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Review article

Leukotriene antagonists and synthesis inhibitors: New directions in asthma therapy

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This article briefly reviews the advances in our understanding of asthma with a particular focus on the role of inflammation, before providing a concise overview of current knowledge of leukotrienes in the pathophysiology of the disease. The development of leukotriene receptor antagonists and synthesis inhibitors is briefly described; and acute exercise, allergen, and aspirin challenge studies with these agents are reviewed. Clinical studies with leukotriene antagonists and inhibitors have confirmed the central role of leukotrienes in asthma pathophysiology. In conclusion, we suggest that the new generation of leukotriene receptor antagonists may be suitable as first-line therapy in patients with mild to moderate asthma. Further studies are required to determine whether the leukotriene synthesis inhibitors will be equally effective or provide any additional antiinflammatory benefit. (J Allergy Clin Immunol 1996;98:1-13.)

Key words: Asthma, leukotrienes, leukotriene D_4 antagonists, 5-lipoxygenase inhibitors, 5-lipoxygenase-activating protein inhibitors, leukotriene B_4 antagonists

Although asthma was once regarded largely as a disease of airway smooth muscle, leading to treatment with oral and inhaled bronchodilators, inves-

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Abbreviations used

BAL:	Bronchoalveolar lavage
FLAP:	5-Lipoxygenase-activating protein
LT:	Leukotriene
PAF:	Platelet activating factor

tigations over the last decade have shown asthma to be a chronic inflammatory disease complicated by periodic acute inflammatory changes.¹ Indeed, it is now clear that even patients with mild asymptomatic asthma have obvious inflammatory changes in their airways, characterized by infiltration of the mucosa and epithelium with activated T cells, mast cells, and eosinophils² (Fig. 1). The

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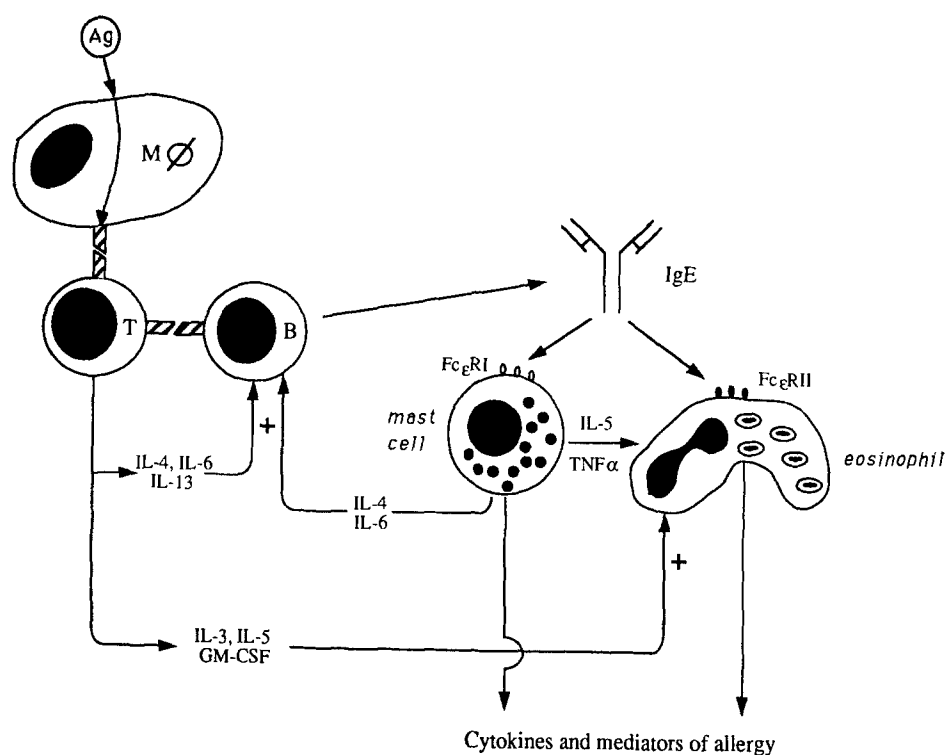


FIG. 1. Schematic diagram of cytokine pathways involved in regulation of the allergic inflammatory response.

recognition that T cells and mast cells secrete an array of cytokines in asthma, the functions of which are to direct eosinophil growth and maturation and to promote the isotype switching of B lymphocytes from IgM to IgE, provides a rational explanation for the inflammatory response of asthma. The consequences of this include increased microvascular permeability, stimulation of local and central neural reflexes, epithelial disruption and stimulation of mucus-secreting glands, smooth muscle hypertrophy, and airway wall remodeling.

Because inflammation plays such a central role in the airway dysfunction of asthma, it is not surprising that national and international guidelines for asthma management have selected this aspect of the disease for specific therapeutic targeting. Corticosteroids are the most effective anti-inflammatory agents currently available for the treatment of asthma, and in many countries of the world, inhaled corticosteroids have become the first-line treatment, with inhaled β_2 -agonists being used on an as-needed basis.³ In line with recent national and international guidelines for asthma

management, earlier and more widespread use of inhaled corticosteroids is recommended.

