Histopathologic difference between sinonasal mucosa and polyp tissue for diagnosing eosinophilic chronic rhinosinusitis

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การศึกษาเปรียบเทียบเซลล์อักเสบระหว่างเยื่อบุโพรงไซนัสและริดสีดวงจมูก ในการวินิจฉัยโรคไซนัสอักเสบชนิดอีโอสิโนฟิล

นางสาววรรณรวี ไทยตระกูล

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วรรณรวี ไทยตระกูล : การศึกษาเปรียบเทียบเซลล์อักเสบระหว่างเยื่อบุโพรงไซนัสและริดสีดวงจมูกในการวินิจฉัยโรคไซนัสอักเสบชนิดอีโอสิโนฟิล (Histopathologic difference between sinonasal mucosa and polyp tissue for diagnosing eosinophilic chronic rhinosinusitis) อ.ที่ปรึกษาวิทยานิพนธ์หลัก: ผศ. ดร. นพ.กรเกียรติ์ สนิทวงศ์, อ.ที่ปรึกษาวิทยานิพนธ์ร่วม: รศ. พญ.สุพินดา ชูสกุล, รศ. นพ.ทรงกลด เอี่ยมจตุรภัทร, 47 หน้า.

การศึกษานี้มีวัตถุประสงค์เพื่อที่จะหาตำแหน่งที่เหมาะสมในการส่งตรวจทางพยาธิวิทยาในการวินิจฉัยโรคไซนัสอักเสบชนิดอีโอซิโนฟิล โดยรวบรวมผู้ป่วยไซนัสอักเสบเรื้อรังที่มีริดสีดวงจมูก เก็บสิ่งส่งตรวจจาก ริดสีดวงจมูกบริเวณยอด ริดสีดวงจมูกบริเวณขั้ว ขูดเซลล์จากยอดริดสีดวงจมูก และเยื่อบุไซนัสส่งตรวจทางพยาธิวิทยาเพื่อเปรียบเทียบว่าบริเวณใดสามารถวินิจฉัยไซนัสอักเสบชนิดอีโอสิโนฟิลได้ดีที่สุด ผลการศึกษาพบว่าผู้ป่วยไซนัสอักเสบเรื้อรังที่มีริดสีดวงจมูกจำนวน 30 ราย พบว่าผู้ป่วย 16 ราย (53.3%) ริดสีดวงจมูกบริเวณยอด บริเวณขั้วและเยื่อบุไซนัสให้ผลการวินิจฉัยโรคไซนัสอักเสบชนิดอีโอสิโนฟิลที่คล้ายคลึงกัน ค่ามัธยฐานของเซลล์อีโอสิโนฟิลบริเวณยอดริดสีดวงจมูก (84, IQR:34-194) และบริเวณขั้วริดสีดวงจมูก (96, IQR:80-320) มากกว่าบริเวณเยื่อบุไซนัส (21, IQR:10-220), p=0.04 ความไวในการวินิจฉัยโรคไซนัสอักเสบชนิดอีโอสิโนฟิลของริดสีดวงจมูกบริเวณยอดเท่ากับ 100% (95%CI:47.8-100) ริดสีดวงจมูกส่วนขั้วเท่ากับ 60% (95%CI:14.7-94.7) เยื่อบุไซนัสเท่ากับ 80% (95%CI:28.4-99.5) พบความสัมพันธ์ระหว่างโรคหอบหืดในผู้ป่วยกับปริมาณเซลล์อักเสบชนิดอีโอสิโนฟิลในเนื้อเยื่อบริเวณขั้วริดสีดวงจมูก (p=0.05) และเยื่อบุไซนัส (p=0.04) แต่ไม่พบความสัมพันธ์กับบริเวณยอดริดสีดวงจมูก (p=0.21) ไม่พบความสัมพันธ์ที่มีนัยสำคัญทางสถิติของปริมาณเซลล์อีโอสิโนฟิลในเนื้อเยื่อกับปริมาณเซลล์อีโอสิโนฟิลในซีรัม และเมือกอักเสบอีโอสิโนฟิล พบความสัมพันธ์ระหว่างปริมาณเซลล์อักเสบชนิดอีโอสิโนฟิลจากการขูดเซลล์ริดสีดวงจมูกกับปริมาณเซลล์อักเสบชนิดอีโอสิโนฟิลในเนื้อเยื่อเพียงบริเวณเดียวคือเยื่อบุไซนัส ดังนั้นความหนาแน่นของเซลล์อักเสบชนิดอีโอสิโนฟิลที่ริดสีดวงจมูกมากกว่าที่เยื่อบุไซนัส บริเวณยอดริดสีดวงจมูกมีความไวในการวินิจฉัยโรคไซนัสอักเสบชนิดอีโอสิโนฟิลมากกว่าบริเวณอื่น บริเวณขั้วริดสีดวงจมูกและเยื่อบุไซนัสมีความสัมพันธ์กับอุบัติการณ์ของโรคหอบหืดในผู้ป่วยโรคไซนัสอักเสบเรื้อรังที่มีริดสีดวงจมูก รวมทั้งปริมาณเซลล์อักเสบชนิดอีโอสิโนฟิลจากการขูดเซลล์ริดสีดวงจมูกมีความสัมพันธ์กับปริมาณเซลล์อักเสบชนิดอีโอสิโนฟิลในบริเวณเยื่อบุไซนัส

สาขาวิชา เวชศาสตร์คลินิก ลายมือชื่อนิสิต .....

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WANRAWEE THAITRAKOOL: Histopathologic difference between sinonasal mucosa and polyp tissue for diagnosing eosinophilic chronic rhinosinusitis. ADVISOR: ASST. PROF.KORNKIAT SNIDVONGS, M.D., Ph.D., CO-ADVISOR: ASSOC. PROF.SUPINDA CHUSAKUL, M.D., ASSOC. PROF.SONGKLOT AEUMJATURAPAT, M.D., 47 pp.

This study aims to assess the appropriate site for diagnosing eosinophilic chronic rhinosinusitis (ECRS) by histopathology. Patients with chronic rhinosinusitis with polyps (CRSwNP) were enrolled. Specimens were collected from polyp apex, polyp pedicle, polyp scraping and ethmoid mucosa. Number of tissue eosinophil of the four samples was assessed with intrapersonal comparison for diagnosing ECRS. Correlations with clinical characteristics of ECRS were assessed for each site. Results showed that thirty patients with CRSwNP were enrolled. Polyp apex, polyp pedicle and ethmoid mucosa gave similar results for diagnosing ECRS in 16 patients (53.3%). Median tissue eosinophil was greater in polyp apex (84, IQR: 34-194) and polyp pedicle (96, IQR: 80-320) than ethmoid mucosa (21, IQR: 10-220), p=0.04. Sensitivity for diagnosing ECRS were 100% (95%CI: 47.8 - 100) for polyp apex, 60% (95%CI: 14.7 - 94.7) for polyp pedicle, 80% (95%CI: 28.4 – 99.5) for ethmoid mucosa. Correlations with asthma were significant for polyp pedicle (p=0.05), and ethmoid mucosa (p=0.04) but not polyp apex (p=0.21). Correlations with serum eosinophilia, and eosinophilic mucin were not significant. Tissue eosinophil from polyp scraping only correlated with ehtmoid mucosa. Consequently, density of tissue eosinophil was greater in nasal polyp than ethmoid mucosa. Polyp apex had greater sensitivity for diagnosing ECRS than others. Polyp pedicle and ethmoid mucosa correlated with asthma. Tissue eosinophil from polyp scraping only correlated with ehtmoid mucosa.

