

IgE produces monocyte superoxide anion release: Correlation with CD23 expression

Comparison of patients with asthma, patients with rhinitis, and normal subjects

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Allergic inflammation involves many different cell types among which mononuclear cells, such as macrophages and monocytes, play an important role. These cells release numerous chemical mediators, including superoxide anion ($O_2^{\cdot-}$). We evaluated the capacity of atopic serum to stimulate peripheral blood monocyte $O_2^{\cdot-}$ release. Thirteen untreated allergic patients (seven with asthma and six with rhinitis), and five nonallergic control subjects were studied. $O_2^{\cdot-}$ was measured in a photon-counting camera with Lucigenin-enhanced (Sigma Chemical Co., St. Louis, Mo.) chemiluminescence. Results were expressed (mean $\pm$ SEM) in relation to basal values (peak/basal chemiluminescence values). Spontaneous production of $O_2^{\cdot-}$ was greater in allergic patients. Moreover, atopic serum stimulated $O_2^{\cdot-}$ production of blood monocytes in all subjects, but this was greater in subjects with allergic asthma than in subjects with allergic rhinitis and normal subjects. Anti-IgE immunoadsorption of atopic serum completely abrogated this effect, which was restored by the IgE-rich eluted fraction. IgE-induced $O_2^{\cdot-}$ release decreased as adherence duration increased and was correlated with surface CD23 expression. These results indicate that monocytes from allergic patients are in an activated state and that binding of IgE to their receptors generates $O_2^{\cdot-}$, possibly by direct activation of blood monocyte reduced nicotinamide adenine dinucleotide phosphate oxidase. (J ALLERGY CLIN IMMUNOL 1994;93:108-16.)

Key words: Allergic asthma, allergic rhinitis, superoxide anion, blood monocytes, IgE, CD23

Allergic diseases (e.g., rhinitis, asthma, conjunctivitis, and atopic dermatitis) are immediate-type hypersensitivity reactions that result from natural exposure to environmental allergens. Their common biologic characteristic is the development of an IgE immune response to low concentrations of these allergens.¹ Bronchial asthma is an inflammatory process characterized by increased vascular permeability and local infiltration of activated cells such as eosinophils, lymphocytes, and mononuclear phagocytic cells.^{2, 3} Cells infiltrating the airways and cells circulating in the

Abbreviations used

AA:	Allergic asthma
AR:	Allergic rhinitis
BMs:	Blood monocytes
Fc ϵ RII (CD23):	Low-affinity IgE receptor
FEV ₁ :	Forced expiratory volume in 1 second
NADPH:	Reduced nicotinamide adenine dinucleotide phosphate
$O_2^{\cdot-}$:	Superoxide anion
PBMCs:	Peripheral blood mononuclear cells

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Received for publication July 15, 1992; revised May 12, 1993; accepted for publication May 28, 1993.

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0091-6749/94 \$1.00 + 0.10 1/1/49021

bloodstream express low-affinity IgE receptors (Fc ϵ RII or CD23) on their surfaces and therefore can be stimulated by IgE-dependent mechanisms, releasing lysosomal enzymes, eicosanoids, cytokines, and superoxide anion ($O_2^{\cdot-}$).⁴⁻⁷ CD23 expression is enhanced in peripheral blood monocytes (BMs)⁸ and alveolar macrophages⁹ from atopic patients.

Mononuclear phagocytic cells (e.g., alveolar macrophages and their circulating precursors, BMs)¹⁰ from allergic patients release greater amounts of oxygen species than normal subjects when stimulated *in vitro* either by phagocytosis¹¹⁻¹³ or by anti-IgE after previous sensitization with allergic sera^{4, 14} or by phorbol esters.^{13, 15} Moreover, using chemiluminescence assays, we have shown¹⁵ that BMs are activated *in vivo* even in very mild asthma and that this activation differs depending on whether the cells adhere to a surface. When appropriately stimulated, these cells reduce oxygen first to superoxide anion ($O_2^{\cdot-}$) (by reduced nicotinamide adenine dinucleotide phosphate [NADPH] oxidase) and subsequently to hydrogen peroxide (by superoxide dismutase). Hydrogen peroxide is then either transformed by myeloperoxidase into hypochlorous acid or hydroxyl radical (through an Fe^{2+} catalyzed reaction).¹⁶ These oxygen species generate chemiluminescence with photon release.¹⁷

In this study we focused on the mechanism by which BMs produce $O_2^{\cdot-}$ in the IgE/anti-IgE system and examined in particular the direct effect of IgE via their binding to CD23. We therefore analyzed the response of BMs directly stimulated by atopic serum and determined the participation of serum IgE in this activation process.