However, despite the well-recognized efficacy and safety of inhaled corticosteroids, many patients will still have poorly controlled asthma and a poor quality of life.⁴ In addition, there are rising trends in asthma prevalence, morbidity, severity, and mortality rates⁵; and there is also concern about the long-term safety of inhaled corticosteroids, particularly in high doses and at the extremes of age. Numerous studies have pointed to poor patient compliance with therapy, especially for inhaled preparations with which many patients also have a poor technique.⁶ These problems have prompted the search for alternative, well-tolerated, and effective pharmacologic agents that target airway inflammatory events.

THE LEUKOTRIENES

In recent years, an increased understanding of the pathophysiology of asthma has revealed a complex network of interactions among a variety of inflammatory cells and the mediators that they

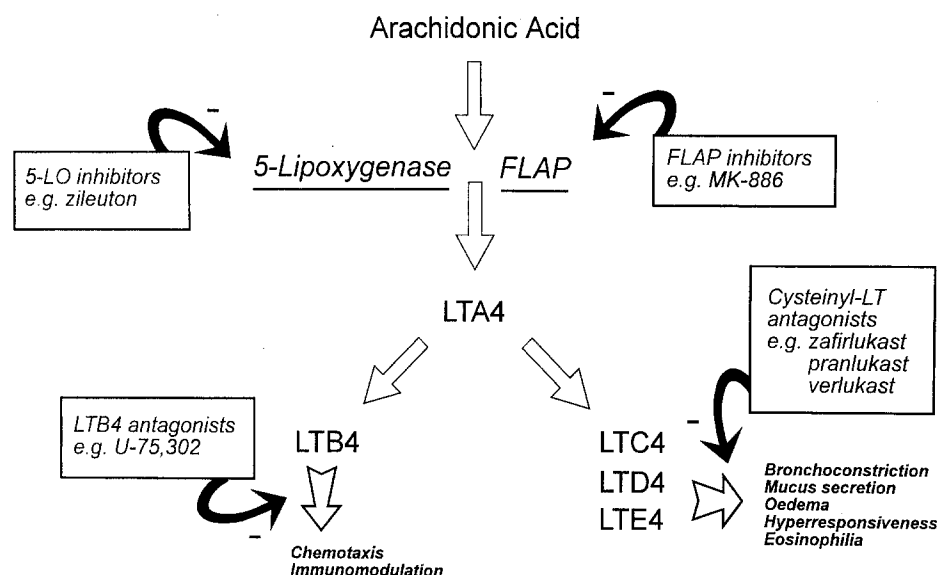


FIG. 2. The four groups of antiasthma and antiinflammatory drugs directed against LT synthesis and activity.

secrete.⁷ The conventional view has been that targeting a single group of these mediators for pharmacologic blockade would have only a limited impact on the disease. This view has had to be revised in the light of the unexpectedly high efficacy in clinical trials of drugs that block the synthesis or activity of the leukotriene (LT) family of mediators.

LTs are metabolites of the fatty acid arachidonate⁸ and are produced by the activity of 5-lipoxygenase (Fig. 2). Arachidonate is presented to the 5-lipoxygenase enzyme by 5-lipoxygenase-activating protein (FLAP),⁹ a cofactor resident in the nuclear membrane, and the interaction leads to the formation of the unstable intermediate LTA₄. Depending on the cell type, further conversion of LTA₄ may occur to the chemotaxin LTB₄, performed by LTA₄ hydrolase, or to the cysteine-containing LTC₄ by the action of the transmembrane enzyme LTC₄ synthase. Further conversion of LTC₄ to LTD₄ and then to LTE₄ occurs by the actions of enzymes ubiquitous in the tissues and circulation.

Four classes of drugs are currently under development as antiasthma or antiinflammatory therapy, which interfere with LT synthesis or activity (Fig. 2 and Table I). First, the 5-lipoxygenase inhibitors directly block the catalytic activity of 5-lipoxygenase itself. Second, the FLAP inhibitors displace arachidonate from its binding site on the FLAP molecule and prevent it from providing arachidonate to 5-lipoxygenase as a substrate for

LT synthesis. Both these classes of drugs block the synthesis of the cysteinyl LTs and of LTB₄. Third, the activity of cysteinyl LTs (LTC₄, LTD₄, and LTE₄) at the LTD₄ receptor can be blocked by competitive cysteinyl LT receptor antagonists, and fourth, blockade of LTB₄ activity may be achieved by competitive antagonists at the LTB₄ receptor.

The cysteinyl LTs have been particularly implicated as important mediators in the pathophysiology of asthma and have become the leading targets for potential new asthma therapies.¹⁰ Several potent competitive cysteinyl LT receptor antagonists have been developed, including zafirlukast (ICI 204,219; Accolate) and pranlukast (ONO-1078). These orally active antagonists are probably the furthest advanced of all the "anti-LT" drugs currently being developed and represent a promising new direction in asthma therapy. The development of LT synthesis inhibitors, such as the 5-lipoxygenase inhibitor zileuton (A-64077) and the FLAP inhibitor MK-0591, is also well advanced. It is too early to say whether these will confer additional benefits by virtue of their blocking LTB₄ synthesis, as well as that of the cysteinyl LTs. Evidence for LTB₄ involvement in asthma is inconclusive, but the issue may be resolved by the further development of experimental LTB₄ receptor antagonists.

