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The Late Phase Response: Clinical Implications*

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Editor's Introduction

Acute hypersensitivity responses to antigenic challenge in sensitized individuals are well known to clinicians. Nevertheless, the late phase response (LPR) which occurs hours or days after challenge has, until recently, received less attention and been less well appreciated. Dr. Lemanske describes the late phase response of various target organs such as the skin, lung, and nose as well as the systemic response. The histopathology and pathogenesis of the LPR, including the role of mediators, are described along with the symptom complex related to each target organ.

Investigation of these late phase responses indicates that inflammation is a major component of the reaction, and this element must be addressed when considering rational treatment.

James J. Leonard, M.D.

Antigen challenge in IgE-sensitized individuals can result in biphasic clinical responses in a variety of tissues including the skin, lung, and nose. The immediate component occurs within minutes following challenge, resolves in 30 to 60 minutes, and may be followed by late phase reactions (LPRs). The LPRs begin 3 to 4 hours following challenge, reach maximal intensity in 6 to 10 hours, and resolve by 24 to 48 hours. Although the immediate consequences of allergic reactions have been appreciated for decades, recent attention has focused on LPRs as a means to explain the more chronic clinical features of various allergic diseases including asthma, allergic rhinitis, urticaria, and atopic dermatitis.

The recognition and description of LPRs actually dates back over a century.¹ In 1873, Blackley reported the development of both upper and lower respiratory tract symptoms that evolved over hours following exposure to grass pollen. In 1922, Cooke skin tested himself to horse dander and described not only the initial wheal and flare reaction, but also the development of edema and erythema, which were still present the next day.²

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Eighty years following Blackley's report, Herxheimer³ emphasized that the "late bronchial reaction" was "of great practical importance" and was associated with more severe asthmatic symptoms than those present in patients without late reactions.

The underlying mechanisms responsible for the second, more delayed, response remained obscure until Pepys et al.,⁴ while working on the pathogenesis of allergic bronchopulmonary aspergillosis, suggested that the dual skin test reactions seen in these patients might be caused by an initial type I reaction followed by a type III or Arthus response. It was considered possible that late responses observed following challenge with other antigens were also the result of local Arthus reactions until Dolovich et al.⁵ in 1973, and Solley et al.⁶ and Umemoto et al.⁷ in 1976, demonstrated that both the early and phase reaction and LPR following skin testing could be mediated solely by IgE-dependent mechanisms. The IgE dependency of these reactions strongly implicated a central role for mast cell and basophil participation in their pathogenesis. With the aid of various animal models and more extensive analyses in humans, a more comprehensive understanding of the LPR is now emerging.

Cutaneous LPR

Incidence and Macroscopic Appearance

Cutaneous antigen challenge can produce a variety of macroscopic and histologic responses that are dependent on the immunologic status of the sensitized individual. Four major types of late-onset dermal reactions to injected antigen have been described in humans and have recently been reviewed by deShazo and coworkers.⁸ These reactions include delayed hypersensitivity (DH), cutaneous basophil hypersensitivity (CBH), Arthus reactions, and allergic LPRs. Although cutaneous LPRs have features in common with DH, CBH, and Arthus reactions, they also have unique characteristics warranting a separate classification. Cutaneous LPRs can be defined macroscopically, histologically, and immunologically.

Allergen skin testing may result in isolated immediate reactions, isolated delayed reactions, or dual reactions in approximately 20%,⁹ 6% to 14%,^{8,10} and 66% to 85%^{8,11} of challenged individuals, respectively. Following cutaneous allergen challenge, wheal and flare reactions develop almost immediately and are characterized by a central area of pale swelling surrounded by a halo of erythema. This macroscopic response, which is intensely pruritic, peaks in 10 to 15 minutes and, if only an isolated immediate reaction develops, will resolve within 30 to 60 minutes. Alternatively, the immediate response may evolve into LPRs, characterized by a burning discomfort, pruritus, erythema, induration, and warmth. Cutaneous LPRs generally peak at 6 to 8 hours and may resolve by 24 hours. In contrast to pulmonary, nasal, and systemic late LPRs, the cutaneous response is not biphasic, but rather a progression from the immediate wheal and flare response.¹²

Although not all skin test-positive individuals manifest clinical LPRs, the intensity of clinically apparent LPRs correlates directly with the intensity of the immediate reaction.^{7,13} DeShazo and coworkers¹³ reported that the smallest wheal diameter noted 15 minutes after skin testing that was associated with a 5-mm or larger LPR was 9.5 ± 1.0 mm.¹³ The development of LPRs may follow either prick or intradermal skin testing.⁷ The size of the initial reaction may not be the only determinant in the development of the late response. Indeed, despite increasing the immediate wheal reaction to 18.2 ± 1.3 mm by admixing ragweed antigen with histamine before skin testing, deShazo and coworkers¹³ were unable to induce LPRs in patients who only produce immediate reactions to allergen challenge. Local tissue innervation, although capable of affecting the flare associated with the immediate response, does not affect the development of the LPR.⁷