METHODS

Study subjects

Seven patients with allergic asthma (AA) (age range, 19 to 58 years; mean $\pm$ SEM, 34 ± 5 years) took part in the study. Asthma was defined as previously described,³ according to the criteria of the American Thoracic Society,¹⁸ and all patients had reversible airway obstruction. The severity of asthma was assessed by the clinical score of Aas¹⁹ and patients' pulmonary function (spirometric measurement of forced expiratory volume in 1 second [FEV₁] and forced vital capacity). None of the subjects had ever smoked, and no subject had had any respiratory tract infection during the month preceding the test. Patients were remote from any acute exacerbation of asthma. Immediate-type hypersensitivity was assessed by the history of asthma, the presence of positive skin prick test results to common aeroallergens, and elevated levels of serum IgE (Phadebas PRIST, Pharmacia Diagnostics AB, Uppsala, Sweden). Patients were receiving only β_2 -agonists, which were withdrawn 12 hours before the study began. They were excluded if they had received systemic corticosteroids in any form during the past 2 months, inhaled corticosteroids during the previous month, or other drugs in the previous 48 hours.

Six patients who had perennial allergic rhinitis (AR) (age range, 25 to 49 years; mean $\pm$ SEM, 32 ± 3 years)

were also investigated. All were nonsmokers, and one had been receiving medication for at least 1 month before the study. They were studied in the same manner as the patients with asthma, allowing us to assess the allergic status and to delete asthma diagnosis according to the criteria of the American Thoracic Society.¹⁸ The study was performed out of the pollen season.

Five normal nonsmokers (age range, 26 to 42 years; mean $\pm$ SEM, 32 ± 3 years) served as a control group. Their pulmonary function was within the normal range, and they were nonallergic and had never had rhinitis or asthma. No subjects had had any respiratory tract infections during the month preceding the test.

The study was performed after informed consent from each subject and approval of the University of Montpellier Ethical Committee were obtained.

Serum from an allergic patient and IgE depletion

The serum from an untreated stable patient with AA (house dust mites) with 1000 U/ml IgE (Phadebas, Pharmacia) was selected. The serum IgE was depleted by solid-phase immunoadsorption with anti-IgE (Caltag, San Francisco, Calif.) Sepharose 4B (Pharmacia).⁴ Briefly, the serum IgE was removed by adsorption with the highly specific anti-IgE coupled to CnBr-activated Sepharose 4B gel (anti-IgE antibodies specific for the constant region). After it was passed through the gel, the serum sample did not contain any IgE (Phadebas, Pharmacia). The bound IgE was eluted with 0.2 N glycine buffer, pH 2.8, and collected into a solution of 2% bovine serum albumin and 0.05 mol/L phosphate buffer, pH 8.0. In this eluted fraction, the amount of IgE was the same as that of the initial serum sample. All procedures were carried out at $+4^\circ C$.

Blood monocyte preparation

Cells were obtained from heparinized blood. Peripheral blood mononuclear cells (PBMCs) were isolated by centrifugation of blood samples over isotonic Percoll solutions (Pharmacia France, St. Quentin en Yveline, France) with densities of 1.097 gm/ml and 1.086 gm/ml at 400g for 20 minutes at room temperature, as previously described.^{14, 15} The mononuclear cell suspension in Percoll was washed once with an equal volume of RPMI 1640 medium (Gibco Chemical, Glasgow, Scotland). Contaminating erythrocytes were lysed by incubation for 10 minutes in a solution of NH_4Cl 130 mmol/L, Tris 10 mmol/L, and K_2CO_3 16 mmol/L and recovered by centrifugation. After two successive 10-minute incubations in 5 ml of fetal calf serum containing 5 mmol/L ethylenediaminetetraacetic acid for removal of platelets as described by Pawlowski et al.,²⁰ the mononuclear cells were washed with RPMI 1640 medium and counted in the initial cell suspension with neutral red stain specific to BMs (neutral red/BM: 3/1).

