

Interleukin-8 secretion in patients with allergic rhinitis after an allergen challenge: interleukin-8 is not the main chemotactic factor present in nasal lavages

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Summary

Background Interleukin-8 (IL-8) is a chemotactic cytokine for neutrophils and primed eosinophils. In allergic rhinitis, allergen exposure triggers leucocyte recruitment.

Objective We evaluated in this study IL-8 secretion and the neutrophil chemotactic activity in nasal lavages collected after a nasal allergen challenge. Moreover, the participation of IL-8 in the neutrophil chemotactic activity was quantified.

Methods Four healthy subjects and 19 patients with allergic rhinitis were exposed to a nasal allergen challenge. As a control, saline challenge was performed in four patients with allergic rhinitis. Concentration of IL-8 was measured by ELISA in nasal lavages collected before and after challenge. Neutrophil chemotactic assay was developed using a 48-well chemotaxis microassembly.

Results After allergen challenge, the healthy subjects, the four patients receiving saline and one patient exposed to allergen did not respond; seven patients presented a single early reaction and 11 patients a dual response. For healthy subjects and the four patients exposed to saline, the level of IL-8 did not increase after challenge in comparison with that at baseline. After allergen challenge, two peaks of IL-8 release were observed for patients with allergic rhinitis during the early (30 min to 1 h 30 min) and the late periods (3 h 30 min to 9 h 30 min), however the difference was not significant for the early period. During the late period, a significant increase in IL-8 concentrations was detected for the patients developing a dual response, whereas the difference was not significant for those presenting only an early reaction. The neutrophil chemotactic activity of nasal lavages from patients with allergic rhinitis collected during the early and the late reactions (17 ± 2.1 and 23.3 ± 2.8 neutrophils per high power field (hpf), respectively) was significantly higher than the activity of lavage fluid collected at baseline (9.2 ± 1.8 neutrophils per hpf). Nevertheless, the addition of a neutralizing anti-IL-8 antibody inhibited weakly the chemotactic activity of lavage fluid from rhinitic patients collected during the early or the late periods (18 and 11% of inhibition) ($P = \text{NS}$).

Conclusion These data show that allergen challenge increased significantly the secretion of IL-8 for the patients with allergic rhinitis. However, neutralization of IL-8 in nasal lavages by a specific antibody revealed that the role of this chemokine in granulocyte infiltrate was limited, suggesting that IL-8 acts in connection with other chemotactic factors in this recruitment.

Keywords: interleukin-8, allergic rhinitis, neutrophils, eosinophils

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Introduction

The late nasal response following allergen challenge is associated with the release of inflammatory mediators and influx of leucocytes into nasal lavages. In contrast with bronchial allergic asthma where the infiltrate is composed of eosinophils and mononuclear cells, the leucocytes infiltrating the nasal mucosa [1] or present in upper airways lumen [2] of patients with allergic rhinitis contained mainly an equal proportion of neutrophils and eosinophils. Immunohistology of the nasal mucosa in allergen-induced rhinitis showed, in addition to granulocytes, a significant increase in CD4⁺-T helper lymphocyte and macrophage populations, an increase level being quite similar for the different types of cells [1]. In nasal lavages, the influx of neutrophils and eosinophils is prominent during the late phase and the migration of mononuclear cells and of basophils was also detected at this period [2–4].

In a previous study we evaluated the release of pro-inflammatory mediators in nasal lavages collected from allergic rhinitic patients challenged with allergen and the relationship with granulocyte influx [5]. Whereas tumor necrosis factor α (TNF) and granulocyte macrophage-colony stimulating factor were undetectable after allergen challenge, the release of interleukins (IL-1 α and IL-6), in addition to mast cell-derived mediators, was noted during the early and the late periods. Using the same model, Sim *et al.* [6] detected different pro-inflammatory cytokines (IL-1 β , IL-5, IL-6 and GM-CSF) after antigen challenge. However, the mediators observed in both studies are weak inducers of neutrophil influx and could not explain the nature of the inflammatory infiltrate.

Interleukin-8 (IL-8) is a chemotactic cytokine identified by its chemotactic activity for neutrophils [7]. This cytokine is a member of the β -thromboglobulin supergene family and is characterized by the presence of four conserved cysteines whose location allows the separation of this family in two groups of molecules [8]: the C-C group and the C-X-C group where IL-8 is considered as the main cytokine. Its properties include also a chemotactic activity for lymphocytes [9], basophils [10] as well as primed eosinophils [11]. In addition, IL-8 activates migrating cells and particularly it induces specific and azurophilic granule exocytosis in neutrophils and in some conditions induces histamine-release by basophils [12,13].

With this in mind, we evaluated the possible involvement of IL-8 in the granulocyte influx observed into the nasal lumen of patients with an allergic rhinitis after allergen challenge. To establish this, IL-8 levels were measured in sequential lavages collected after an allergen challenge. The role of IL-8 in the chemotactic activity of nasal lavages was also specified by neutralization of IL-8 with an anti-IL-8 neutralizing antibody.

