

- mucosa of patients with coeliac disease. *J Exp Med* 175: 293-6.
49. Azzawi M, Bradley B, Jeffery PK, et al. Identification of activated T lymphocytes and eosinophils in bronchial biopsies in stable atopic asthma. *Am Rev Respir Dis* 1990; 142:1407-13.
 50. Hynes RO. Integrins: versatility, modulation and signalling in cell adhesion. *Cell* 1992;69:11-25.
 51. Dri P, Cramer R, Spessotto P, Romano M, Patriarca P. Eosinophil activation on biologic surfaces. *J Immunol* 1991;147:613-20.
 52. Anwar ARE, Cromwell O, Walsh GW, Kay AB, Wardlaw AJ. Adhesion to fibronectin primes eosinophils via $\alpha 4/\beta 1$. *Immunology* 1994;82:222-8.
 53. Anwar ARE, Moqbel R, Walsh GW, Kay AB, Wardlaw AJ. Adhesion to fibronectin prolongs eosinophil survival. *J Exp Med* 1993;177:839-43.
 54. Mould AP, Wheldon A, Komoriya EA, Wayner EA, Yamada KM, Humphries MJ. Affinity chromatic isolation of the melanoma adhesion receptor for the IIICS region of fibronectin and its identification as the integrin $\alpha 4/\beta 1$. *J Biol Chem* 1990;265:4020.
 55. Vignola AM, Campbell AM, Chanez P, et al. HLA-DR and ICAM-1 expression on bronchial epithelial cells in asthma and chronic bronchitis. *Am Rev Respir Dis* 1993; 148:689-94.
 56. Altman LC, Ayars GH, Baker C, Luchtel DL. Cytokines and eosinophil-derived cationic proteins upregulate intercellular adhesion molecule-1 on human nasal epithelial cells. *J ALLERGY CLIN IMMUNOL* 1993;92:527-36.
 57. Bloeman PGM, van den Tweel MC, Henricks PA, et al. Expression and modulation of adhesion molecules on human bronchial epithelial cells. *Am J Respir Cell Mol Biol* 1993;9:586-93.
 58. Tosi MF, Stark JM, Smith CW, Hamedani A, Gruenert DC, Infeld MD. Induction of ICAM-1 expression on human airway epithelial cells by inflammatory cytokines: effects on neutrophil-epithelial cell adhesion. *Am J Respir Cell Mol Biol* 1992;7:214-21.
 59. Robbins RA, Koyama S, Spurzem JR. Modulation of neutrophil and mononuclear cell adherence to bronchial epithelial cells. *Am J Respir Cell Mol Biol* 1992;7:19-29.
 60. Cox G, Ohtoshi T, Vancheri C, et al. Promotion of eosinophil survival by human bronchial epithelial cells and its modulation by steroids. *Am J Respir Cell Mol Biol* 1991;4:525-31.
 61. Devalia JL, Sapsford RJ, Rusznak C, Davies RJ. The effect of human eosinophils on cultured human nasal epithelial cell activity and the influence of nedocromil sodium in vitro. *Am J Respir Cell Mol Biol* 1992;7:270-7.

Distinguishing features of basophil and neutrophil activation by major basic protein

Larry L. Thomas, PhD, Michelle D. Haskell, Emmanuel U. Sarmiento, MD, and Yasmeen Bilimoria, MD *Chicago, Ill.*