TABLE I. Patients' characteristics

Subjects (n = 18)	Sex	Age (yr)	Aas score	Allergy	Serum IgE (U/L)	FEV ₁ (% predicted)
Patients with AA (n = 7)						
1	F	58	1	+	37	100
2	M	21	1	+	189	100
3	M	51	2	+	258	110
4	F	30	2	+	370	70
5	M	34	2	+	61	83
6	M	19	2	+	492	85
7	M	25	3	+	520	95
Mean ± SEM		34 ± 5			275 ± 73	92 ± 5
Patients with AR (n = 6)						
8	M	27		+	142	105
9	M	30		+	590	95
10	M	33		+	95	89
11	M	49		+	650	100
12	F	25		+	151	95
13	F	29		+	269	102
Mean ± SEM		32 ± 3			316 ± 99	98 ± 2
Normal subjects (n = 5)						
14	M	42		—	27	95
15	M	26		—	46	90
16	M	35		—	19	110
17	F	32		—	54	102
18	M	26		—	98	98
Mean ± SEM		32 ± 3			49 ± 14	99 ± 3

TABLE II. Blood monocyte O₂^{•-} production ratio in patients with AA (n = 7), patients with AR (n = 6), and normal subjects (n = 5)

Stimulation	Normal subjects	Patients with AR	Patients with AA
Two-hour adherence			
Controls	1.10 ± 0.04	1.4 ± 0.1†	1.34 ± 0.09
IgE	5.4 ± 0.8*	14 ± 1.3*†	29 ± 9.7*†
IgE immunoadsorption	ND	1.3 ± 0.08	1.15 ± 0.07
Eluted fraction	ND	13 ± 1.2*	30 ± 8.3*
Three-hour adherence			
Controls	1 ± 0.03	1.7 ± 0.3†	1.4 ± 0.1†
IgE	5.8 ± 0.4*	8.7 ± 0.7*†‡	11 ± 4.2*†‡

Results represent the ratio of duplicates for each subject, peak chemiluminescence/basal chemiluminescence values, and are expressed as means ± SEM.

**p* < 0.05 by comparison with controls.

†*p* < 0.05 by comparison with normal subjects.

‡*p* < 0.05 by comparison with 2-hour adherence.

Neutral red positive cells (5×10^5 per Petri dish) (Becton Dickinson, Grenoble, France) were then incubated with 2 ml of RPMI 1640 medium with 20% fetal calf serum for 2 or 3 hours at 37° C in a humid atmosphere of 95% air and 5% CO₂. The monolayers

were then washed three times with warm medium to remove nonadherent cells such as lymphocytes, and the same medium was added to cells before stimulation. The BM preparation was more than 95% pure as assessed by May-Grünwald-Giemsa and neutral red