This review briefly outlines the evidence for LT involvement in asthma, before describing the insights into their role, which have arisen from clinical trials of each of the four classes of anti-LT drugs.

TABLE I. The principal anti-LT compounds in clinical trials since 1989, with their generic and trade names

Generic name	Compound	Trade name	References
LTD ₄ antagonists			
Zafirlukast	ICI-204,219	Accolate	14, 36, 41-44, 46, 47, 51-55, 59, 61, 63-65
Pobilukast	SK&F 104353-Q		14, 33, 60
Pranlukast	ONO-1078		37, 38, 49, 66
Tomelukast	LY171883		32
Verlukast	MK-679		56
	MK-0476		40, 45, 62
	MK-571		14, 34, 35, 50, 57, 58
	RG-12525		39
5-Lipoxygenase inhibitors			
Zileuton	A-64077	Leutrol	75-82
	ABT-761		83
	Z-D2138		84
FLAP inhibitors			
	MK-886		85, 86
	MK-0591		87, 88
	BAY x1005		89

PROPERTIES OF LTs RELEVANT TO THE PATHOPHYSIOLOGY OF ASTHMA

Cysteinyl LTs

Originally described by their biologic activity as "slow-reacting substance of anaphylaxis" (SRS-A), the cysteinyl LTs can be produced by a variety of inflammatory cells in the airways, including eosinophils, mast cells, macrophages, monocytes, and basophils.¹⁰ Although no single mediator is responsible for all the clinical features and pathologic changes of asthma, cysteinyl LTs exert many effects that mimic the clinical expression of the disease, including bronchoconstriction, increased airway responsiveness, increased microvascular permeability, and hypersecretion of mucus. These activities are mediated through an interaction between the agonist and specific cell-surface receptors, with LTD₄ being the most active of the three components and the partial agonist LTE₄ having the most long-lived activity.¹¹

Among the most important pharmacologic actions of the cysteinyl LTs are their effects on the contraction of airway smooth muscle. These mediators are among the most powerful bronchoconstrictor agents, being approximately 100 to 1000 times more potent than histamine on a molar basis. In addition, bronchoconstriction produced by cysteinyl LTs is more prolonged than that produced by histamine both in vitro and in vivo. When

administered by inhalation to either healthy volunteers or patients with asthma, all three cysteinyl LTs cause bronchoconstriction, with LTD₄ being the most potent. In addition, asthmatic airways are 14, 15, 6, 9, and 219 times more responsive to histamine, methacholine, LTC₄, LTD₄, and LTE₄, respectively, than are the airways of normal subjects.¹² However, in both normal and asthmatic subjects, there is wide interindividual variability in the sensitivity of the airways to the contractile effects of LTs.¹⁰

Asthma is characterized by enhanced responsiveness of the airways to a variety of stimuli, whether pharmacologic or physical. When inhaled by patients with asthma, all three cysteinyl LTs produce an approximately threefold increase in airway responsiveness to histamine, which persists for several days after provocation.¹² Among the three cysteinyl LTs, LTE₄ seems to be particularly active in this regard. Thus if cysteinyl LTs are involved in active asthma, then antagonism of their pharmacologic effects or blockade of their synthesis might be expected to reduce bronchial hyperresponsiveness.

The formation of mucus plugs by an interaction of mucus with plasma protein and cell debris is a major feature of asthma, particularly in severe forms of the disease. Cysteinyl LTs are particularly potent stimulators of mucus secretion from human

airways in vitro and probably act both at the level of the goblet cell and the submucous gland.¹³ In addition, these mediators have the powerful effect of increasing microvascular permeability with exudation of plasma into the airway wall and lumen, leading to tissue edema, which again may contribute to reduced airway caliber and increased bronchial hyperresponsiveness.¹³

The relative contribution of smooth muscle contraction to airflow obstruction, compared with changes in vascular permeability with edema of the airway wall and increased mucus production, remains unknown. However, the fact that cysteinyl LTs can produce all of these effects supports the view that they are major candidate targets for pharmacologic intervention in asthma. This mediator class may also exhibit complex interactions with other components of the inflammatory process. For example, they may stimulate the synthesis and release of other inflammatory mediators¹⁴ or may themselves be the final common pathway for the inflammatory activity of other mediators, such as platelet activating factor (PAF).

Recently, it has been recognized that LTD₄ is an extraordinarily potent and specific chemoattractant for human eosinophils in vitro.¹⁵ This activity may underlie the recruitment of eosinophils into the airway mucosa observed after LTE₄ inhalation in human beings.¹⁶ Long-term production of cysteinyl LTs may therefore have a hand in directly producing all three cardinal characteristics of the asthmatic lung, namely airflow obstruction, bronchial hyperresponsiveness, and eosinophilia.