Eosinophils play a prominent role in the pathogenesis triggered by the inflammatory late-phase reactions in allergic disease.^{1, 2} Although eosinophil release of mediators such as leukotriene C₄ and platelet activating factor clearly contributes to the pathogenesis,¹⁻³ a body of evidence has accumulated to demonstrate that the cationic proteins present within eosinophil granules are of at least comparable importance to the observed

pathogenesis.³ Most extensively studied among the various cationic proteins is major basic protein (MBP), a highly charged (isoelectric point = 10.9) 13.8 kd polypeptide that comprises a large portion of the eosinophil granule protein.^{3, 4} Results from a number of laboratories suggest that the severity of allergic late-phase reactions, and in particular airway hyperreactivity in asthma, is correlated with presence of MBP in sputum and at the reaction sites.^{2, 3} Although the pathogenic activity of MBP has been attributed most frequently to the well-described cytotoxic properties of the molecule,³ other findings have demonstrated that MBP activates various inflammatory cells in a noncytotoxic fashion. Specifically, micromolar concentrations of MBP stimulate basophil histamine release,⁵ neutrophil release of superoxide anion and specific granule enzymes,⁶ and

From the Department of Immunology/Microbiology, Rush Medical College, Chicago.

Reprint requests: Larry L. Thomas, PhD, Department of Immunology/Microbiology Rush Medical College, 1653 W. Congress Pkwy., Chicago, IL 60612.

J ALLERGY CLIN IMMUNOL 1994;94:1171-6.

Copyright © 1994 by Mosby-Year Book, Inc.

0091-6749/94 \$3.00 + 0 1/0/59913

Abbreviations used

[Ca ²⁺] _i :	Concentration of cytosolic free calcium
EGTA:	Ethyleneglycol-bis-(β-aminoethylether)-N,N,N',N'-tetraacetic acid
FMLP:	N-formyl-methionyl-leucyl-phenyl-alanine
GM-CSF:	Granulocyte-macrophage colony-stimulating factor
IL:	Interleukin
MBP:	Major basic protein
raMBP:	Reduced and alkylated major basic protein

platelet release of alpha granule contents.⁷ Given the participation of basophils and neutrophils in late-phase reactions in both cutaneous and pulmonary tissue,⁸⁻¹¹ it is postulated that activation of basophil and neutrophil mediator release by MBP may contribute to the inflammatory events in allergic late-phase reactions. Accordingly, delineation of the mechanisms for basophil and neutrophil activation by MBP may provide insights into new approaches for the therapeutic control of allergic disorders.

CHARACTERISTICS OF BASOPHIL AND NEUTROPHIL ACTIVATION BY MBP

The possibility that MBP stimulates basophil mediator release was predicated on the high (14%) arginine content of the MBP molecule⁴ and the finding that poly-L-arginine is a potent stimulus for basophil histamine release.¹² Consistent with the different percentages of charged arginine residues in the two molecules, stimulation of basophil histamine release by MBP occurs at approximately 10-fold higher molar concentrations (1 to 5 μmol/L) than does stimulation by a 13.9 kd poly-L-arginine.⁵ However, additional results suggest that the positive charge of the MBP molecule is not solely responsible for the activity of MBP. First, reduction and alkylation of the MBP molecule diminishes both the potency and efficacy of the native MBP molecule. At concentrations (1 to 3 μmol/L) optimal for native MBP, raMBP does not stimulate histamine release.¹³ The raMBP does stimulate histamine release at 10-fold higher concentrations, but the level of histamine release is lower than that obtained with native MBP.⁵ Second, eosinophil cationic protein, which like MBP has a high isoelectric point (10.8)

and arginine content (15%), does not stimulate basophil histamine release at the concentrations optimal for MBP.¹³ Third, basophil histamine release stimulated by MBP and by poly-L-arginine differs in sensitivity to pharmacologic agents, including theophylline and the calmodulin antagonist W-7, as well as in sensitivity to pertussis toxin treatment.¹⁴ MBP-stimulated histamine release is inhibited by these agents, whereas release stimulated by poly-L-arginine is inhibited minimally or not at all.