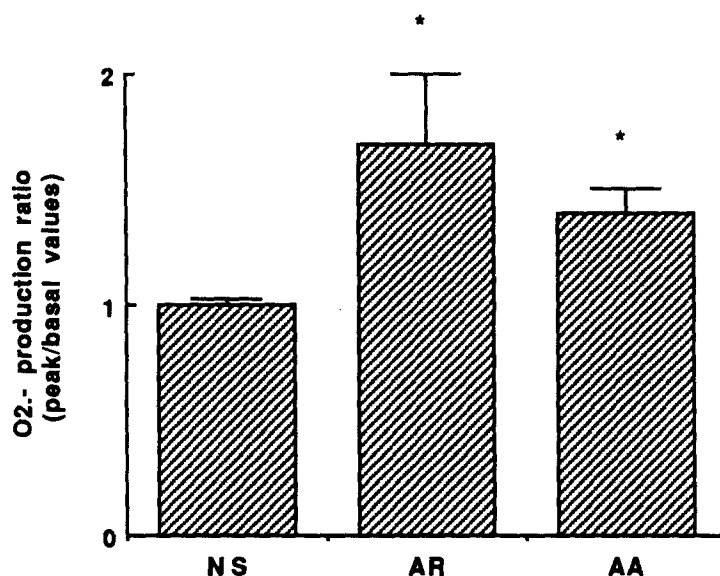


FIG. 1. O₂⁻ production ratio (chemiluminescence peak/basal values, mean ± SEM) obtained for unstimulated BMs from patients with AA and AR and normal subjects after 3-hour adherence. The statistical analysis showed a significant difference (**p* < 0.05) among patients with AA (1.4 ± 0.1), patients with AR (1.7 ± 0.3), and normal subjects (1 ± 0.03).

staining. Cell viability was greater than 98%, as determined by trypan blue exclusion. There was no difference in the monocyte number in the Petri dishes as assessed by DNA measurement²¹ (data not shown).

Blood monocyte stimulation

O₂⁻ release was induced by adding 250 µl of the atopic serum (i.e., 250 U of IgE) to the reaction mixture for 5 × 10⁵ cells. Control experiments were performed with 250 µl of anti-IgE immunoabsorbed atopic serum. In addition, some studies were performed with serum from healthy nonsmokers. The stimulation procedure was done after 2 hour preincubation, optimal time for a good adherence, and with the knowledge that cell responses could be different in their intensity related to duration of adherence after 3 hour preincubation.

Chemiluminescence assay

Before stimulation, 100 µl of Lucigenin (Sigma Chemical Co., St. Louis, Mo.) was added to the reaction mixture (10⁻⁵ mol/L final concentration), and the final volume was adjusted to 1 ml. Lucigenin (5 mg) was dissolved in 100 ml of medium 199 (Gibco Chemical) containing 476 mg N-2-hydroxyethylpiperazine-N-2-ethanesulfonic acid (Sigma Chemical Co.) and 100 mg gelatin, heated to obtain a 10⁻⁴ mol/L concentration. Lucigenin (bis-N-methylacridium nitrate) was used to enhance the luminescence induced by O₂⁻ production. Photons were counted by an ultrasensitive photon-counting imaging camera, monitored by a computer-assisted image processor (Argus 100, Hamamatsu Photonics, Japan).¹⁴ The variability inside the measure-

ments was 10%. The assay was conducted in the dark at room temperature under sterile conditions. Measurements with 5-minute integration time were carried out before (basal values) and after stimulation until the chemiluminescence peak (maximum value) was reached. Viability of the cells at the end of the experiments was greater than 98% as assessed by lactate dehydrogenase determination.²²

Immunohistochemistry

The percentage of Fc_εRII (CD23) positive PBMCs was assessed with immune complexes of alkaline phosphatase and monoclonal anti-alkalin phosphatase.²³ Briefly, cytocentrifugated PBMCs were fixed in methanol/acetone (vol/vol, +4° C) and incubated for 45 minutes at 37° C with 1:50 final dilution of anti-CD23 monoclonal antibody 25 (Immunotech, Marseille, France).²⁴ Slides were washed three times in Tris-buffered saline/bovine serum albumin (10 minutes each) and then incubated for 30 minutes with a rabbit anti-mouse IgG (final dilution 1:100) (Boehringer Mannheim, Meylan, France). Slides were again washed three times and then incubated with the anti-alkalin phosphatase antibody (final dilution 1:50) (Dako, Glostrup, Denmark). Finally the alkaline phosphatase substrate, mixed with the indicator Fast Red dye (Dako) was added, and the slides were washed and counterstained with hematoxylin and mounted in glycerol (Dako). Control slides were prepared by substitution of the primary monoclonal antibody with the same dilution of an irrelevant mouse antibody of the same isotype (Dako). Positive PBMCs were counted per 100 cells.