Substances that have been reported to induce LPRs include various allergens, such as ragweed,^{6,14} grass, and tree pollens⁷; molds, such as *Alternaria*, *Hormodendrum*, and *Aspergillus*; cat and dog dander; feathers; dust⁹ and *Dermatophagoides pteronyssinus* (house dust mite)¹⁵; *Bacillus subtilis* enzyme preparations¹⁶; and insulin¹⁷ (Table 1). Other agents known to induce mast cell secretion, such as monospecific antiserum to IgE, its F(ab')₂ fragment,⁵ or compound 48/80,^{5,6} may also induce LPRs. Codeine, thought to induce mast cell secretion,¹⁸ produces immediate but not late skin test responses in normal individuals and slight LPRs in patients with angioedema.¹⁹

Histopathology

The histopathologic changes seen following cutaneous allergen challenge in allergic individuals have been evaluated by a number of investigators and are illustrated in Figure 1. Overall, human cutaneous LPRs are characterized by a mixed cellular infiltrate. Recent work by Charlesworth et al.²⁰ has demonstrated a more substantial influx of both eosinophils and basophils during the late response in comparison with mononuclear cells and neutrophils. Although one report indicates their presence,²¹ immunoglobulins (other than IgE) and complement do not appear to contribute significantly to the pathogenesis of LPRs.⁶ Although somewhat controversial, fibrin deposition may be a distinguishing feature of skin biopsies following allergen challenge in individuals who develop LPRs.¹²

Skin Blister Fluid Studies

To determine more precisely the mediator profile temporally associated with the development of LPRs, skin blister fluid studies have been performed.^{20,22-37} In this model, antigen or control buffer is placed in a chamber over an area of denuded skin, and fluid from the chamber is collected at various times afterward. A variety of mediators have been measured in these secretions, and their levels appear to be influenced by the time point studied, the concentration of antigen, and whether the skin is

TABLE 1.
Agents Reported to Cause LPRs in Various Target Organs

Skin	Lung	Nose	Systemic*
Pollens	Pollens	Pollens	Rabies vaccine
Ragweed	Ragweed	Ragweed	Pollen mold and dust
Grass	Molds	Grass	extracts
Tree	<i>Alternaria</i>		Yellow jacket venom
Molds	<i>Candida</i>		Antimicrobial drugs
<i>Alternaria</i>	<i>Aspergillus</i>		→ Penicillins
<i>Hormodendrum</i>	Animal danders		→ Cephalosporins
<i>Aspergillus</i>	Cat		→ Sulfamethoxazole-
Animal dander	Laboratory animals		trimethoprim
House dust and	Insects		Radiocontrast material
dust mite	Housedust mite		Heterologous
(<i>D pteronyssinus</i>)	(<i>D pteronyssinus</i>)		antibodies
<i>B subtilis</i> enzyme	Sewer flies		Aspirin
Insulin	Cockroaches		Zomepirac
	Dusts		
	House dust		
	Wood dust		
	Grain dust		
	Soybean dust		
	Industrial chemicals		
	Plicatic acid		
	Platinum salts		
	Piperazine		
	hydrochloride		
	Ethanolamines		
	Papain		
	Chromium and		
	nickel salts		
	Diphenylmethane		
	diisocyanate		
	Toluene diisocyanate		
	Exercise		
	Distilled water		

*Biphasic or protracted

continuously or intermittently exposed to antigen.³⁷ Chamber-fluid histamine levels tend to be greatest 30 minutes to 1 hour following challenge but can be elevated for as long as 9 hours.^{22, 24, 31, 34, 35, 37} The quantity of histamine released correlates with the intensity of the LPR.²⁶ A biphasic pattern of histamine release has also been described.²⁰

Since histamine alone cannot cause LPRs nor act as a gatekeeper,¹³ it

may be a marker for prolonged release from the mast cell of other mediators that are responsible for the LPR. In this regard, Dorsch and Ring²³ demonstrated that skin blister fluid obtained 30 and 60 minutes following allergen challenge could produce macroscopic LPRs if injected elsewhere. High doses of skin blister fluid obtained from sites not exposed to allergen produced smaller LPRs than those produced by blister fluid taken from allergen-provoked sites developing dual skin reactions. These results indicate that factors are released or generated within the first hour following allergen challenge that can passively induce LPRs.

	ANTIGEN CHALLENGE	
5 minutes		Vessel engorgement (120)
		Edema of papillary dermis (120)
10 minutes		Perivascular neutrophils (90%)
		and eosinophils (10%) (120)
30 minutes		90% eosinophils (120)
1 hour		Mononuclear cells initially (6)
3 hours		10% eos, 82% neuts, 8% monos (120)
4 hours		7% eos (121)
6 hours		Edema, mast cell degranulation (5)
		Perivascular eos; No IgG, IgM, IgA,
		complement or fibrin (5)
6-8		Mixed infiltrate + fibrin (13)
8 hours		Mixed cellular infiltrate:
		48% lymphs; 7% monos, 27% eos;
		9% neuts; 3% basos (6)
		No immunoglobulins or complement (6)
		Eosinophil granules (6)
		Mixed infiltrate with basophils 14x,
		eosinophils 6x, monos 2x, neuts
		2x control (20)
24 hours		10% eos, 82% neuts, 8% monos (120)
		No fibrin (11)
48 hours		Dense perivascular infiltrate-eos
		and monos (10)

FIG 1.

