The enzymology of nasal secretion

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SUMMARY

Normal human nasal fluid contains several enzymes of the intermediary metabolism as well as a specific protease inhibitor, which inhibits trypsin, chymotrypsin and leucocytic proteases. During the course of acute and chronic nasal and paranasal sinus infections the inhibitor level varies. The inhibitor level is an indicator of poor healing. It is possible too, to differentiate viral rhinitis from bacterial or allergic or atrophic rhinitis by a significant increase of the activities of the enzymes GOT, LDH and CPK.

HUMAN nasal secretion has been generally regarded as biochemically empty. It has therefore remained of little interest for basic research. In most relevant studies it is simply stated that it consists of 95% water and contains only a few electrolytes, the long-kown lysozyme, immunoglobulins and some glycoproteins difficult to define.

At the Münich ENT Dept. we have been concerned for some time with the enzymology of human nasal secretion, the term "enzymology" includes the processes which eventually regulate the actual enzymatic action. The action of the enzymes is influenced not only by the pH value and the ion strength of the milieu but also by specific effectors which can activate or inhibit enzymatic action. Human nasal secretion has a high antiproteolytic activity, comparable with the antiproteolytic activity of the serum. At our clinic we succeeded in demonstrating that 80% of this inhibition comes from a specific inhibitor which apparently is produced in the mucous membrane and given off into the secretion. It has not been possible so far to demonstrate this particular inhibitor in the serum or in other tissues. In contrast to the serum inhibitors it is acid-stable and has a molecular weight of about 14.000.

The physiological function of this inhibitor is based on the fact that it can inactivate trypsin and chymotrypsin as well as proteolytic enzymes from breaking-down leucocytes.

In infections of the mucous membranes leucocytes pass from the intravasal space into the secretion. After phagocytosis of infectious matter these leucocytes break down. The liberated proteases are powerful inflammation mediators which increase inflammation if the natural pool of inhibitors is not sufficient. As Reichert and Hochstrasser at our clinic were able to show, there actually occurs

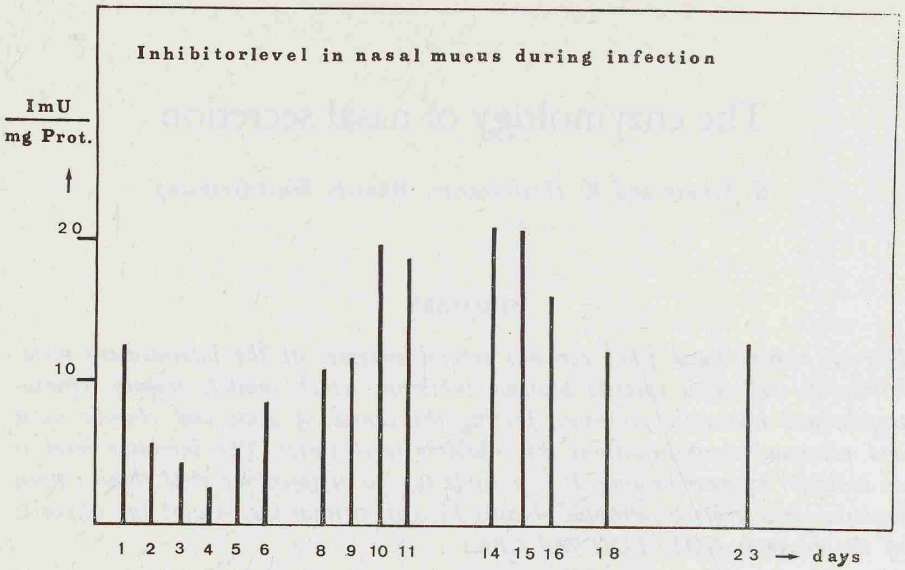


Figure 1. Inhibitor level in nasal secretion in the course of acute rhinitis.

an analytically demonstrable fall of the inhibitor concentration in the course of various rhinopathies (Figure 1).

The lowest amount of inhibitors in the nasal secretion, e.g. in acute coryza, was found at a time corresponding to the maximum of subjective clinical symptoms, when the inhibitor may completely disappear and free proteolytic activity may occur. When there is a poor tendency to healing and the infection does not clear up despite aimed physical and chemotherapeutic treatment, the inhibitor level remains low. Estimation of the low-molecular, mucosaspecific inhibitor can thus be adduced for diagnosis in the assessment of cases of rhinitis running a chronic course, so as to objectivate the symptoms of these patients.

