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The roles of histamine, leukotriene C_4 and bradykinin on nasal vascular permeability in experimental nasal allergy of guinea pigs

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SUMMARY

The releases of histamine, leukotriene C4 (LTC_4) and bradykinin into the nasal cavity were measured following nasal antigenic challenge in ovalbumin (OA) sensitized guinea pigs, or following nasal stimulation with one of these chemical mediators in GA-non-sensitized animals. In sensitized animals, increased vascular permeability of nasal mucosa was recognized immediately after antigenic stimulation and lasted for 90 minutes. The release of histamine into the nasal lavage fluid was observed only immediately after the antigenic stimulation. The releases of LTC_4 and kinins into the nasal lavage fluid were augmented not only immediately after the antigenic challenge, but also 60 to 90 minutes after the stimulation. Nasal stimulation with one of these chemical mediators also increased nasal vascular permeability, but lasted for less than 40 minutes. These results suggest that the antigen-induced release of these chemical mediators might play some important roles in early increase of nasal vascular permeability, and that the increase of LTC_4 and kinin levels might be involved in the prolonged nasal vascular permeability after nasal allergic response.

INTRODUCTION

It has previously been reported that many chemical mediators were involved in anaphylactic reactions (O'Flaherty et al., 1983). Application of histamine (Weiss et al., 1982), peptide leukotrienes (Hamel et al., 1982; Weiss et al., 1982) and bradykinin (Herxheimer et al., 1961) via an aerosol cause bronchoconstriction in humans and guinea pigs. Application of histamine to the nasal mucosa can induce nasal symptoms such as allergic rhinitis in humans and animals (Okuda et al., 1982). Recently, it was clarified that the levels of these chemical mediators in

the nasal secretions markedly elevated after nasal antigenic challenge in patients with allergic rhinitis (Creticos et al., 1984; Kojima et al., 1989). It was reported that a late phase increase of the nasal symptoms and/or concentration of chemical mediators in the nasal secretion occurred several hours after a nasal antigenic challenge in patients with allergic rhinitis (Naclerio et al., 1985; Kojima et al., 1989).

In this study, we examined the chronological changes of nasal vascular permeability and mediator release into the nasal cavity after nasal antigenic challenge in ovalbumin-sensitized guinea pigs. We also examined the effects of nasal application of these chemical mediators on the nasal vascular permeability in OA-non-sensitized guinea pigs.

MATERIALS AND METHODS

Male Dunkin Hartley guinea pigs (200-250 g) were sensitized with ovalbumin (OA) as reported by Sohma et al. (1986). Each guinea pig was injected intraperitoneally with OA (10 μg) and propranolol (20 mg/kg). Two weeks later, a second immunization with the same doses was performed. One week later, they were placed in a perspex box and exposed to OA (1 mg/ml) aerosol for 1 min every day for one month. The method of Kojima et al. (1986) was used for the nasal antigenic challenge in OA-sensitized guinea pigs, and nasal stimulation with one of the chemical mediators in OA-non-sensitized animals. Under general anaesthesia with sodium pentobarbital (30 mg/kg, i.p), a tracheostomy was performed and a dwelling tube connected with a peristaltic pump was inserted into the cboana from the tracheostoma. Intravenous injection of 1 ml of 4% brilliant blue was used as an indicator of the exudative reaction. After perfusing the nasal cavity with saline (warmed to 37 °C) for 20 minutes, OA (1 mg/ml), histamine (20 μg/ml), LTC_4 (500 ng/ml) or bradykinin (100 μg/ml) dissolved in saline was perfused for 3 minutes into the nasal cavity. Each guinea pig was perfused with only one stimulation. Saline was subsequently perfused for 90 to 150 minutes. Each perfusion was performed at the rate of 0.25 ml/min. The nasal lavage fluid dropping from the nostril was collected every 10 minutes in an ice-cooled plastic tube containing 0.1 ml of 500 mM EDTA. Kinin-generating and destroying enzyme inhibitors (aprotinin 1,000 KIU, soy-bean trypsin inhibitor 80 μg, polybrene 0.4 mg, 1,10-phenanthroline 1 mg, EDTA 2 mg/0.1 ml) were added to 0.5 ml of each sample, and the concentration of kinin was measured by the radioimmunoassay method previously reported by Ishida et al. (1986). The remaining samples were centrifugated at 3,000 rpm for 10 minutes, and the supernatants were collected to quantitatively analyze dye, histamine and LTC_4. The amount of dye was measured by using a spectrophotometer (620 nm). The concentrations of histamine were directly measured by using commercial histamine radioimmunoassay (RIA) kits (Immunotech, Marseille, France). This

RIA-based histamine assay utilized a monoclonal antibody raised to acylated histamine. The sensitivity and specificity of this assay system were previously reported (McBride et al., 1988). Similarly, the concentrations of LTC_4 were directly measured by using commercial LTC_4-specific RIA kits (TRK.905, Amersham, UK). The antiserum reacts by 1.6% with LTD_4, and crossreactivity for other ligands is reported to below 0.6% by the manufacturer. The standard curve was constructed in the range of 10 pg to 500 pg/0.1 ml.