Materials and methods

Patients

Twenty-three allergic subjects (seven women and 16 men, 19 to 30 years old) with a history of house dust mite perennial (13 patients) or grass pollen seasonal (10 patients) rhinitis and a positive prick test to the appropriate antigen extract, were included in this study. Patients with seasonal allergic rhinitis were studied out of season. On the day of the experiment, all subjects had been free of symptoms for at least 1 month and received no treatment for at least 2 weeks.

Four healthy subjects (one woman and three men, 25 to 36 years old) were also studied. Two out of four were exposed to dust mite antigen and the two others to grass pollen extract (Stallergènes Laboratories). All subjects gave their informed consent, and the study was approved by the Lille U.F.R. Ethic Committee.

Antigen challenge

The protocol of nasal allergenic challenge was performed according to the procedure described by Naclerio *et al.* [14] and detailed elsewhere [5]. After oxymetazoline hydrochloride treatment (Iliadine®, Merck-Clevenot, Nogent sur Marne, France), three doses of dust mite or grass mixed extracts (2, 20 and 70 reactivity index units) (Stallergènes Laboratories, Fresnes, France) were nebulized into the nose by a Fisons nebulizer at 15-min intervals. Nasal secretions were collected by lavage with 10 mL of saline solution. After four prewashes, the protocol involved, 10 min after administration of oxymetazoline, a control challenge with diluent and a lavage which was used to determine the baseline level, followed 10 min later by antigen challenge. Subsequent nasal lavages were performed 15 min after each dose of antigen, then after the last challenge, 30 min, 1 h and every hour until the 9th hour. The clinical response was assessed for the duration of the challenge and the symptom scores were determined as previously described [15]. Symptom scores represented the sum of scores for sneezing, rhinorrhea, nasal congestion, pruritis of the nose, pruritis of the palate or ear, and conjunctivitis. Allergen challenge was considered as positive when the symptom score sum was ≥ 5 . The early period (EP) took place between 15 and 90 min and the late period (LP) between 3 h 30 min and 9 h 30 min. One patient with allergic rhinitis did not develop clinical symptoms, seven exhibited a single early reaction (single responders: SR) and 11 presented an early and late reaction (dual responders: DR). Four patients with allergic rhinitis were exposed to a saline challenge in order to evaluate the effect of this protocol. These four patients did not present any clinical symptoms.

Nasal lavage analysis

Nasal lavages were centrifuged at 1000g for 10 min. The cell pellet was resuspended in phosphate buffer saline and the total cell number and the differential cell count were evaluated. The supernatants were aliquoted and stored at -20°C . IL-8 concentrations were measured by ELISA method (Medgenix, Fleurus, Belgium). Reproducibility of this assay with nasal lavages was $6 \pm 0.5\%$ as determined by the percentage of variation. Moreover, 110.7% of human recombinant IL-8 (RD Systems, Minneapolis, Minnesota) was recovered after addition into nasal lavages demonstrating that IL-8 ELISA was unaffected by nasal lavage fluid. Cytokine levels and cell counts were assessed during three critical periods: (1) at baseline, (2) during the early period (between 15 and 90 min, five lavages evaluated separately) and (3) the late period (between 3 and 9 h 30 min, seven lavages evaluated separately). Values obtained during the early or the late period were expressed as the peak value (the higher value) detected during both periods or as the mean of the increases calculated as followed: $\Sigma ([\text{IL-8}]_t - [\text{IL-8}]_{t_0})/n$; where $[\text{IL-8}]_{t_0}$ means the IL-8 concentration at baseline and $n = 5$ or 7 for the early and the late period, respectively.

Neutrophil chemotaxis assay

Neutrophils obtained from peripheral venous blood from controls using the Boyüm's technique [16] were suspended in RPMI at the cell concentration of 1×10^6 cells/mL.

The chemotaxis assay was developed with a 48-well microchemotaxis assembly (Neuro Probe, Cabin John, Maryland) with a $3 \mu\text{m}$ pore polycarbonate filter (Nuclepore Corp., Pleasanton, California) [17]. The nasal lavages were diluted at least five-fold in RPMI before the chemotaxis assay in order to minimize the effect of the saline used for the lavage procedure. Nasal lavages collected at baseline, 30 min and 5–6 h after allergen challenge were used in chemotaxis assay. The chamber was incubated for 30 min at 37°C . The number of neutrophils that had migrated through the filter was determined microscopically with a magnification of 1000-fold. The results were expressed as the difference between the neutrophil number/high power field (hpf) with the sample or the positive control minus the number obtained with medium alone (22.5 ± 5.4 neutrophils/hpf). Positive control experiments consisted in migration towards 10^{-7} M solution of N-formyl-methionyl-leucyl-phenylalanine (Sigma Chemical Co., St Louis, Missouri); it induced an increase of migration equal to 42 ± 6.1 neutrophils/hpf.