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LIST OF ABBREVIATION

AERD = aspirin exacerbated respiratory disease

CRS = chronic rhinosinusitis

CRSsNP = chronic rhinosinusitis without nasal polyps

CRSwNP = chronic rhinosinusitis with nasal polyps

CT = computer tomography

ECRS = eosinophilic chronic rhinosinusitis

EPOS = European position paper on rhinosinusitis and nasal polyps

HPF = high power field

Ig = immunoglobulin

IL = interleukin

IQR = interquartile range

NA = no analysis

nECRS = non eosinophilic chronic rhinosinusitis

ROC curve = receiver operation characteristic curve

SD = standard deviation

SIT = smell identification test

Th cell = T helper cell

μm = micrometer

CHAPTER I INTRODUCTION

Background & rationale

Chronic rhinosinusitis (CRS) is an important healthcare problem in Thailand. At Chulalongkorn Memorial Hospital, there is an increasing number of patients with chronic rhinosinusitis (CRS) each year. In 2014, there were up to 313 CRS, and out of this, there were 128 chronic rhinosinusitis with nasal polyps (CRSwNP) patients visiting Chulalongkorn Memorial Hospital.

CRS is an inflammatory disorder involving the mucosa of the nose and paranasal sinuses. It can be further classified by phenotypic presentation as CRS without nasal polps (CRSsNP) or CRS with nasal polyps (CRSwNP). Over the last decade, there are many hypotheses that try to explain the pathophysiology of CRS. Both dysregulation of the individual host and multi-factors exogenous to the host have been hypothesized. Theories on pathogenesis of CRS are such as the fungal hypothesis, the super antigen hypothesis, the biofilm hypothesis, the microbiology hypothesis, the eicosanoid hypothesis and the dysregulation of immune barrier hypothesis.[1] A single common pathway in the etiology of CRS is yet to be explained. A histological examination of CRS is an essential step in understanding the potential mechanism of a pathogenesis of CRS and its subtype.

The underlying inflammatory profile of CRS is classified into eosinophilic CRS and non-eosinophilic CRS.[2] These two subtypes have distinct pathogenesis pathways. Eosinophilic inflammation appears to mount T-helper2(Th2) mediated inflammation while non-eosinophilic inflammation is associated with T-helper1(Th1) pathways. Therefore, underlying inflammatory profile explains pathogenesis of CRS. CRS with different underlying inflammatory profile have different pathogenesis, comorbidities, clinical severity, prognosis and require different treatment aspect. Non-eosinophilic CRS patients tend to have a better response to macrolides as macrolides have interleukin8 (IL-8) modifying anti-neutrophilic activity, while eosinophilic CRS patients tend to respond well to oral or aggressive local corticosteroids[3, 4]. Recently, there are new

medications aiming for eosinophilic CRS such as Mepolizumab which has anti-interleukin 5 (IL-5) activity. Mepolizumab brings good outcomes when used in patients with eosinophilic CRSwNP. Eosinophilic CRSwNP is associated with TH2 pathway and the increase of IL-5 and IgE levels. According to the work of Philippe Gevaert and Claus Bachert et al., they found that Mepolizumab reduced the size of nasal polyps in eosinophilic CRSwNP patients, and improved of endoscopic and CT scan score.[5] Traditional sinus surgery aiming to correct osteomeatal complex obstruction may not be effective in an eosinophilic CRS subgroup because generalized severe inflammation found in eosinophilic CRS is not caused by osteomeatal complex obstruction. Different technique of sinus surgery aiming to bring the maximum access of topical corticosteroid to all units of sinonasal mucosa should be more appropriate and bring better therapeutic outcmes for eosinophilic CRS[3].

CRSwNP may have either eosinophilic or non-eosinophilic inflammatory profile. Non-eosinophilic polyps were considered associated with an incomplete treatment or unresolved bacterial infection, while eosinophilic polyps were regarded as a noninfectious disorder linked to atopy[1]. Although the effects of eosinophilic inflammatory pattern on treatment outcomes have been acknowledged, there is no consensus yet with regard to the diagnostic criteria of eosinophilic CRS. Basically eosinophil is the key inflammatory cell of eosinophilic CRS, tissue eosinophil count is therefore widely used by researchers[6]. Eosinophilic subgroup of patients with in CRSwNP, defined by tissue eosinophilia correlates significantly with co-morbidity asthma, lower smell identification test scores(SIT), more severe sinus disease, more severe endoscopic and radiologic scores,[7, 8]less improvement in quality-of-life outcomes after endoscopic sinus surgery[9], less improvement in disease-specific and general quality of life[10], and higher polyps recurrence rates.[11]

To assess tissue eosinophil count, tissue from various locations have been studied. While several researchers studied polyp tissue[6, 8, 11-15], others studied sinonasal mucosa[2, 9, 10, 16, 17]. When studied tissue was taken from different locations, the cellular density should be different. The histopathology of polyp tissue describes

mucosal sac composing of edematous tissue, hyperplastic mucous glands, fibrous tissue, vessels, and inflammatory cells, while the histopathology of sinus mucosa reports more density of inflammatory cells with less edematous. This greatly affects the assessment of the severity of inflammation and tissue eosinophil count.

While European polyps are acknowledged as eosinophilic inflammation, Asians polyps are acknowledged as neutrophilic inflammation.[18] The inflammatory pattern of CRSwNP has been reported differently even in the same country. The work of Kirtsreesakul et al. proposed that Thai polyps were eosinophilic[15], The work of Katotomichelakis et al reported that Thai polyps were neutrophilic. When Katotomichelakis et al assessed Thai polyps again twelve years later, Thai polyps were proposed eosinophilic and the researchers proposed that the inflammatory pattern of Thai polyp may change over time.[19] These different findings may be caused by different locations of studied tissue taken. Studied tissue from two different locations may report two different findings. When it is essential to assess tissue eosinophil count for an appropriate plan of management, it is crucial to take tissue from the right location. At present, it is inconclusive on which source of the studied tissues, either from nasal polyps or sinonasal mucosa, will show better correlation with tissue eosinophilia and disease severity.

The investigators aim to investigate the histopathologic difference between sinonasal mucosa and polyp tissue on diagnosing eosinophilic rhinosinusitis. We hypothesize that tissue eosinophil count from these two locations of the same person should be different. Sinus mucosa should report greater tissue eosinophil count and reflect the true inflammatory pattern. Eosinophilic CRS is driven by Th2 inflammation which originates from sinus mucosa and subsequently results in polyp formation.[20] Sinus mucosa therefore is hypothesized the better location for diagnosing eosinophilic rhinosinusitis.

Presently, studies of CRS tend to focus on histopathology. Knowledge about underlying inflammatory profile makes us understand the difference in pathogenesis pathways of CRS and the different therapeutic options. Studies of inflammatory cells

infiltration in sinonasal tissue brought the studied tissues from various sites in sinonasal cavity. But there is yet a clear consensus about where is the best source of tissue to diagnose eosinophilic CRS.

We aim to compare tissue eosinophil count between the sinonasal mucosa, polyp tissue and polyp scrubbing exudates. Inflammatory pattern between the pedicle and the apex of the same polyp will be also investigated. The knowledge from this study will help clinicians in the step of diagnosis and treatment planning in CRSwNP patients. It will guide clinicians to tailor medical and surgical management for individual patient for the most efficient and best treatment result.

Research Questions

1. Which is the best location between sinus mucosa and polyp tissue for histopathologically diagnosing eosinophilic CRS?

2. Is the inflammatory pattern different between the pedicle and the apex of the same nasal polyp?

3. Is nasal polyps scrubbing exudate correlate with histopathology of polyp tissue?

Hypothesis

As eosinophilic CRS is driven by Th2 inflammation which originates from sinus mucosa and subsequently results in polyp formation, we hypothesize that tissue eosinophil count between sinus mucosa and polyps of the same person should be different. Sinus mucosa should report greater tissue eosinophil count and reflect the true inflammatory pattern. Sinus mucosa therefore is hypothesized the better location for diagnosing eosinophilic rhinosinusitis than polyp tissue.

In addition, we hypothesize that tissue eosinophil count between the pedicle and the apex of the same polyps should be different. Polyp pedicle should report greater tissue eosinophil count and reflect the true inflammatory pattern.