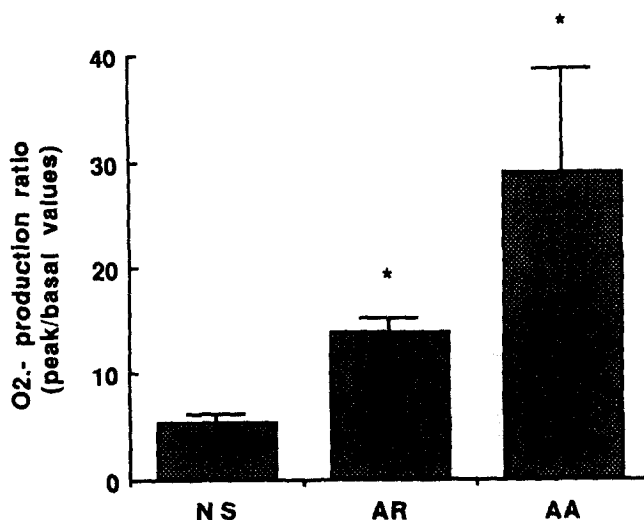


FIG. 2. O₂⁻ production ratio (chemiluminescence peak/basal values, mean ± SEM) obtained for atopic serum-stimulated BMs from patients with AA (29 ± 9.7), patients with AR (14 ± 1.3), and normal subjects (5.4 ± 0.8) after 2-hour adherence. Statistical analysis showed that there were significant differences among patients with AA, patients with AR, and normal subjects: $p < 0.05$ and $p < 0.01$, respectively.*

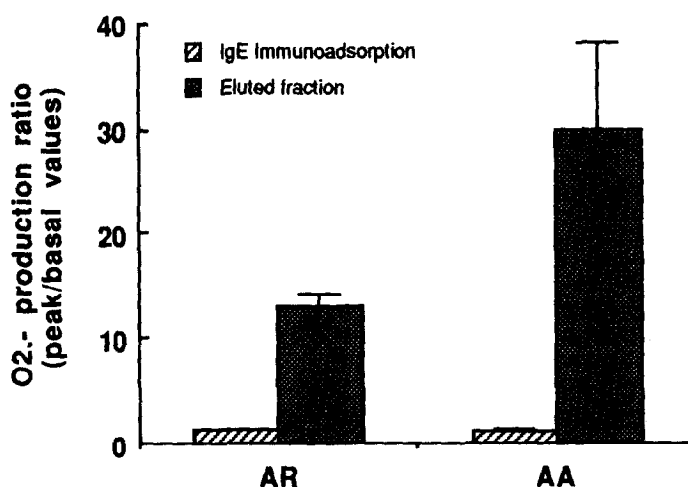


FIG. 3. O₂⁻ production ratio (chemiluminescence peak/basal values, mean ± SEM) obtained for anti-IgE immunoabsorbed serum-stimulated BMs for patients with AA (1.15 ± 0.07) and patients with AR (1.3 ± 0.08) after 2-hour adherence and for eluted fraction serum-stimulated BMs from patients with AA (30 ± 8.3) and patients with AR (13 ± 1.2) after 2-hour adherence.

Statistical analysis and expression of results

The chemiluminescence measurements were performed in duplicate for each subject. The nonparametric Wilcoxon matched-paired signed-rank test was used for statistical comparisons between stimuli for each patient group. The Mann-Whitney U test was used for statistical comparisons between patient groups for each stimulus (independent data). Spearman's rank-order correlations were established to compare O₂⁻ production after stimulation by the atopic serum versus the number of PBMCs with CD23 expression. Results are given as means ± SEM of the O₂⁻ production ratio:

chemiluminescence peak/basal values, which could be considered as an index of stimulability. Times to reach peak values are expressed in minutes (means ± SEM). The SAS version 6.06 software package (SAS Institute, Cary, N.C.) was used.