To determine the role of IL-8 in the migration of neutrophils towards nasal lavages, they were preincubated for 1 h at 37°C with a goat- or a rabbit-anti human IL-8 neutralizing antibodies (RD Systems, Minneapolis, Minnesota;

Endogen, Boston, Massachusetts, respectively) at a final concentration of $50 \mu\text{g/mL}$. Their neutrophil chemotactic activity was then evaluated as described and compared with the corresponding control (with non-immune serum). The results were expressed in inhibition per cent defined as followed: $(100 \times ((\text{nasal lavage} - \text{baseline level}) - (\text{nasal lavage with anti-IL-8} - \text{baseline level})) / (\text{nasal lavage} - \text{baseline level}))$. Both antibodies neutralized about 80% of the neutrophil chemotactic activity of hr-IL-8 ($0.5 \mu\text{g/mL}$).

Statistical analysis

The results are expressed as mean $\pm$ SEM or median with interquartile range for IL-8 levels. The data analysis was performed using non-parametric tests: Mann-Whitney and Wilcoxon rank-sum test; $P < 0.05$ was considered as significant. Correlation was estimated by Spearman correlation procedure statistics. Variations of IL-8 levels in kinetic study were measured by analysis of variance for repeated data (ANOVA) with «protected least significant difference (PLSD)» of Fisher to compare group data. Statistical analysis was tested using a software (Statview F 4.11, Abacus Concept, Berkeley, California, USA).

Results

Nasal response in healthy subjects

The results of IL-8 measurements in nasal lavages obtained at baseline were not significantly different between the controls and the patients with allergic rhinitis (Table 1). After allergen challenge, IL-8 levels in nasal lavages from healthy subjects did not increase in comparison with baseline except for one case that peaked at 19 pg/mL (expressed as the difference between the peak and baseline level, Table 1). These healthy subjects did not develop clinical symptoms and the differential cell counts in the nasal lavages showed only a weak neutrophil influx (peak value during the early period = $1.1 \pm 0.26 \times 10^3$ neutrophils/mL and during the late period = $0.68 \pm 0.54 \times 10^3$ neutrophils/mL).

Allergen challenge in patients with allergic rhinitis

IL-8 levels at baseline were similar in patients exposed to diluent and to allergen, whatever the type of response to allergen: SR and DR (Table 1). Instillation of control diluent in patients with allergic rhinitis did not induce a leucocyte influx (data not shown) nor an increase of IL-8 concentration (Table 1). In patients exposed to allergen, an increase of IL-8 secretion was detected during the early period in five out of seven SR ($P = \text{NS}$) and in all DR ($P < 0.05$) as well as during the late period in all patients ($P < 0.05$); however, there was a great range in the level of increase (Table 1) and

the peak of IL-8 secretion was detected at different time-points. Statistical analysis of the kinetic study revealed that, for the DR, the levels of IL-8 were significantly increased during the late period (at 5 h 30 min) compared with concentrations at baseline (Fig. 1); in SR, no significant difference was detected (Fig. 2). The means of increase levels and the peak values were not significantly different between the different groups of allergic patients during both periods of

time (Table 1). Concerning the leucocyte influx, a migration of eosinophils and neutrophils was observed in the SR and the DR during the early (19 ± 8.3 and $31 \pm 17.5 \times 10^3$ neutrophils/mL; 13.2 ± 6.7 and $11.2 \pm 14 \times 10^3$ eosinophils/mL, respectively) and at a higher level during the late periods (55.3 ± 17.8 and $103 \pm 39.1 \times 10^3$ neutrophils/mL; 19.1 ± 8.6 and $51.6 \pm 16.8 \times 10^3$ eosinophils/mL, respectively). Although it was higher during the late period, the

Table 1. IL-8 concentrations in nasal lavages from patients with allergic rhinitis or healthy controls. Nasal lavages were collected either at baseline or during the early period (EP, from 15 min to 1 h 30 min) and the late period (LP, from 3 h 30 min to 9 h 30 min) following instillation of allergen or control diluent (patient with diluent). In patients with allergic rhinitis challenged with allergen, one did not respond (NR) whereas seven patients presented a single early reaction (SR) and 11 patients a dual response (DR). The means of increase in IL-8 levels were reported after subtraction of baseline level (for details see nasal lavage section in materials and methods). * $P < 0.05$ compared with the concentrations at baseline by the use of the Wilcoxon test

