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Microbiologic and cytologic analysis of nasal polyps

Polyposis in the nose arises as a result of complicated process in which inflammatory changes of upper and lower airways, as well as the specific properties of the ethmoid and the anatomical conditioning of the middle nasal meatus, are extremely important. Nasal polyps formation due to previous sinus infection creates the conditions to make the inflammation persistent. Some investigators who cultured polyps for aerobic and anaerobic bacteria and other microorganisms stress the close relation of the number of bacteria and neutrophilia in polyp's tissue (4). On the other hand, some authors found the eosinophils rather than neutrophils in stroma of nasal polyps and indicated the significance of allergic inflammation in their generation (8, 10). However, local eosinophilia found histologically are not usually allergy dependent (11). In both, neutrophilic and eosinophilic nasal polyps the multivariety of microorganisms was observed rather than one predominant type (2, 13). We determined in this study the microbiology of nasal polyps removed during endoscopy and compared the quantity of the inflammatory cells infiltrating the polyp tissue with the frequency of microorganisms.

MATERIAL AND METHODS

C u l t u r e s. Polyps collected from 22 patients who underwent nasal polypectomy were analyzed microbiogically and histologically for the presence of inflammatory cells. One part of nasal polyps was homogenized and inoculated onto bacteriologic media such as chocolate agar, Mac Conkey's agar, blood agar, mannitol salt agar, Sabouraud's dextrose agar and agar incubated in aerobic conditions for aerobic organisms and fungi. The second part of homogenated tissue was diluted in thioglicolate broth and incubated at 37°C for 4 days and then inoculated onto Schaedler agar for the anaerobic bacteria. Colonies of bacteria were identified according to conventional procedures and determined by species according of cfu (colony forming unit) per gram of tissue (12).

P a t i e n t s. In the group of 20 patients, 17 suffered from bilateral nasal polyps. Thirteen patients gave the history of bronchial asthma, among them 5 had the symptoms of aspirin intolerance. Eight patients underwent nasal polypectomy at least 2 times in the past and one of them – 10 times. The age of patients ranged from 28 to 67 years, the median was 51.8 years. The examined group consisted of 14 males and 8 females. All patients were off antibiotics for at least 2 weeks at the time of polypectomy.

C y t o l o g i c a n a l y s i s o f n a s a l p o l y p s. We performed semiquantitative cytologic analysis of nasal polyps sections stained with H+E with conventional light microscopy at magnification x 400. The cells were evaluated in 10 randomly selected fields and their quantity was graded from 0 to 3.

RESULTS

The results of cultures of nasal polyps are displayed on Table 1. Bacteria were isolated from 20 patients. The culture was sterile in 2 samples. The predominant microorganisms were: *S. aureus* present in 10 samples, *S. epidermidis* – in 8 samples and *Corynebacterium sp.* – in 6 samples. Gram-negative bacteria have been found less frequently in single samples of nasal polyps. No anaerobes were found in polyps' tissue. Of these positive 20 polyps 1 homogenate demonstrated the growth of *H. parainfluenzae* at levels $13x10^3$cfu/gm, 1 – *S. epidermidis* at $9x10^3$cfu/gm, 1 – *Corynebacterium sp.* at $8x10^3$cfu/gm, 1 – *S. aureus* at $6x10^3$cfu/gm, 1 – *S. epidermidis* at $3x10^3$cfu/gm, 1 – *Corynebacterium sp.* at $3x10^3$cfu/gm, 1 – *S. aureus* at $2x10^3$cfu/gm, 5 – *S. aureus* at $1x10^3$cfu/gm, 3 – *S. epidermidis* at $1x10^3$cfu/gm, 4 – *Corynebacterium sp.* at $1x10^3$cfu/gm, 1 – *P. aeruginosa* at $1x10^3$cfu/gm and 1 – *Citrobacter sp.* at $1x10^3$cfu/gm.