Study design

diagnostic study

Objectives

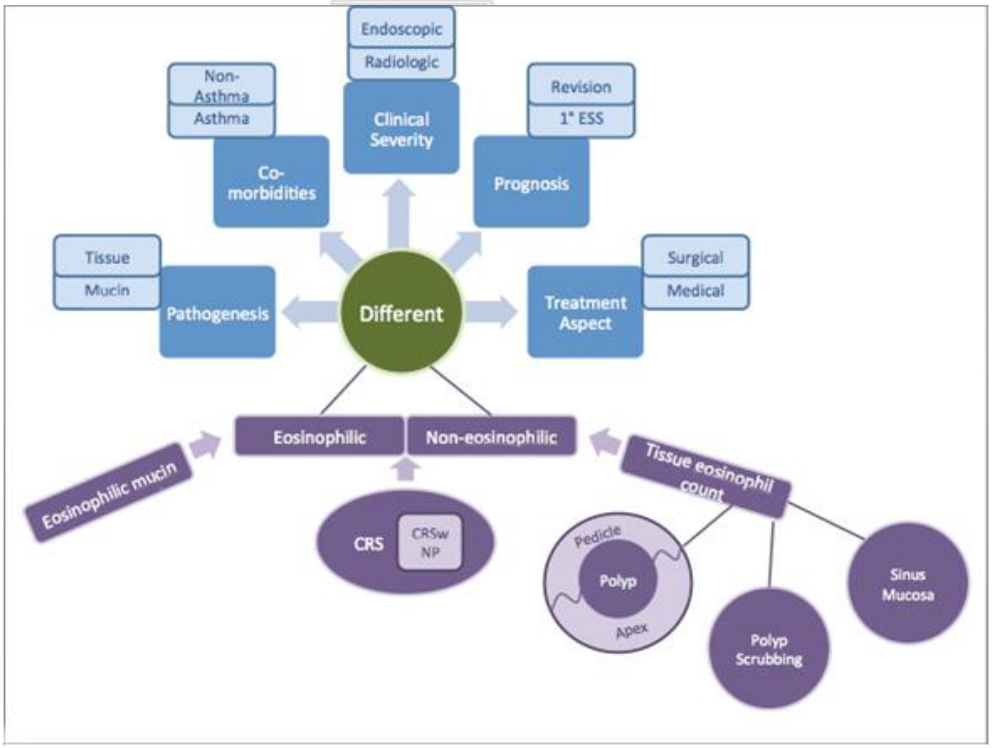
1.) To compare sinus mucosa to polyp tissue with regard to tissue eosinophil count and its sensitivity for diagnosing eosinophilic CRS

2.) To assess the correlation between tissue eosinophil count and the characteristics of eosinophilic CRS in terms of co-morbidity asthma, history of previous surgeries, serum eosinophilia, endoscopic score severity, and radiologic score severity

3.) To compare tissue eosinophil count between the pedicle and the apex of the same polyp

4.) To assess histopathological correlation between polyp scrubbing exudates and polyp tissue

Conceptual framework



Key words

sinusitis, ethmoid sinus, nasal polyps, nasal mucosa, histology

CHAPTER II LITERATURE REVIEW

Chronic rhinosinusitis (CRS) is an inflammatory disorder involving the mucosa of the nose and paranasal sinuses. CRS in adult is defined by criteria of European position paper on rhinosinusitis and nasal polyps 2012 (EPOS 2012) that CRS is an inflammation of nose and paranasal sinuses. CRS patients must have more than 12 weeks of two or more symptoms, one of which must be either nasal obstruction, nasal blockage, nasal congestion or nasal discharge (may be anterior or posterior nasal drip) and may have facial pressure and/or reduction of smell symptoms. CRS patients must have either endoscopic signs or CT changes. Endoscopic signs are polyps or mucopurulent discharge from middle meatus or mucosal edema in middle meatus. CT changes are mucosal changes within sinuses or osteomeatal complex.[21] Phenotypic classification of chronic rhinosinusitis is Chronic Rhinosinusitis without nasal polyps (CRSsNP) or Chronic Rhinosinusitis with nasal polyps (CRSwNP). Definition of Chronic Rhinosinusitis with nasal polyps (CRSwNP) is Chronic Rhinosinusitis that has bilateral polyps, endoscopically visualized in middle meatus.[21] Etiology of CRS is multifactorial. There is no common single pathway of pathogenesis of CRS. The current hypothesis suggests that CRS pathogenesis is the abnormal host immune response against multi-environmental factors. Systemic host factors are innate immunity, adaptive immunity, airway hypereactivity, allergy, mucociliary dysfunction, immunodeficiency, etc. Local host factors are odontogenic inflammation, anatomic abnormalities, bone inflammation, acquired mucociliary dysfunction, etc. Environmental factor are viruses, bacteria, superantigen, biofilms, fungi, air pollution, smoking, etc. The above multi-factors explain the pathogenesis of CRS that is associated with many immune systems, cytokines and inflammatory mediators.[22] Histopathological examination of CRS is therefore an essential step in understanding the pathogenesis. The inflammatory pattern of CRS is classified into non-eosinophilic CRS (nECRS) and eosinophilic CRS (ECRS)[2]. These two subtypes have distinct pathogenesis pathways. Non-eosinophilic inflammation is associated with T-helper1 and T-helper17 pathways while eosinophilic inflammation appears to mount T-helper2 mediated inflammation. Therefore,

underlying inflammatory profile explains pathogenesis of CRS. CRS with different underlying inflammatory profile have different pathogenesis, comorbidities, clinical severity, prognosis and require different treatment aspect. Eosinophilic CRS subgroup is associated with aspirin exacerbated respiratory disease (AERD), allergic fungal rhinosinusitis, nasal polyposis with Staphylococcus Aureus-induced superantigen. Eosinophils release many of toxic and pro-inflammatory mediators composed of eosinophil-derived neurotoxin, major basic protein and eosinophil cationic protein. These toxic and pro-inflammatory mediators intoxicate nasal mucosa and surrounding tissues. In addition, they are associated with airway hyperactivity, vascular leakage, mucus secretion overproduction, epithelial injury, tissue remodeling factors and large production of cytokines including interleukin 4, interleukin 5, leukotrienes and eotaxin[23, 24].

The work of Kornkiat Snidvongs et al. has found that eosinophilic chronic rhinosinusitis patients, as defined by sinonasal tissue eosinophilia >10 eosinophils/HPF, have worsening endoscopic and radiological CT score when compared to non-eosinophilic chronic rhinosinusitis group.[2] The work of Zachary M. Soler et al. has found that these patients also have less improvement in quality of life outcome after surgery when compared to non-eosinophilic chronic rhinosinusitis group.[9] This is in coherent with the study of relationship between clinical measure and histopathologic findings in chronic rhinosinusitis by Zachary M.Soler et al., which proposed that mucosal eosinophilia in sinonasal tissue correlates with sinonasal disease severity in term of CT score, endoscopic score and smell identification test (SIT) score. The higher the number of tissue eosinophilia, the higher the severity score is. [10]

Mucosal eosinophilia also correlates with the less improvement after surgical treatment of CRS patients. Mucosal eosinophilia is the predictor of less improvement in general quality of life and disease-specific quality of life in CRS patients who undergo endoscopic sinus surgery compare with non-eosinophilic CRS group.[9] Furthermore, tissue eosinophilia also correlates with higher recurrent rate of nasal polyps. According to study of predictive significance of tissue eosinophilia for nasal polyp recurrence in Chinese population by Hongfei Lou et al., (i) the percentage of tissue eosinophil to

other inflammatory cells at 27% or (ii) absolute total eosinophil count per HPF in nasal polyps tissue more than 55 eosinophils/HPF is the cut-off point for a good predictor of nasal polyps recurring after endoscopic sinus surgery in CRSwNP patients.[11] The above is in harmony with the work of Marko Velimir Grgic' et al. which proposes that high eosinophilic infiltration in nasal polyps tissues correlates with high occurrence of nasal polyps recurrence and lower symptomatic and functional improvement after endoscopic sinus surgery in CRSwNP patients.[8]

