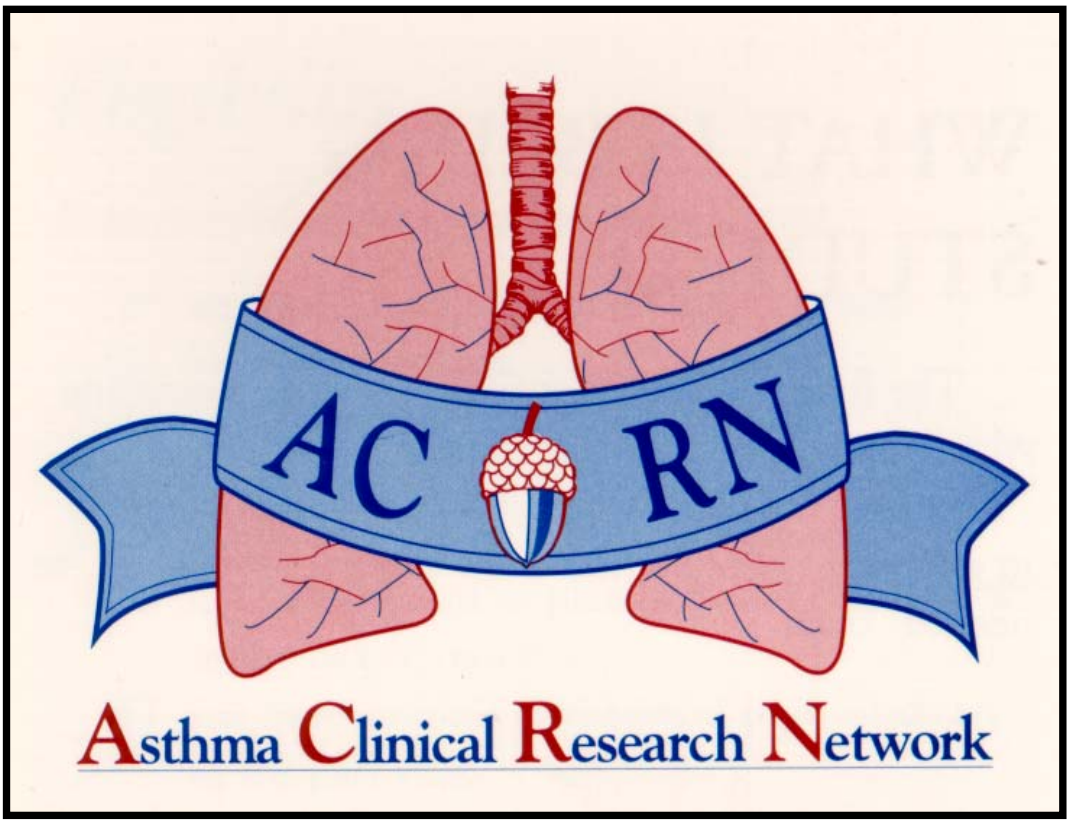

Smoking Modulates Outcomes of Glucocorticoid Therapy in Asthma

The "SMOG" Study

A study of subjects with persistent asthma comparing the effect of 32 weeks of inhaled corticosteroid therapy in subjects who smoke cigarettes with that in subjects who do not smoke.

May 6, 2002

Version 1.9

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I. HYPOTHESES:

A. Principal Hypothesis to be Tested:

Smoking activates mechanisms in asthmatics that reduce the response to inhaled corticosteroids.

Proposed Null Hypothesis: In subjects with persistent asthma who smoke, the effect of continuous daily treatment with an inhaled corticosteroid on FEV_1 does not differ from the effect observed in subjects with persistent asthma who do not smoke.

B. Additional Null Hypotheses to be Tested:

- The effect of continuous daily treatment with an inhaled corticosteroid on PC_{20}Methacholine does not differ between subjects who smoke and those who do not smoke.

- The effect of continuous daily treatment with an inhaled corticosteroid on markers of inflammation in induced sputum does not differ between subjects who smoke and those who do not smoke.

- The effect of continuous daily treatment with an oral leukotriene receptor antagonist on FEV_1, PC_{20}Methacholine, and markers of inflammation in induced sputum does not differ between subjects who smoke and those who do not smoke.

- The effect of continuous daily treatment with an oral leukotriene receptor antagonist does not differ from the effect of continuous daily treatment with an inhaled corticosteroid.

II. BACKGROUND AND RATIONALE:

A. Epidemiology of Smoking and Asthma

Epidemiologic studies of the role of smoking in the development of adult asthma have found either no association (1), a positive association (2, 3), or a negative association (4). However, the association between smoking and asthma severity is clear: Asthmatic patients who smoke have more severe respiratory symptoms (5, 6), and demonstrate a greater loss of lung function over time than do asthmatics who do not smoke (7). In a cohort study of adult asthmatics followed by telephone interviews over 18 months, Eisner and colleagues found that those who smoked throughout the 18-month follow-up had more asthma symptoms, worse quality of life, more emergency department visits, and more hospitalizations than asthmatics who did not smoke, or those who quit smoking during the study (5).