RESULTS Subjects

In the present study six patients had mild asthma (Aas score = 1 to 2) and one had moderately severe asthma (Aas score = 3). Pulmo-

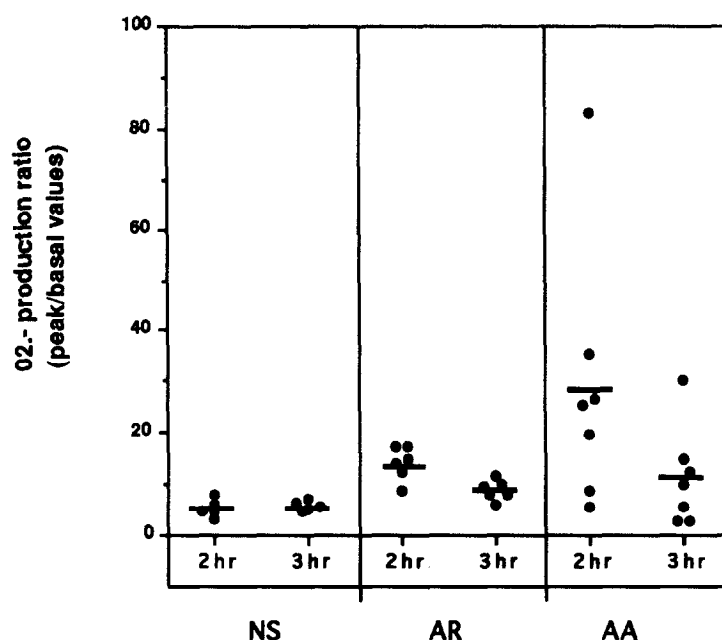


FIG. 4. Comparison of O_2^- production after BM stimulation by atopic serum maintained for 2 and 3 hours in adherence from patients with AA, patients with AR, and normal subjects. The statistical analysis showed a significant difference among the three groups (mean $\pm$ SEM at 2-hour adherence; normal subjects: 5.4 ± 0.8 , patients with AA: 29 ± 9.7 ; $p < 0.05$, and patients with AR: 14 ± 1.3 ; $p = 0.01$). There was a significant decrease in the adherence time for the AA group (2-hour adherence: 29 ± 9.7 , 3-hour adherence: 11 ± 4.2 ; $p < 0.05$) and for the AR group (2-hour adherence: 14 ± 1.3 , 3-hour adherence: 8.7 ± 0.7 ; $p = 0.05$).

nary function of the patients as assessed by FEV_1 ranged from 70% to 110% of predicted values. All patients with asthma and rhinitis were allergic (to house dust mites and/or pollen and/or molds) with 275 ± 73 U/L and 316 ± 99 U/L serum IgE, respectively (vs 49 ± 14 U/L for normal subjects). Characteristics of the patients are listed in Table I.

Measurement of superoxide release

The spontaneous production of O_2^- by unstimulated BMs (O_2^- production ratio) was greater in allergic subjects and reached a statistically significant difference after 3-hour adherence ($p < 0.05$): 1.4 ± 0.1 , 1.7 ± 0.3 , and 1 ± 0.03 in patients with AA, patients with AR, and normal subjects, respectively (Table II, Fig. 1). The kinetics of release were also significantly different, with a peak chemiluminescence later in allergic subjects (patients with AA: 35 ± 6 minutes, patients with AR: 27 ± 4.5 minutes) than in normal subjects (11.5 ± 1.5 minutes, $p = 0.02$).

The atopic serum stimulated the BM O_2^- release (Fig. 2) in all patients. The O_2^- production ratio was greater in patients with AA (29 ± 9.7 ; $p < 0.05$) and patients with AR (14 ± 1.3 ;

$p < 0.01$) as compared with normal subjects (5.4 ± 0.8). The kinetics of this release were not significantly different, with a peak chemiluminescence of 25 ± 1.5 , 28 ± 5 , and 33 ± 5 minutes in normal subjects, patients with AR, and patients with AA, respectively. Similarly, the time to reach the peak of O_2^- production induced by atopic serum did not significantly differ from that of unstimulated BMs. The serum from a healthy nonsmoker did not induce O_2^- release (data not shown).

Anti-IgE immunoadsorption of the atopic serum completely abrogated its effect. O_2^- production ratios were almost identical for anti-IgE-immunoadsorbed serum and controls: 1.3 ± 0.08 , 1.15 ± 0.07 in patients with AR and patients with AA, respectively. But this production was restored almost completely with the eluted fraction: 13 ± 1.2 and 30 ± 8.3 in patients with AR and patients with AA, respectively (Table II, Fig. 3).

