◎総 説

Basophil response to antigen and anti-IgE 3. Ca^{2+} influx and histamine release

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Abstract : The release mechanism of chemical mediators from basophils and mast cells was discussed when these cells were stimulated by different antigens and anti-IgE. 1. Ca^{2+} influx into mast cells increased after stimulation by antigen. The increased Ca^{2+} uptake by mast cells was inhibited by antiallergic agents, disodium cromoglycate (DSCG) and tranilast, and calcium antagonists, nifedipine and nicardipine. 2. The dose-response curve of histamine release by antigen was different from that by anti-IgE. The maximum release of histamine by house dust was, however, similar to that by anti-IgE. House dust- and anti-IgE-induced release of histamine increased with higher serum IgE levels. On the contrary, the release of histamine by Candida albicans was not releated to serum IgE levels, and the release by C. albicans did not parallel the release by anti-IgE.

Key words : Ca^{2+} influx, histamine release, house dust, Candida albicans, anti-IgE

Introduction

In is well known that extracellular Ca^{2+} is required for release of histamine from basophils and mast cells in response to antigen-antibody reaction[1)]. When mast cells are stimulated by antigen, Ca^{2+} influx into the cells significantly increases[2)]. A positive correlation between the magnitude of the release of histamine and the amonut of ^{45}Ca uptake has been observed in antigen-stimulated mast cells[3)]. Later, Ishizaka et al. reported that phospholipid methylation in mast cells is at first induced by bridging of IgE receptors on cell surface, and that this increased methylation of phospholipids sets the stage for Ca^{2+} influx and the subsequent release of histamine[4)].

In the present study, a release of histamine from basophils stimulated by antigen and anti-IgE was discussed by observing differences in the release mechanism among stimulating agents.

The role of calcium ions

Calcium ions uptaken by the cells play an important role in two different ways of the release mechanism. In first one, calcium ions

induce phosphorylation of proteins, leading to degranulation of the cells. In this release mechanism, chemical mediators included in the granules such as histamine, heparin, eosinophil chemotactic factor (ECP), neutrophil chemotactic factor (NCF), arylsulfatase and lysosomal enzymes (preformed mediators), are released with degranulation. In the second one, calcium ions activate phospholipase A_2, which increases the synthesis of arachidonic acid mainly from phosphatidylcholine. The arachidonic acid generated is metabolized by cyclo-oxgenase and lipoxygenase. The products of arachidonic acid such as prostaglandins (PGD_2, PGE_2 and $PGF_{2\alpha}$)[5], thromboxane A_2, leukotrienes (LTB_4, LTC_4, LTD_4) are called newly generated mediators (Fig. 1).

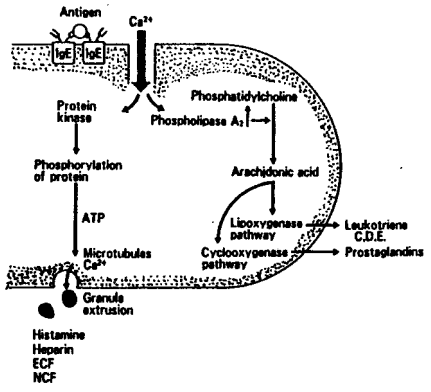

Fig. 1. Release mechanisms of chemical mediators from basophils and mast cells

Inhibition of Ca^{2+} influx into mast cells.

When Ca^{2+} influx into basophils or mast cells is inhibited by certain agent, histamine release from the cells is also inhibited. Therefore, agents preventing Ca^{2+} influx into mast cells might inhibit histamine release from the cells. Antiallergic agent, disodium cromoglycate (DSCG) and a calcium channel antagonist, nifedipine, inhibit ^{45}Ca uptake and histamine release in mast cells stimulated by antigen. Several reports have suggest that inhibitory effect of DSCG on histamine release takes place by blocking Ca^{2+} uptake by mast cells[6~8]. The comparison of the effect of DSCG and nifedipine on ^{45}Ca uptake and histamine release shows some differences in the inhibitory action. The effect of DSCG on ^{45}Ca uptake and histamine release was compatible at each employed concentration of the drug, and the inhibitory effect was dose-dependent and monophasic. While the inhibition by nifedipine is somewhat inconsistent in the sense that the degree of inhibition is different in ^{45}Ca uptake and in histamine release. Nifedipine is more effective on ^{45}Ca uptake inhibition than histamine release[8~10].

