ARAŞTIRMA RESEARCH ARTICLE

ROLE OF IMPRESSION CYTOLOGY METHOD IN THE ASSESSMENT OF FOLLOW UP AND SEVERITY OF SYMPTOMS FOR ALLERGIC RHINITIS

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ABSTRACT

Objective: *Impression cytology (IC) is an easily repeated technique for investigating several pathologies of conjunctiva and nasal mucosa. We aimed to find an easy reliable and objective method to evaluate severity of symptoms in allergic rhinitis. For this purpose we investigated usefulness of impression cytology as an diagnostic and follow up tool in patients with allergic rhinitis.*

Material and Method: *Thirty patients with allergic rhinitis were evaluated with both impression cytology and nasal biopsy. After medication, impression cytology and excisional biopsies were repeated, and results were compared.*

Results: *: A correlation was observed between biopsy and impression cytology slides before (r=0.72, p=0.000) and after medication (r=0.66, p=0.000). Following the therapy, biopsy scores (z=-4.11, p=0.000) and impression cytology scores (z=-3.50, p=0.0005) were significantly lower than to the earlier scores of the same patients. But no correlation was observed between the severity of pre and post-treatment symptoms and IC and nasal biopsy scores (p>0.05).*

Conclusion: *The results of this study is compatible with the allergic rhinitis and it's Impact on Asthma (ARIA) documentation on clinical evaluation of the severity of allergic rhinitis (AR) symptoms. However, in cases where optional nasal biopsy is considered, it shows that IC can be used reliably. Impression cytology is a quick, simple, painless, and noninvasive technique. It can be preferred to biopsy*

Key Words: *Allergic rhinitis, cytology, impression cytology, diagnosis.* ***Nobel Med 2011; 7(3): 18-21***

ALLERJİK RİNİTTE TAKİP VE SEMPTOM ŞİDDETİNİN DEĞERLENDİRİLMESİNDE İMPRESYON SİTOLOJİSİ METODU

ÖZET

Amaç: İmpresyon sitolojisi nazal mukoza ve konjunktiva patolojilerini araştırmada kolay tekrar edilebilen bir yöntemdir. Allerjik rinit semptomlarının şiddetini değerlendirmek için kolay, güvenilir ve objektif bir yöntem bulmayı amaçladık. Bu amaçla impresyon sitolojisinin allerjik rinitli hastalarda tanısal ve takip yöntemi olarak yararlılığını araştırdık.

Materyal ve Metod: Allerjik rinitli 30 hasta, hem impresyon sitolojisi hem de nasal biyopsi ile birlikte değerlendirildi. Tedaviden sonra impresyon sitolojisi ve eksizyonel biyopsiler tekrarlandı ve sonuçlar karşılaştırıldı.

Bulgular: Tedavi öncesi (r= 0,72, p= 0,000) ve sonrası (r= 0,66, p= 0,000) biyopsiler ve impresyon sitolojisi arasında korelasyon saptanmıştır. Tedavi sonrası, biyopsi skorları (z=-4,11, p=0,000) ve impresyon sitolojisi skorları (z=-3,50, p= 0,0005) aynı hastaların önceki skorlarına göre daha düşüktür. Fakat tedavi öncesi ile sonrası semptomların şiddetleri arasında ve impresyon sitolojisi ile biyopsiler arasında korelasyon saptanmadı (p>0,05).

Sonuç: Bu çalışmanın sonuçları allerjik rinit semptomlarının şiddetinin klinik olarak değerlendirmesi ilişkili alerjik rinitler ve astım üzerine etkisi (ARIA) dokümanları ile uyumludur. Nazal biyopsi düşünülen olgularda impresyon sitolojisi güvenle kullanılabilir. İmpresyon sitolojisi hızlı, basit, ağrısız ve invaziv olmayan bir yöntemdir. Bu teknik biyopsiye tercih edilebilir.

Anahtar Kelimeler: Allerjik rinit, sitoloji, impresyon sitolojisi, tanı **Nobel Med 2011; 7(3): 18-21**

INTRODUCTION

Allergic rhinitis (AR) is a global health problem affecting at least 10-25% of the world population and its prevalence is increasing.[1] AR is an immune-mediated, Th2-type disease of nasal mucosa. In AR, as a result of cytokine or mediator release, the nasal mucosa becomes infiltrated with inflammatory cells, leading to the characteristic symptoms of AR.[2-6]

The Allergic Rhinitis and its Impact on Asthma (ARIA) initiative has been developed in collaboration with World Health Organization for revision of diagnosis methods, treatments and management of the disease. Diagnosis of AR is based on patient's history, symptoms and the clinical findings. For asthma, there are objective measures of severity, such as pulmonary function tests, and well defined criteria for symptom severity. For atopic dermatitis there are clinical scores of severity, such as scoring atopic dermatitis.[1] However, for rhinitis there is no accepted objective measure of nasal symptoms. For this reason we investigated an objective method for evaluation of symptom severity in AR. Nasal biopsy and impression cytology (IC) are objective methods that can be used in AR. IC, an extensively used technique in ophthalmopathology, is not common in assessment of AR. [3,7-13]

In this study, we aimed to investigate concordance of symptom severity and objective diagnostic tools in AR to find a new, reliable objective criterion of follow up for management of the disease.

