

Tryptase in nasal fluid is a useful marker of allergic rhinitis

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Tryptase is a mast cell-specific marker of degranulation. To investigate the possible diagnostic value of tryptase in allergic rhinitis, we measured the levels in both serum and native nasal fluid with a sandwich RIA-assay (Pharmacia). Twenty-three allergic patients and five patients with chronic ethmoidal sinusitis were included. Eighteen of the 23 allergic patients were tested within the pollen season or had perennial rhinitis; the remainder were tested at least 1 month out of the pollen season. None of the patients had detectable serum tryptase (>0.1 ng/ml). Also patients with chronic ethmoidal sinusitis showed no tryptase in nasal fluid. One of seven allergic patients tested out of season had slightly increased nasal tryptase of 1.8 ng/ml. In patients with active nasal allergy, the tryptase in nasal fluid ranged from 6.4 ng/ml to 640 ng/ml with a mean of 101 ng/ml and SD 173. These results show a clear distinction between active and non-active nasal allergy and other non-mast-cell-related nasal disease. Further, nasal tryptase release by natural allergen exposure is even higher than that observed in allergen challenge tests.

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The role of nasal mast cells in the immediate hypersensitivity reaction of allergic rhinitis patients has been well documented. Mast cell degranulation leads to a biphasic mediator release. In the early phase, a histamine peak is shown in many patients, and at least 50% also show a late phase reaction with a second histamine peak following at least 4 h after allergen challenge. In addition to the immediate release of mediators in the early phase and the secondary mast cell activation in the late phase, there exists a third mechanism of slow release of proteins from mast cells during this time. In the early 1960s, mast cells were reported to have trypsin-like activity (3). In 1981, this activity was shown to belong to a tryptic protease and was called tryptase (6). Tryptase, as a major protein component of mast cells, was found in both connective tissue and mucosal mast cells (2). It was also found in trace amounts in basophils, but not in other human cell types (4). Tryptase is a serine endoprotease that cleaves at the carboxy-site of lysine or arginine residues. The TAME (*N* α -*p*-tosyl-L-arginine methyl ester) hydrolytic activity is known and has often been reported in nasal secretions. Tryptase may account for most of this activity. In its physiological form, tryptase is a tetrameric enzyme with a molecular mass of 134 kDa, which is only stable in the presence of sulfated proteoglycans. Under reducing conditions, tryptase dissociates into subunits of

34 000 and 37 000 molecular weight. This protease cannot be inhibited by any of the known protease inhibitors.

The physiological role of tryptase is not yet clear. Among its numerous functions is a strong interaction between tryptase and heparin which may drain the diffusion away from the site of inflammation. It can also destroy the clotting ability of fibrinogen, leading to anticoagulant function. Further, tryptase can cleave C3 to produce C3a anaphylatoxin (5), which increases vascular permeability and causes bronchospasm. Also, it has been demonstrated that tryptase activates latent collagenases from rheumatoid synovial tissue. An elevated level of tryptase in human serum has been observed after acute anaphylaxis and in systemic mastocytosis (7). The enzyme is also increased in a skin chamber model following cutaneous testing (8). In the nasal lavage fluid following allergen provocation, tryptase was found to be elevated in allergic, but not in healthy, persons (1). The levels of nasal tryptase correlate with the intensity of nasal symptoms in the patients. Previously, studies of tryptase in nasal fluid have been undertaken solely with allergen challenge. In the present inquiry, we investigated serum and nasal fluids of allergics and non-allergics under natural allergen exposure, in order to show natural mast cell activity in disease.

Material and methods

Nasal fluid and blood samples were taken from 23 allergics, and five subjects with chronic ethmoidal sinusitis of the infectious type were used as a control group. All patients underwent tests for allergy including medical history, skin prick-test, total and specific IgE tests and nasal challenge tests. Serum was taken from venous blood and stored at -80°C after centrifugation. Native nasal fluid was obtained by inserting cotton gauze strips beneath the lower turbinate for 10 min. After removal, the gauze was centrifuged off in special tubes. The resulting nasal fluid was also stored at -80°C for the tests. Tryptase levels were determined by a sandwich RIA test assay as described (9) using ^{125}I -labeled antibodies in coated tubes (Pharmacia, Uppsala, Sweden). Briefly, the fluid was incubated in the tube, washed, incubated with the second antibody, washed again and the activity measured in a gamma counter using standards for calibration. Eighteen of the 23 investigated allergic subjects were tested within the pollen season (birch and/or grass pollen) or had perennial allergic rhinitis, mostly attributed to house-dust mites. Five of the allergics sensitive to pollen (birch or grass) were tested at least 1 month after the pollen season had finished in our region and were found to be free of symptoms.

Results

Tryptase levels in serum were not found to exceed 0.1 ng/ml in all patients. None of the patients with chronic sinusitis demonstrated a detectable tryptase level in nasal fluids. In patients with pollen allergy out of the pollen season, four of five also showed no detectable tryptase levels; one of these patients showed an only slightly increased level of 1.8 ng/ml (mean 0.5 ng/ml , SD 0.85). In contrast, nasal tryptase levels from patients with active allergy ranged from 6.4 ng/ml to 640 ng/ml with a mean of 101 ng/ml and SD 173 (Fig. 1). Tryptase levels in nasal fluid showed no correlation to the total IgE level in either serum (Fig. 2) or nasal fluid.