In addition to eosinophilia in sinonasal tissue being associated with more severe sinuses disease, it also has distinct character of disease and pathogenesis. Difference in underlying tissue inflammation also affects the treatment options. Non-eosinophilic CRS patients tend to have a better response to macrolides as macrolides have interleukin8 (IL-8) modifying anti-neutrophilic activity, while eosinophilic CRS patients tend to respond well to oral or aggressive local corticosteroids[3, 4]. Intranasal corticosteroid reduces tissue eosinophilia but has no effect in neutrophil recruitment or interleukin-8 expression.[4] This is coherent with the work of Weipin Wen et al. which found that increasing in number of tissue neutrophilia correlates with reducing in response of oral corticosteroid treatment in patients with CRSwNP interm of polyp size, nasal congestive symptoms, total nasal symptom scores, and nasal airflow resistance.[13] Recently, there are new medications aiming for eosinophilic CRS such as Mepolizumab which has anti-interleukin 5 (IL-5) activity. Mepolizumab brings good outcomes when used in patients with eosinophilic CRSwNP. Eosinophilic CRSwNP is associated with TH2 pathway and the increase of IL-5 and IgE levels. According to the work of Philippe Gevaert and Claus Bachert et al., they found that Mepolizumab reduced the size of nasal polyps in eosinophilic CRSwNP patients, and improved of endoscopic and CT scan score.[5] Traditional sinus surgery aiming to correct osteomeatal complex obstruction may not be effective in an eosinophilic CRS subgroup because generalized severe inflammation found in eosinophilic CRS is not caused by osteomeatal complex obstruction. From the work of Kornkiat Snidvongs et al. "eosinophilic rhinosinusitis is not a disease of osteomeatal occlusion", they proposed that osteomeatal complex occlusion is not associated with draining sinuses

of eosinophilic CRS and CRSwNP patients and the osteomeatal complex manipulation has no effect in these groups of patients. Different technique of sinus surgery aiming to bring the maximum access of topical corticosteroid to all units of sinonasal mucosa should be more appropriate and bring better therapeutic outcmes for eosinophilic CRS[3].

Eosinophilic mucin is an important character of eosinophilic CRS. Eosinophilic mucin demonstrates the eosinophil activation evidence in eosinophilic CRS.[25] The typical characteristic of eosinophilic mucin is sticky mucous secretion. The color varies from light brown to dark green.[26, 27] Histological findings to make diagnosis of eosinophilic mucin comprise of i) eosinophils aggregation and ii) Charcot-Leyden crystals which are the eosinophil decomposing by-product.[25, 26, 28-30] In year 2012 Snidvongs et al. proposed that eosinophilic CRS traditional phenotypic features are not the good markers for tissue eosinophilic infiltration. They suggested that the best predictors are tissue eosinophilia along with eosinophilic mucin which is the evidence of eosinophil activation.[2] Phenotypic features are not accurate for diagnosing ECRS as tissue eosinophilia can be found in patients with CRS without polyps (CRSsNP) with a percentage of 19%[2]. Eosinophilic infiltration with viscous eosinophilic mucin and eosinophils aggregation with Charcot-Leyden crystals[25, 26, 28-30], are manifestations of eosinophil activation[2, 6) (10]. CRS with polyps (CRSwNP), although associated with eosinophilic inflammation, may present with diverse inflammatory patterns[18]. Understanding the underlying inflammatory process of CRS suggests individualized effective therapeutic options. However, tissue from various locations have been studied to assess tissue eosinophil count. While several researchers studied polyp tissue[6, 8, 11-15], others studied ethmoid mucosa[2, 9, 10, 16, 17]. The inflammatory pattern of CRSwNP has been reported differently even in the same country. While Songkhla University proposed that Thai polyps were eosinophilic[15], Siriraj hospital reported that Thai polyps were neutrophilic. When Siriraj hospital assessed Thai polyps again twelve years later, Thai polyps were proposed eosinophilic and the researchers proposed that the inflammatory pattern of Thai polyp may change over time.[19] These different findings may be caused by different locations of studied tissue taken. Studied tissue

from two different locations may report two different findings. When it is essential to assess tissue eosinophil count for an appropriate plan of management, it is crucial to take tissue from the right location. There is no consensus yet regarding an appropriate source of tissue taken for histopathologic assessment. Different sources for tissue biopsy have different density of inflammatory cell infiltration and this should affect sensitivity and specificity for evaluating CRS endotype.

Nasal polyp is not a disease but it is a physical finding. The most common site of nasal polyps occurrence is in the middle meatus. Stammburger analyzed nasal polyps from 200 patients undergoing endoscopic sinus surgery. He found that 80% of nasal polyps originated from middle meatal mucosa, uncinate process, and infundibulum. 65% of nasal polyps originated from ethmoid bulla and hiatus semilunaris. 48% of nasal polyps originated from frontal recess.[31] The work of Benamara A. et al. about "The site of origin of nasal polyposis in the ethmoid subcompartments assessed from clinical observation of ninety-four nasal cavities", found that the most common site of nasal polyps occurrence is in the middle meatus with 98%, follow by 75% in the posterior olfactory fossa, 61% in the superior meatus, 50% on middle turbinate and 41% in the anterior olfactory fossa.[32] Nasal polyps are mucosal sac composing of edematous tissue, fibrous tissue, vessels, inflammatory cells and glands. There is fluid, gland blood vessels and inflammatory cells in nasal polyps stroma. Nasal polyps are mostly covered with pseudostratified columnar cylindric epithelium with goblet and ciliary cells. The covered epithelium and inflammatory cells of nasal polyps change during their growth and developmental process. There is an analytic study that examined the serially-sectioned polyp halves. There are various types of epithelium in nasal polyp, which are pseudostratified epithelium of various height, transitional epithelium and squamous nonkeratinized epithelium. There are larger areas of transitional epithelium and lower globet cell density in an anterior polyp halves than in the posterior one. The previous findings may be caused by the effect of the air current flow on the anterior halves of the polyps.[33] Nasal polyps infiltrate with various types of inflammatory cells and mediators such as eosinophil, neutrophil, lymphocyte, mast cell, plasma cell. These inflammatory cells are mostly located

around subepithelial, perivascular and periglandular areas. Among the inflammatory cells infiltrated in nasal polyps, the most predominant cell is eosinophil. Eosinophils are mostly distributed around superficial layer rather than in stromal layer. The air current flow on nasal polyps brings larger areas of transitional epithelium and lower goblet cell density to polyp apex[33]. Histopathology assessment of superficial layer of polyp apex may be more accurate than stromal layer according to the work of Rafal Pawliczak et al. They found that distribution of eosinophils and mast cells in nasal polyps are more abundant in superficial layer[34]. In contrast, Saseki Y. et al. found higher density of mast cells and degranulated mast cells infiltrate in nasal polyp pedicle than in polyp apex. They proposed that polyp pedicle and degranulated mast cell should play an important role in pathogenesis of nasal polyps formation[35]. Presently, there are many studies about underlying inflammatory profiles of CRSwNP, the degree of infiltration of eosinophils in sinonasal tissues and the correlation between underlying inflammatory profiles with the severity of sinus diseases. In these studies, the studied tissues came from various sites either sinus mucosa[2, 9, 10, 16, 17] or nasal polyps[6, 8, 11-15]. Jacqueline Ho and Richard Harvey, et al. studied cellular comparison of sinus mucosa VS polyp tissue from a single sinus cavity in chronic rhinosinusitis. They found that cellular composition such as CD4 T cell, activated CD4 T cell, CD8 T cell, T follicular helper cell, regulatory T cell, B cell and IgA+ B cell in nasal polyps and sinus mucosa are quite similar. They found the elevation of ILC2s, activated CD8 T cells, pDCs, plasma cells and IgG+B cells in nasal polyps, which they proposed that these cells are associated with developmental process of nasal polyps. Finally, they concluded that nasal polyps tissue biopsy is sufficient for investigation of CRS and it can be used interchangeably with sinus mucosa biopsy.[20] In addition, there is a study that compared eosinophils in nasal exudates from lateral nasal wall scrubbing and eosinophils in polyp tissue. They found that percentage of eosinophils in the exudate has the strong correlation with the one in the polyp tissue. So, they recommended that nasal cytology is a convenient and non- traumatic technique to evaluate the inflammatory profile in CRSwNP patients.[6] But there is yet a consensus on the good source of diagnosing eosinophilic CRS. When investigators assess histopathology of tissue taken from different locations for studying ECRS, different

findings are reported. This study, therefore aims to assess histopathology and its sensitivity and specificity on diagnosing ECRS of three sources of tissue; polyp apex, polyp pedicle and ethmoid mucosa. And to assess the histopathologic difference in term of eosinophil count between sinonasal mucosa, polyp tissue and polyp scraping.