Summary of histopathologic findings in the skin at various times following antigen challenge. Numbers in parentheses are reference numbers. eos = eosinophils; neuts = neutrophils; monos = mononuclear leukocytes; lymphs = lymphocytes; basos = basophils.

Kallikrein, Hageman factor,³² and thromboxane B₂²⁴ activity have also been measured in skin blister fluid. Leukotriene C₄ (LTC₄) levels are increased following challenge.²⁹ The pattern of release of LTC₄ differs from histamine, since peak levels occur 2 to 6 hours following challenge.³⁷ Since intradermal injections of LTC₄ in humans can induce wheal and flare responses that persist for hours, these findings support a possible role for LTC₄ in the pathogenesis of the LPR. The major cyclooxygenase pathway product formed is prostaglandin D₂ (PGD₂), with levels that reach maximum at 5 to 6 hours after challenge and that correlate with the intensity of the late response.³⁷

Tryptase is the primary neutral protease found in mast cells and in much smaller amounts in basophils. The ability to detect histamine and tryptase within the first hour following allergen challenge, but only significantly elevated levels of histamine and not tryptase later, suggests a potential participation of basophils in the cutaneous late response.³⁴ Indeed, basophil influx during the late response has recently been reported.²⁰ However, the late rise in PGD₂ levels and the continued elevations in histamine levels are also consistent with mast cell involvement.³⁷

With the use of membrane filters, skin blister techniques have revealed that eosinophils,^{20, 22} neutrophils,²⁵ and basophils²⁰ appear following allergen challenge; no correlations between degrees of local histamine release and neutrophil or eosinophil accumulation can be demonstrated. Eosinophil influx tends to precede that of the basophil.²⁰

Pathogenesis

A number of investigators have demonstrated that cutaneous LPRs can be mediated by IgE antibody. Dolovich and coworkers⁵ produced immediate phase responses and LPRs following skin testing with heterologous whole anti-human IgE or its F(ab')₂ fragment.⁵ Solley and coworkers⁵ extended these initial observations through a series of elegant passive transfer experiments that demonstrated that IgE antibody alone could produce LPRs. The ability of IgE antibody to induce LPRs, coupled with the fact that skin testing with the mast cell secretagogue compound 48/80 also produced biphasic reactions, suggested that mast cells play a central role in their development.

Mast cells are capable of releasing a variety of preformed or generated mediators that could potentially contribute to the development of tissue inflammation. Histamine, abundant in large quantities and preformed within the mast cell granule, can elicit immediate wheal and flare reactions but does not cause macroscopic LPRs.¹³ Further, histamine injected concomitantly with antigen will not induce LPRs in nonresponders despite increasing the size of the initial wheal and flare reaction.¹³ Histologically, histamine produces dilation of dermal vessels, the formation of endothelial pores, and perivascular edema, but no cellular reaction unless repeated challenges at the same site are performed. These results indicate that histamine alone is not responsible for the tissue inflammation observed in LPRs.

A number of preformed mediators capable of activating the coagulation, fibrinolytic, and bradykinin pathways have been described.³⁸ Kallikrein, when injected intradermally, produces macroscopic and histologic inflammation similar to the LPR observed with compound 48/80.³⁹ Bradykinin, which causes vasodilation and vasopermeability when injected intradermally, does not cause inflammation.⁶ Tryptase, a mast cell granule protease, is fibrinogenolytic and, along with heparin, may contribute to the prevention of clotting at sites of mast cell activation.⁴⁰ These results may help to explain why fibrin deposition in LPRs has been seen by some¹³ but not all investigators.⁶

Mediators generated as a result of activation of the arachidonic cascade, either by mast cells themselves or by infiltrating leukocytes, may contribute to LPRs. Leukotrienes possess a variety of potent biologic actions. After injection into human skin, LTD₄ and LTE₄ generate a burning, erythematous wheal-and-flare response persisting for up to 4 hours, with a characteristic central pallor thought to be caused by arteriolar constriction. Leukotriene C₄ causes an asymptomatic version of the same lesion. Histologically, the LTD₄-induced wheal and flare is accompanied by dermal edema and marked dilation of venules and capillaries.⁴¹ Leukotriene B₄ elicits a transient wheal and flare, followed in 3 to 4 hours by induration that is characterized by a dermal infiltrate composed predominantly of neutrophils. An interstitial array of fibrin deposition is also noted.⁴¹ Prostaglandin D₂, the major prostaglandin generated by mast cells but not basophils, elicits an asymptomatic wheal-and-flare reaction that is dissipated 2 hours following skin testing. Histologically, PGD₂ injections produce dermal edema and vessel dilation; neutrophil infiltration is evident from 2 to 6 hours following challenge.⁴¹

