasal polyps in smokers and non-smokers. J. Craniofac. Surg. 25, 946-949.

Lildholdt T., Rundcrantz H. and Lindqvist N. (1995). Efficacy of topical corticosteroid powder for nasal polyps: a double-blind, placebo-controlled study of budesonide. Clin. Otolaryngol. Allied Sci. 20, 26-30.

Lund V.J. and Mackay I.S. (1993). Staging in rhinosinusitus. Rhinology 31, 183-184.

Mendelsohn D., Jeremic G., Wright E.D. and Rotenberg B.W. (2011). Revision rates after endoscopic sinus surgery: a recurrence analysis. Ann. Otol. Rhinol. Laryngol. 120, 162-166.

Mortuaire G., Vandeville S. and Chevalier D. (2010). Psychometric evaluation of the SinoNasal Outcome Test-16 for quality of life in chronic rhinosinusitis with nasal polyps. Eur. Ann. Otorhinolaryngol. Head Neck Dis. 127, 91-96.

Payne S.C., Early S.B., Huyett P., Han J.K., Borish L. and Steinke J.W. (2011). Evidence for distinct histologic profile of nasal polyps with and without eosinophilia. Laryngoscope 121, 2262-2267.

Polzehl D., Moeller P., Riechelmann H. and Perner S. (2006). Distinct features of chronic rhinosinusitis with and without nasal polyps. Allergy 61, 1275-1279.

Simon H.U., Yousefi S., Schranz C., Schapowal A., Bachert C. and Blaser K. (1997). Direct demonstration of delayed eosinophil apoptosis as a mechanism causing tissue eosinophilia. J. Immunol. 158, 3902-3908.

Soler Z.M., Sauer D.A., Mace J. and Smith T.L. (2009). Relationship between clinical measures and histopathologic findings in chronic rhinosinusitis. Otolaryngol. Head Neck Surg. 141, 454-461.

Tos M. and Mogensen C.(1997). Pathogenesis of nasal polyps. Rhinology 15, 87-95.

Watelet J.B., Bachert C., Claeys C. and Van Cauwenberge P. (2004). Matrix metalloproteinases MMP-7, MMP-9 and their tissue inhibitor TIMP-1: expression in chronic sinusitis vs nasal polyposis. Allergy. 59, 54-60.

Zhang N., Van Zele T., Perez-Novo C., Van Bruaene N., Holtappels G., DeRuyck N., Van Cauwenberge P. and Bachert C. (2008). Different types of T-effector cells orchestrate mucosal inflammation in chronic sinus disease. J. Allergy Clin. Immunol. 122, 961-968.

Zhang Z., Linkin D.R., Finkelman B.S., O'Malley B.W. Jr, Thaler E.R., Doghramji L., Kennedy D.W., Cohen N.A. and Palmer J.N. (2011). Asthma and biofilm-forming bacteria are independently associated with revision sinus surgeries for chronic rhinosinusitis. J. Allergy Clin. Immunol. 128, 221-223.

Accepted May 19, 2015