CHAPTER III MATERIAL AND METHODS

This study was approved by the Institutional Review Board of the Faculty of Medicine, Chulalongkorn University number 560/58. We enrolled patients with CRSwNP who presented at the Endoscopic Nasal and Sinus Surgery Excellence Center, King Chulalongkorn Memorial Hospital and scheduled for endoscopic sinus surgery from January 2016 to January 2017.

Patient population

The diagnosis of CRSwNP was based on the European Position Paper on Rhinosinusitis and Nasal Polyps (EPOS 2012)[21]. All patients signed the informed consents before participation in the study. Patients with unilateral nasal polyposis were excluded. Other exclusion criteria were cystic fibrosis, immunodeficiency, primary ciliary dyskinesia, fungal rhinosinusitis, systemic vasculitis and granulomatous diseases, cocaine abuse, and any other neoplasia. Comorbidity of asthma was recorded if presence; defined as clinically using inhaled β-agonist or corticosteroid. Preoperative Lund-Kennedy endoscopy score and Lund-Mackay CT score were recorded. Endoscopic score severity is assessed using Lund-Kennedy endoscopic scoring system. Lund-Kennedy endoscopic scoring system assesses nasal polyps, mucosal edema and secretion. Each left and right nasal cavity is scored separately and finally the two scores are summed up to determine the overall endoscopic score, the maximum overall score is 12.

Radiologic score severity is scored using Lund-Mackay CT scan scoring system. Lund-Mackay CT scan scoring system assesses individual sinus involvement including maxillary, anterior ethmoid, posterior ethmoid, sphenoid and frontal. The degree of sinus involvement is scored by degree of opacification in the individual sinus. In score 0, there is no abnormalities in the sinus. In score 1, there is partial opacification in the sinus. In score 2, there is total opacification in the sinus. Lund-Mackay CT scan scoring system also assesses osteomeatal complex status. In score 0, there is no obstruction at the osteomeatal complex from CT scan. In score 2, there is obstruction at the osteomeatal complex. Each left and right nasal cavity is scored separately and finally the two scores are summed up to determine the overall CT scan score, the maximum

overall score is 24.

Histopathology assessment

To assess inflammatory pattern of CRSwNP, any antibiotics, topical corticosteroids or systemic corticosteroids were not allowed within 4 weeks before endoscopic sinus surgery. Tissue specimens were collected intra-operatively from three sources; polyp apex, polyp pedicle and ethmoid mucosa. All studied tissues were processed using a standard pathological laboratory technique. The studied tissues were fixed and processed for tissue paraffin embedding. The paraffin embedded tissues were sectioned at 5 μm thickness, processed by hematoxylin/ eosin staining and then collected in the slides bank for histopathological review. This was assessed by a single certified pathologist in a blind fashion, where the pathologist knew neither patient history, and characteristics nor the source of tissue specimens. All studied slides were evaluated for histopathology profiling described in a published study[2]. The absolute number of tissue eosinophil was counted in all three sources of tissue. Low power microscopic magnification was used to scan and to identify three non-overlapping most intense areas of inflammatory cell infiltration. These three areas were then assessed by bright-field light microscope at x400 magnification. Tissue eosinophils were counted as numbers of eosinophils per HPF in three areas and the median of eosinophil counts was used. High tissue eosinophilia was defined when the median of eosinophil counts was ≥10/HPF. The work of Kornkiat Snidvongs et al. has found that eosinophilic chronic rhinosinusitis patients, as defined by sinonasal tissue eosinophilia >10 eosinophils/HPF, have worsening endoscopic and radiological CT score[2] and the work of Zachary M. Soler et al. has found that these patients also have less improvement in quality of life outcome after surgery when compared to non-eosinophilic chronic rhinosinusitis group.[9] Histopathological data were recorded[2] for each source of the studied tissues (polyp apex, polyp pedicle and ethmoid mucosa). Mucin was collected in formalin or fixed immediately onto slide with 95% alcohol if small quantity. The slides were stained with hematoxylin-eosin for evaluation of inflammatory pattern and with Gomeri methenamine-silver stain for identification of fungal hyphae. Mucin was evaluated by bright-field light microscope at x400

magnification for presence or absence of eosinophil aggregates, Charcot-Leyden crystals and fungal elements. After mucin collection, nasal polyp's scraping is collected by scraping the nasal polyp using rhino-probe, placing the scraping specimen onto slide and fixing with 95% alcohol immediately. The polyp's scraping will be performed before removing the polyp from the nasal cavity to mimic the in-office procedure and provide for clinical applicability in the future. The polyp's scraping slides were processed by hematoxylin/ eosin staining and then collected in the slides bank for histopathological review.

Statistical Analysis

Descriptive data were presented as means ± standard deviation (SD), median ± interquartile range (IQR) and percentages. Friedman and Wilcoxon sign rank test were used for comparing different non-parametric variables. McNemar test was used to compare diagnostic ability between mucin and tissue eosinophil counts on diagnosing eosinophilic CRS. Receiver operation characteristic curve (ROC curve) was used to evaluate ability of tissue eosinophilia from ethmoid mucosa and polyp tissue in diagnosing ECRS. Pearson correlation coefficients were performed for linear relationship of scale variables. The P value of ≤ 0.05 was defined as statistical significance. Statistical analyses were performed using STATA version 15.0 (Stata Corp., Texas, USA)

CHAPTER IV RESULTS

Thirty patients with a mean age of 48.6±16.7 years old were enrolled. Seventeen (56.7%) patients were male. Nine (30.0%) patients were asthmatic. Five (16.7%) patients had received previous endoscopic sinus surgery. Four (13.3%) patients had serum eosinophilia. Median percentage of eosinophil in the serum was 5.3% (1.6 - 7.6). Median Lund-Kennedy endoscopy score was 11 (7 - 12). Median Lund-Mackay CT scan score was 17.5 (14 - 21). When tissue eosinophilia was assessed from polyp apex, polyp pedicle and ethmoid mucosa, ECRS was diagnosed when having high tissue eosinophilia ≥10/HPF. The three sites gave similar diagnosis in sixteen (53.3%) patients. Eleven (36.7%) patients were diagnosed as ECRS and five (16.6%) patients were diagnosed as nECRS consistently.

Density of tissue eosinophil in ECRS patients by sources of tissue specimens

To analyze density of tissue eosinophil in ECRS, eleven patients consistently diagnosed by all three sources as ECRS were assessed. For multiple comparison, tissue eosinophil count was significantly different (p=0.04) among polyp apex (median 84, IQR 34-194), polyp pedicle (median 96, IQR 80-320) and ethmoid mucosa (median 21, IQR 10-220). Figure 1 displays intrapersonal difference in tissue eosinophil count among three sites of tissue sample. For pair comparison, tissue eosinophil count from polyp pedicle was significantly greater than from ethmoid mucosa, p=0.05. Difference of density of tissue eosinophil between polyp apex and ethmoid mucosa did not reach significance. Density of eosinophils in polyp apex and polyp pedicle was not statistically different. Data are displayed in Table 1.