A direct proof that the inhibitor is really required in pathological cases is easily adduced. Owing to its low molecular weight of 14.000 the inhibitor is stable against protein-precipitating reagents, i.e. it can be liberated in an active form from the inactive protease inhibitor complexes. In purulent nasal secretions which show no antiproteolytic activity, inhibitor can be demonstrated again after removal of protein. Direct proof that these inhibitor complexes exist through reaction between the specific mucosa inhibitor and leucocyte proteases was adduced by immunological methods (Figure 2). It was possible to enrich these complexes from pathological secretions. The complexes react with specific antiinhibitor serum as well as with specific antileucocyte elastase serum.

In the case of extreme mucosal atrophy, as in ozaena, it was found that the secretions contain neither free nor complexed inhibitor. Investigations are in process to establish whether chronic infections of the nasal sinuses can be treated by installation of inhibitors with a suitable inhibitory spectrum.

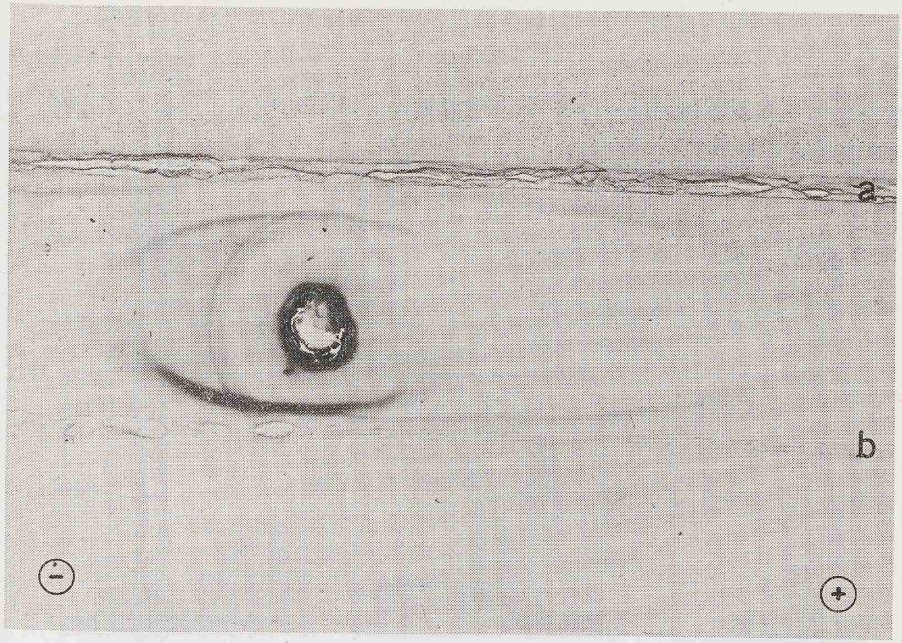


Figure 2. Immunoelectrophoresis of purified, bound mucosa inhibitor. In channel (a) anti-mucosa inhibitor serum, in channel (b) antileucocyte protease serum.

It was also found at our clinic that the humoral protease inhibitor alpha-1-anti-trypsin, which exercises an important protective function in the lung, also occurs in the nasal secretion but has no great significance.

Besides these enzyme effectors we are also intensively occupied with the enzymes occurring in nasal secretion. It was our aim in the investigation of the enzymes in the nasal secretion to find further biochemical parameters for the functional condition of the nasal mucosa. Our attention was fixed in the first place on the enzymes of the intermediary metabolism which also occur in the serum and considerably contribute to organic diagnosis in internal medicine. These are mainly GOT, GPT, LDH, CPK, acid and alkaline phosphatase.

We were, in fact, able to demonstrate these enzymes in human nasal secretion in not inconsiderable amounts. By estimation of the enzyme activities in a fair number of nasal secretions of patients without pathological nasal findings it was possible to determine an approximate normal distribution of these enzymes (Figure 3).

In order to prove that these enzymes originate in the cells of the nasal mucosa and do not get into the nasal secretion by transudation from the serum, we determined the enzymes of patients with diseases which manifest themselves by a high enzyme level in the serum, like GOT and GPT in patients with hepatitis, LDH and CPK in patients with myocardial infarction, and acid and alkaline phosphatase in patients with prostatic carcinoma or cholelithiasis.