Table 1. Results of aerobic cultures of 22 nasal polyps

Patient no	Organism	cfu/gm	Patient no	Organism	cfu/gm
1	*S. epidermidis*	$1x10^3$	3	*P. aeruginosa*	$1x10^3$
2	*H. parainfluenzae*	$13x10^3$	7	*S. xylosus*	$1x10^3$
	S. aureus	$1x10^3$			
4	*S. epidermidis*	$9x10^3$	13	negative	
	Corynebacterium sp.	$8x10^3$			
	Citrobacter sp.	$1x10^3$			
5	*S. epidermidis*	$1x10^3$	14	*Str. mitis*	$1x10^3$
6	*S. aureus*	$1x10^3$	15	*S. epidermidis*	$1x10^3$
8	*S. epidermidis*	$1x10^3$	16	*S. aureus*	$1x10^3$
9	*S. aureus*	$2x10^3$	18	negative	
	Corynebacterium sp.	$1x10^3$			
10	*S. epidermidis*	$1x10^3$	19	*Corynebacterium sp.*	$1x10^3$
	S. aureus	$1x10^3$			
	Corynebacterium sp.	$1x10^3$			
11	*S. epidermidis*	$3x10^3$	20	*S. aureus*	$6x10^3$
12	*S.epidermidis*	$1x10^3$		*Corynebacterium sp.*	$3x10^3$
	S. aureus	$1x10^3$			
17	*S. aureus*	$1x10^3$			
21	*S. aureus*	$1x10^3$			
22	*S. aureus*	$1x10^3$			

The evaluated quantity of the eosinophils and neutrophils is showed on Table 2. Our findings indicate that the most numerous cell infiltrations were observed in the samples with the greatest number of microorganisms and the eosinophils predominated among nflammatory cells of polyp's tissue. We noticed that nasal polyps of patients with a history of asthma demonstrated more often positive cultures and the number of cultured microorganisms was greater than in polyps of patients without asthma. The nasal polyps taken from patients who underwent polypectomy more than one time presented a larger variety and number of microorganisms.

Table 2. Cytologic analysis of nasal polyps

Patient no	Eosinophils	Neutrophils
1	2	1
2	3	0
3	1	1
4	2	1
5	1	1
6	2	1
7	1	1
8	1	1
9	3	0
10	2	1
11	1	1
12	1	1
13	1	1
14	1	2
15	1	1
16	0	2
17	3	0
18	2	0
19	1	1
20	1	1
21	1	1
22	3	0

0 – the lack or single cells, 1 – few cells, 2 – numerous cells, 3 – dense cell infiltrations

DISCUSSION

The studies on microbiology of nasal polyps show the high presence of aerobes and low anaerobes (4). A lower isolated rate of anaerobes was found in the bacteriology of the ethmoid sinus (3, 6, 7, 9). We cultured no anaerobes in our experiment and *S. aureus* was predominant bacteria. Similar results presented some authors (1, 5). *S. aureus* represents a risk factor for subsequent invasive infections. A higher cultural rate of *Staphylococcus epidermidis*, a possible nasal contaminant, presented some of our samples. Similar data were published in work of other authors (3, 6, 7, 9). We found two samples with no bacterial growth. The presence of sterile cultures among others is explained by antimicrobial activity of nasal polyps (4).

When comparing the pathogens obtained from nasal polyps of patients with asthma and without asthma, we can state that a greater variety and number of bacteria was accompanied in polyps of patients with asthma (Tab. 1). Data from another study show that the number of neutrophils correlates with the number of bacteria (4). Our findings do not confirm this relationship and indicate that the most numerous cell infiltrations containg eosinophils, were observed in the samples with the greatest number of pathogens in the group of patients who underwent more than one polypectomy. These patients had simultaneously a history of asthma.

CONCLUSIONS

In summary, we can conclude that the polymicrobe flora can be associated with nasal polyps and the number of cell infiltrations (containing lot of eosinophils) correlates well with the number of bacteria cultured.