Platelet activating factor (PAF), a newly formed lipid generated by a variety of cell types, is a potent inflammatory mediator with a variety of effects that may contribute to LPRs.⁴² Intradermal injections of PAF produce a biphasic cutaneous response characterized by an immediate wheal-and-flare reaction and a delayed (3 to 6 hours) reaction characterized by erythema and hyperalgesia.⁴³ In addition to its ability to cause platelet aggregation *in vitro*, and neutrophil and monocyte aggregation *in vitro* and *in vivo*, injections of PAF can produce dermal leukocyte infiltrations.⁴⁴

Pulmonary LPRs

Patterns of Response

A variety of patterns of pulmonary allergic reactions can develop following appropriate challenge in sensitized individuals.⁴⁵ Clinical patterns of response can be characterized temporally, clinically (fever, chills, leukocytosis, rales vs. wheezing, pharmacologic modulation, etc.), and immunologically.

Following antigen challenge, isolated immediate, isolated late, and

dual reactions have been reported by a variety of investigators, with incidences ranging from 9% to 53%, 7% to 50%, and 18% to 84%, respectively.^{3, 14, 46-48} Isolated immediate reactions generally occur within minutes following antigen exposure or challenge and are characterized by airway obstruction; they generally resolve 30 to 60 minutes later. Isolated late reactions begin approximately 3 to 4 hours following challenge, airway obstruction reaches maximal intensity by 4 to 8 hours, and the response (airway obstruction) resolves in 12 to 24 hours.

Clinically, dual asthmatic reactions are characterized by a biphasic period of asthmatic symptoms consisting of wheezing, coughing, and shortness of breath. Despite a single antigen exposure or challenge 4 to 8 hours earlier, the intensity of the airway obstruction developed during the late response may be more prolonged and severe than that observed during the immediate reaction. Fever, chills, and leukocytosis, seen in hypersensitivity pneumonitis, are not clinical features of LPRs.¹⁴

A variety of antigens have been noted to cause late asthmatic reactions, including ragweed^{14, 49}; *B subtilis* enzyme preparations¹⁶; cat dander and pollen³; molds³ including *Alternaria*,⁴⁶ *Candida*, and *Aspergillus*⁵⁰; plicatic acid (red cedar asthma)⁴⁸; house dust mite (*D pteronyssinus*)⁵¹; sewer flies⁵²; cockroaches⁵³; wood dust⁵⁴; laboratory animals⁵⁵; soybean dust⁵⁶; platinum salts⁵⁷; piperazine hydrochloride⁵⁸; ethanolamines used in soldering flux,⁵⁹ paint,⁶⁰ and film development⁶¹; papain⁶²; grain dust⁶³; chromium and nickel salts^{64, 65}; diphenylmethane diisocyanate⁶⁶; and toluene diisocyanate.⁶⁵ In addition to antigen provocation, exercise challenge can also produce dual asthmatic reactions in some⁶⁷ but not all patients.⁶⁸ Recent preliminary reports suggest that late responses observed following exercise may actually reflect normal diurnal variability in pulmonary functions.⁶⁸ Finally, ultrasonically nebulized distilled water has been reported to produce immediate and late bronchial responses⁶⁹ (see Table 1).

The temporal and clinical features of dual bronchial responses were initially used to formulate hypotheses as to their immunopathogenesis. In his studies of patients with allergic bronchopulmonary aspergillosis, Pepys⁷⁰ concluded from his biopsy examination of dual cutaneous responses that the immediate response was IgE mediated and the late response was due to an Arthus or type III reaction. While true for the pulmonary response to *Aspergillus* challenge in these patients, the following observations indicate that late responses that develop following challenge with other antigens are primarily mediated by IgE-dependent mechanisms. First, both the immediate and late reactions can be blocked by premedication with the mast cell-stabilizing agent, cromolyn sodium,⁵¹ or antihistamines⁴⁷ before challenge. Second, many patients who develop dual responses to bronchial challenge have positive immediate skin test and IgE antibody responses, but no precipitating antibody in their serum.¹⁴ Third, patients with dual asthmatic responses tend to have the highest levels of IgE-mediated sensitivity.¹⁴ Fourth, immunotherapy, thought to raise levels of IgG-blocking antibody, has been found to protect against the development of the LPR.^{15, 71} Fifth, elevations in plasma⁷² (but not urinary⁷³) histamine levels

have been noted during the late phase of the dual response. Sixth, neutrophil chemotactic factor of anaphylaxis (NCF-A), a putative mast cell mediator, has been reported in serum samples obtained during both the immediate phase response and the LPR.⁷⁴ Seventh, complement levels are not decreased during the development of the LPR.⁷⁵ Finally, inhalation of anti-IgE antibody can induce biphasic responses.⁷⁶ Taken together, these observations suggest that immediate and dual responses are immunologically mediated primarily by IgE-dependent mechanisms.