Neutrophil activation by MBP displays the same concentration requirements and the same profile of responsiveness as established for stimulation of basophil histamine release. MBP in concentrations of 1 to 5 μmol/L stimulates neutrophil oxidative metabolism, specific granule discharge, and increased surface expression of CR3.^{6, 15} However, neither raMBP nor eosinophil cationic protein, in concentrations that are optimal for activation by MBP, stimulates neutrophil oxidative metabolism as measured by luminol-enhanced chemiluminescence.⁶ The same concentrations of raMBP and eosinophil cationic protein, likewise, do not mimic the ability of MBP to stimulate increases in neutrophil expression of CR3.¹⁵

CALCIUM SIGNALING AND MBP ACTIVATION

Calcium plays an integral role in the regulation of numerous cellular responses. An increase in the concentration of cytosolic free calcium ([Ca²⁺]_i) is a critical event in intracellular signaling cascades, triggered through either G protein-linked by tyrosine kinase-associated receptors.¹⁶ Previous studies have shown that stimulation of basophil histamine release and neutrophil oxidative metabolism by MBP are dependent on the presence of extracellular calcium.^{5, 6} However, results of recent studies indicate distinct differences in the nature of calcium signaling in the two cells in response to activation by MBP.

Studies by MacGlashan et al.^{17, 18} have demonstrated two phases for the stimulated increase in basophil [Ca²⁺]_i: an initial mobilization of intracellular calcium stores and a subsequent elevation dependent on the presence of extracellular calcium. Both phases are evident during basophil activation by optimal or supraoptimal concentrations of anti-IgE,¹⁸ as well as by N-formyl-methionyl-leucyl-phenylalanine (FMLP).¹⁷ In contrast, activation by C5a¹⁷ or platelet activating factor¹⁹ is accompanied only by the initial elevation in [Ca²⁺]_i. Release of calcium from the intracellular

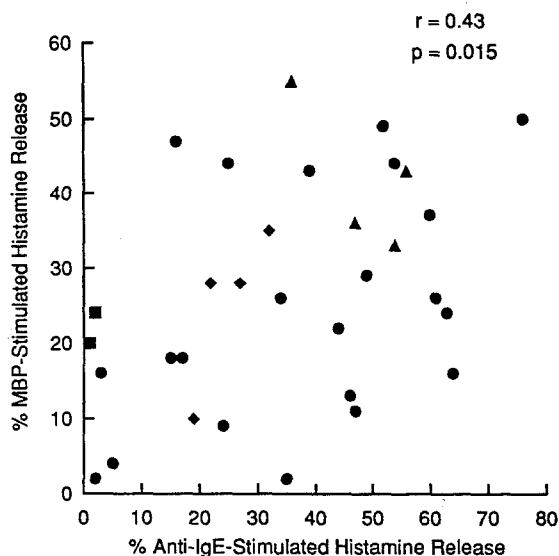


FIG. 1. Relationship between the levels of basophil histamine release stimulated by 1.5 or 2 $\mu\text{mol/L}$ MBP and 0.1 $\mu\text{g/ml}$ anti-IgE. Histamine release was measured as described previously.¹⁴ Significance was established by regression analysis.

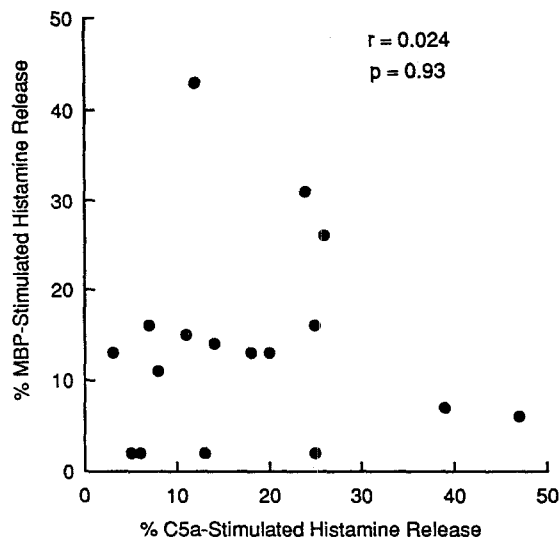


FIG. 2. Relationship between the levels of basophil histamine release stimulated by 1.5 or 2 $\mu\text{mol/L}$ MBP and 100 ng/ml rC5a. Histamine release was measured as described previously.¹⁴ Significance was established by regression analysis. Each value is the result of a single experiment with cells from a different donor. Three of the experiments and 10 of the donors are the same as those used for the results presented in Fig. 1.