MATERIAL and METHOD

Patients

Thirty patients (12 males and 18 females; median age 42 years, ranging from 17 to 63 years) with 14 perennial and 16 seasonal AR who were followed up in the department of Ear, Nose and Throat at the Adnan Menderes University, School of Medicine, Aydın, Turkey, were included in this study.

A detailed clinical history and a complete physical examination were carried out for each patient. The diagnosis of AR was made, according to international guidelines, on the basis of history and skin prick test (SPT) positivity for allergens.[1] Each patient was questioned for rhinorrhea, nasal obstruction, nasal itching and sneezing before and after the treatment. Patients were expected to classify their complaints as "mild", "moderate" or "severe" based on the quality of life parameters.[1] There was no systemic diseases in any of the patients. No medication was taken at the time of investigation (with the exception of the study medication). All patients gave their informed consent before the study.

Impression cytology specimens

At first appointment, nasal mucosal impression material were collected with a strip of cellulose acetate paper (0.22mm, Millipore products Catalog GSWP04700) of approximately 5x5 mm in size. Before the procedure, the strip was soaked in distilled water for 8 hours and dried at room temperature. Strip was then pressed the anterior one-third of inferior turbinates and was held there for about 5 seconds. IC materials were stained with hemotoxylin and eosine (HE) and evaluated under bright light microscopy by a pathologist. Four microscopy fields were taken into consideration and the specimens were grouped (Table 1). IC and punch biopsy results were compared.

Nasal mucosal biopsy specimens

After handling impression cytology material, punch biopsies from the anterior one-third of inferior turbinate of nasal mucosa, were taken following the lidocaine injection. They were fixed in 10% formaline solution, embedded in paraffin, taken on slides and stained with HE and evaluated by a pathologist. Biopsies contained mucosa and submucosa. Biopsy slides were grouped according to the Table 2.

Medication

Patients were treated with mometasone furoate nasal sprays (50mg/puff). One puff of the medication was sprayed into each nostril from a metered-dose pump spray (delivering 50mg/ puff x 2) for a period of 28 days.

Pathologist didn't know about the medication, she evaluated IC and punch biopsy slides blindly.

Twenty-eight days later, IC and punch biopsies were repeated, and results were re-compared. Initial IC slides and nasal biopsies were also compared with after treatment IC slides and nasal biopsies.

Statistical analysis

Differences between two groups were evaluated using Wilcoxon Matched-Pairs Signed-Ranks test (SPSS, Chicago, IL, USA). Correlations between IC, nasal biopsy results and severity of clinical symptoms were assessed by the Kendall's rank-order correlation coefficient (Kendall tau-b, SPSS). Differences with a p-value of less than 0.05 were considered significant.

RESULTS

All patients were symptomatic. Severity of the symptoms before and after the treatment are summarized in Table 3. Patients with severe symptoms before the treatment (Figure 1-2) were 86% whereas it decreased to 17% following the treatment (Figure 3-4.) →

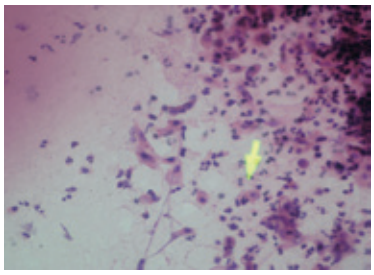

Figure 1. Infiltration of eosinophils in the epithelial cells in impression cytology slide.

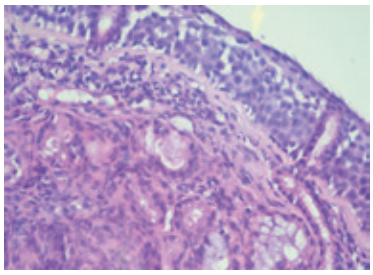

Figure 2. Infiltration of eosinophils in nasal biopsy in the same case.

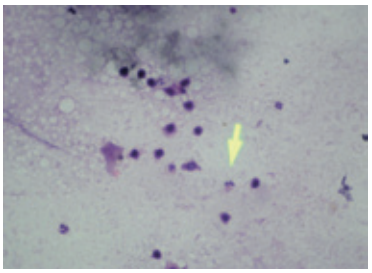

Figure 3. A few eosinophils in IC in the same case after topical glucocorticoid treatment.