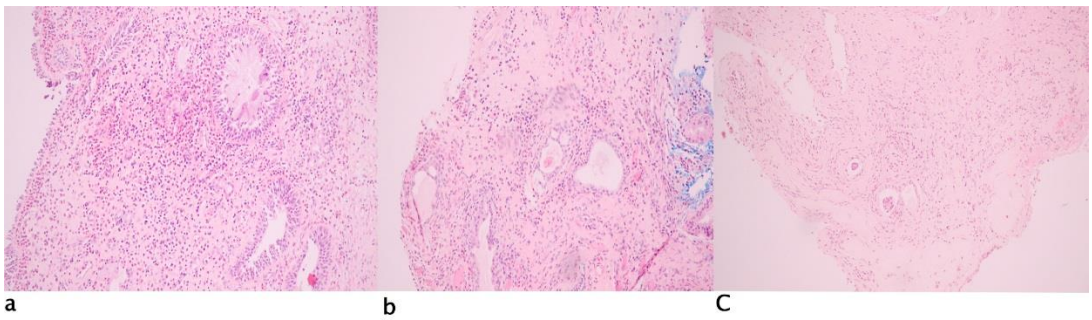


Figure 1: Difference in tissue eosinophil count among three sites of tissue sample: polyp apex (a), polyp pedicle (b) and ethmoid mucosa (c)

Source of tissue specimen	Median tissue eosinophil (IQR)	Multiple comparison P-value[a]	Pair comparison with apex P-value[b]	Pair comparison with pedicle P-value[b]
polyp apex	84 (34-194)	0.04	NA	0.15
polyp pedicle	96 (80-320)		0.15	NA
ethmoid mucosa	21 (10-220)		0.28	0.05

Table 1 Comparison of tissue eosinophil count among 3 sites

Footnote: P-value[a] analyzed with Friedman, P-value[b] analyzed with Wilcoxon sign rank test, NA= no analysis

Performance of each source of tissue specimens on diagnosing ECRS

Using presence of eosinophilic mucin as a reference, polyp apex had sensitivity of 100% (95%CI: 47.8 - 100), specificity of 36% (95%CI: 18-57.5), positive predictive value of 23.8% (95%CI: 8.2-47.2) and negative predictive value of 100% (95%CI: 66.4-100). The area under the curve of ROC was 0.68 (95%CI: 0.58-0.78). Youden index was 0.36.

Polyp pedicle had sensitivity of 60% (95%CI: 14.7 - 94.7), specificity of 40% (95%CI: 21.1 - 61.3), positive predictive value of 16.7% (95%CI: 3.6-41.4) and negative predictive value of 83.3% (95%CI: 51.6-97.9). The area under the curve of ROC was 0.5 (95%CI: 0.24-0.76). Youden index was 0.

Ethmoid mucosa had sensitivity of 80% (95%CI: 28.4 – 99.5), specificity of 48% (95%CI: 27.8 – 68.7), positive predictive value of 23.5% (95%CI: 6.8-49.9) and negative predictive value of 92.3% (95%CI: 64-99.8). The area under the curve of ROC was 0.64 (95%CI: 0.42-0.86). Youden index was 0.28. Data are displayed in Table 2.

Tissue eosinophil from polyp apex	**Eosinophilic mucin**		**Number of patient**
	presence	**absence**	
≥10	5	16	21
<10	0	9	9
Total	5	25	30
Tissue eosinophil from polyp pedicle	**Eosinophilic mucin**		**Number of patient**
	presence	**absence**	
≥10	3	15	18
<10	2	10	12
Total	5	25	30
Tissue eosinophil from ethmoid mucosa	**Eosinophilic mucin**		**Number of patient**
	presence	**absence**	
≥10	4	13	17
<10	1	12	13
Total	5	25	30

Table 2 Comparison of three sources of tissue specimen: polyp apex, polyp pedicle and ethmoid mucosa on diagnosing ECRS

Performance of each source of tissue specimens on correlation with characteristic of ECRS

A correlation between high tissue eosinophilia and comorbid asthma was revealed when histopathology was assessed from polyp pedicle and ethmoid mucosa but this correlation was not seen when polyp apex was analyzed. There was no significant correlation seen between tissue eosinophilia and other ECRS characteristics

including previous endoscopic sinus surgeries, eosinophilic mucin, serum eosinophilia, high endoscopy score, and high CT score when histopathology was assessed from any source of tissue specimen. Data are displayed in Table 3.

ECRS characteristics	Total number	Polyp apex				Polyp pedicle				Ethmoid mucosa			
		nECRS (n=9)	ECRS (n=21)	P-value	R	nECRS (n=12)	ECRS (n=18)	P-value	R	nECRS (n=13)	ECRS (n=17)	P-value	R
Asthma (number of patient)	9	1	8	0.21	NA	1	8	0.05	NA	1	8	0.04	NA
Previous endoscopic sinus surgeries (number of patient)	5	2	3	0.62		1	4	0.62		2	3	0.87	
High serum eosinophilia (number of patient)	4	0	4	0.29		2	2	0.66		0	4	0.11	
Eosinophilic mucin (number of patient)	5	0	5	0.29		2	3	0.99		1	4	0.25	
Median percent of serum eosinophil (IQR)	30	3.2 (1.4-6.2)	5.6 (2-8.8)	0.21	0.28	3.9 (1.5-7)	5.6 (2-8.5)	0.68	0.06	4.5 (2.2-6.4)	5.6 (1.6-8.9)	0.57	0.20
Median endoscopic score (IQR)	30	12 (11-12)	10 (7-11)	0.06	-0.30	11 (10-12)	10 (7-12)	0.39	-0.21	11 (7-12)	11 (9-11)	0.51	-0.07
Median CT score (IQR)	30	20 (18-21)	17 (13-20)	0.23	-0.23	20 (16-21.5)	16.5 (13-20)	0.13	-0.29	18 (13-21)	17 (15-20)	0.78	-0.04

Table 3 Performance of each source of tissue specimens on correlation with characteristic of ECRS

Footnote: ECRS= eosinophilic chronic rhinosinusitis, IQR= interquartile range, n=number, NA= no analysis

Performance of eosinophilic mucin on diagnosing ECRS

Figure 2 displays histopathology of eosinophilic mucin. To analyze the performance of eosinophilic mucin on diagnosing ECRS, histopathology criteria of having high tissue eosinophil ≥ 10/HPF was used as a reference. Sixteen patients undeviatingly diagnosed by all three sources (as ECRS or nECRS) were assessed. Eosinophilic mucin had sensitivity of 27.3% (95%CI: 6-61) and specificity of 100% (95%CI: 47.8-100). It had positive predictive value of 100% (95%CI: 29.2-100) and negative predictive value of 38.5% (95%CI: 13.9-68.4). The area under the curve of ROC was 0.64 (95%CI, 0.49-0.77). Data are displayed in Table 4.