stores is associated with histamine release stimulated by FMLP or C5a,¹⁷ but IgE-mediated release is associated with the second phase of the elevation of $[\text{Ca}^{2+}]_i$.¹⁸ Addition of ethyleneglycol-bis(β -aminoethylether)-N, N, N', N'-tetraacetic acid (EGTA) to the incubation buffer abolishes both histamine release and the second, but not the initial, increase in $[\text{Ca}^{2+}]_i$ stimulated by anti-IgE. Recent results,²⁰ obtained with Fura-2-loaded basophils and digital video microscopy, indicate that MBP also triggers elevations in basophil $[\text{Ca}^{2+}]_i$, with the pattern of the elevations similar to that observed with IgE-mediated activation. The increase in $[\text{Ca}^{2+}]_i$ occurs in a concentration-dependent fashion that parallels the increase in histamine release. Both phases of the increase in $[\text{Ca}^{2+}]_i$ that are observed with other stimuli^{17, 18} are also evident after stimulation by MBP, although the immediate transient increase associated with mobilization of intracellular calcium is blunted in comparison with that observed with FMLP or supraoptimal concentrations of anti-IgE.^{17, 18} Like IgE-mediated histamine release,¹⁸ MBP-stimulated histamine release is also dependent on the second phase of the $[\text{Ca}^{2+}]_i$ increase, because both histamine release and the second phase of the $[\text{Ca}^{2+}]_i$ increase are abolished by the addition of EGTA to the incubation buffer.²⁰

Additional results indicate that there is also a loose but statistically significant correlation between the levels of histamine release stimulated by optimal concentrations of MBP and anti-IgE. A plot of the levels of histamine release stimulated by 1.5 or 2 $\mu\text{mol/L}$ MBP in 32 experiments with cells of 18 donors versus the levels of histamine release stimulated by 0.1 $\mu\text{g/ml}$ of anti-IgE in the same experiments is shown in Fig. 1. In contrast, a similar evaluation indicates that no correlation exists between the level of histamine release stimulated by 1.5 or 2 $\mu\text{mol/L}$ MBP and the level of release stimulated by an optimal concentration (100 ng/ml) of rC5a (Fig. 2). As shown in Fig. 1 by the results obtained with cells of three donors (triangles, diamonds, and squares, respectively), the relationship between the levels of histamine release stimulated by MBP and those stimulated by anti-IgE for a single individual remains fairly constant when examined in multiple experiments. In general, the results shown in Fig. 1 indicate a tendency for basophils that respond well to IgE-mediated activation to also respond well to activation by MBP as measured by histamine release. It is clear, however, that responsiveness to anti-IgE certainly does not predict a priori responsiveness to MBP. MBP can