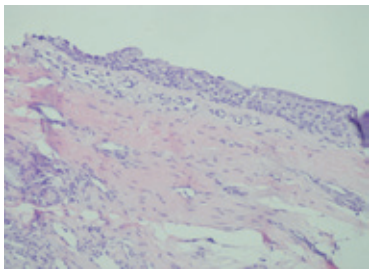

Figure 4. Nasal biopsy in the same case after topical glucocorticoid treatment

Table 1: Groups of the impression cytology.	
Group I	No free or intraepithelial eosinophils
Group II	Free eosinophils in the mucus secretion but no intraepithelial infiltration
Group III	Intraepithelial eosinophils and free eosinophils in the mucus secretion

Table 2: Groups of the nasal biopsies	
Group I	No eosinophils
Group II	Eosinophils in the submucosa but no intraepithelialial infiltration
Group III	Intraepithelial eosinophils and submucosal eosinophils

Table 3: Severity of the symptoms before and after the treatment		
	Before medication n (%)	After medication n (%)
Mild	2 (7)	11 (36)
Moderate	2 (7)	14 (47)
Severe	26 (86)	5 (17)

IC specimens were successfully obtained from all of the 30 patients. No local or systemic reactions were observed. Predominant leukocytes in the nasal biopsy and IC slides were eosinophils. Presence of mononuclear cells, mast and goblet cells were seen in both IC and nasal biopsy slides. Some of the IC slides had free mucus at the background. In these patients, mucus secretion was more dense and most of them had eosinophilic infiltration in the mucus. Patients' IC and nasal biopsy evaluations are summarized in Table 4. No correlation was observed between the severity of pre and post-treatment symptoms and IC and nasal biopsy scores. (p>0.05)

A correlation was observed between biopsy samples and IC before (r=0.72, p=0.000) and after medication (r=0.66, p=0.000). Following the therapy, biopsy scores were significantly lower than the ones before medication (z=-4.11, p=0.000). IC results were also decreased after therapy compared to the earlier scores of the same patients before medication (z=-3.50, p=0.005). Topical glucocorticoid treatment significantly inhibited accumulation and groups of eosinophils.

DISCUSSION

We aimed to find an easy and reliable and objective method to evaluate severity of symptoms in AR. There were a few studies indicating correlations between subjective nasal complaints and objective diagnostic methods.[1] Comparison of the biopsy, IC and severity of symptoms was not reported before.

Inflammatory cellular infiltrates of eosinophils and basophilic cells are hallmark of atopic nasal responses in AR. Nasal smears are widely used for the detection of eosinophilia in patients with AR. In addition, nasal lavage and biopsies are also performed.[3,14-17]

In this study, we used an alternative method, IC with nasal biopsy procedures and investigated the value of the IC according to nasal biopsy.

IC has been used to detect individual cellular changes on ocular surface or nasal mucosa. This technique was first described by Ebert et al. and Thatcher et al, in 1977.[14-15]

By means of IC technique, cells denuded of epithelial surfaces can be collected with a strip of paper. It is an evaluation of cluster of cells can give fruitful results. Cell infiltration into the intraepithelial areas and even alteration in epithelial cells can directly be observed. However, photographic illustration yields a nonhomogeneous appeareance because of differences of layers of the clustered epithelial cells. Thus, nonhomogeneous distribution of eosinophils is one of the disadvantages of the IC evalution. To eliminate this disadvantage, four microscopy fields were taken into consideration. Future studies are needed to be tried various techniques of actually counting eosinophils per highpower field, as is done in counting WBCs and RBCs.[3,18,19]

IC technique allows us to see not only the inflammatory cells but the infiltration and its relation to the epithelial cells. In this study, we saw intraepithelial eosinophils in the mucosal epithelial cells.(Group III) Intraepithelial cell infiltration is a sign of active infection which can also be used in the therapy follow-up. →

Indeed, in this study severity of the symptoms and objective diagnostic methods (nasal biopsies and IC) were not related (p>0.05) but IC and nasal biopsies were significantly related (r=0.72, p=0.00). We think that if there is any necessity for optional biopsy, IC method is reliable to be used.

CONCLUSION

In summary, IC, a quick, simple, painless and noninvasive technique, is particularly appropriate not only for the diagnosis of eosinophilia in AR, but also for serial evaluations of the follow-up therapy, in outpatient settings.

Table 4: Patients and their impression cytology and nasal biopsy results

	Impression cytology		Nasal biopsy	
	Before medication n (%)	After medication n (%)	Before medication n (%)	After medication n (%)
Group I	3 (10)	14 (47)	1 (3)	15 (50)
Group II	13 (43)	11 (36)	12 (40)	10 (33)
Group III	14 (47)	5 (17)	17 (57)	5 (17)

The result of this study is compatible with ARIA document on clinical evaluation of the severity of AR symptoms. However, in cases where optional nasal biopsy is considered, it seems that IC can be used reliably.

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