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In vitro histamine release from nasal mucosa upon bacterial antigens

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SUMMARY

Polyposis of the nose has been discussed as to be caused by hypersensitivity. Therefore, in vitro histamine release from nasal mucosa upon different bacterial and synthetic antigens was measured using a radioenzymatic assay. Also anti-human IgE mediated and basic release from specimens were determined. In total, 12 patients with polyposis and 21 patients with septal deviation or conchal hyperplasia were tested.

Histamine release was significantly increased upon anti-IgE in polyposis only. The incubation of mucosal biopsies with the antigens on the average caused a slight elevation of histamine release in the same group. In contrast, specimens of patients with other diseases did not respond to anti-IgE or bacterial antigens.

The specific increase of histamine-release from polyps after challenge with anti-IgE and similar spontaneous release in all tested individuals indicate a different pattern of mucosal reactivity in patients suffering from nasal polyposis.

INTRODUCTION

Hypersensitivity is an important immunological disease of the respiratory tract. This is particularly true with regard to IgE-mediated release of substances from mast cells. Some forms of rhinitis (Albegger, 1979; Fuchs, 1979) are typical examples for such an allergy. An allergic component has also been stressed in the development of nasal and sinusal polyposis. Evidence for this idea is provided by the elevated release of IgE and histamine from the mucosa of polyps in allergic patients. Moreover, clinical links between allergic rhinitis, sinusitis, polyposis and bronchial asthma support this idea (Baenkler et al., 1983; Calenoff et al., 1983; Marks, 1982). Finally, infiltration of lymphocytes and plasma cells regularly found in mucosal biopsies of patients suffering from polyposis may be a sign of an immunological event (Bussutil et al., 1976). However, by morphological examination of polyps, it is impossible to obtain evidence for allergy as a pathogenetic factor in a given case (Baumgarten et al., 1980; Waller et al., 1976). There are some doubts about allergy as a general pathogenetic factor of nasal polyposis (Small et al., 1982). Because of difficulties in establishing assay systems, only limited information about functional aspects is available. The

demonstration of histamine release from biopsy or polyps upon antigens in vitro is an approach for the elucidation of possible allergic mechanics (Baenkler et al., 1983; Bork and König, 1981). Therefore, this study was designated to evaluate particular patterns of histamine release in patients suffering from polyposis and other diseases focusing upon bacterial components as a possible causative agent.

MATERIALS AND METHODS

Patients

In total, 33 patients were studied. Twelve of them suffered from polyposis which was confirmed during surgical intervention and by histological examination. The remaining 21 patients had septal deviation, conchal hyperplasia or other diseases, but they had never experienced polyposis. Patients with any proved allergy or with inflammatory diseases of the nose as well as patients treated with corticosteroids or antihistaminics were excluded from this group.

Processing of the biopsy

Biopsy material was obtained from all patients using systematic or local anesthesia. In patients with nasal polyposis, biopsies were taken from hyperplastic mucosa or polyps, in patients with septal deviation, nasal mucosa – mostly of the inferior turbinate – was biopsied. Six specimens were obtained from each patient. Each specimen of the mucosa was immediately placed into Hanks' solution. Antigenic solutions (Bencard, London) containing staphylococcus aureus, Streptococcus viridans or mixed streptococcus as well as synthetics were individually added to each sample. Specimens mixed either with anti-human IgE or medium alone, omitting antigenic solutions, served as control. All samples were incubated at 37° C for 30 minutes in a shaking water bath. The liquid phase was then separated and stored at $-20°$ C until further processing. The dry weight of the particles was obtained after lyophilization.

Determination of histamine

Histamine was assayed using a radioenzymatic technique (Bork and König, 1981; Subramanian and Mitznegg, 1978). Sensitivity was 1 ng/ml. The amount of histamine within the liquid phase was corrected according to the dry weight of the particles. Also the results were normalized to the basic release.

RESULTS

Basic and anti-IgE-induced histamine release

Patients with polyposis: Biopsy specimens in pure medium released in the mean 5.92 ng histamine from each mg of mucosa. This amount increased to 12.6 ng after challenge with anti-human-IgE. The difference was significant ($p < 0.05$). Patients with conchal hyperplasia and other diseases: The biopsies released in

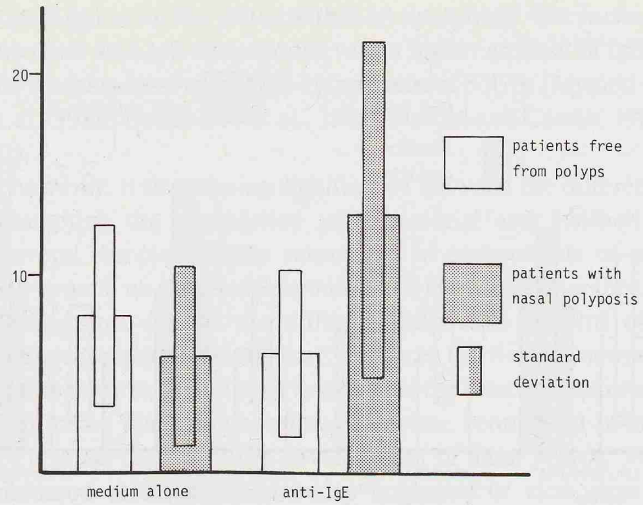

Figure 1. Histamine release by anti-human IgE and medium alone of patients with different diseases. Mean value of standard deviation are listed in ng histamine/mg tissue.

the mean 7.74 ng histamine per mg of mucosa during incubation with pure medium. After challenge with anti-human IgE 5.95 ng were assayed. There was no significant difference. Also separate evaluation of patients suffering from conchal hyperplasia, septal deviation or other diseases showed no significant differences.