Pulmonary Function Alterations

Sites of Airflow Obstruction.—Using methodology to localize the sites of involvement, a number of investigators have demonstrated that, following allergen or exercise challenge, large airways are the primary site of narrowing during the early response and small-airway narrowing predominates during the late reaction.⁷⁷

Airway Responsiveness.—A major clinical characteristic of asthma is airway hyperresponsiveness to a wide range of stimuli, including allergens, methacholine, histamine, and exercise. Cockcroft and his colleagues⁷⁸ were one of the first groups to report an interesting relationship between the experimental induction of dual responses and the development of increased airway responsiveness in patients allergic to ragweed. Airway responsiveness to histamine was measured both before and 24 hours following allergen challenge. Airway response to histamine did not change in patients following the development of an isolated immediate response. However, patients who developed dual responses demonstrated significant increases in histamine reactivity 24 hours following challenge. This increase in airway responsiveness was noted to persist in some patients for weeks following challenge. These observations indicated that the late response could potentially serve as a useful model for the study of the pathogenesis of asthma.

However, subsequent investigations have indicated that if measurements of airway responsiveness are made sooner following allergen challenge, increases in responsiveness can be demonstrated even in patients who develop isolated immediate responses only. Thorpe and coworkers⁷⁹ reported more than threefold changes in histamine responsiveness soon after the early response in seven subjects challenged with specific allergens, although the presence and severity of any subsequent late asthmatic responses was not documented. Durham and colleagues⁸⁰ noted increases in airway histamine responsiveness 3 hours following challenge with occupational agents. In subsequent studies, they have also characterized the changes in airway responsiveness that occur following the development of both the immediate and late response after allergen challenge.⁸⁰ Increases in airway responsiveness were noted following both the immediate and late response. However, increases in histamine reactivity for the definite late responders were significantly more than those observed for subjects who had equivocal or no late responses at both 3 and 24 hours following

challenge. Interestingly, the magnitude of the immediate response was similar in both groups as was the baseline airway responsiveness. Finally, a significant relationship was noted between the changes in airway responsiveness and the magnitude of the late asthmatic response. Taken together, these data indicate that, in comparison with isolated immediate reactions, the development of late asthmatic responses is associated with more significant and persistent changes in airway responsiveness.

Mediators

The inability to study the dynamics of LPRs using isolated lung tissue has necessitated alternative approaches for study. Investigators have used the following techniques: (1) measurement of mediators contained within serum during both the immediate and late response; (2) analysis of bronchoalveolar lavage (BAL) fluid for both mediators and cells; (3) analysis of the mediators released by pulmonary mast cells *in vitro* to determine (a) if they are released following *in vivo* antigen challenge and (b) if their known biologic effects could contribute to the pathogenesis of pulmonary late asthmatic responses.

As indicated in the previous discussion, analysis of cutaneous LPRs suggested that the mast cell played a critical role in the immunopathogenesis of these responses. Thus, investigations of late asthmatic responses focused initially on the detection of mediators within serum that were mast cell derived and whose presence paralleled observed changes in airway mechanics following antigen challenge. In humans, the majority of histamine is localized within mast cells and circulating basophils. Elevations in plasma histamine during the early response and the late response have been found by some,⁷² but not all,⁷³ investigators. Further, levels of urinary N-methylhistamine, a unique histamine metabolite, have only been detected in samples obtained during the early response following allergen challenge.⁷³

Neutrophil chemotactic activity has been detected in serum samples obtained during both the early and late response following both allergen⁷⁴ and exercise⁸¹ challenge. The observed activity is thought to be due to a large molecule, NCF-A, which has an estimated molecular weight of 600,000 daltons. The physicochemical characteristics of this activity are similar following either exercise or allergen challenge or during either the early or late response. The appearance of the NCF-A activity parallels the rise and fall of the pulmonary function parameters seen during the development of the dual response.⁷⁴ Further, leukocyte activation, assessed by measuring neutrophil and monocyte complement rosettes, is also noted to parallel the levels of NCF-A activity.⁸² While the precise cell source of NCF-A is unknown, the ability to detect its presence following a number of known triggers of immediate hypersensitivity reactions, including challenge of sensitized lung tissue, indicates that it may be mast cell derived.

In addition to the analysis of serum, the potential participation of mast cell activation in the pathogenesis of late asthmatic responses has been

evaluated indirectly through the study of pulmonary mast cell mediator release *in vitro*, by analyzing bronchial alveolar lavage fluid following allergen challenge for the presence of mast cell mediators, and by the evaluation of what effects these mediators may have on airway function following inhalation or local instillation. Mediators formed as a direct consequence of membrane activation and the formation of arachidonic acid have received considerable attention recently. Arachidonic acid is the pivotal point for two important pathways: the cyclooxygenase and the lipoxygenase pathway. Following mast cell activation *in vitro*, the predominant cyclooxygenase pathway product formed is PGD₂. Interestingly, increased levels of PGD₂ can be found in BAL fluid following local antigen challenge.⁸³ Further, inhalation of PGD₂ can produce bronchoconstriction.⁸⁴ Thus, since pulmonary late responses are associated with airway obstruction and inflammation, it is possible that PGD₂ plays a role in the pathogenesis of these responses. Although PGD₂ is the major cyclooxygenase pathway product formed by activated mast cells, plasma levels of thromboxane B₂ have been noted to be elevated during both the immediate and late asthmatic responses. These results indicate that additional cells, recruited to sites of mast cell degranulation by various chemotactic factors, may also participate in the pathogenesis of these reactions. Anti-IgE stimulation of mast cells can also result in the formation of LTC₄ and LTD₄. Both LTC₄ and LTD₄ can induce bronchospasm,⁸⁵ enhance vascular permeability, and promote mucus secretion.⁸⁶