LTB₄

In contrast, the possible role of LTB₄ in asthma is unclear. In the human lung, it is a potent chemoattractant for neutrophils,¹⁷ which are implicated in sudden-onset fatal asthma¹⁸ and in nocturnal asthma¹⁹; and in vitro LTB₄ is also a chemoattractant for eosinophils, particularly if they have been primed by a specific factor such as IL-5.²⁰ LTB₄ induces T lymphocyte production of IL-5,²¹ which is implicated in eosinophilia in asthma and atopy, and promotes the effects of IL-4 on IgE production by B lymphocytes.²² LTB₄ has been detected in the bronchoalveolar lavage (BAL) fluid of patients with asthma,²³ and altered production of LTB₄ has been described in the BAL cells and blood leukocytes of asthmatic subjects.²⁴⁻²⁶

RELEASE OF LTs IN ASTHMA

Early studies clearly established that IgE-dependent stimulation of sensitized lung tissue from a

variety of animals generated slow-reacting substance of anaphylaxis, an observation that was later confirmed on isolated human lung fragments. More recent studies have shown that allergen- or anti-IgE-induced contractions of lung isolated from normal subjects or patients with asthma are entirely mediated by cysteinyl LTs together with a minor histamine-dependent component.²⁷

The potency of LTs means that only small amounts are generated in vivo, and this makes their assay in biologic fluids difficult. Nevertheless, cysteinyl LTs have been found by chromatography and mass spectrometry in the lavage fluid of patients with asthma.²³ In addition, increased levels of cysteinyl LTs may be recovered from lavage fluid or in the form of LTE₄ in urine after exposure to inhaled allergen²⁸ or aspirin,²⁹ during an acute spontaneous attack of asthma,³⁰ and for some weeks after clinical resolution of such an attack.³¹ These observations, both in experimental and naturally occurring asthma, together with their known pharmacologic effects, make the cysteinyl LTs a major target for new classes of drugs active in asthma.

CYSTEINYL LT ANTAGONISTS

Cysteinyl LT antagonists block the actions of these mediators at the receptor level. Early antagonists, including FPL 55712, L-649,923, REV 5901, and tomelukast (LY171883), although having variable inhibitory effects in vitro against LTD₄ contraction, demonstrated poor efficacy and poor tolerability when administered to human beings. These compounds had only small effects on the early asthmatic response to inhaled allergen in sensitized asthmatic subjects and failed to influence the late asthmatic response or the acquired increase in airway responsiveness. Studies with these drugs in chronic asthma were also disappointing. For example, in a large scale study in which asthmatic patients were treated with tomelukast for 6 weeks, only a small improvement in FEV₁ (7%) and minor reductions in as-needed β_2 -agonist use could be documented.³² It is now generally thought that these somewhat disappointing results were due to a combination of lack of potency and specificity.

The clear demonstration that antagonists could be developed against the cysteinyl LT receptor nevertheless spurred on further research and led to the development of a new generation of drugs. These included pobilukast (SK&F 104 353-Q)³³ and MK-571.^{34, 35} MK-571 is no longer being developed on account of side effects and has been

succeeded by verlukast (MK-679). Zafirlukast is a potent, selective, competitive, reversible cysteinyl LT receptor antagonist.³⁶ It is active both orally and when administered by inhalation and is among the most potent oral cysteinyl LT antagonists currently under clinical development. Another orally active compound, pranlukast, became the first cysteinyl LT antagonist to be used in clinical practice when it was marketed in Japan in mid 1995.^{37, 38} Other LT antagonists at various stages of clinical development include RG-12525,³⁹ Ro-245913, CGP 45715A, BAYx 7195, and MK-0476.⁴⁰

Zafirlukast, MK-571, and pobilukast were at least 200 times more potent than the early LTD₄ antagonists when assessed by tritiated-LTD₄ binding assays¹⁴ and caused a shift of up to 100-fold in the bronchoconstrictor dose-response curve when administered before LTD₄ inhalation.⁴¹ Early studies have shown that these compounds are effective and well tolerated when given orally, intravenously, or by aerosol.¹⁴ Efficacy of each one has been clearly demonstrated by prevention or reduction of LTD₄-induced bronchoconstriction and attenuation of the early and late phase response after allergen exercise challenge.

EFFECT OF CYSTEINYL LT ANTAGONISTS ON AGONIST-INDUCED BRONCHOCONSTRICTION

When administered orally to healthy volunteers, zafirlukast is very active in inhibiting bronchoconstriction provoked by inhaled LTD₄.⁴¹ Thus a single oral dose of 40 mg produced a 100-fold shift of the LTD₄-induced bronchoconstrictor dose-response curve 2 hours after dosing, with significant antagonism (fivefold) being maintained for up to 24 hours. MK-0476 also produces a 100-fold shift in the curve.⁴⁰ For comparison, tomelukast and pobilukast produced only three- to fourfold shifts of the LTD₄ dose-response curve, whereas MK-571 displaced the dose-response curve 30 to 40-fold.¹⁰ In patients with mild asthma, oral administration of zafirlukast produced a dose-related inhibition of LTD₄-induced bronchoconstriction, as would be expected by a competitive receptor antagonist.⁴² These compounds seem to be equally active at antagonizing the cysteinyl LT receptor in normal and asthmatic airways, with the drug given orally being on the whole more effective than when administered by inhalation.