	Patients	Level at baseline	Peak value (pg/mL)		Mean of increase (pg/mL)	
			EP	LP	EP	LP
Controls	1	225	230	221	0	0
	2	650	637	220	0	0
	3	430	305	290	0	0
	4	105	95	124	0	4.6
	Median $\pm$ IR	327.5 $\pm$ 375	267.5 $\pm$ 308	220.5 $\pm$ 83.5	0	0 $\pm$ 4.6
Patients tested with diluent	1	138	130	132	0	0
	2	380	280	240	0	0
	3	420	498	464	19.5	23
	4	320	220	322	0	0.3
	Median $\pm$ IR	350 $\pm$ 171	250 $\pm$ 210	281 $\pm$ 207	0 $\pm$ 9.7	0.15 $\pm$ 11.6
NR.	1	226	184	318	0	13.1
SR.	1	358	298	686	0	110.3
	2	350	338	1138	33.6	271.7
	3	164	162	430	0	95.7
	4	472	3950	1246	1244.4	423.3
	5	610	1818	1418	384	200.9
	6	580	1884	1088	484	170
	7	250	628	490	104.8	140.8
	Median $\pm$ IR	358 $\pm$ 278	628 $\pm$ 1559	1088 $\pm$ 680*	104.8 $\pm$ 451	170 $\pm$ 136.1
DR.	1	384	1084	1334	216.4	317.7
	2	402	516	530	43.6	40.3
	3	184	440	1060	89.6	604.3
	4	374	860	1570	160	444
	5	608	750	620	28.4	1.4
	6	118	299	196	47.2	83
	7	148	318	682	34.4	131
	8	684	696	2040	1.7	832.4
	9	100	162	660	20.8	57.5
	10	40	70	180	42	79.7
	11	22	128	150	54.8	230.9
	Median $\pm$ IR	184 $\pm$ 293	440 $\pm$ 540*	660 $\pm$ 986*	43.6 $\pm$ 51	131 $\pm$ 349.4