Platelet activating factor can be produced by a number of cells that may infiltrate sites of mast cell degranulation. Interestingly, although human pulmonary mast cells generate PAF, they do not release it.²⁶ The biologic effects of PAF are numerous and include platelet activation, neutrophil stimulation, vasoactivity, smooth-muscle contraction, and cardiovascular effects.²⁶ When inhaled, PAF can produce airway obstruction and enhance bronchial hyperresponsiveness in humans⁸⁷ and a biphasic airway obstructive response in a rabbit model of the late asthmatic response.⁸⁸ These results, coupled with the observations that a number of the biologic effects of PAF can be blocked by drugs that also affect the late response (cromoglycate, glucocorticoids, ketotifen, and theophylline), indicate that this mediator may contribute to the pathogenesis of LPRs.

BAL Studies

Bronchoalveolar lavage has been recently utilized in an attempt to characterize the airway inflammatory response following allergen challenge.⁸⁹⁻⁹¹ A variety of leukocytes, including eosinophils, neutrophils, T lymphocytes, and alveolar macrophages, have been noted in BAL fluid obtained during the late response. These studies indicate that airway inflammation occurs following allergen challenge and that certain features of this inflammatory reaction, such as eosinophil infiltration⁸⁹ and a diminution in numbers of T suppressor cells,⁹² may be characteristic of late phase responders.

Pathogenesis

From the previous discussion regarding the evidence implicating mediator release and inflammatory cell infiltration, it should be apparent that the pathogenesis of the late asthmatic reaction and the subsequent development of increased airway responsiveness no doubt involve multiple factors. Figure 2 depicts some of these potential relationships. Pulmonary mast cells appear to initiate both an immediate bronchospastic response and a late inflammatory response that can result in biphasic alterations in pulmonary function and airway hyperresponsiveness. Mast cell activation can result from antigen interaction with IgE antibody, exercise, or non-IgE-mediated release resulting from a variety of stimuli such as complement fragments or histamine releasing factors produced by a number of different cell types. Mast cell-derived mediators, such as histamine and LTC_4 and LTD_4 , can produce bronchoconstriction that seems to be the major feature of the immediate response. Chemotactic factors, such as NCF-A, eosinophil chemotactic factor of anaphylaxis, monohydroxyeicosatetraenoic acids, and LTB_4 , can promote the ingress of both eosinophils and neutrophils. Eosinophils can generate both LTC_4 and LTD_4 and release both PAF and major basic protein. Major basic protein can damage airway epithelium and induce mast cell mediator release.

The importance of the airway epithelium in a number of airway functions has only been recently recognized.⁹² The epithelium appears to make a number of relaxing factors that promote bronchial smooth-muscle relaxation. Once destroyed, the epithelium can no longer produce these factors, resulting in bronchial smooth muscle contraction and further airway obstruction. In addition, sloughing of the epithelium also exposes irritant receptors to the airway lumen, which in turn enhances airway responsiveness.

Neutrophils also appear to contribute to the late response. In rabbits, neutrophil depletion ablates the late response.⁹³ In dogs, inhalation of ozone results in enhanced airway responsiveness, which is associated with the influx of neutrophils into the airway.⁹⁴ Pretreatment of these dogs with indomethacin blocks the induction of airway hyperresponsiveness but has no effect on the neutrophil migration. These results, coupled with the additional observation that thromboxane antagonists also block the development of this response, suggest that neutrophils produce thromboxanes, which are influential in promoting airway hyperresponsiveness.⁹⁵ Neutrophil and eosinophil generation of PAF may further contribute to the airway hyperresponsiveness that accompanies the development of the LPR. Both neutrophils and alveolar macrophages, once activated, can generate respiratory burst metabolites, which can further damage airway epithelium and promote airway edema and inflammation. Recently, interest has centered around the potential role of the alveolar macrophage in IgE-dependent processes. Alveolar macrophages possess a low-affinity IgE receptor that, when stimulated, can result in the release of toxic oxygen species and lysosomal enzymes.⁹⁶