EFFECT OF CYSTEINYL LT ANTAGONISTS IN ACUTE CHALLENGE STUDIES

Cysteinyl LT antagonists reduce the bronchoconstriction induced by several natural triggers of

asthma including exercise, cold air, allergen, and aspirin.

Exercise

Bronchoconstriction in asthma provoked by exercise is, to a large extent, mediated through the release of bronchoconstrictor substances from inflammatory cells in the airway wall. LTs seem to play a particularly important role in this response. A single 20 mg oral dose of zafirlukast produces marked protection against exercise-induced bronchoconstriction in the majority of patients. Thus in the study by Finnerty et al.,⁴³ the mean maximum percent fall in FEV₁ after exercise was 36% in patients treated with placebo and 21.6% in patients treated with the active drug, with a maximum effect being observed 5 to 30 minutes after stimulation. When 400 µg of the drug was administered by inhalation, it also produced a similar degree of protection against bronchoconstriction, although this varied among individuals.⁴⁴ When compared with placebo, intravenously administered MK-571 also reduced exercise-induced bronchoconstriction, decreasing the maximum fall in FEV₁ by 68% and resulting in a reduced time to recovery³⁴; and once-daily oral dosing with MK-0476 for only 2 days produced a similar inhibition of fall in FEV₁ and reduced the recovery time after exercise from 62 to 7 minutes.⁴⁵ Pobilukast was at least as effective as sodium cromoglycate in protecting against exercise-induced bronchoconstriction.³³

Another marker of airway hyperresponsiveness related to exercise-induced asthma is bronchoconstriction provoked by cold air. In the only report to date on the effect of cysteinyl LT antagonists on cold air-induced bronchoconstriction in asthma, Glass et al.⁴⁶ showed that inhaled zafirlukast attenuated bronchoconstriction in patients with mild to moderate disease.

PAF

Another stimulus that is shown to be LT-dependent is the bronchoconstriction produced by inhalation of PAF. Single doses of inhaled PAF increase the excretion of the cysteinyl LT metabolite LTE₄ in the urine. Thus a single dose of 40 mg of zafirlukast inhibited PAF-induced bronchoconstriction in normal volunteers by 59% when compared with placebo.⁴⁷

Allergen challenge

Provocation of sensitized asthmatic airways results in a mast cell-dependent early asthmatic response, which peaks at 15 minutes and recovers over the next hour. In about 50% of patients this is

followed by a late asthmatic response 6 to 8 hours after challenge, which may last up to 24 hours, and is accompanied by an acquired increase in bronchial responsiveness to agents such as histamine and methacholine. The late reaction is accompanied by swelling of the airway wall and infiltration of the airway with inflammatory leukocytes, predominantly neutrophils, eosinophils, and T cells.⁴⁸

The first-generation cysteinyl LT antagonists L-649,923 and tomelukast had only small inhibitory actions on the early and late phase responses to inhaled allergen, whereas pranlukast significantly inhibited the early response to allergen after 1 week of treatment.⁴⁹ The greatest effect on the early response was between 20 and 60 minutes after challenge, which is consistent with de novo generation of the cysteinyl LTs, whereas the rapid release of stored histamine from mast cell granules probably accounts for most of the contraction during the first 20 minutes.

MK-571, when administered intravenously, blocked both early and late responses, achieving 88% and 63% inhibition respectively,⁵⁰ and an oral dose of 40 mg of zafirlukast, administered 2 hours before allergen challenge reduced the early and late responses by 81% and 55%, respectively, when measured as the maximum decrease in FEV₁.⁵¹ The close concordance between these results and those of other studies demonstrates that cysteinyl LTs are responsible for the major proportion of the airflow obstruction observed in the early response and that at least half of the late response is also cysteinyl LT-dependent. In addition, the subsequent allergen-induced increase in responsiveness to inhaled histamine was also attenuated by zafirlukast.⁵²

Zafirlukast, when administered by inhalation in a dose of 200 µg, blocked the early phase of the allergen-induced airway response⁵³ but had a lesser effect on the late response. A second study showed that a larger 1600 µg inhaled dose of zafirlukast also inhibited the early reaction but again failed to influence the late response.⁵⁴ These results suggest that cysteinyl LTs released in the lungs during the early response may have a systemic role, possibly promoting leukocyte chemotaxis during the late response, which in turn is associated with a further bout of airway obstruction. Evidence for this view comes from a recent study in which oral zafirlukast was given to 11 patients with mild to moderate asthma, who had a segmental allergen challenge performed after 5 days of treatment, and BAL after an additional 2 days.⁵⁵ Compared with placebo, zafirlukast produced a dramatic reduction in basophil count in

BAL fluid, accompanied by significant reductions in lymphocyte count and histamine level, a trend toward reduction in eosinophil count, and a reduction in superoxide production in alveolar macrophages *ex vivo*. This is the first published report of a cysteinyl LT antagonist altering cellular infiltration and activation after allergen challenge of patients with asthma.