RESULTS

Following the antigenic challenge, the concentrations of dye in the nasal lavage fluid rapidly increased (32.5 ± 22.9 μg/ml, $p < 0.005$) and remained for 90 minutes at the levels which were significantly higher than the prechallenge levels (Figure 1). As shown in Figure 2, increased release of histamine into the nasal lavage fluid was also recognized soon after antigenic stimulation (59.2 ± 22.1 ng/ml, $p < 0.05$), but lasted less than 30 minutes. The concentrations of immunoreactive LTC_4 (i-LTC_4) significantly increased not only soon after the antigenic stimulation (409 ± 333 pg/ml, $p<0.05$), but also between 50 minutes (478 ± 383 pg/ml, $p<0.05$) and 90 minutes (421 ± 237 pg/ml, $p<0.01$) after the stimulation. Similarly, the concentrations of immunoreactive kinins (i-kinins) significantly increased not only soon after the antigenic stimulation (4.70 ± 3.22 ng/ml, $p<0.05$), but also between 60 minutes (2.10 ± 1.56 ng/ml, $p<0.05$) and 90 minutes (1.01 ± 0.64 ng/ml, $p < 0.05$) after the stimulation. Nasal stimulation with histamine, LTC_4 or bradykinin in OA-non-sensitized guinea pigs increased excretion of dye into the nasal lavage fluid only soon after these stimulations (Figure 3).

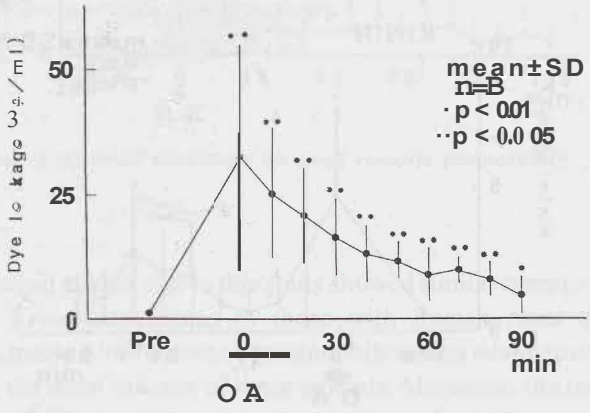

Figure 1. Excretion of dye into nasal lavage fluid before and after topical antigenic challenge.

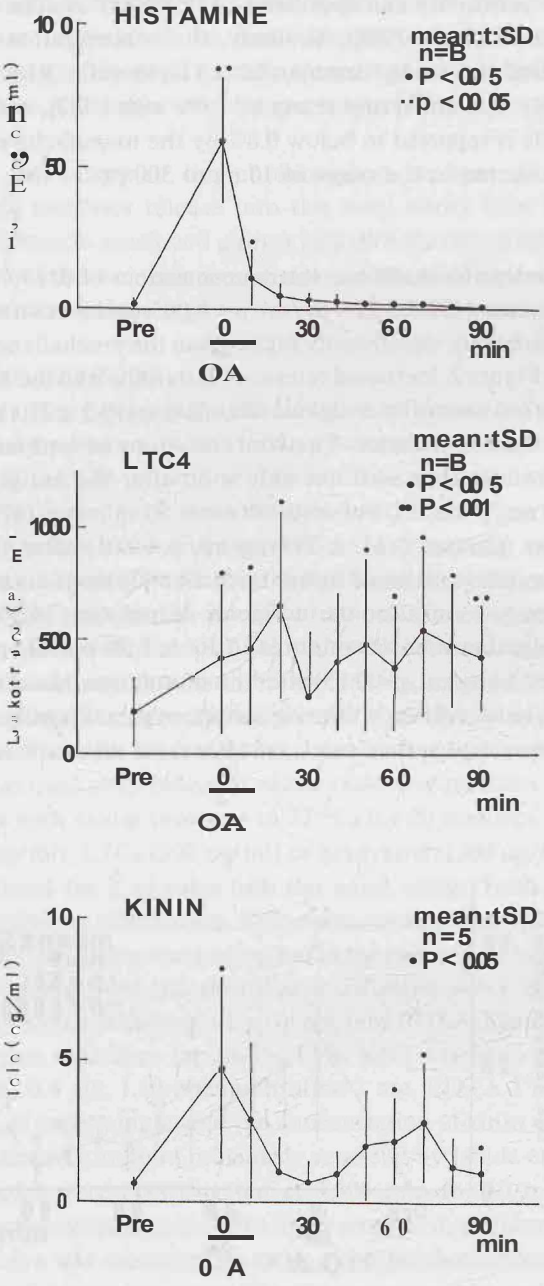

Figure 2 Release of chemical mediators into nasal lavage fluid before and after topical antigenic challenge.