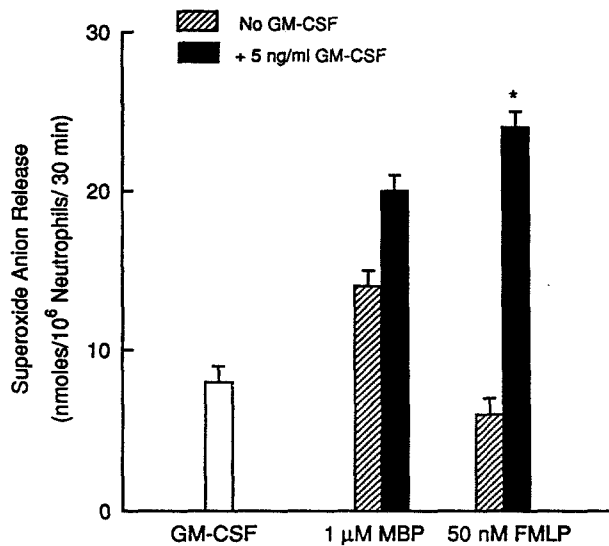


FIG. 3. Neutrophils (10^6 /ml) in Hanks' balanced salt solution, pH 7.4, containing 10 mmol/L N-2-hydroxyethylpiperazine-N-2-ethane sulfonic acid, 0.003% human serum albumin, 0.6 mmol/L CaCl_2 , and 1 mmol/L MgCl_2 were preincubated for 75 minutes at 37°C in the presence and absence of GM-CSF. MBP or FMLP and 80 $\mu\text{mol/L}$ ferricytochrome c were added, and the cells were incubated for 30 minutes at 37°C . Final incubation volume was 0.5 ml. The reactions were stopped by centrifugation at 200 g for 10 minutes at 4°C , and the $A_{550\text{ nm}}$ of the supernatant was measured after a 1:6 dilution in saline solution. Superoxide anion release was calculated as nanomoles cytochrome c reduced per 10^6 cells, as described previously.⁶ Results are expressed as the means $\pm$ SEM after subtraction of spontaneous release. * $p < 0.05$.

stimulate histamine release from basophils that are unresponsive to anti-IgE (note the filled squares in Fig. 1), and conversely, MBP may cause little release from cells that respond well to stimulation by anti-IgE. Recent findings²¹ underscore the complexity of the parameters that control the level of histamine release obtained with optimal concentrations of anti-IgE. Thus, it is likely that the relationship shown in Fig. 1 simply reflects overlap in one of these parameters, possibly at the level of the increase in $[\text{Ca}^{2+}]_i$ required for histamine release by anti-IgE or by MBP.

Other results²² indicate that in contrast to basophil activation by MBP, neutrophil activation by MBP concentrations (1 to 3 $\mu\text{mol/L}$), which are optimal for stimulating neutrophil superoxide anion and lysozyme release,⁶ is not accompanied by the immediate increase in $[\text{Ca}^{2+}]_i$ associated with calcium release from intracellular stores. MBP does, however, stimulate a gradual increase in $[\text{Ca}^{2+}]_i$ in cytochalasin B-treated neutrophils,

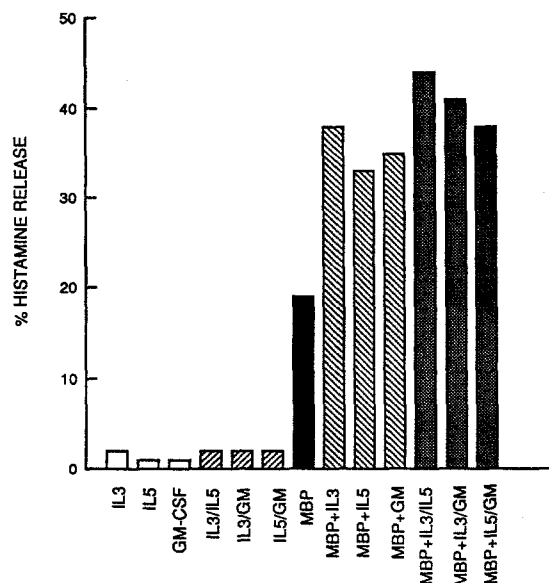


FIG. 4. Enhancement of MBP-stimulated histamine release by IL-3, IL-5, and GM-CSF. Basophil-containing mononuclear cells were preincubated with 5 ng/ml of the cytokines alone and in combination, as indicated, for 15 minutes at 37°C . The cells were then incubated with 1.5 $\mu\text{mol/L}$ MBP for 30 minutes at 37°C , and histamine release was measured as described previously.¹⁴ The results of a single experiment are presented.