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SUMMARY

The retrospective analysis of microbial flora and inflammatory cells of polyp's tissue in 22 patients who underwent nasal polypectomy was carried out. One part of nasal polyps was homogenized and inoculated onto bacteriologic media (chocolate agar, Mac Conkey's agar, blood agar, mannitol salt agar, Sabouraud's dextrose agar) and incubated in aerobic conditions for aerobic organisms and fungi. The second part of polyps was placed in thioglycolate broth and incubated at 37°C for 4 days and then inoculated onto Schaedler agar for the anaerobic bacteria. Bacteria were isolated from 20 patients. The culture was negative in 2 samples. The predominant microorganisms were: *Staphylococcus aureus* present in 7 samples, *Corynebacterium sp.* – in 6 and *Staphylococcus epidermidis* – in 5 samples. Gram-negative bacteria have been found less frequently, only in single samples of nasal polyps. No anaerobes were found in the specimens. We evaluated the quantity of the inflammatory cells infiltrating the polyp and compared it with the number of bacterial colonies. Our findings indicate that the most numerous cell infiltrations were observed in the samples with the greatest number of microorganisms and the eosinophils predominated among inflammatory cells.

Analiza mikrobiologiczna i cytologiczna polipów nosa

Przeprowadzono retrospektywną analizę mikrobiologiczną i cytologiczną polipów nosa 22 pacjentów po przebytej polipektomii. Próbki polipów nosa I grupy pacjentów po homogenizacji były hodowane na odpowiednich podłożach w warunkach tlenowych celem wyizolowania bakterii tlenowych i grzybów. Polipy nosa II grupy były umieszczane w podłożu z tioglikolem sodu i inkubowane w temperaturze 37°C przez 4 dni, a następnie wysiewane na agar Schaedlera w celu wyizolowania bakterii beztlenowych. W dwóch przypadkach nie uzyskano wzrostu bakterii. Dominującym patogenem był *Staphylococcus aureus* – obecny w siedmiu próbkach. Z pozostałych próbek polipów nosa wyhodowano: *Corynebacterium sp.* – w sześciu i *taphylococcus epidermidis* – w pięciu przypadkach. Gram-ujemne bakterie hodowano znacznie rzadziej niż bakterie Gram-dodatnie. Nie wyhodowano żadnych bakterii beztlenowych. Z 20 próbek polipów nosa w jednym przypadku wyhodowano *Haemophilus parainfluenzae* w ilości $13x10^3$cfu/gm, w jednym – *Staphylococcus epidermidis* w ilości $9x10^3$cfu/gm, w 1 – *Corynebacterium sp.* w ilości $8x10^3$cfu/gm, w jednym – *Staphylococcus aureus* w ilości $6x10^3$cfu/gm, w jednym – *Staphylococcus epidermidis* w ilości $3x10^3$cfu/gm, w jednym – *Corynebacterium sp.* w ilości $3x10^3$cfu/gm, w jednym – *Staphylococcus aureus* w ilości $2x10^3$cfu/gm, w pięciu – *Staphylococcus aureus* w ilości $1x10^3$cfu/gm, w trzech – *Staphylococcus epidermidis* w ilości $1x10^3$cfu/gm, w czterech – *Corynebacterium sp.* w ilości $1x10^3$cfu/gm, w jednym – *Pseudomonas aeruginosa* w ilości $1x10^3$cfu/gm i w jednym – *Citrobacter sp.* w ilości $1x10^3$cfu/gm. Metodą półilościową oznaczaliśmy liczbę komórek zapalnych w tkance polipa i porównywaliśmy ją z liczbą kolonii bakteryjnych. Stwierdziliśmy, że najliczniejsze nacieki komórkowe występowały w próbkach polipów nosa z największą liczbą wyhodowanych mikroorganizmów. Wśród komórek zapalnych dominowały eozynofile.