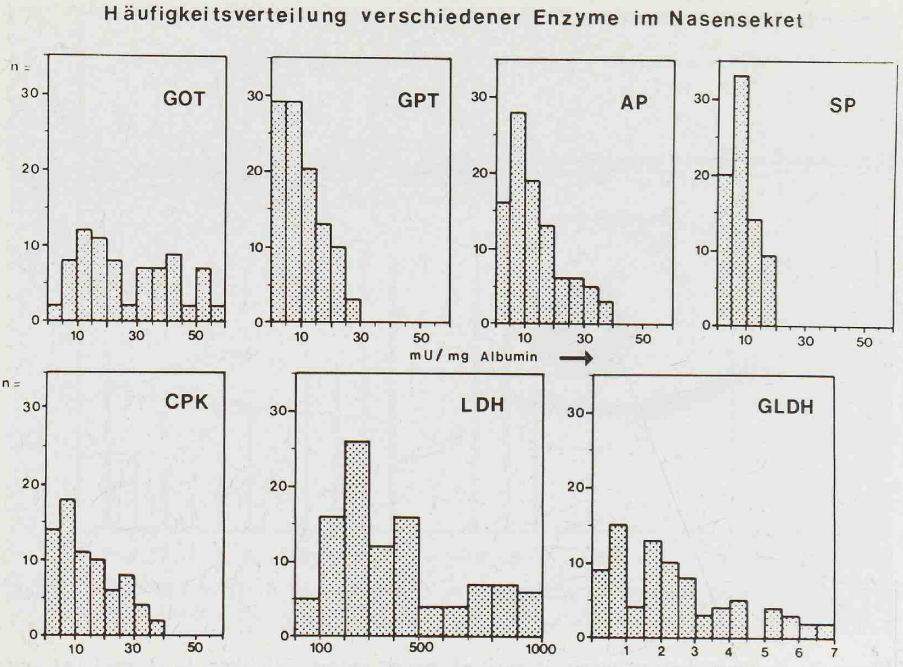


Figure 3. Frequency distribution of various enzymes in nasal secretion.

As an example of this (Figure 4) I am showing you some cases of patients with hepatitis which impress by high serum levels of GOT and GPT. As you can see in the illustration, there occurs no rise of the relevant enzyme activities in the nasal secretion. It is therefore very likely that the enzymes of the intermediary metabolism do not get into the nasal secretion by transudation from the serum but by excretion from the epithelium of the nasal mucosa.

After these preliminary investigations we examined the enzyme activities in the secretion in pathological conditions of the nasal mucosa and nasal sinuses. The test subjects were divided according to the following clinical pictures: acute maxillary sinusitis, chronic maxillary sinusitis, virus rhinitis, allergic rhinitis and atrophic rhinitis. In addition we examined patients with tracheal stenosis, nasal obstruction, state after laryngectomy and with tumours of the nasopharynx and the oral cavity, in order to recognise possible effects of these diseases on the concentration of enzymes in the mucosal secretion. The values found for the activities of acid and alkaline phosphatase show no correlation with any definite clinical picture. Only in acute maxillary sinusitis did we find GPT values which make a significant rise of enzyme activity in this condition appear possible. Remarkable results however were produced by the GOT, LDH and CPK values (Figures 5-7). In the group of rhinitis cases of various aetiology, only in virus rhinitis we found a strong rise of all 3 enzymes up to 5 to 8 times the determined normal values. The values in allergic rhinitis are far below the norm and in