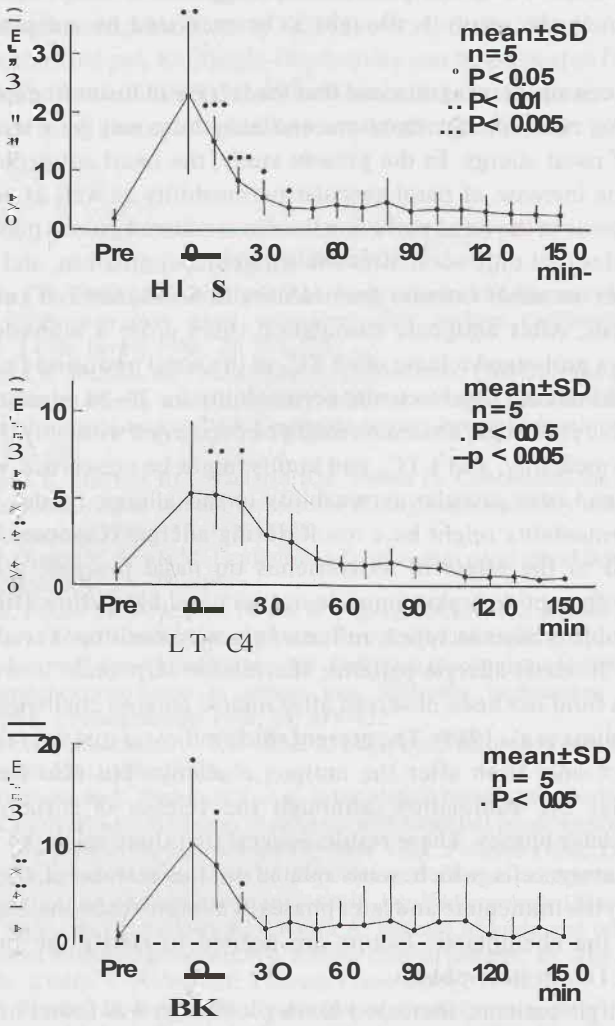

Figure 3. Effect of chemical mediators on nasal vascular permeability.

DISCUSSION

The GA-sensitized guinea pigs in this study showed similar symptoms (sneezing, stiffness and hyperrhinorrhoea) as those with human nasal allergy. Also, histologically, marked infiltrations of eosinophils and an edematous change were recognized in the nasal mucosa of these animals. Moreover, the increased nasal vascular permeability was induced by the injection of rabbit anti-guinea pig lgE into the nasal mucosa in these sensitized animals (Sohrna et al., 1986). Therefore,

the increased nasal vascular permeability triggered by nasal topical challenge with antigen in this study is thought to be med_iated by antigen-specific IgE antibody.

There has been a general agreement that the release of histamine from mast cells occurs during nasal allergic reactions and histamine may be a most important mediator of nasal allergy. In the present study, the nasal antigenic stimulation provoked the increase of nasal vascular permeability as well as an increase of histamine levels in the nasal perfusion fluid in sensitized guinea pigs. The level of histamine elevated only soon after the antigenic stimulation, and the effect of histamine on the nasal vascular permeability in non-sensitized guinea pigs was only transient. After antigenic stimulation there arose a biphasic increase of i-kinins and a prolonged release of i-LTC_4 in the nasal perfusion fluid. LTC_4 and kinins could increase nasal vascular permeability for 20-30 minutes. Therefore, after antigenic challenge, histamine might be concerned with only the immediate vascular permeability, and LTC_4 and kinins might be concerned with both the immediate and later vascular permeability in this allergic model. A prolonged vascular permeability might be a result during allergic response.

With regard to the effect of leukotrienes on nasal mucosa, a nasal topical challenge with peptide leukotrienes increases nasal blood flow (Bisgaard et al., 1984) and induces nasal secretion and nasal mucosa! swelling (Terada et al., 1987) in humans. In nasal allergic patients, the release of peptide leukotrienes into nasal lavage fluid has been observed after topical antigen challenge (Creticos et al., 1984; Kojima et al., 1989). The present study indicates that the release of LTC_4 occurred not only soon after the antigen challenge but also between 60-90 minutes after the stimulation, although the release of histamine was not observed in later phases. These results suggest that there might be several types of inflammatory cells which were related to the release of these chemical mediators in the immediate and later phases of allergic reactions. Further studies concerning the chemotactic factors are needed to clarify the mechanism of release of LTC_4 in later phases.

In nasal allergic patients, increased kinin generation was found in nasal lavage fluid following nasal challenge with allergen. The kinins in these nasal washes were revealed to be a mixture of lysyl-bradykinin and bradykinin by high-performance liquid chromatography analysis (Proud et al., 1983). Kininogen in nasal wash was also revealed to be increased after antigenic challenge. And there arose a hypothesis that, during an allergic reaction, an increased vascular permeability might allow a transudation of kininogens from plasma into nasal secretions and provide a substrate for kinin-forming enzymes (Baumgarten et al., 1985). Similarly, plasma kallikrein and glandular kallikrein were both reported (Baumgarten et al., 1986) to be elevated after antigenic challenge. In the present study, a release of i-kinins into the nasal lavage was revealed to be augmented

after the nasal antigenic challenge. But the mechanisms of the immediate and later increases in the concentrations of i-kinins after the nasal antigenic challenge have not been clarified yet. Kallikrein-like activity can be generated from human basophils (Newball et al., 1979), therefore we will have to obtain more data, especially concerning the cytological aspects of allergic reaction.

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