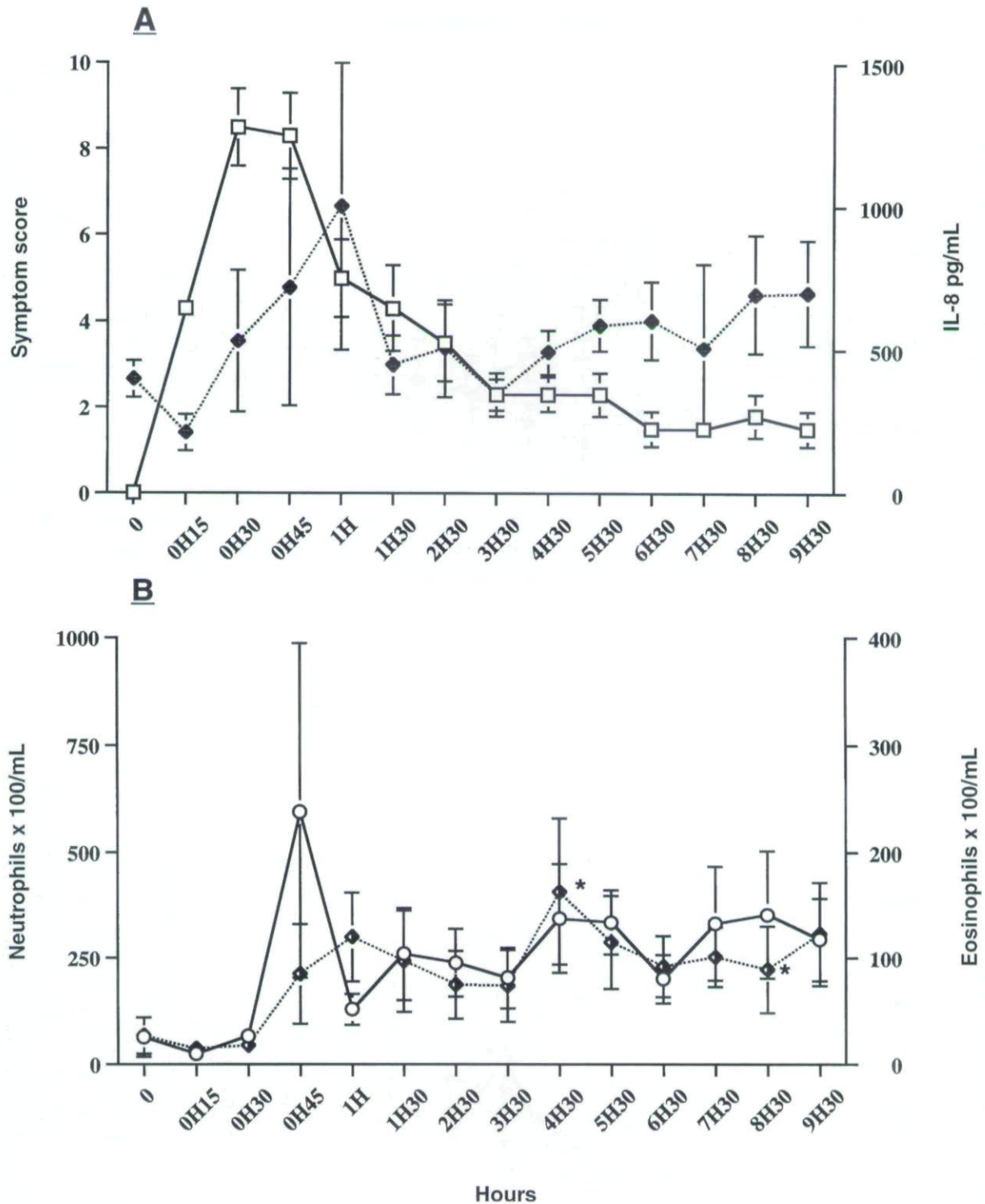
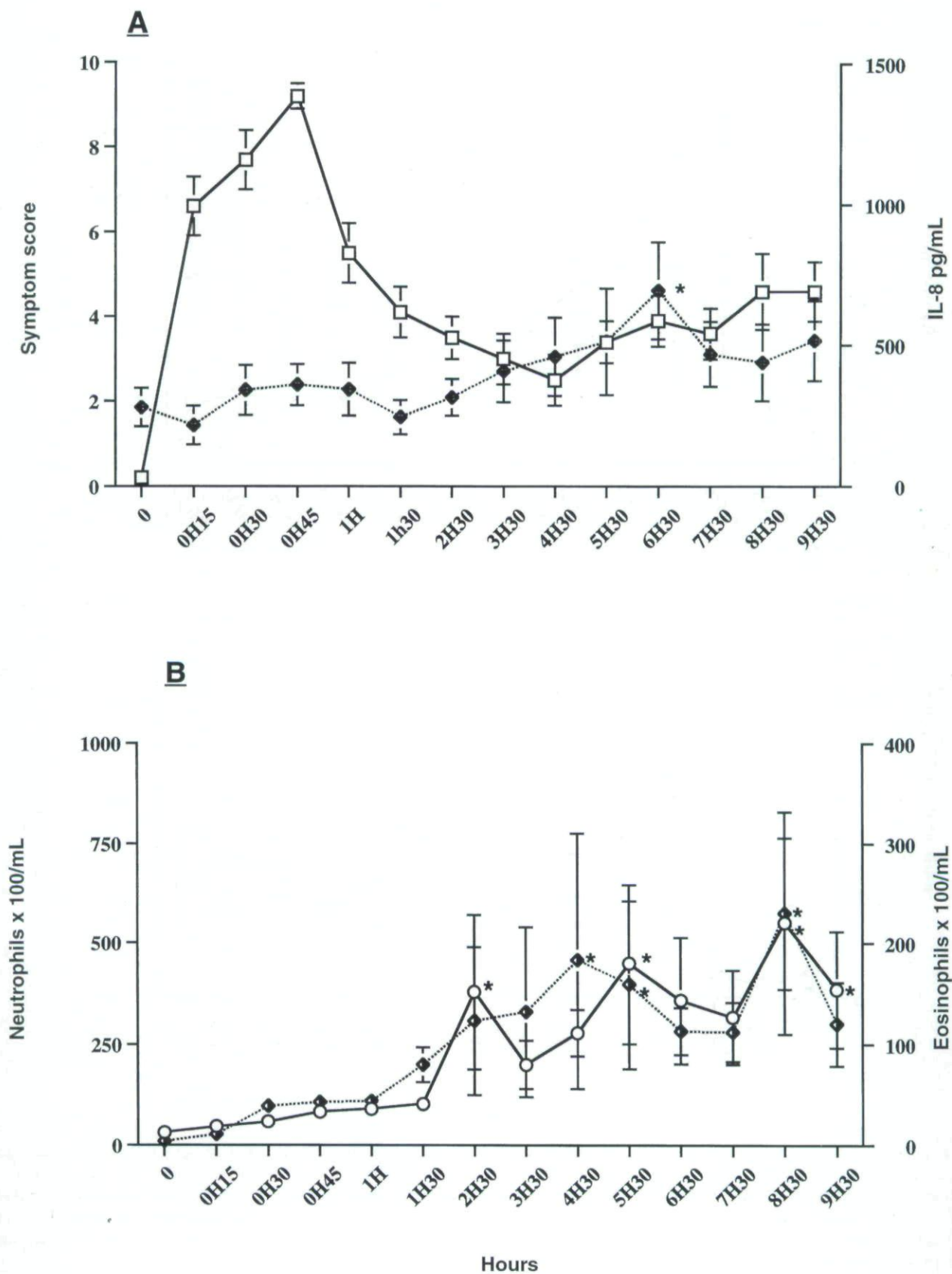


Fig. 1. Time course of symptom score and IL-8 concentration (part A) as well as granulocyte influx (part B) in the patients challenged with allergen who developed a single early reaction (single responders). Results were expressed as mean $\pm$ SEM except for IL-8 (median $\pm$ interquartile range). The values of symptom scores were always significantly higher ($P < 0.05$) than at baseline; for IL-8: * = $P < 0.05$ compared with baseline (by analysis of variance of Fisher with PLSD). The numbers of neutrophils and eosinophils were significantly different ($P < 0.05$) from the results obtained at baseline except for the neutrophils at 15, 30 and 45 min. A: □ = symptom score; ◆ = IL-8 pg/mL. B: ○ = neutrophils; ◆ = eosinophils.



intensity of granulocyte influx for DR was not significantly different from that obtained for the SR.