An antiallergic agent, tranilast, and a calciuim channel antagonist, nicardipine, also inhibit ^{45}Ca uptake and histamine release in mast cells stimulated by antigen. The inhibitory effect of tranilast, as well DSCG, on ^{45}Ca uptake and histamine release reachs the peak after 20 min preincubation with the agent, and the inhibition is reduced by prolonged preincubation with the agent[11, 12].

The inhibition of nicardipine on ^{45}Ca uptake of mast cells increases lineally as the duration of preincubation with the agent extends, peaking at 30 min. The effect of the agent on histamine release also increases in accordance with the duration of the preincubation time[13]. Nicardipine also inhibits both antigen- and anti-IgE-induced histamine release from basophils[14]. Similar results showing the inhibitory effect of verapamil and nifedipine on histamine release from human basophils induced by antigen, anti-IgE and Ca iono-

phore A23187 have been demonstrated by Jensen, et al[15].

Histamine release from whole blood

The release of histamine from whle blood has been applied for clinical evaluation of allergy[16~18]. The studies by Redermecker demonstrate the difference in the kinetics of histamine release between whole blood and washed leucocytes ; the rate of release from whole blood with the challenge of allergens is faster than from washed leucocytes. Our previous studies also showed that whole blood histamine release is rapidly elicited and attained the maximum within 15 min. While release from washed leucocytes is much slower. The release of histamine from washed leucocytes has been observed by many investigators[20~24].

Release from whole blood has been noticed by some researchers to simplify the method for measurement of histamine release[16~19]. Siraganian, et al. suggested that the whole blood is better for observation of the in vivo immunological situation, because the amount of histamine released when allergen is added to whole blood is due to the interaction of all factors which participate in allergic reaction[17]. It has been shown that the release of histamine from whole blood is usually less than the release from washed leucocytes. There is, however, a close correlation between the two methods.

Histamine release by house dust

House dust extract induces a significant amount of histamine release from basophils in atopic subjects sensitive to the allergen. The dose-response curve of histamine release by house dust shows that the amount of hiatamine release increases as the antigen concentrations are higher. House dust-induced release of histamine from whole blood correlates with the levels of specific IgE antibodies expressed as a RAST acore[25]. The maximun histamine release by house dust is higher in cases with a RAST score of 3+ than that in cases with a RAST score of 2+. While no significant release of histamine by house dust is observed in cases with a RAST score of 0+ to 1+[26~28]. These findings demonstrate that house dust-induced histamine release is elicited by bridging of IgE receptors[29, 30].

Relationship to skin reactivity

Histamine release by an antigen correlates with skin reactivity to the antigen. Skin reactivity to house dust is lower in cases with basophils low reactive to anti-IgE and house dust. Skin sensitivity is high with higher reactivity of basophils[25]. These findings show that IgE-mediated reactivity of basophils correlates with skin sensitivity to the antigen.

Histamine release by anti-IgE

It has been shown that there are significant differences in the dose-response curve of histamine release from washed leucocytes between allergen and anti-IgE. Allergen-induced release is elicited over a wide range of concentrations, while release by anti-IgE occurres within a limited concentration range[31]. Previous studies, using washed leucocytes, demonstrated that there was a significant correlation between anti-IgE-induced release of histamine and serum IgE levels[32~34]. Our previous studies, using whole blood, also revealed similar results showing a positive correlation between anti-IgE-induced histamine release and the serum IgE levels[27].

Anti-IgE-induced release of histamine is

different between the two groups of asthmatics with peripheral eosinophilia (more than 8%), who are classified by skin reactivity and RAST score to house dust. The dose-response curve of histamine release in group I (positive skin test and positive RAST score to house dust) shows a consistent increase as the anti-IgE concentrations increase. While the dose-response couve of histamine release in group II (negative skin test and negative RAST score to house dust) is very low and symmetric at the concentration causing the maximum percent release. The curve in group I (atopic) is much higher than that in group II (nonatopic) in spite of peripheral eosinophilia in both groups[36].

In the experiments for histamine release from whole blood, the interaction with a serum factor has to be considered. Effect of a serum factor is found in dose-response curves of anti-IgE-induced histamine release, but not in those of allergen-induced release. Basophils from cases with high serum IgE levels require much more anti-IgE to produce maximum histamine release compared to cases with low serum IgE levels[36].