which can be detected after 3 minutes of activation and reaches a maximum of 50 to 100 nmol/L after 6 minutes of stimulation, the latest time point examined.²² Like the second phase of the MBP-induced elevation in $[\text{Ca}^{2+}]_i$ in basophils, the delayed increase in $[\text{Ca}^{2+}]_i$ in cytochalasin B-treated neutrophils is abolished by the addition of EGTA to the incubation medium. The MBP-induced increase in $[\text{Ca}^{2+}]_i$ in cytochalasin B-treated neutrophils, however, is not necessary for activation of neutrophil superoxide anion or lysosomal enzyme release. MBP does not stimulate an increase in $[\text{Ca}^{2+}]_i$ in neutrophils in the absence of cytochalasin B treatment,²² whereas the levels of MBP-stimulated superoxide anion and lysozyme release are not different in the presence and absence of cytochalasin B.^{6, 22} Thus, the calcium requirement for neutrophil activation by MBP appears to be unrelated to the signaling events for oxidative burst activity or for granule enzyme release.

CYTOKINE EFFECTS ON MBP ACTIVATION

Of concomitant interest to delineation of the signaling events that mediate cell activation by

TABLE I. Summary of basophil and neutrophil responses to MBP

Response	Basophil	Neutrophil
	Histamine release, leukotriene C ₄ release 1-5 μ mol/L	Superoxide anion and lysozyme release, CR3 expression 1-5 μ mol/L
MBP concentration requirements		
Activated by raMBP (2 μ mol/L)	No	No
Activated by eosinophil cationic protein (2 μ mol/L)	No	No
Ca ²⁺ -dependent activation	Yes	Yes
Increase in [Ca ²⁺] _i	Yes	(Yes)*
Mobilization of intracellular Ca ²⁺	Yes	No
Enhanced by IL-3, IL-5, and GM-CSF	Yes	No

*With cytochalasin B pretreatment.

MBP is the impact of other mediators present at sites of allergic reactions on activation. In situ hybridization and protein measurement have documented the presence of interleukin (IL)-5, IL-3, and granulocyte-macrophage colony-stimulating factor (GM-CSF) at sites of pulmonary and cutaneous allergic disease²³⁻²⁵ and thus have implicated the cytokines in the pathogenesis of these disorders. The roles of IL-5, IL-3, and GM-CSF in promoting eosinophil growth and differentiation, prolonging eosinophil survival in vitro, and enhancing a number of eosinophil effector functions, including degranulation, are well defined.²⁶ In addition, GM-CSF primes neutrophils for enhanced responsiveness, including superoxide anion release, to activation by chemoattractants^{27, 28}; whereas IL-3, IL-5, and GM-CSF each prime basophils for enhanced responsiveness to IgE-mediated activation, as well as to activation by IgE-independent stimuli such as C5a.^{29, 30} Together, these findings suggested that IL-3, IL-5, and GM-CSF, in addition to promoting eosinophil function, may also enhance neutrophil and basophil activation by MBP.

Initial results, however, indicate that priming neutrophils by a 75-minute preincubation with 5 ng/ml of GM-CSF at 37° C, conditions that are optimal for enhancing superoxide anion release stimulated by chemoattractants such as FMLP, does not potentiate superoxide anion release stimulated by 1 μ mol/L MBP (Fig. 3). In these experiments preincubating the neutrophils with GM-CSF also increased spontaneous superoxide anion release, but the combined effect with MBP was a less than additive increase in the amount of superoxide anion release. In contrast, preincubation with GM-CSF potentiated superoxide anion release stimulated by 50 nmol/L FMLP, as noted by the synergistic increase in FMLP-induced su-

peroxide anion production after neutrophil priming with GM-CSF in the same experiments (Fig. 3). No superoxide anion production was detected for any of the experimental conditions in the presence of 50 μ g/ml of superoxide dismutase (data not shown).