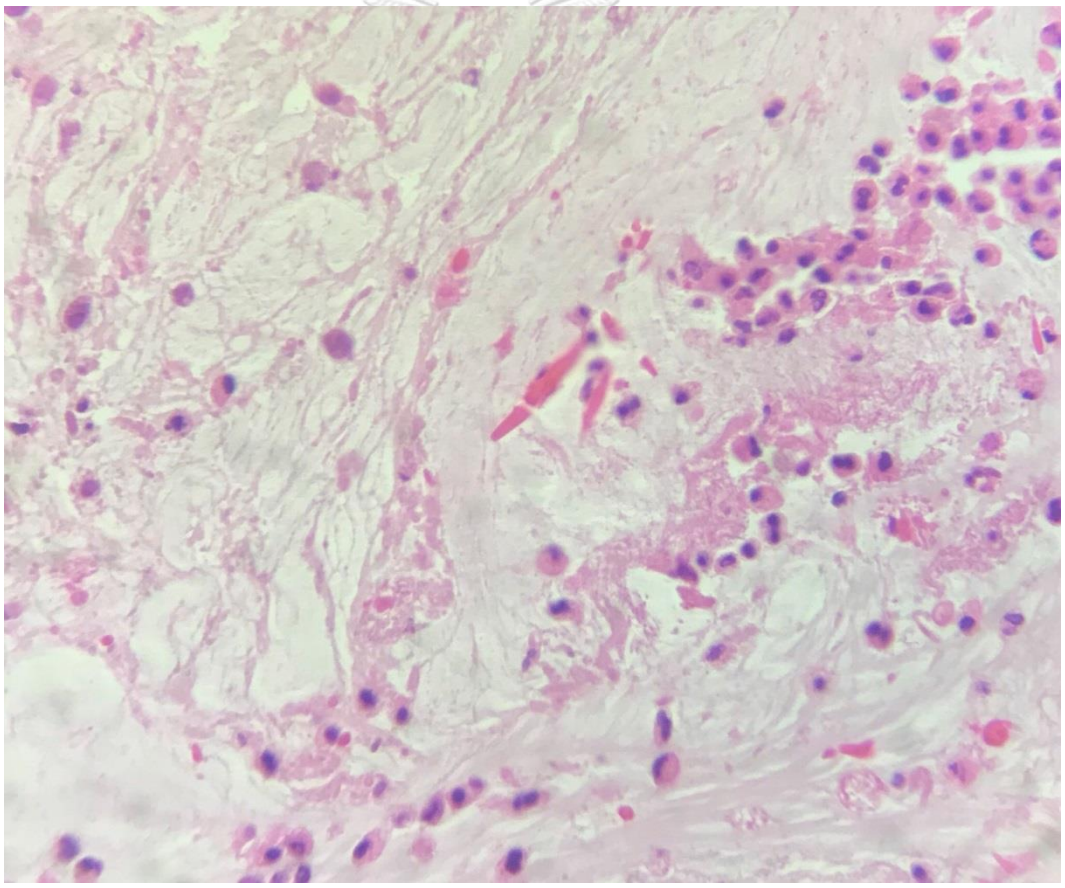


Figure 2: Histopathology of eosinophilic mucin

Mucin	Tissue eosinophil count/ HPF		Total number of patient
	≥ 10	< 10	
Eosinophilic	3	0	3
Non eosinophilic	8	5	13
Total number of patient	11	5	16

Table 4 Performance of eosinophilic mucin on diagnosing ECRS

Correlation of tissue eosinophil between polyp's scraping and each source of tissue specimens

There was a correlation between polyp's scraping and tissue eosinophil count at sinus mucosa. In two other sites, there were no statistical correlation between tissue eosinophil count and polyp's scraping. Figure 3, 4 and 5 demonstrate correlations between polyp's scraping and tissue eosinophil count at sinus mucosa, polyp's pedicle and polyp's apex respectively.

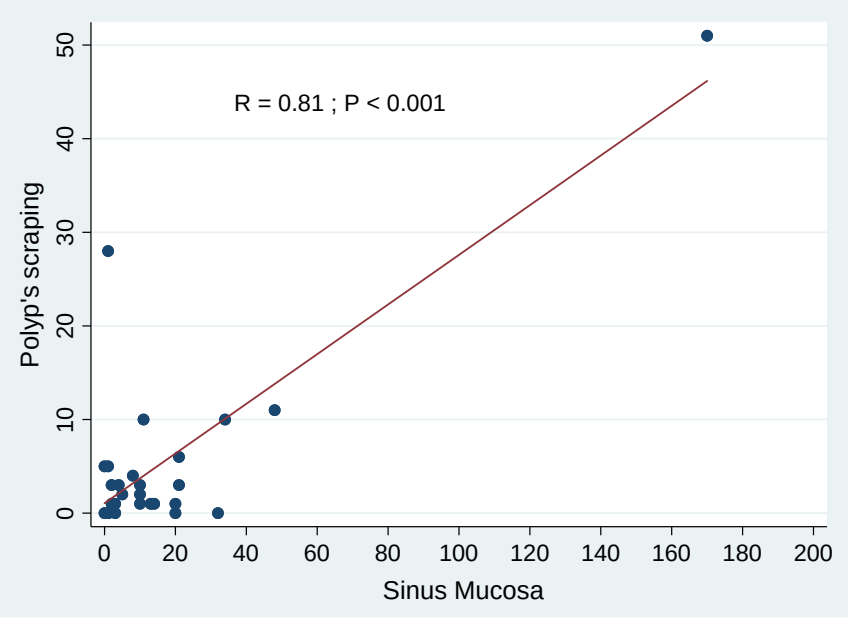


Figure 3: Correlations between polyp's scraping and tissue eosinophil count at sinus mucosa

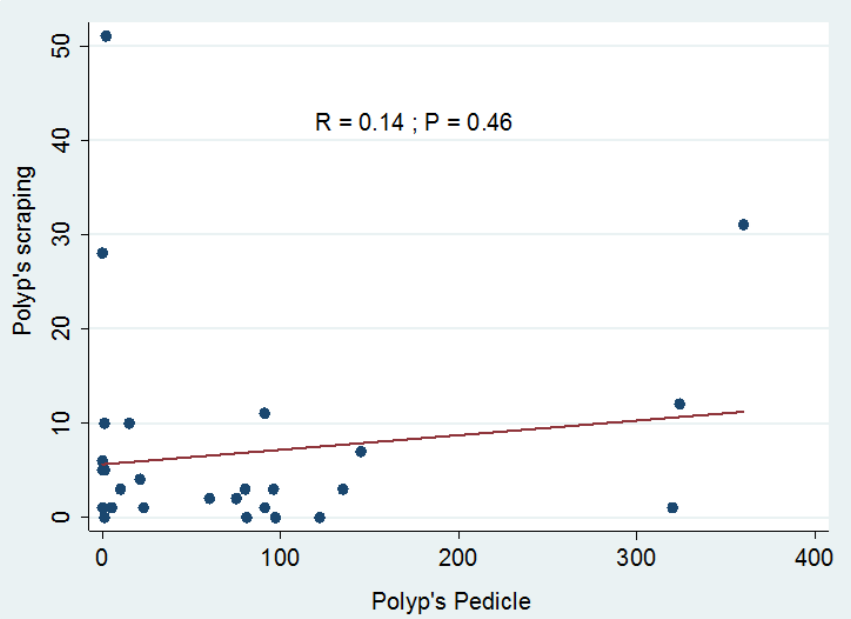


Figure 4: Correlations between polyp's scraping and tissue eosinophil count at polyp's pedicle

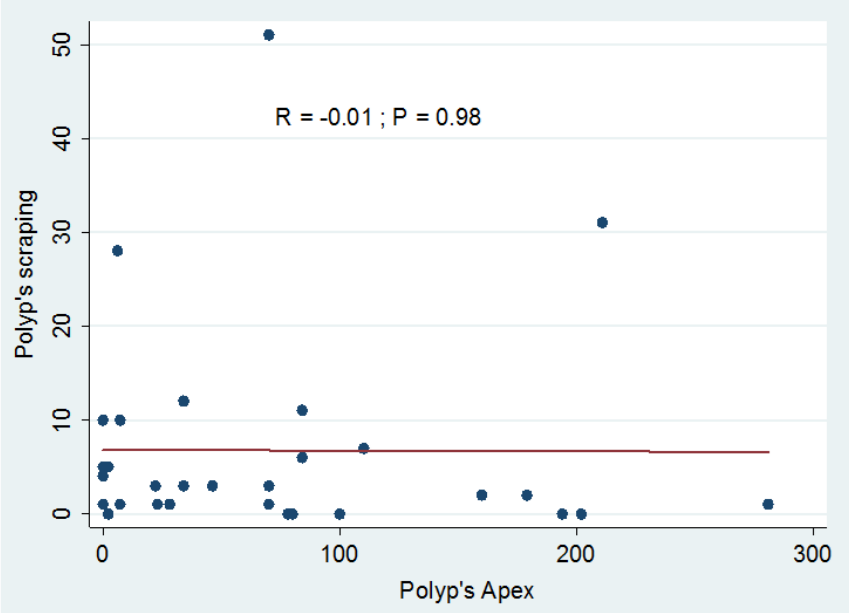


Figure 5: Correlations between polyp's scraping and tissue eosinophil count at polyp's apex