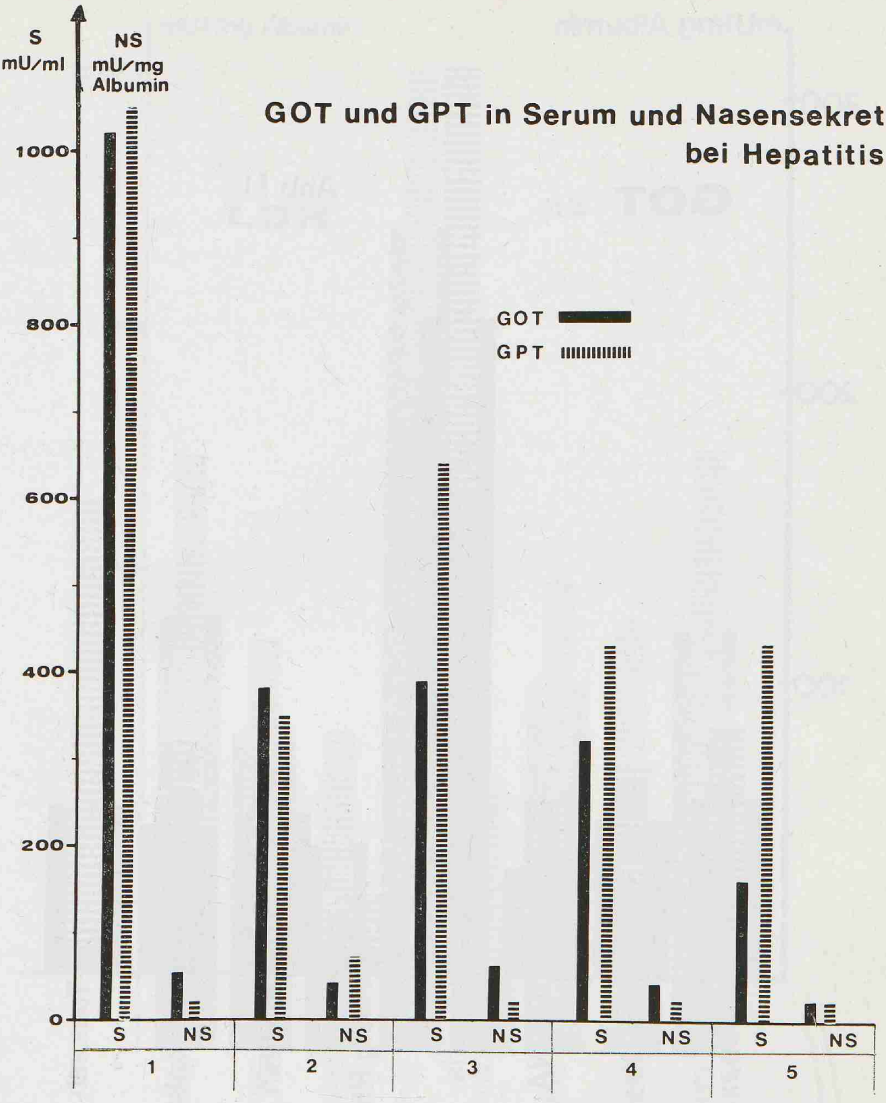
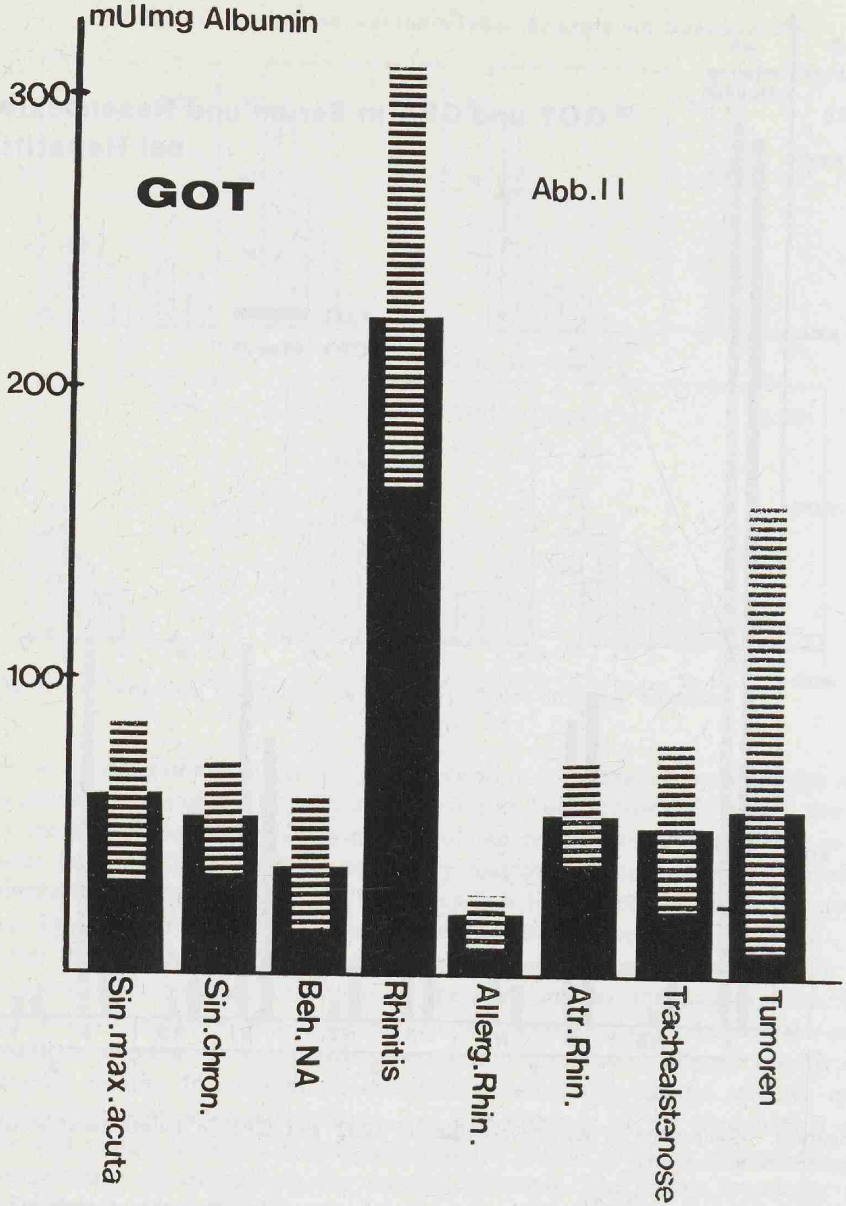