In contrast to the lack of a GM-CSF effect on neutrophil superoxide anion release stimulated by MBP, a brief exposure of basophils to IL-3, IL-5, or GM-CSF primes them for enhanced responsiveness to MBP.³¹ In agreement with results obtained with other basophil stimuli,^{29, 30} the results in Fig. 4 show that a 15-minute preincubation of the basophils with 5 ng/ml of the cytokines enhances histamine release stimulated by 1.5 μ mol/L MBP. Consistent with the receptors for the three cytokines having a shared β -chain,³² preincubation of the cells with suboptimal concentrations of the cytokines in combination produces levels of enhancement that are less than the additive effects obtained with the individual cytokines alone.

CONCLUSION

Table I provides a summary of the results described here and illustrates that although basophil and neutrophil activation by MBP have many characteristics in common, the two cells differ in calcium signaling and sensitivity to priming by cytokines. Activation of neutrophil superoxide anion release by MBP proceeds independently of an elevation in [Ca²⁺]_i and is not enhanced by exposure of the neutrophils to GM-CSF. In contrast, an elevation in [Ca²⁺]_i appears to be integral to basophil activation by MBP. Significantly, basophil mediator release stimulated by MBP is enhanced after a brief preincubation with IL-3, IL-5, or GM-CSF. These results suggest that avenues may exist for the differential regulation of basophil

and neutrophil activation by MBP. In addition, the results indicate that MBP stimulation of basophil mediator release may be influenced by the presence of IL-3, IL-5, and GM-CSF at sites of allergic late-phase reactions.