CHAPTER V DISCUSSION AND CONCLUSION

Nasal polyps are mucosal sac composed of edematous tissue, fibrous tissue, vessels, inflammatory cells and glands. There is fluid, gland, blood vessels and inflammatory cells in nasal polyps stroma. Nasal polyps are mostly covered with pseudostratified columnar cylindric epithelium with goblet and ciliary cells. The covered epithelium and inflammatory cells of nasal polyps change during their growth and development[33]. Our study shows that density of tissue eosinophilia was greater in nasal polyps than ethmoid mucosa. Histopathology assessed from polyp apex had greater sensitivity for diagnosing ECRS than from polyp pedicles and ethmoid mucosa and its negative predictive value is 100%. This may be an effect of air current flow on the polyp apex resulting in further histopathological change which explains these findings. Therefore, the polyp apex should be the most sensitive site to define inflammatory patterns of chronic inflammatory disease of paranasal sinuses. Nasal polyp is simply one clinical feature of chronic inflammation other than a disease. According to the pathogenesis, nasal polyp originates from Th2 inflammation, so it well represents underlying inflammatory process of CRS. This is in line with a previous analytic study of serially-sectioned nasal polyps showing larger areas of transitional epithelium and lower goblet cell density in an anterior polyp halves[33]. In addition, the site of nasal polyps occurrence is more common in the area exposed to the air current flow like the middle meatal mucosa, uncinate process, and infundibulum[31-33]. Jacqueline Ho et al. studied cellular comparison of polyp tissue versus sinus mucosa from a single sinus cavity in chronic rhinosinusitis. They found the elevation of ILC2s, activated CD8 T cells, pDCs, plasma cells and IgG+B cells in nasal polyps when compared to sinus mucosa. They hypothesized that these cells are associated with the developmental process of nasal polyps and suggested nasal polyp tissue biopsy for investigation of CRS which can be used interchangeably with sinus mucosa

biopsy[20]. However, our findings contrast with the work of Saseki Y. et al. They reported higher density of mast cells and degranulated mast cells infiltrate in nasal polyp pedicle than in nasal polyp apex[35]. We believe degranulated mast cell may play an important role in pathogenesis of allergic rhinitis other than rhinosinusitis. Eosinophil should be a key inflammatory cell which better represents Th polarization of chronic inflammatory disease.

Both polyp tissue[6, 8, 11-15], and ethmoid mucosa[2, 9, 10, 16, 17] were assessed for inflammatory pattern of CRS. Findings from this study showed that histopathology assessment from different sources of tissue specimen gave similar diagnosis in only about half of the patients. Histopathology assessment of different source of tissue specimens may bring different findings and unreliable interpretation. Although total IgE, specific IgE to Staphylococcus aureus enterotoxins and eosinophil cationic protein were shown elevated[36], Chinese nasal polyp tissue has been acknowledged neutrophilic polyp. While one study reported higher prevalence of eosinophilic polyps[15], the other study group, within the same country, reported more neutrophilic inflammation[19]. After the histopathology of nasal polyps was re-evaluated within the same geographic location, there was a change over time of the inflammatory pattern from neutrophilic to eosinophilic[19]. Various criteria used to define ECRS without consensus should be the key reason for different findings that were reported. However, different locations of studied tissues taken without consensus should be the other factor of reporting various inconsistent findings.

Eosinophilic mucin is an important character of eosinophilic CRS. It demonstrates the eosinophil activation evidenced in eosinophilic CRS.[25] Nasal exudates from lateral nasal wall was assessed by Armengot et al. Eosinophilic inflammation found in the exudate had a strong correlation with the inflammatory pattern in the polyp tissue[6]. In our study, mucin had high diagnostic specificity and

positive predictive value of 100%. However, it had low sensitivity of 27.3% and modest accuracy with area under the ROC of 0.64. Therefore, clinicians should use mucin for histopathology assessment with cautions. Mucin when found is reliable to diagnose ECRS but it is not sensitive enough to exclude ECRS when absent.

Our study is the first that compared the correlation between nasal polyp's scraping with tissue eosinophil count at nasal polyp and sinus mucosa. We found that polyp's scraping only correlated with sinus mucosa. There is a previous study that compared eosinophils in nasal exudates from lateral nasal wall scraping and eosinophils in polyp tissue. They found that percentage of eosinophils in the lateral nasal wall scraping had a strong correlation with the one in the polyp tissue. Therefore, they recommended that nasal cytology is a convenient and non-traumatic technique to evaluate the inflammatory profile in CRSwNP patients.[6]

Although polyp apex is sensitive to define ECRS, it does not correlate with co-morbidity asthma. Instead we found a correlation when tissue eosinophilia was assessed from ethmoid mucosa and polyp pedicle. This is in agreement with a study of Weibman et al. They studied CRSwNP patients and found that eosinophilia in uncinate process tissue mucosa was a more coherent biomarker of co-morbidity asthma than eosinophilia in nasal polyp[37]. These findings may support the hypothesis of one airway disease[38, 39] as ethmoid mucosa, and uncinate process have the same epithelial lining of respiratory epithelium with the lower airway.

To date, number of tissue eosinophil is acknowledged the gold standard for diagnosing eosinophilic chronic rhinosinusitis. When this study aimed to assess difference of tissue eosinophil between sinonasal mucosa and polyp tissue, one reference was required and eosinophilic mucin was used. As eosinophilic mucin was not the gold standard, we acknowledged this should be a limitation of this study. A

deprivation of positive eosinophilic mucin patients may impact the accuracy of data interpretation. Sensitivity of the three locations of specimens was calculated based on the use of eosinophilic mucin as a reference. Thus, findings of this study should be used with precautions.

Our findings should suggest an appropriate site of tissue specimens taken for histopathology. The greatest density of eosinophil in polyp apex suggests that it should be taken because of high sensitivity and negative predictive value which is useful for screening purpose. When polyp apex biopsy can be used interchangeably with ethmoid mucosa biopsy, polyp biopsy can be performed at the clinic without the need for endoscopic sinus surgery. Histopathology assessment prior to surgery is helpful to predict further clinical severity and prognosis. When eosinophilic inflammation defined by high tissue eosinophilia together with eosinophilic mucin is present, aggressive topical corticosteroids should be considered[3, 4]. When eosinophilic inflammation is absent, low-dose long term macrolide therapy is suggested[40, 41]. Molecular therapy should be kept in mind in the future. Mepolizumab may be prescribed to patients with ECRS who have high tissue eosinophilia together with high serum eosinophilia as it has anti-interleukin 5 and specific anti-eosinophil activity[5]. When failed to medication, patients with ECRS require a big common cavity which all paranasal sinuses have been combined for maximum penetration of topical corticosteroid to sinonasal mucosa and for mechanical removal of thick eosinophilic mucin by nasal saline irrigation in the long term[42].

Conclusion

Based on findings of this study, polyp apex biopsy is suggested to assess histopathology of chronic rhinosinusitis with polyps. This is because polyp apex has the greatest density of eosinophil and gives the highest sensitivity for diagnosing eosinophilic chronic rhinosinusitis when using eosinophilic mucin as a reference. In

addition, when tissue was taken during endoscopic sinus surgery for histopathology study, polyp tissue should be chosen other than sinonasal mucosa. Taking the whole polyp including the polyp pedicle is suggested because the polyp pedicle is well correlated with co-morbidity asthma. Based on the principle of one airway, treating the co-morbidity asthma together with eosinophilic chronic rhinosinusitis has an impact on long term disease control.

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