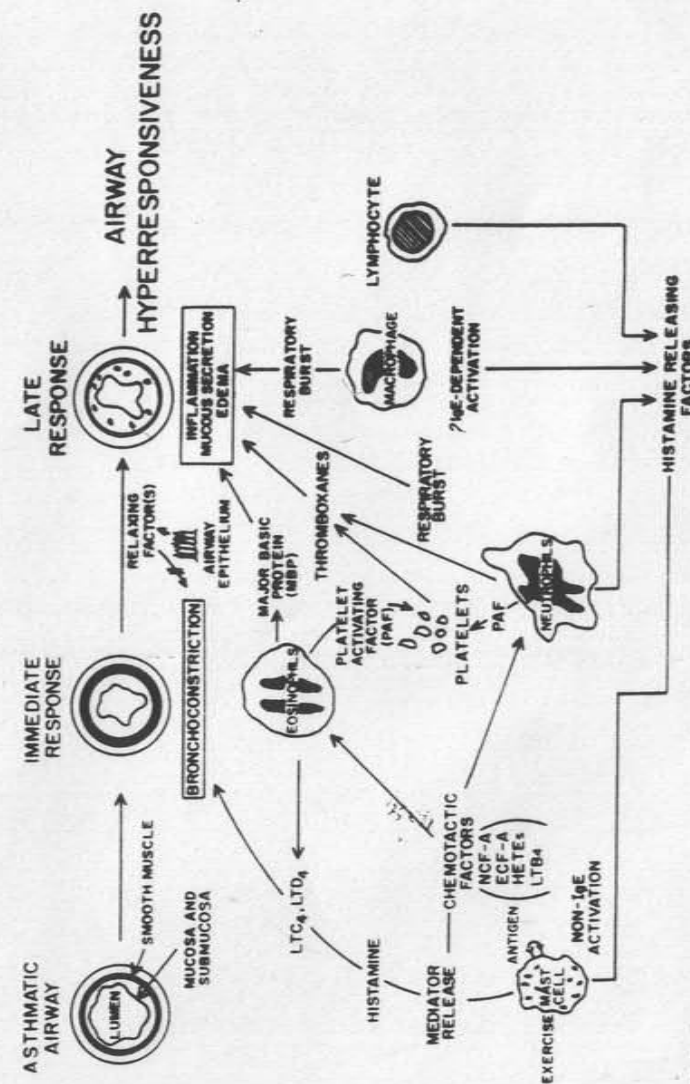


FIG 2. Potential involvement of various mediators and cells in pulmonary late phase reactions. LTC_4 , LTD_4 , and LTB_4 = leukotrienes C_4 , D_4 , and B_4 ; NCF-A = neutrophil chemotactic factor of anaphylaxis; ECF-A = eosinophil chemotactic factor of anaphylaxis; HETE's = monohydroxyeicosatetraenoic acids.

Finally, a variety of cell types including neutrophils, alveolar macrophages, and lymphocytes have all been demonstrated to produce histamine-releasing factors, which may feed back to resident mast cells to amplify or perpetuate mast cell secretion.⁹⁷ In summary, as noted in Figure 2, with the complexity of the interplay among the various biologic substrates and cells found within the lung, it appears unlikely that any one cell type or mediator is alone responsible for the pathogenesis of the late asthmatic response.

Nasal LPRs

Symptoms and Incidence

Allergen nasal provocation in sensitive patients results in the appearance of symptoms within minutes. The classic symptoms provoked include sneezing, rhinorrhea, and pruritus associated with the development of nasal congestion due to edema formation. Isolated immediate, isolated late, and dual responses have been described using symptom evaluation, measurements of nasal airway resistance, and quantitation of mediators and cells using nasal lavage.^{98,99} The clinical symptoms associated with LPRs differ from those appreciated with immediate responses. The majority of patients experiencing LPRs complain of nasal congestion as the predominant symptom, with a minority of subjects complaining of rhinorrhea and sneezing.^{100,101} Pruritus is usually not part of the constellation of symptoms. The reported incidence of nasal LPRs (30% to 50%) varies depending on the methods of analysis.^{100,101}

Pathogenesis

Insight into the pathogenesis of nasal LPRs has come primarily from evaluations of nasal lavage fluid at various times following allergen challenge. Mediators that have been described include histamine^{102,103}, PGD₂¹⁰²⁻¹⁰⁴, tosyl-L-arquinine (TAME)-esterase¹⁰², kinins¹⁰⁵, LTB₄, LTC₄, LTD₄, and LTE₄^{106,107}, and tryptase.¹⁰⁸ Each of these inflammatory mediators is measurable in nasal washings shortly after allergen challenge. Interestingly, although antihistamines are widely used in the treatment of allergic rhinitis, levels of PGD₂¹⁰⁴ and tryptase¹⁰⁸ in lavage fluid correlate better with the development of clinical symptoms than do histamine levels.