The IgE-induced O_2^- production decreased with the time of adherence (Table II, Fig. 4) in both allergic patient groups. In the AA group the O_2^- production ratio was 29 ± 9.7 and 11 ± 4.2 for 2-hour and 3-hour adherence, respectively

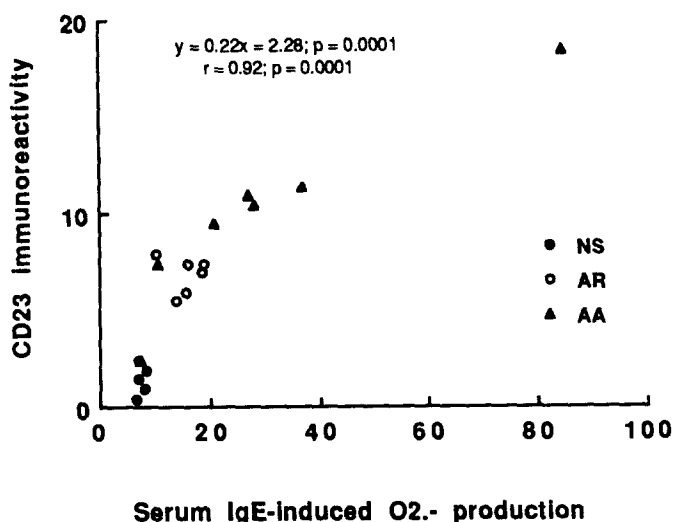


FIG. 5. Comparison of PBMC CD23 expression versus serum IgE-induced O₂⁻ production by BMs from all patients showed a linear correlation ($r = 0.92$; $p = 0.0001$; $y = 0.22x + 2.28$; $p = 0.0001$).

($p < 0.05$). In the AR group it was 14 ± 1.3 and 8.7 ± 0.7 for 2-hour and 3-hour adherence, respectively ($p = 0.05$). Moreover, the peak chemiluminescence induced by serum IgE stimulation was delayed with increased adherence time for all patients: 30 ± 2.7 minutes and 36.5 ± 2.7 minutes for 2-hour and 3-hour adherence, respectively ($p < 0.01$).

PBMC CD23 expression

Normal subjects had very low levels of PBMC CD23 expression (mean $\pm$ SEM, $1\% \pm 0.3\%$). Allergic patients had significantly elevated PBMC CD23 expression ($6.4\% \pm 0.4\%$ and $9.6\% \pm 1.8\%$ for patients with AR and patients with AA, respectively, $p < 0.01$). We did not find any statistically significant differences in PBMC CD23 expression between patients with AR and patients with AA ($p = 0.07$). Comparison of PBMC CD23 expression versus serum IgE-induced O₂⁻ production by BMs showed a linear correlation ($r = 0.92$; $p = 0.0001$; $y = 0.22x + 2.28$; $p = 0.0001$) (Fig. 5). No statistical correlations ($p < 0.07$) were found between patients' serum IgE levels and IgE-induced O₂⁻ production or between patients' serum IgE levels and CD23 expression ($r = 1.2$; $p = 0.2$).

DISCUSSION

In allergic diseases peripheral BMs are activated with respect to oxygen metabolite production¹²⁻¹⁵ and bear the CD23 low-affinity receptor for IgE.⁸ In this study we hypothesized that the

binding of IgE to CD23 on adherent BMs could generate reactive oxygen species, in particular O₂⁻. We examined this previously unknown pathway for NADPH activation by using a highly sensitive endotoxin-free chemiluminescence assay. We began our study with atopic serum and demonstrated that it induced O₂⁻ production by BMs and that serum IgE is the responsible factor in serum of atopic patients. In addition, we correlated this activation with the number of CD23-positive PBMCs from the same patients and analyzed the effect of the adherence time. We found that both spontaneous and serum IgE-stimulated BMs from allergic subjects produced greater amounts of O₂⁻ and that there was a linear correlation between CD23-positive PBMCs and O₂⁻ production induced by serum IgE.