Inhibition of the allergen-induced early and late responses by the oral drugs and their attenuation of allergen-acquired hyperresponsiveness provides further evidence for the clinical potential of this class of compound in asthma.

Aspirin-induced asthma

For many years it has been known that in 5% to 10% of patients with asthma cyclooxygenase inhibitors, including aspirin, produce profound bronchoconstriction, particularly in patients with so-called late-onset intrinsic disease. Recent studies have shown that patients with aspirin-sensitive asthma have an increased production of cysteinyl LTs and increased airway sensitivity when challenged with these agents.¹² The cysteinyl LT receptor antagonists pobilukast and verlukast improve lung function and inhibit bronchoconstriction induced by aspirin in patients with aspirin-sensitive asthma,^{12, 56} supporting the view that activation of the 5-lipoxygenase pathway for the enhanced formation of cysteinyl LTs is an important component of this type of asthma.

EFFECT OF CYSTEINYL LT RECEPTOR ANTAGONISTS ON BASELINE LUNG FUNCTION IN ASTHMA

A particularly interesting observation made in several studies investigating the efficacy of cysteinyl LT antagonists in stable asthma has been the ability of these drugs to produce bronchodilatation, suggesting that cysteinyl LTs contribute to the basal airway tone in this disease. Gaddy et al.^{57, 58} have shown that intravenously administered MK-571 resulted in acute bronchodilatation, which was additive to that induced by the inhaled β_2 -agonist salbutamol in patients with mild to moderate asthma. The FEV₁ increased by 20% when compared with placebo. A second study⁵⁹ demonstrated that a single oral dose of 40 mg of zafirlukast produced a mean 8% increase in FEV₁ in 10 patients with mild to moderate asthma, despite concurrent treatment with inhaled corticosteroids. The bronchodilator effect was sustained for several hours and again was additive to that achieved by inhaled salbutamol. The oral compound pobilukast also produced a moderate de-

gree of bronchodilatation in patients with stable mild asthma.⁶⁰ In a further study, zafirlukast failed to induce significant bronchodilatation when given by inhalation at a dose of 1600 μ g, nor did it have an additive effect with salbutamol in patients with moderate asthma.⁶¹ This result appears to suggest that these drugs need to achieve significant circulating levels before this beneficial effect can be produced.

Pranlukast, given orally for 1 week to patients with stable asthma, produces a small but significant reduction in bronchial responsiveness to methacholine,³⁸ showing that cysteinyl LTs are involved in hyperresponsiveness in chronic asthma, as well as asthma induced after allergen challenge.

EFFECTS OF CYSTEINYL LT ANTAGONISTS IN CLINICAL ASTHMA

Preliminary studies in patients with mild to moderate disease showed a definite therapeutic benefit with cysteinyl LT antagonists. Oral RG 12525 at a dose of 200 mg per day for 10 days caused a small but significant (8%) increase in FEV₁ and a parallel increase in peak flow.³⁹ MK-571 (now discontinued), when administered orally for 6 weeks, resulted in an 8% to 14% increase in baseline FEV₁ and a 30% decrease in daily symptom scores, paralleled by a 30% reduction in β -agonist use and diurnal variation in peak flow³⁵; and its successor, MK-0476, produced similar improvements after 10 days of treatment in 29 patients with asthma.⁶² MK-0476 has the advantage of requiring only once-daily dosing, compared with twice-daily dosing for zafirlukast.

A double-blind, randomized, placebo-controlled, parallel-group study in outpatients with mild to moderate asthma examined the effects of oral zafirlukast at three doses (5, 10, and 20 mg twice daily) for 6 weeks; approximately 60 patients were randomized to each treatment group. Efficacy of objective and subjective measures in patients with symptoms was dose-dependent, and the greatest improvements were achieved with the 40 mg total dose. When compared with placebo, significant improvements in evening peak flow, a 30% reduction in rescue use of inhaled β_2 -agonist, a 46% reduction in night waking, and a 26% improvement in morning asthma and daily symptoms were observed; these observations have increased confidence in the efficacy of these drugs for treatment of clinical asthma.⁶³ Zafirlukast doses, up to 80 mg twice daily, have been given to 285 patients with asthma for 13 weeks, with beneficial effects increasing still further at the higher doses.⁶⁴

Zafirlukast and pranlukast have been well toler-

ated in clinical trials to date; the main adverse effects reported, which may or may not be related to the active treatment, were headache, dry mouth, and somnolence.^{63, 65, 66} Although further clinical studies are clearly necessary, evidence to date supports the concept that cysteinyl LT antagonists are safe and efficacious in the treatment of asthma, whether assessed in various challenge models or in the clinic.