To illustrate the time course of IL-8 secretion and its relationship with the clinical symptoms and the granulocyte influx, values from SR (Fig. 1) and from the DR (Fig. 2) were reported. In SR, the occurrence of the early reaction was visualized by a clear increase of symptom scores ($P < 0.01$) (Fig. 1A), simultaneously, IL-8 level increased two-fold compared with the baseline level ($P = \text{NS}$). During the late period, the symptom score decreased slowly and the IL-8 production was lower than before ($P = \text{NS}$) (Fig. 1A). An influx of neutrophils and eosinophils (Fig. 1B) occurred at the end of the early reaction and was sustained during the late period. The symptom score of the DR showed two peaks at 1 h and between 6 h 30 min and 10 h (Fig. 2A), the last being lower because the time course of the symptom score largely differed according to the patients tested. During the early reaction, a weak release of IL-8 (Fig. 2A) was detected ($P = \text{NS}$). During the late phase, the concentration of IL-8 increased slowly until 6 h 30 min where it reached its maximum ($P < 0.05$). The granulocyte influx detected in DR appeared during the early period and then augmented progressively during the late period until 8 h 30 min (Fig. 2B).

No close relationship was found between IL-8 levels and other parameters. For example, Fig. 3 showed the results of the correlation between IL-8 concentrations and either the neutrophil count ($r = 0.37$) and eosinophil count ($r = 0.25$).

Neutrophil chemotactic activity of the nasal lavages

Analysis of the neutrophil chemotactic activity of nasal lavages from controls and patients with allergic rhinitis showed that nasal lavages collected at baseline induced a low but significant migration of neutrophils (Fig. 4). After allergen challenge in healthy subjects (Fig. 4) and saline challenge in patients with allergic rhinitis (data not shown), no modification of the neutrophil chemotactic activity was observed in nasal lavages collected at 30 min and 5–6 h. In patients with allergic rhinitis exposed to allergen, the neutrophil chemotactic activity of nasal lavages collected during the early and the late periods was significantly

increased compared with the activity obtained in fluids collected at baseline ($P < 0.05$). No significant difference was found between the neutrophil chemotactic activity detected in nasal lavages from DR compared with SR.

Addition of two different neutralizing anti-IL-8 antibodies into nasal lavage specimens collected during either the early and the late period poorly affected their neutrophil chemotactic activity (18 ± 11 and 11 ± 5 % inhibition, respectively) ($P = \text{NS}$, in both cases); similar results were obtained with both antibodies. Positive controls of this neutralization assay were performed by measuring the neutrophil chemotactic activity of hr-IL-8 ($0.5 \mu\text{g/mL}$) incubated with nasal lavages in the presence or not of anti-IL-8 antibodies: anti-IL-8 antibodies neutralized 94.6% of the neutrophil migration induced by hr-IL-8 ($n = 5$).

Discussion

Allergen challenge in patients with allergic rhinitis induced a granulocyte infiltrate. As shown in this study, it also provokes an enhancement of the chemotactic activity found in nasal lavage fluids, an activity which is partly dependent on IL-8 secretion. The involvement of allergen in the secretion of IL-8 was suggested by the lack of response in healthy subjects and patients challenged with diluent. These results differed from those obtained after segmental saline challenge in allergic asthmatics: a neutrophil influx and a secretion of IL-8 were reported [18], whereas both are not observed in the nasal lavages after saline instillation. Moreover, the kinetic study showed that increase in IL-8 release was mainly detected at the same time as the clinical symptoms (during the early and the late phases). These data demonstrated that increase in IL-8 secretion was associated with the nasal allergic reaction.

The cell source of IL-8 in allergic rhinitis might potentially involve different cell types. First, IL-8 production by IgE-stimulated mast cells was recently described [19] and their activation in allergic rhinitis was shown by the release of histamine and tryptase [5,20]. Second, histamine- and TNF-stimulated epithelial cells synthesize large amounts of IL-8 [21,22]; moreover, spontaneous production of IL-8 by bronchial epithelial cells is increased in patients with allergic asthma [23]. Third, mononuclear cells, eosinophils

Fig. 2. Time course of symptom score and IL-8 concentration (part A) as well as granulocyte influx (part B) in the patients challenged with allergen who developed an early plus a late reaction (dual responders). Results were expressed as mean $\pm$ SEM except for IL-8 (median $\pm$ interquartile range). The values of symptom scores were significantly higher ($P < 0.05$) than at baseline except for 6 h 30 min, 7 h 30 min and 9 h 30 min; for IL-8: * = $P < 0.05$ compared with baseline (by analysis of variance of Fisher with PLSD). The numbers of neutrophils and eosinophils were significantly higher ($P < 0.05$) at the following periods: for 45 min and between 4 h 30 min and 9 h 30 min and for 1 h, 1 h 30 min, 7 h 30 min and 8 h 30 min concerning neutrophils and eosinophils respectively, compared with the results obtained at baseline. A: $\square$ = symptom score; $\blacklozenge$ = IL-8 pg/mL. B: $\circ$ = neutrophils; $\blacklozenge$ = eosinophils.