Figure 1 summarizes histamine release upon anti-human IgE and medium alone of patients with different diseases.

Bacterially induced histamine release

Patients with polyposis: Stimulation with bacterial solutions caused a various pattern of histamine release. Each patient released significant histamine at least from one specimen. Because of this individual response, no general difference was objectivated, although the mean histamine release was increased to 7.03, 7.86 and 8.0 ng per mg of mucosa after incubation with staphylococcus aureus, mixed streptococci or synthetics respectively.

Patients with conchal hyperplasia: Upon different bacterial solutions, histamine release from biopsy was 5.42, 4.57 and 4.44 ng per mg of mucosa, using streptococcus aureus, mixed streptococci or synthetics respectively. Separate analysis of the different groups of patients showed no typical pattern. There was no significant difference neither to basic nor to anti-human IgE-triggered histamine release.

Figure 2 summarizes the histamine release upon different antigenic solutions.

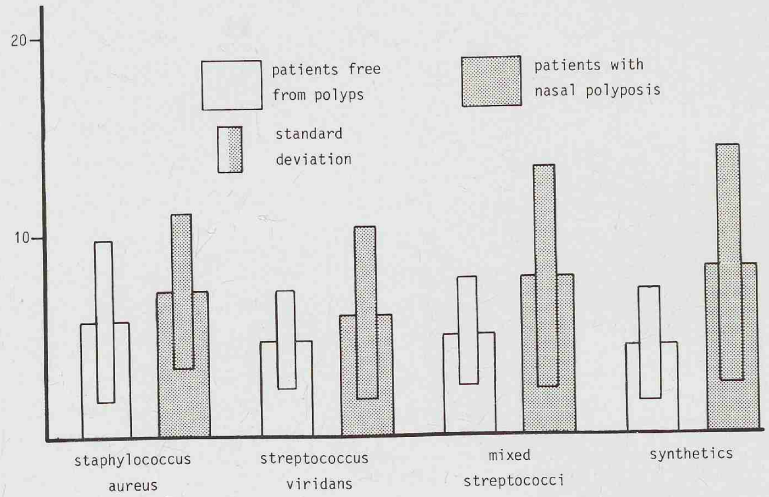

Figure 2. Histamine release upon different antigenic solutions of patients with different diseases. Mean value and standard deviation are listed in ng histamine/mg tissue.

Comparison of histamine release from corresponding speciments

A significant difference ($p < 0.05$) between patients suffering from conchal hyperplasia and those with polyposis was objectivated with regard to the challenge with anti-human IgE, as mucosa of polyps responded regularly, whereas mucosa of hyperplastic concha did not. In addition, histamine release upon mixed streptococci and synthetics was significantly higher when using specimens of polyps than in the case of hyperplastic conchae biopsies being used.

DISCUSSION

As demonstrated in former experiments, the reactivity of polyps is different from abnormalities like hyperplasia or septal deviation (Baenkler et al., 1983). However, this was significant in one series only.

Most important is the increase of histamine release upon anti-IgE from mucosa of the polyps as compared to the spontaneous output. In contrast, mucosa of other patients did not release more histamine after challenge with anti-IgE. It is of further importance that there was a similar basic histamine release from mucosa no matter whether the patient was suffering from polyposis or from other diseases. Therefore, the difference between positive and negative controls, i.e. basic and anti-IgE triggered controls, represents the crucial point of our experiments.

Since basic histamine release corresponds to the number of mast cells, their content of mediators and their instability to mechanical or biological irritation,

similar results suggest similar status within all specimens. The increase of histamine release upon anti-IgE corresponds with a higher amount of IgE-molecules bound to the membrane of mast cells in the tissue of polyps (Mygind et al., 1979; Calenoff et al., 1983; Drake-Lee et al., 1984; Marom and Casale, 1983; Wassermann, 1983).

Surprising, however, is the missing significance between the different groups of patients concerning the stimulation with bacterial and synthetic antigens. Although several samples clearly responded to components of at least one bacterium, in general no significance could be objectivated after incubation with identical components due to the differing individual patterns of histamine release. Moreover, negative results may be due to the limited number of tested antigens. In this context, the missing response to the selected bacteria in patients suffering from other diseases underlines the close connection of an increased releasability of mast cells with the development of nasal polyps. The question whether increased releasability precedes polyposis or vice versa cannot be answered by our experiments.

ZUSAMMENFASSUNG

Als eine der Ursachen einer Polyposis nasi werden allergische Reaktionen diskutiert. Daher wurden Biopsiepartikel in vitro mit verschiedenen Antigenen inkubiert und die dabei freigesetzte Histaminmenge mittels eines Radioenzymassay gemessen. Zugleich wurden anti-IgE-vermittelte und spontane Ausschüttungen ermittelt. Von den insgesamt 33 untersuchten Patienten litten 12 an einer Polyposis nasi, die übrigen an einer Septumdeviation, Muschelhyperplasie oder anderen nicht-entzündlichen Erkrankungen.

Nur aus der Schleimhaut von Patienten mit Polyposis nasi wurde durch anti-IgE eine signifikante Steigerung der Histamin-Ausschüttung induziert. Auch war bei ihnen die Histaminfreisetzung auf die verschiedenen bakteriellen und synthetischen Antigene durchschnittlich geringfügig vermehrt.

Die Ergebnisse zeigen ein unterschiedliches Verhalten der Schleimhaut bei Polyposis nasi, verglichen mit anderen nasalen Erkrankungen. Ursache hierfür ist die unterschiedliche Reagibilität der Mediator-haltigen Zellen in den untersuchten Geweben.

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