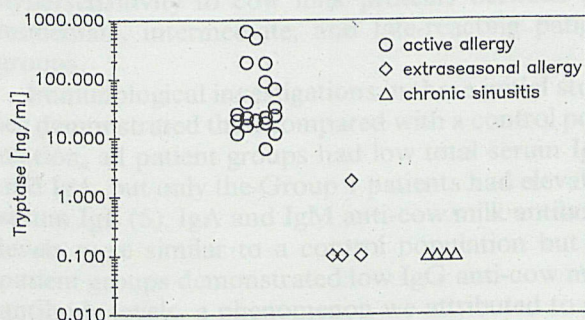


Fig. 1. Tryptase levels in native nasal fluid measured in patients with different diseases as shown within the plot.

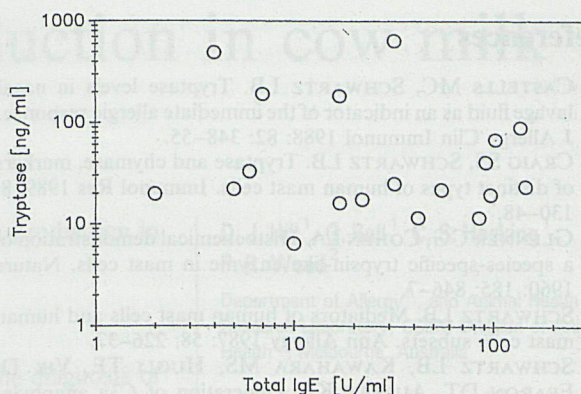


Fig. 2. There is no correlation between tryptase levels in nasal fluid and total IgE levels in serum in 18 patients with active nasal allergy.

Discussion

Tryptase has been demonstrated to be elevated in nasal fluid following allergen challenge. In this study we investigated patients with or without active nasal allergy and patients with chronic sinusitis as controls. The results show that active nasal allergy is associated with elevated tryptase levels in nasal fluid. These levels proved to be even higher than those found by allergen challenge tests (1). Whether this can be attributed to the use of native nasal fluid instead of nasal lavage or is indeed due to greater mast cell activation has yet to be determined in a comparative study. On the other hand, allergic patients tested out of the pollen season showed no increase or only slightly increased nasal tryptase levels. Patients with severe inflammatory nasal diseases without an allergic component show no elevated nasal tryptase levels. Thus, we conclude that elevated nasal tryptase levels can be provoked not only by allergen challenge as described (1), but also by natural allergen exposure, which seems to induce continuous mast cell degranulation and release of tryptase into nasal fluid, independently of the individual patient's specific allergen. This study also demonstrates that isolated pollen allergy leads to an at least partly reversible nasal disease and that mast cell activity is not detectable out of the pollen season. Therefore, we can further conclude that if an allergic patient has nasal symptoms out of the pollen season, these problems should not be related to enhancing mast cell activity, but must be explained by different mechanisms. Further, our results have shown that mast cell activation can also be demonstrated without artificial allergen challenge; in other words, simply by means of natural allergen exposure in untreated patients. Thus, we have found an objective method to measure natural allergen challenge in addition to the patient's medical history. Further studies are necessary to establish normal values for healthy persons in a greater population.

References

- CASTELLS MC, SCHWARTZ LB. Tryptase levels in nasal lavage fluid as an indicator of the immediate allergic response. *J Allergy Clin Immunol* 1988; 82: 348-55.
- CRAIG SS, SCHWARTZ LB. Tryptase and chymase, markers of distinct types of human mast cells. *Immunol Res* 1989; 8: 130-48.
- GLENNER CG, COHEN LA. Histochemical demonstration of a species-specific trypsin-like enzyme in mast cells. *Nature* 1960; 185: 846-7.
- SCHWARTZ LB. Mediators of human mast cells and human mast cells subsets. *Ann Allergy* 1987; 58: 226-37.
- SCHWARTZ LB, KAWAHARA MS, HUGLI TE, VIK D, FEARON DT, AUSTEN KF. Generation of C3a anaphylatoxin from human C3 by human mast cell tryptase. *J Immunol* 1983; 130: 1891-5.
- SCHWARTZ LB, LEWIS RA, AUSTEN KF. Tryptase from human pulmonary mast cells: purification and characterization. *J Biol Chem* 1981; 256: 11939-43.
- SCHWARTZ LB, YUNGINGER JW, MILLER J, BOKHART R, DULL D. Time course of appearance and disappearance of human mast cell tryptase in the circulation after anaphylaxis. *J Clin Invest* 1989; 83: 1551-5.
- SHALIT M, SCHWARTZ LB, ALLMEN C, ATKINS PC, LAVKER RM, ZWEIMAN B. Release of histamine and tryptase during continuous and interrupted cutaneous challenge with allergen in humans. *J Allergy Clin Immunol* 1990; 86: 117-25.
- WENZEL S, IRANI AM, SANDERS JM, BRADFORD TR, SCHWARTZ LB. Immunoassay of tryptase from human mast cells. *J Immunol Methods* 1986; 86: 139-42.

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