In previous work, we demonstrated that BMs from patients with AA have an enhanced capacity to release reactive oxygen species when stimulated by phorbol esters and that they could thus be considered as primed *in vivo*.¹⁵ Our present study deals with direct activation of the respiratory enzyme NADPH oxidase when IgE binds to CD23. We were interested not only in the increased number of CD23 receptors on BMs from allergic patients but also in the functional status of these receptors, because IgE-CD23 binding could have been ineffective. We thus demonstrated that both aspects were enhanced and strongly correlated in allergic patients. As expected, there was no correlation between patients' serum IgE levels with the number or with

the functional status of the CD23 on BMs, because it is unlikely that IgE could modulate CD23 expression on BMs.^{8, 25} Moreover, this finding strongly suggests that the simple binding of IgE to low-affinity receptors activates the BMs and could participate in general activation of the respiratory-burst enzyme in allergic diseases, which is in agreement with many previous well-supported hypotheses.¹¹⁻¹⁵ It is noteworthy that the binding of monomeric IgE to high-affinity receptors, present on mast cells and basophil surfaces, does not seem to activate these cells, because a small proportion of these receptors need to be aggregated to activate the cell.²⁶ However, this may be dependent on the sensitivity of the technique used to assess cell activation and on the number of CD23 receptors on the cell surface. Moreover, using our chemiluminescence assay, we have also demonstrated the effect of anti-IgE after 30-minute IgE preincubation,¹⁴ as previously reported for alveolar macrophages.^{4,7} In comparison, the peak chemiluminescence stimulation by IgE alone was four fifths and one fifth of that of IgE/anti-IgE and phorbol esters, respectively (personal data). These results will be confirmed with other methodologies in a further work.

Our results on $O_2^{\cdot-}$ spontaneous production by unstimulated BMs from allergic patients suggests that these cells are activated in vivo to release reactive oxygen species. Because IgE alone generated $O_2^{\cdot-}$ and because the number of cells bearing the $Fc_\epsilon RII$ (CD23) is enhanced on BMs from atopic patients,^{8, 13} the greater production of $O_2^{\cdot-}$ by unstimulated BMs could be due in part to the increased number of these receptors on these cells. This latter observation has been widely published, and it appears that the number of CD23-positive peripheral BMs increases with the severity of the disease.^{8, 13, 25} Indeed, our patients had mild chronic allergic disease, which may explain our relatively low results.

Moreover, we showed that adherence participates in the response modulation, decreasing the IgE-induced $O_2^{\cdot-}$ production and delaying the peak chemiluminescence. We recently reported that adherence directly activates BM NADPH oxidase.¹⁵ Thus BM adherence on Petri dishes could be considered a physical stimulation of cells,²⁷ suggesting that BM margination in vessel and bronchoalveolar walls is associated with oxygen species release. In the present study the decrease cannot be explained by experimental problems because the viability of cells at the end of the chemiluminescence assay was greater than

98% and because the DNA measurement did not show any differences in the number of cells in the dishes of 2-hour adherent BMs versus 3-hour adherent BMs. This could be due in part to the participation of CD23 in the adherence mechanisms.²⁵ Indeed, the CD23 protein contains the consensus RGD amino-acid sequence (Arg311-Gly-Asp), which enables its adherence to fibronectin,²⁵ possibly involving changes in protein conformations leading to modulations of IgE-CD23 binding and then to delay and decrease in the $O_2^{\cdot-}$ production.

Our data suggest not only that BMs from patients with AA and AR with chronic stable allergic disease present an enhanced capacity to produce superoxide anion, but also that the IgE-CD23 binding on BMs may activate NADPH oxidase. Thus CD23-positive BMs may play an important role in chronic allergic diseases because of their ability to release reactive oxygen species partially through IgE-CD23 binding.

We thank Jean-Claude Nicolas, PhD, for his helpful discussions (INSERM U58) and Cécile Bonifacj, MD, for performing the statistical analysis (Département d'Information Médicale, Hôpital Lapeyronie, Université de Montpellier).

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