Human C5a Induces a Substantial Histamine Release in Human Basophils but Not in Tissue Mast Cells

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Abstract

The histamine-releasing effect of human C5a was compared with that of concanavalin A in blood basophils and isolated adenoidal mast cells from the same donors. In basophils, C5a (10 ng/ml) induced a significant histamine release (7.5 ± 2.1 %, corrected value). In isolated adenoidal mast cells C5a had only a marginal effect (2% histamine release), although the cells responded markedly to concanavalin A stimulation (about 13% release). The results support the view that the heterogeneity of mast cells and basophils is also reflected in the expression of anaphylatoxin receptors on their surface.

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Friedberger's classical anaphylatoxin [1] was first characterized as a histamine liberator in the guinea pig [2]. Later, the spasmogenic and chemotactic principles of the anaphylatoxins were identified as the complement peptides C3a and C5a [3]. Since C5a has been reported to release histamine from rat mast cells and human basophils [4–6], anaphylatoxin has been regarded as a general histamine liberator in various conditions in man and laboratory animals. Only very recent studies have reemphasized the species-specific action of C5a [7]. In view of the lately appreciated heterogeneity of blood basophils and tissue mast cells [8], it seemed possible that C5a was considerably more active in some of these cell populations than in others. We here wish to report comparative studies of the effects of concanavalin A (Con A) and human C5a in human adenoidal mast cells and blood basophils obtained simultaneously from the same donors.

The donors were generally healthy children of 3–7 years. The adenoidal cells were isolated using the mechanical method described previously [9]. Each sample contained about 30,000 mast cells. Basophils were obtained from 10 ml venous blood according to Hook et al. [4]. Each sample contained about 10,000 basophils. The cells were equilibrated in Hanks' solution for 5 min at 37 °C. Human C5a (10⁻⁹ or 10⁻⁸ g/ml) or, as a positive control, Con A (50 µg/ml; Sigma, München, FRG) was added and the incubation was continued for another 10 min. The histamine content was determined in the supernatant and in the cell pellet using the double-isotopic assay of Beaven et al. [10]. C5a was prepared from human plasma, biologically assayed through its chemotactic activity and quantitated by radioimmunoassay (Upjohn Diagnostics, Kalamazoo, Mich.) [11].

For the statistical evaluation the Student t test for paired samples was used, $\alpha = 5\%$ was considered significant.

Pilot experiments with basophils confirmed the histamine-releasing effect of the C5a preparation. Figure 1 shows the results of experiments in both cell types from the same donors. They demonstrate that the spontaneous histamine release is appreciably higher from tissue mast cells when compared with basophils. A higher histamine release from the tissue mast cells was also observed following stimulation with Con A. The relation of Con-A-induced release to spontaneous release, however, was about equal in both the mast cell and basophil preparations. In the basophils, C5a was ineffective at 10^{-9} g/ml, but induced a significant histamine release at 10^{-8} g/ml. This effect corresponds to that of Con A (50 μ g/ml). In the tissue mast cells, C5a, in contrast to Con A, was only marginally effective (1.9 ± 1.1 %; corrected release). This was found to be true also if the C5a concentrations in the mast cell samples were increased up to 10^{-7} g/ml (table I).

The data reported here for human basophils are in good agreement with those reported by others [6]. The values for the adenoidal mast cells are almost identical with those recently obtained using purified hog C5a [12]. The results support the view that human and porcine C5a, in spite of their structural differences, are similarly effective at least in vitro. They might fur-

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Table I. Effect of human C5a (10^{-8} – 10^{-7} g/ml) on histamine release in three preparations of human adenoidal mast cells

30-

~ 20-

10-

Con A 50 μ g/ml

Saline

10^{-9} 10^{-7}

C5a, g/ml

1.9 ± 1.1 13.0 ± 2.2

Corrected histamine release, % 0.7 ± 0.6

40-

of 30

10-

20-

10^{-8} C5a, g/ml

Saline

0J

Corrected histamine release, % 0.1 ± 1.0

Fig. 1. Effect of human C5a in isolated human adenoidal mast cells (a) and in human basophils (b), each from the same of 7 different donors. Con A was used as a reference stimulans. The histamine release is given in percent of the total. Vertical bars represent single values. Means $\pm$ SEM of the 7 donors are also given. Stars represent $\alpha = 5\%$ (compared to the saline control).

ther explain older observations to the effect that anaphylatoxin exerts its main activity with formed blood elements in the living individual [13]. This might be regarded as a consequence of differences in the expression of anaphylatoxin receptors in human tissue mast cells and in basophils.

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