REFERENCES

- Weller PF. The immunobiology of eosinophils. *New Engl J Med* 1991;324:1110-8.
- Gleich GJ, Adolphson CR, Leiferman KM. The biology of the eosinophilic leukocyte. *Ann Rev Med* 1993;44:85-101.
- Sur S, Adolphson CR, Gleich GJ. Eosinophils. Biochemical and cellular aspects. In: Middleton E Jr, Reed CE, Ellis EF, Adkinson NF Jr, Yunginger JW, Busse WW, eds. *Allergy: principles and practice*. 4th ed. St Louis: Mosby, 1993:169-200.
- Wasmoen T, Bell MP, Loegering DA, Gleich GJ, Prendergast FG, McKean DJ. Biochemical and amino acid sequence analysis of human eosinophil granule major basic protein. *J Biol Chem* 1988;263:12559-63.
- O'Donnell MC, Ackerman SJ, Gleich GJ, Thomas LL. Activation of basophil and mast cell histamine release by eosinophil granule major basic protein. *J Exp Med* 1983;157:1981-91.
- Moy JN, Gleich GJ, Thomas LL. Nontoxic activation of neutrophils by eosinophil granule major basic protein: synergistic influence on superoxide anion generation and lysosomal enzyme release. *J Immunol* 1990;145:2626-32.
- Rohrbach MS, Wheatley CL, Slifman NR, Gleich GJ. Activation of platelets by eosinophil granule proteins. *J Exp Med* 1990;172:1271-4.
- Lichtenstein LM, Bochner BS. The role of basophils in asthma. *Ann NY Acad Sci* 1991;629:48-61.
- Charlesworth EN, Kagey-Sobotka A, Schleimer RP, Norman PS, Lichtenstein LM. Prednisone inhibits the appearance of inflammatory mediators and the influx of eosinophils and basophils associated with the cutaneous late-phase response to allergen. *J Immunol* 1991;146:671-6.
- Lemanske RF, Kaliner M. Late-phase IgE-mediated reactions. *J Clin Invest* 1988;81:1-13.
- Zweiman B, Shalit M, Atkins PC. Release of lactoferrin and elastase in human allergic skin reactions. *J Immunol* 1990;144:3953-60.
- Foreman JC, Lichtenstein LM. Induction of histamine secretion by polycations. *Biochim Biophys Acta* 1980;629:587-603.
- Zheutlin LM, Ackerman SJ, Gleich GJ, Thomas LL. Stimulation of basophil and rat mast cell histamine release by eosinophil granule-derived cationic proteins. *J Immunol* 1984;133:2180-5.
- Thomas LL, Zheutlin LM, Gleich GJ. Pharmacological control of human basophil histamine release stimulated by eosinophil granule major basic protein. *Immunology* 1989;66:611-5.
- Moy JN, Thomas LL, Whisler LC. Eosinophil major basic protein enhances the expression of neutrophil CR3 and p150,95. *J ALLERGY CLIN IMMUNOL* 1993;92:598-606.
- Berridge MJ. Inositol triphosphate and calcium signaling. *Nature* 1993;361:315-25.
- MacGlashan DW Jr, Warner J. Stimulus-dependent leukotriene C4 release from human basophils: a comparative study of C5a and F met-leu-phe. *J Leukoc Biol* 1991;49:29-40.
- MacGlashan DW Jr, Botana LM. Biphasic Ca^{2+} responses in human basophils. Evidence that the initial transient elevation associated with the mobilization of intracellular calcium is an insufficient signal for degranulation. *J Immunol* 1993;150:980-91.
- Columbo M, Casalaro V, Warner JA, MacGlashan DW Jr, Kagey-Sobotka A, Lichtenstein LM. The mechanism of mediator release from human basophils induced by platelet-activating factor. *J Immunol* 1990;145:3855-61.
- MacGlashan DW Jr, Gleich GJ, Thomas LL. Increases in cytosolic calcium ($[Ca^{2+}]_i$) accompany basophil activation by eosinophil major basic protein. (submitted for publication).
- MacGlashan DW Jr. Releasability of human basophils: cellular sensitivity and maximal histamine release are independent variables. *J ALLERGY CLIN IMMUNOL* 1993;91:605-15.
- Haskell MD, Moy JN, Gleich GJ, Moy JN. Neutrophil stimulation by eosinophil major basic protein is associated with increased tyrosine activity. (submitted for publication).
- Kay AB, Ying S, Varney V, et al. Messenger RNA expression of the cytokine gene cluster, interleukin 3 (IL-3), IL-4, IL-5 and granulocyte/macrophage colony stimulating factor, in allergen-induced late-phase cutaneous reactions in atopic subjects. *J Exp Med* 1991;173:775-8.
- Sedgwick JB, Calhoun WJ, Gleich GJ, et al. Immediate and late airway response of allergic rhinitis patients to segmental antigen challenge. Characterization of eosinophil and mast cell mediators. *Am Rev Respir Dis* 1991;144:1274-81.
- Massey W, Friedman B, Kato M, et al. Appearance of granulocyte-macrophage colony-stimulating factor activity at allergen-challenged cutaneous late-phase reaction sites. *J Immunol* 1993;150:1084-92.
- Abu-Ghazaleh RI, Kita H, Gleich GJ. Eosinophil activation and function in health and disease. In: Coffey RG, ed. *Granulocyte responses to cytokines: basic and clinical research*. New York: Marcel Dekker, 1992:137-67.
- Lopez AF, Williamson DJ, Gamble JR, et al. Recombinant human granulocyte-macrophage colony-stimulating factor stimulates in vitro mature human neutrophil and eosinophil function, surface receptor expression, and survival. *J Clin Invest* 1986;78:1220-8.
- Weisbart RH, Kwan L, Golde DW, Gasson JC. Human GM-CSF primes neutrophils for enhanced oxidative metabolism in response to the major physiologic chemoattractants. *Blood* 1987;69:18-21.
- Hirai K, Morita Y, Misaki Y, et al. Modulation of human basophil histamine release by hematopoietic growth factors. *J Immunol* 1988;141:3958-64.
- Bischoff SC, Brunner T, deWeck A, Dahinden CA. Interleukin 5 modifies histamine release and leukotriene generation by human basophils in response to diverse agonists. *J Exp Med* 1990;172:1577-88.
- Sarmiento EU, Gleich GJ, Thomas LL. IL-3, IL-5, and GM-CSF potentiate basophil mediator release stimulated by eosinophil granule protein. (submitted for publication).
- Kishimoto T, Taga T, Akira S. Cytokine signal transduction. *Cell* 1994;76:253-62.