Naclerio and coworkers¹⁰² have sequentially followed allergen challenges with nasal washings through the appearance of clinical late responses. Nasal washings obtained during the LPR contain increased amounts of histamine and TAME-esterase, much as seen during the early response. However, PGD₂ levels are not increased in nasal washings obtained during the LPR. As PGD₂ is the major cyclooxygenase product of mast cells but is not produced in measurable amounts by basophils,¹⁰⁹ these data have been interpreted to suggest that basophils might participate in LPRs by appearing in the nasal mucosa and releasing their mediators. In this regard, superficial scrapings of nasal mucosa are devoid of ba-

sophils before antigen challenge but contain basophils after a positive challenge.¹¹⁰ Therefore, it is possible that basophils are attracted during nasal LPRs and that once they migrate into the nasal mucosa, they release histamine and other mediators.

In addition to allergen challenge, cold dry air challenge also induces a late reaction.⁹⁹ These findings indicate mast cell and/or basophil mediator release may also play a role in nonallergic rhinitis.

As seen in both the skin and the lung, nasal allergen challenge results in tissue inflammation. Increased numbers of lavage eosinophils, neutrophils, and mononuclear cells have been described,¹¹⁰⁻¹¹³ as well as evidence of mast cell involvement.¹¹⁴ Alcian blue-positive cells, in which more than 65% are basophils, can also be seen temporally associated with the clinical late response.¹¹⁰ Treatment with topical corticosteroids prevents the immediate and late response as well as the influx of inflammatory cells.¹¹⁰ Interestingly, oral corticosteroids have no effect on the immediate reaction (presumably owing to their inability to block mast cell mediator release) but block the development of the late response and the influx of eosinophils but not of neutrophils or mononuclear cells.¹¹² These models of nasal allergen challenge appear to be relevant clinically as both inflammatory cells¹¹³ and mediators^{115,116} have been noted to be present during natural allergen exposure.

Systemic LPRs

Late systemic reactions may occur following a number of different stimuli (see Table 1).¹¹⁷⁻¹¹⁹ Stark and Sullivan¹¹⁹ performed a prospective study of anaphylaxis in 25 consecutive patients. Three distinct clinical patterns were observed: uniphasic, biphasic, and protracted anaphylaxis. Thirteen (52%) experienced a single episode, while 5 patients (20%) developed biphasic reactions consisting of two episodes of hypotension or laryngeal edema separated by asymptomatic intervals of 1 to 8 hours. Seven (28%) suffered hypotension, lower respiratory tract obstruction, or laryngeal obstruction that persisted 5 to 32 hours despite vigorous therapy that included systemic glucocorticoids. Recurrent or prolonged reactions were 2.8-fold more likely if the onset was 30 or more minutes after exposure to the stimulus or if the offending agent had been administered by mouth.

Thus, patterns of systemic anaphylactic responses have temporal similarities to those seen in the lung following inhalation challenge. It is unclear whether or not immunologic features of these events are also similar. It is tempting to speculate that anaphylaxis might be followed by systemic LPRs. Importantly, unlike pulmonary LPRs, which can be attenuated by prior administration of corticosteroids, both biphasic^{117,119} and protracted¹¹⁹ reactions cannot be prevented despite the use of aggressive corticosteroid therapy. Although corticosteroids or other forms of therapy used for anaphylaxis (epinephrine and antihistamines) do not prevent the late systemic responses from occurring, it is unclear if they are capable of modulating these responses in any way. These studies have obvious important implications regarding the wisdom of outpatient management of anaphylaxis.

TABLE 2.
Effect of Various Treatment Regimens on LPRs in Different Tissues*

Target Organ	Antihistamines				Cromolyn	Nedocromil	Ketotifen	Steroids	NSAIDs	Sympatho-mimetics	Xanthine Derivatives	Immunotherapy
	H ₁	H ₂	H ₁ + H ₂									
Skin	+	NE	+	+	±	?	+	++	±	NE	NE	+
Lung	+	?	?	?	++	+	+	++	±	NE	±	+
Nose	?	?	?	?	+	?	?	+	?	?	?	?
Systemic	NE	NE	NE	NE	?	?	?	NE	?	?	?	?

* + = mild effect; ++ = significant effect; ± = variable effect; NE = no effect; ? = not established.

Pharmacologic Modulation of Dual Reactions

The effects of various medications on the development of both immediate phase responses and LPRs following allergen challenge in various target organs have been evaluated by a number of investigators and are summarized in Table 2. Such data have been invaluable in providing insight into potential pathophysiologic mechanisms, in treating patients with various allergic diseases and asthma, and in promoting research in the development of new pharmaceuticals that would be useful in treating these types of patients.

Conclusion

The LPR has provided important and relevant clinical information in a number of ways. First, it has emphasized that hypersensitivity reactions must be viewed in terms of hours, and perhaps days, rather than minutes. Thus, treatment of allergen exposure in sensitized patients should reflect this temporal relationship. Second, investigations into the pathophysiology of late reactions in the skin, lung, and nose have indicated that tissue inflammation is a major feature of allergic reactions. Thus, treatment of such diseases as atopic dermatitis, asthma, and allergic rhinitis should be focused on this inflammatory component. Third, analysis of the late response has provided insight into the interrelationships among cell-derived mediators, inflammatory cells, and such clinical features as airway hyperresponsiveness in asthma and the priming effect in allergic rhinitis.

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