LTB₄ RECEPTOR ANTAGONISTS

The verdict on a possible role of LTB₄ in asthma is yet to be delivered. LTB₄ is a potent chemotaxin for a range of inflammatory leukocytes, is a complete secretagogue for neutrophils, and can induce or promote secretion by lymphocytes of a range of inflammatory cytokines including IL-2, IL-4, IL-5, and IL-8.^{21, 22} In guinea pigs and dogs, LTB₄ inhalation causes an increase in bronchial responsiveness accompanied by neutrophil and eosinophil influx.^{67, 68} LTB₄ released during the early response to allergen challenge may conceivably be responsible for the nonspecific leukocyte influx during the late response. Some of the efficacy of LT synthesis inhibitors in human clinical trials may be due to their blockade of LTB₄ production, as well as that of the cysteinyl LTs, but the demonstration of a long-term involvement of LTB₄ in clinical asthma will require trials of specific antagonists acting at the LTB₄ receptor in human beings. No such trials have yet been reported.

In guinea pig and rat models of asthma, however, a specific LTB₄ receptor antagonist (U-75,302) reduces the allergen-induced increase in bronchial responsiveness⁶⁹ and specifically reduces eosinophil influx to the lungs.⁷⁰ More potent and selective LTB₄ antagonists, such as CP-105,696, are being developed.⁷¹ It is possible that such compounds will first enter clinical trials for inflammatory disease in which a role for LTB₄ is more widely accepted, including ulcerative colitis, psoriasis, and adult respiratory distress syndrome.

LT SYNTHESIS INHIBITION

An alternative approach to targeting activity of LTs in asthma is to block their production with drugs that inhibit either 5-lipoxygenase or FLAP enzymatic activity. Blockade of 5-lipoxygenase-FLAP interaction blocks the synthesis of both LTB₄ and the cysteinyl LTs.

The 5-lipoxygenase inhibitors

Lipoxygenase and cyclooxygenase enzymes contain a catalytically important iron atom, which is

susceptible to inhibition by compounds with a low redox potential, such as the prototype drugs in this class, BW755c and phenidone, which had little selectivity for 5-lipoxygenase. Trials showed the early, selective 5-lipoxygenase inhibitors nafazotrom, piriprost (U-60,257), and docebenone (AA-861) to be ineffective in reducing bronchial responsiveness or allergen-induced bronchoconstriction in human beings,⁷²⁻⁷⁴ probably because of low inherent potency or poor bioavailability as a result of a short half-life in vivo. AA-861 was the prototype of a large group of hydroxamic acid derivatives, which share antioxidant properties but which have enhanced potency and selectivity for 5-lipoxygenase compared with other lipoxygenases and cyclooxygenase. One of the best studied of this group is the 5-lipoxygenase inhibitor zileuton.⁷⁵

Urinary LTE₄ excretion is a marker for whole-body synthesis of the cysteinyl LTs, and this can be used to monitor the bioavailability and efficacy of the LT synthesis inhibitors. In a placebo-controlled study of nine patients with asthma, oral zileuton (800 mg) inhibited the early response to allergen challenge and reduced LT synthesis in vivo (measured as urinary LTE₄ concentration) by 50% and ex vivo (measured as ionophore-stimulated LTB₄ synthesis in blood) by 80%.⁷⁶ Zileuton ameliorated the response to cold air challenge in 13 patients with asthma⁷⁷ and to aspirin challenge in eight patients with aspirin-sensitive asthma.⁷⁸ A clinical trial of zileuton, 1.6 to 2.4 gm/day, in 139 patients with mild to moderate asthma demonstrated an increase in FEV₁ of 15% within an hour of treatment, which was maintained at the end of an additional 4 weeks of treatment.⁷⁹ Zileuton is as effective as theophylline in treatment of moderate asthma; 600 mg four times daily produced a sustained increase in FEV₁ of around 20% for up to 13 weeks without the need for blood monitoring of drug concentrations.⁸⁰ Zileuton has also been given to patients with moderate asthma (mean FEV₁, 63%) for 6 months⁸¹; FEV₁ improved with zileuton compared with placebo after day 8 and continued to improve to a maximum of 19% after 120 days. Asthma symptoms were reduced by 31% to 38%, and β -agonist and inhaled steroid use fell by 30% and 22%, respectively. The number of acute asthma exacerbations was only 76 in the zileuton group compared with 134 in the placebo group. Zileuton has recently been shown to have a remarkable steroid-sparing effect in patients with severe asthma (FEV₁ <50% of predicted value), who required 80% lower doses of corticosteroid when treated for 13 weeks with zileuton (600 mg

four times daily).⁸² These results support the concept that LTs are involved in asthma across the whole range of disease severity and that the greater the severity, the more the patient may gain from anti-LT therapy.

Zileuton appears to be safe in clinical trials, but with compounds of this class there is theoretically more cause for concern because they are antioxidants able to interfere with redox reactions in other metalloenzymes, and some early compounds were withdrawn because of methemoglobinemia. Nevertheless, zileuton has produced sustained improvement in lung function in a number of clinical trials, and its steroid-sparing activity strongly suggests an antiinflammatory effect. These properties make zileuton the leading compound among this group of drugs for introduction to clinical practice as asthma therapy. Further studies are required to examine the possible effect of zileuton and other LT synthesis inhibitors on cellular influx and activation markers after allergen challenge and in clinical asthma.