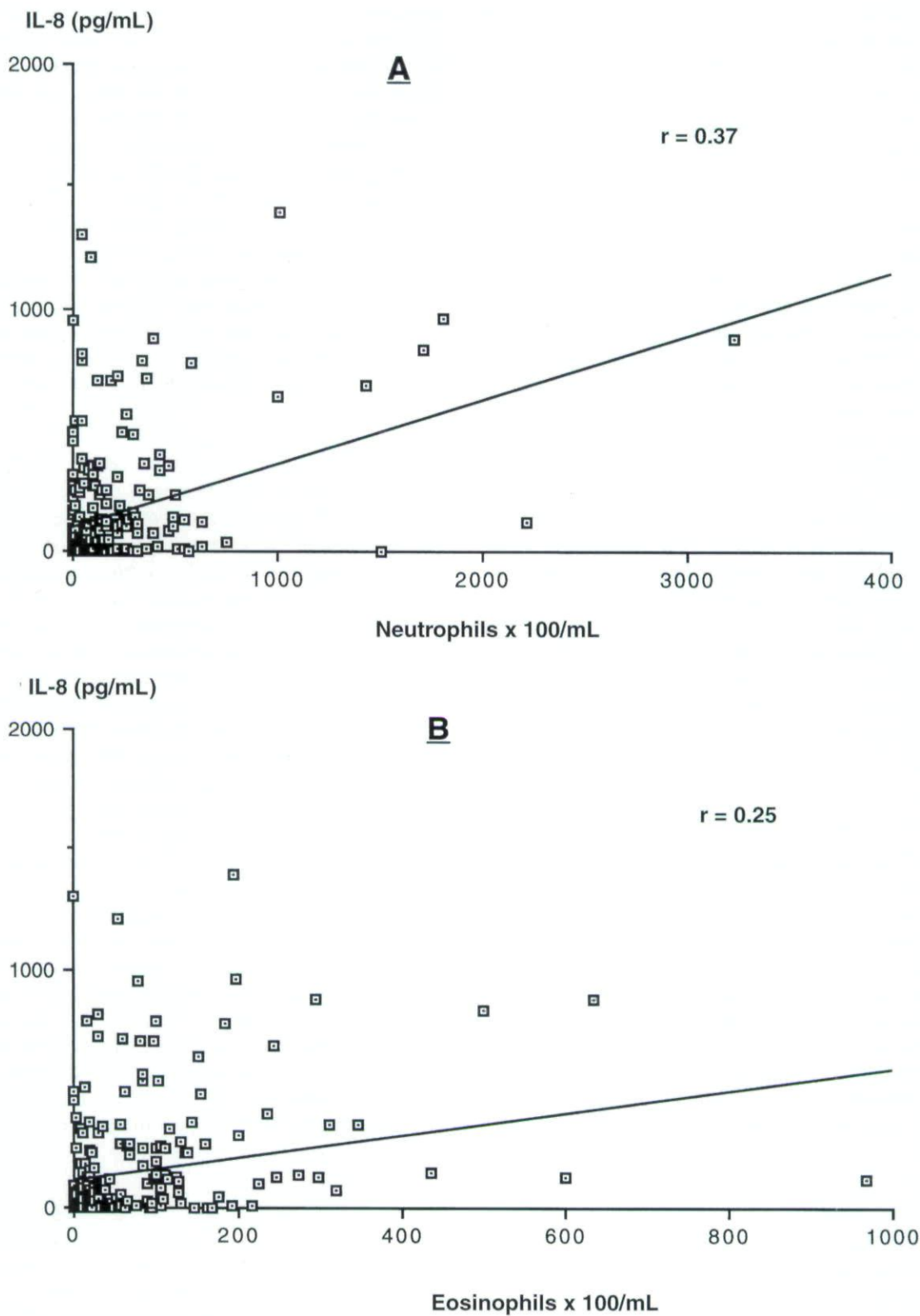


Fig. 3. Correlation between the IL-8 level and either the neutrophil count (A) or the eosinophil count (B) observed in nasal lavages from patients with allergic rhinitis exposed to allergen. The values reported in this figure expressed the difference between values obtained in nasal lavages and those at baseline.