Comparison between house dust- and anti-IgE-induced histamine release

In comparison of histamine release between house dust and anti-IgE stimulation, a positive correlation between the two agents can be observed in asthmatics with RAST score of 2+ or more ($r=0.66$, $p<0.001$). The results show that basophil reactivity demonstrated through IgE receptors is similar, even if the cells are incubated with different stimulating agents ; basophils more sensitive to house dust are more sensitive to anti-IgE, and less to house dust less to anti-IgE in cases sensitive to the allergen. While there is no correlation between the two agents in cases with RAST score of 0+ and 1+[27].

The release of histamine by house dust increases as the RAST score goes up from 0+ to 3+. In cases with RAST score of 0+ and 1+, the release of histamine by anti-IgE is higher than the release by house dust. Basophil reactivity to anti-IgE varied widely in these cases, while the reactivity of basophils to house dust is consistently low. Basophils from cases with RAST score of 2+ and 3+ show a strong reactivity to both house dust and anti-IgE[37].

Histamine release by Candida albicans

Candida albisans induces histamine release from basophils of patients with bronchial asthma. The release of histamine induced by C.albicans correlates with serum levels of specific IgE to the allergen. Our previous studies revealed that a significant amount of histamine release was observed by C.albicans in cases with the RAST score 2+ or more. There was, however, no correlation between serum concentration of specific IgE and histamine release. A significant amount of histamine release was elicited by C.albicans in all cases with serum specific IgG concentrations between 0 and 0.5, who were all sensitive to the allergen. It is likely from the results that C.albicans-induced histamine release may be inhibited in some cases with high serum concentrations of specific IgG antibodies. In fact, no significant release was elicited in 3 out of the seven cases positive in C.albicans-RAST, with the levels of specific IgG more than 0.5[38].

Comparison among house dust-, anti-IgE- and Candida albicans-induced histamine release

The release of histamine induced by house

dust and the release by C.albicans are closely related to the serum levels of specific IgE antibodies to each allergen as expressed by RAST score. A correlation of house dust-induced histamine release with the release elicited by anti-IgE is generally observed in cases sensitive to house dust. On the contrary, any correlation is not found between C.albicans- and anti-IgE-induced histamine release in cases sensitive to C.albicans[39,40].

House dust- and anti-IgE-induced histamine release show a tendency to increase with higher serum IgE levels. There is, however, no correlation between C.albicans-induced histamine release and serum IgE levels. In cases showing a significant amount of histamine release by C.albicans, basophils are less reactive to house dust and anti-IgE when their serum IgE levels are low (0－300 IU／mℓ), and the cells are more reactive to both agents, house dust and C.albicans when the IgE levels are high (more than 1001 IU／mℓ). These results reveal that serum IgE levels show a close correlation with basophil reactivity to house dust and anti-IgE[41].

Conclusion

Histamine releases from basophils is clinically significant to evaluate allergy situation in patients with bronchial asthma and other allergic diseases. In further studies on experiments for release of chemical mediators, it is more desirable to use leucocytes separated into basophils, neutrophils and eosinophils by counterflow centrifugation elutriation[42].

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抗原および抗ヒトIgEに対する抗塩基球の反応性. 3. Ca^{2+}の細胞内流入およびヒスタミン遊離

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ハウスダスト, カンジダなどの抗原および抗ヒトIgE刺激時の, 抗塩基球および肥満細胞からの化学伝達物質遊離機序について, 若干の検討を行った。1. 肥満細胞へのCa^{2+}の流入は, 抗原刺激後に増加する傾向を示した。この肥満細胞のCa^{2+}取り込みの増加は, 抗アレルギー剤であるクロモリン(DSCG)やトラニラストやCa^{2+}拮抗剤であるニフェジピンやニカルジピンによって抑制された。2. 抗原(ハウスダスト)によるヒスタミン遊離の濃度依存性カーブは, 抗ヒトIgEによるカーブとは異なっていた。しかし, ハウスダストが抗原である症例では, 抗原による最高ヒスタミン遊離値は, 抗ヒトIgEによる遊離値とある程度の相関を示した。そして, 抗原(ハウスダスト)および抗ヒトIgEによる好塩基球からのヒスタミン遊離は, 血清IgE値が上昇するにつれて, 増加する傾向が見られた。一方, カンジダによるヒスタミン遊離は, 血清IgE値との相関は無く, また, 抗ヒトIgEによるヒスタミン遊離との間にも関連は見られなかった。

キーワード：Ca^{2+} 流入, ヒスタミン遊離, ハウスダスト, カンジダ, 抗ヒトIgE