Figure 4. Comparison of enzyme activities of GOT and GPT in human nasal secretion and serum in hepatitis.

cases of atrophic rhinitis within the normal range. In the present state of the investigations there already results from this for clinical practice a simple laboratory-diagnostic demarcation of virus rhinitis from allergic or bacterial rhinitis by determination of CPK, GOT and LDH.

Still more interesting for the diagnosis, however, appears to be the determination of the isoenzyme pattern of the enzymes mentioned. With regard to LDH, which in human secretions and tissues occurs in 5 different forms distinguishable elec-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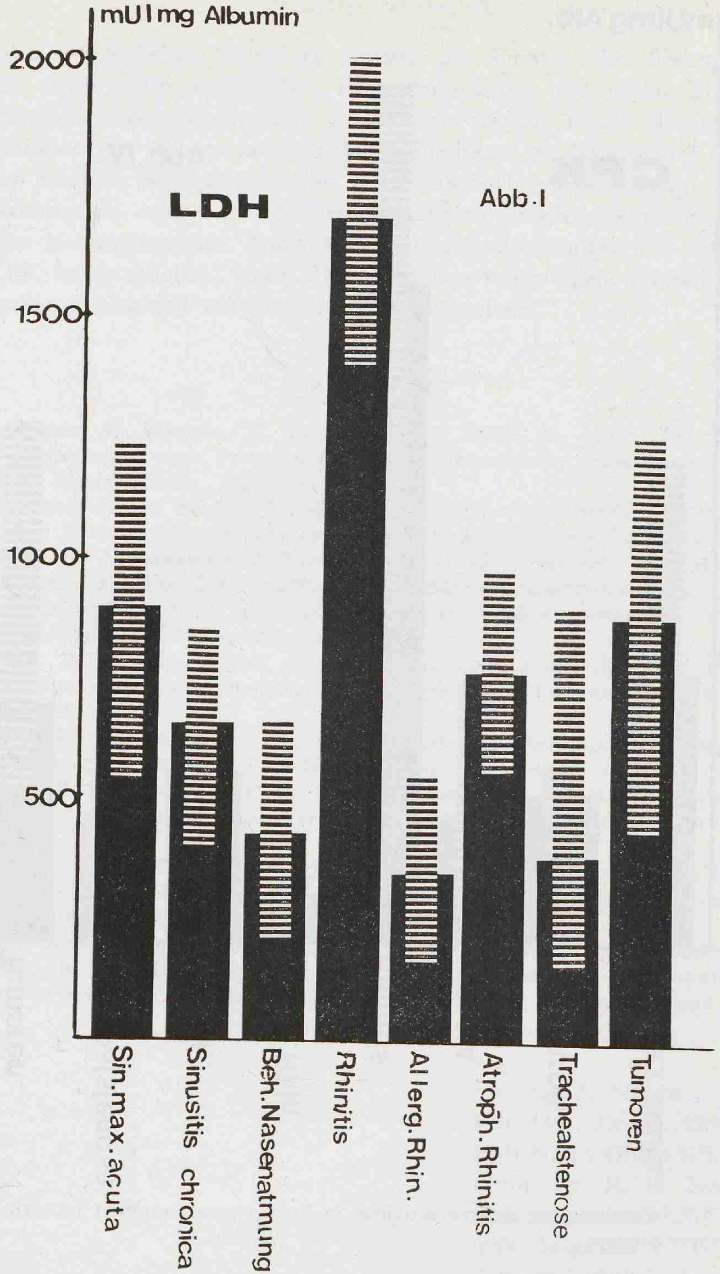
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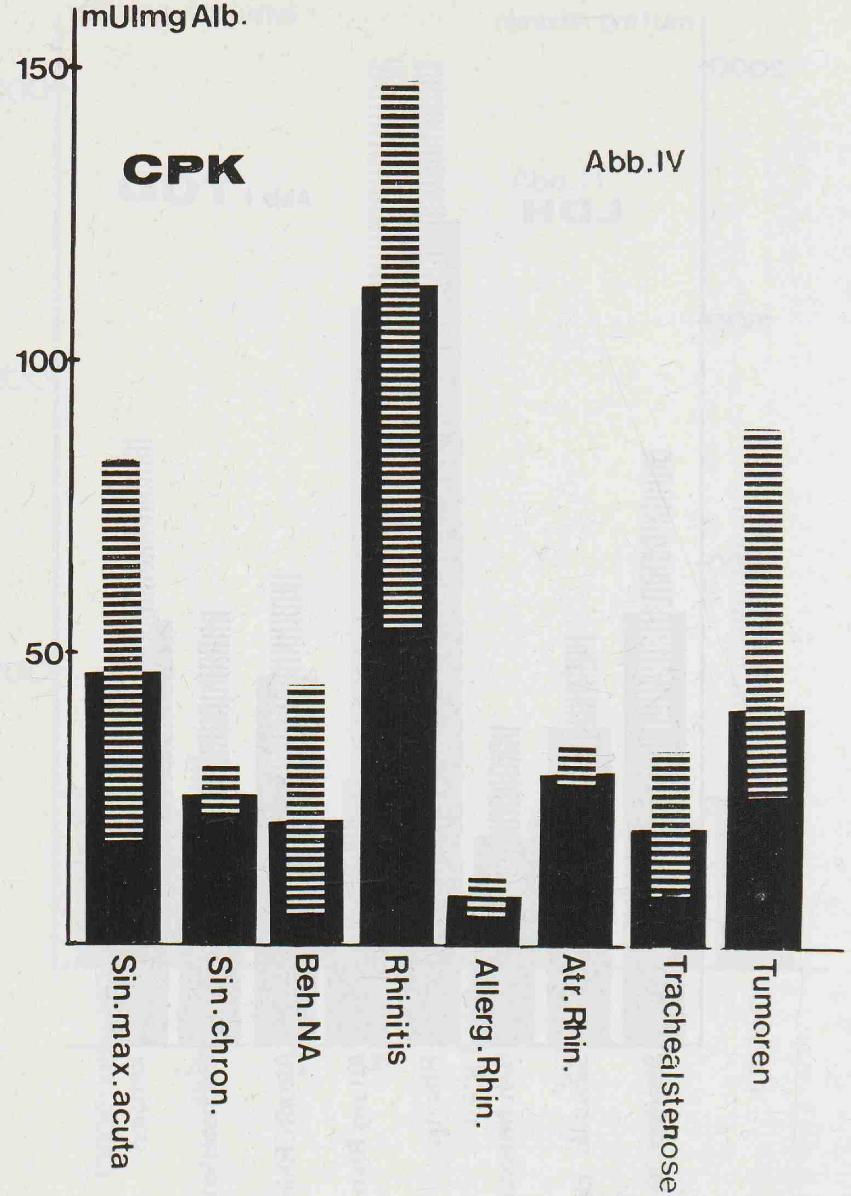

Figures 5-7. Frequency of enzyme activities in human nasal secretion in various diseases of the upper airways.

trophoretically, we were able to show that in normal nasal secretion there occurs an entirely different enzyme pattern from that in the serum and that in pathological secretions other enzyme patterns occur than in normal secretions. Details will be published elsewhere.

ZUSAMMENFASSUNG

Normales menschliches Nasenkret enthält alle Enzyme des Intermediärstoffwechsels sowie einen spezifischen Proteaseninhibitor, der neben Trypsin und Chymotrypsin Leukozytenproteasen zu hemmen vermag. Durch Bestimmung der Konzentration des schleimhautspezifischen Inhibitors im Nasensekret kann nicht nur das Stadium einer akuten oder einer chronischen Entzündung im Bereich der Nasenhaupt- oder -nebenhöhlen objektiviert werden, sondern auch eine schlechte Heilungstendenz. Anhand signifikanter Erhöhungen für GOT, LDH und CPK ist es möglich, virale Rhinitiden von bakteriellen, atrophischen oder allergischen Rhinitiden enzymatisch zu differenzieren.

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