Zileuton's relatively low potency and short half-life in vivo (2.5 hours) mean that dosing four times a day is required. More potent and selective 5-lipoxygenase inhibitors have been developed, including ABT-761 and Z-D2138, which have much longer half-lives in vivo (12 hours) than zileuton, allowing the possibility of a once-daily formulation. ABT-761 has been shown to protect patients with asthma from allergen challenge,⁸³ and Z-D2138 reduces responses to cold air challenge.⁸⁴ Results of further clinical trials are keenly awaited. In particular, it would be of interest to determine whether the relative lack of efficacy of zileuton in blocking the late response to allergen is a function of potency or bioavailability or whether it is a general feature of this class of drug.

The FLAP inhibitors

LT synthesis depends on the interaction between 5-lipoxygenase and FLAP. Unlike 5-lipoxygenase, however, FLAP bears no structural resemblance to other enzymes, except, curiously, to LTC₄ synthase.⁸⁵ As a result, inhibitors of FLAP are perhaps inherently less likely to interfere with other metabolic pathways and cause side effects than the direct 5-lipoxygenase inhibitors. The archetypal drug in this group was MK-886. Although MK-886 blocks LTC₄ synthase, as well as FLAP,⁸⁵ it is not clear whether its efficacy is enhanced by this blockade of cysteinyl LT production at two successive points in the synthetic pathway.

In eight atopic subjects, MK-886 reduced the

early response to allergen by 58%, and this was accompanied by a reduction in the production of urinary LTE_4 of 52%. The late response was inhibited by 44%, accompanied by a reduction in urinary LTE_4 of up to 80%.⁸⁶ MK-886 has now been withdrawn from further clinical trials, but two new FLAP inhibitors, MK-0591 and BAY x1005, have become the current leading compounds in this class of drug. MK-0591 was given to eight men with asthma in three equal doses of 250 mg over 24 hours preceding allergen challenge; the drug very effectively blocked LT synthesis *in vivo*, as shown by a near-total (98%) block of ionophore-stimulated blood synthesis of LTB_4 *ex vivo* and by an 87% inhibition of the rise in urinary LTE_4 .⁸⁷ Moreover, MK-0591 blocked the early response to allergen by 79% and the late response by 39%. The close similarity of these figures to those achieved by a number of cysteinyl LT receptor antagonists suggests that they represent the upper limits of the proportions of each response mediated by cysteinyl LTs and that even more potent drugs are unlikely to have a greater effect in this model.

In clinical asthma, a recent study of 6 weeks of treatment with MK-0591 in 239 subjects showed improvements in lung function, reduction in β_2 -agonist use, fewer asthma symptoms, and no adverse effects.⁸⁸ The only trial of BAY x1005 in clinical asthma reported to date⁸⁹ demonstrated small (6%) but significant increases in FEV_1 after 4 weeks of treatment in 67 patients with moderate, chronic asthma who were also receiving corticosteroids.

CONCLUSIONS

There is now overwhelming evidence that the cysteinyl LTs play an important role as inflammatory mediators in the pathogenesis of asthma. Clinical studies with selective cysteinyl LT receptor antagonists have been critical in confirming this role. Both cysteinyl LT antagonists and LT synthesis inhibitors reduce the symptoms of asthma triggered by physiologic, environmental, and allergenic stimuli. The most widely investigated antagonist, zafirlukast, has been shown to protect against bronchoconstriction provoked by inhaled LTD_4 , exercise, cold air, PAF, and allergen. In addition, cysteinyl LT antagonists inhibit the late asthmatic response to inhaled allergen and the subsequent increase in bronchial responsiveness, suggesting that they may have effects on the inflammatory cascade. Preliminary studies in patients with mild, moderate, and severe asthma have shown that LT antagonists and inhibitors exert significant therapeutic benefit. Improvements in

lung function are additive to β_2 -agonists, suggesting that endogenous LTs are doing much more than simply constricting bronchial smooth muscle. In addition, the continued clinical improvement over several weeks of treatment and the evidence that LT antagonists and inhibitors have a steroid-sparing effect suggest that they also have some underlying impact on modifying the disease process.

With these factors in mind, the new generation of cysteinyl LT receptor antagonists may be suitable as first-line therapy for patients with mild to moderate asthma when used alone or in combination with β -agonists used as required. They may be of special benefit in patients with exercise- or aspirin-induced asthma. As an oral medication, they may be useful in patients with poor compliance with inhaled drugs because of poor inhaler technique. Their use in severe asthma has yet to be fully evaluated, but it is possible that they may be effective in allowing reduction of inhaled or oral steroid doses, although it is not likely that they will replace steroids in this group. They may also be a safer alternative to theophylline as an oral prophylactic with bronchodilator and mild antiinflammatory properties. However, until more clinical studies are done, the precise positioning of this class of compound in the armamentarium of asthma therapy remains to be demonstrated.

Cysteinyl LT receptor antagonists represent a step forward and a new direction in targeted drug therapy for asthma and will be the first new class of antiasthmatic drugs introduced in nearly 25 years. Time will tell whether the newer LT synthesis inhibitors will become an equally safe and effective route to blocking cysteinyl LT action and whether they may demonstrate additional antiinflammatory effects by virtue of their blockade of LTB_4 synthesis.

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