Neutrophils/HPF

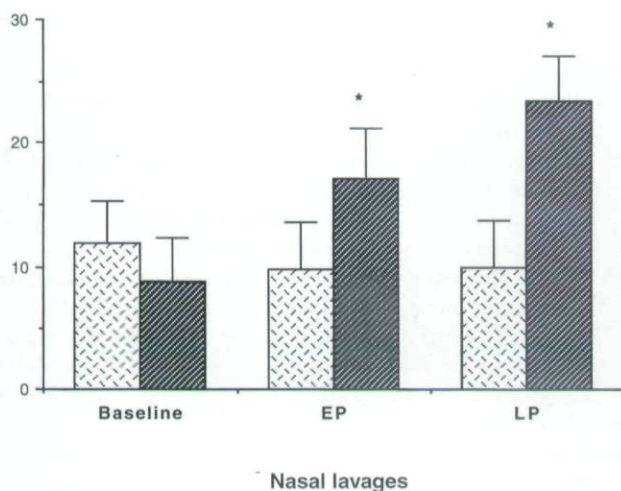


Fig. 4. Neutrophil chemotactic activity in nasal lavages from healthy subjects (controls) and patients with allergic rhinitis performed at baseline and after an allergen challenge at 30 min (early period: EP) and 5–6 h (late period: LP). The results were expressed as the difference between the neutrophil number/high power field (hpf) obtained with the sample minus the number obtained with medium alone (22.5 ± 5.4 neutrophils/hpf). N-formyl-methionyl-leucyl-phenylalanine (the positive control) induced an increase of migration equal to 42 ± 6.1 neutrophils/hpf. ▨ = controls; ■ = allergic rhinitis.

and the neutrophils are able to produce IL-8 [7,24,25]. We may suspect that mast cells produce IL-8 during the early reaction since they are directly stimulated by the allergen. However, we cannot exclude the involvement of other cell types activated by an indirect mechanism like epithelial cells. During the late period, all these cells might participate in IL-8 secretion. It is interesting to note that IL-8 levels were correlated with IL-6 and IL-1 concentrations produced in nasal lavages obtained in the same conditions [5] (data not shown). These relationships suggest that the production of these three cytokines are linked or that they may be the consequence of the same process. Since epithelial cells secrete *in vitro* the same pattern of cytokines, we may hypothesize that these cytokines, detected during the late period in nasal lavages from patients with allergic rhinitis, are mainly produced by epithelial cells.

A role for IL-8 in the granulocyte recruitment observed in allergic rhinitis is suggested by different observations: (1) IL-8 is a potent chemotactic factor for neutrophils [7] and primed eosinophils [11]; (2) IL-8 is produced just before and at the same time as the granulocyte infiltrate (Figs 1 and 2), and (3) the nasal lavages, for which an increase in IL-8 release was detected, have an higher chemotactic activity than the nasal lavages obtained at baseline. However, this

effect observed in lavages collected during the early and the late periods was poorly inhibited (18 and 11%, respectively) by addition of a neutralizing anti-IL-8 antibody showing that other chemotactic factors for neutrophils and probably other chemokines of the C-X-C family are present in these lavages.

Another property of IL-8 suggests that it may be implicated in the recruitment and the activation of basophils which may be involved in the development of the late nasal reaction [3]. The effect of IL-8 on histamine-release is different depending on its concentration: at a very low concentration (10^{-9} M), IL-8 inhibits histamine-release induced by cytokines and histamine releasing factors [26]. In contrast, IL-8 induces histamine-release, either at high concentrations (10^{-6} M) [12] or at lower level on IL-3 primed basophils (10^{-7} and 10^{-8} M) [13]. In allergic patients, the IL-3 priming of basophils can be hypothesized. In the same manner, the local concentration of IL-8 in the nasal mucosa reaching 10^{-8} M (the method of lavage dilutes the nasal secretions 10 to 100 times), we can hypothesize that IL-8 plays a role in basophil activation observed during the late nasal reaction. Indeed, a significant increase of IL-8 secretion during the late period was only found in DR. However, during the late period, an IL-8 secretion was observed in nearly all the SR and there was no significant difference between the mean of increase in IL-8 secretion in DR compared with SR (Table 1). Moreover, during this late period, the patients belonging to the SR had a granulocyte influx in the nasal mucosa and developed mild clinical symptoms. Thus, the groups of the SR and the DR are not opposed entities but differed by the intensity of the symptoms during the late period. In this context, it is not surprising that the IL-8 secretion observed in the two groups of patients with allergic rhinitis are not statistically different.

Our study shows that IL-8 is produced during the early and the late periods in patients with allergic rhinitis after a nasal allergen challenge. IL-8 might be involved in the development of the late nasal reaction both through the granulocyte recruitment towards the nasal mucosa and upper airways lumen but also through basophil activation. However, other chemotactic factors are certainly implicated in the neutrophil influx, as demonstrated by the low inhibitory effect of an anti-IL-8 neutralizing antibody on the chemotactic activity in nasal lavage fluid. Identification of the other chemotactic factors participating in leucocyte influx into the nose during allergen challenge and their exact hierarchy is presently under investigation.

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