The data on the rate of loss of pulmonary function among asthmatic patients who smoke is particularly alarming. Lange et al (7) followed pulmonary function for 15 years in 17,506 subjects from the general population of Copenhagen, of whom 6.2% had asthma. The yearly decline in forced expiratory volume in one second (FEV_1) was 22 ml for non-asthmatic subjects and 38 ml for asthmatic subjects. Moreover, the decline was greatest among the asthmatic subjects who smoked. These data suggest that cigarette smoking may accelerate the mechanisms of irreversible airway wall "remodeling," which are attributed to the effects of chronic airway inflammation (8).

The U.S. Centers for Disease Control surveyed a nationally representative sample of 32,440 civilians aged ≥18 years in the 1998 National Health Interview Survey. Based on the results of this questionnaire, they estimated that 47.2 million adults (24.1% of the population) were current smokers (9). Other studies, in the United States and abroad, have reported that the prevalence of active cigarette smoking among adults with asthma is approximately the same as in the general adult population (10-13). Moreover, even non-smoking asthmatics may have significant exposure to passive smoke. One recent study in Northern Italy reported that 55% of children and adolescents with asthma lived in a home where at least one parent was an active smoker (14).

B. Effect of Corticosteroids on Airway Disease in Smokers

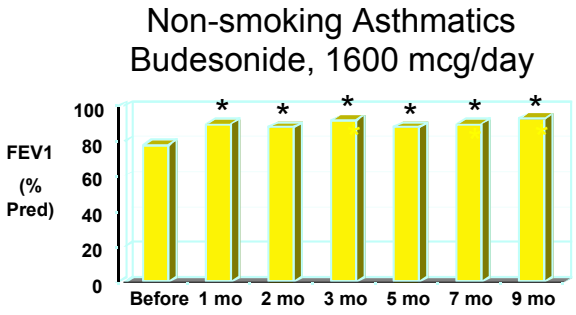
1. COPD: The effect of corticosteroids on airway disease in people who smoke has been studied almost exclusively in COPD. There are data that support the use of systemic corticosteroids in the treatment of acute exacerbations of COPD (15, 16). However, long-term use of corticosteroids to manage chronic COPD has not been demonstrated to be beneficial. In particular, there have been no data to suggest that chronic use of inhaled corticosteroids will alter the rate of progression of COPD. Recently, 4 large, multi-national, multi-center studies of the chronic use of corticosteroids in COPD have been completed (The Copenhagen City Lung Study, The EUROSCOP Study, The ISOLDE Study, and The Lung Health Study II). The results of these studies as regards FEV_1 are summarized in tabular form below, along with data from the Lung Health Study I, showing the decrease in FEV_1 in active smokers with COPD vs. those who quit:

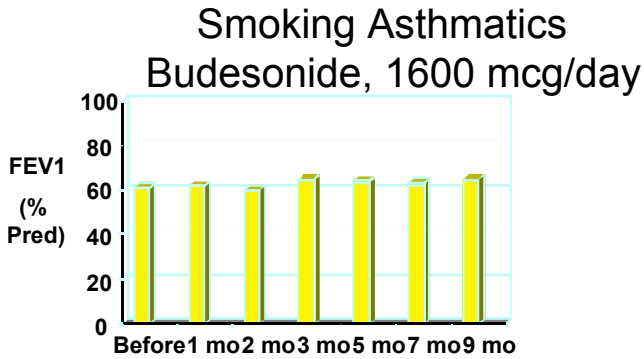
			Decrease in FEV_1/year	
STUDY	**SITES**	**COMPARISON**	**SMOKERS**	**QUITTERS**
Lung Health Study I	US + Canada	Smokers vs. Quitters	62 mL	32 mL
STUDY	**SITES**	**COMPARISON**	**PLACEBO**	**STEROID**
Copenhagen City Lung Study	Denmark	BUD vs. PBO	42 mL	42 mL
EUROSCOP	Europe	BUD vs. PBO	69 mL	57 mL
ISOLDE	Europe	FP vs. PBO	59 mL	50 mL
Lung Health Study II	US + Canada	TAC vs. PBO	47 mL	44 mL

The conclusions from these 4, large, carefully conducted studies are clear: Inhaled corticosteroids do not modify the long-term decline in FEV_1 in people with COPD.

2. Asthma: Far less is known about the effects of corticosteroids in asthmatic patients who smoke. Nearly all clinical studies on the pathogenesis and treatment of asthma have specifically excluded subjects who smoke in order to avoid the confounding effects of enrollment of patients with COPD. In an extensive search of the literature, we have found only two studies of the effects of corticosteroids in asthma patients who smoked cigarettes, both of which utilized a *post hoc* analysis to examine subgroups of patients who smoked.

Pedersen and colleagues (17) compared the effect of two doses of inhaled budesonide with that of theophylline in 85 asthmatics, in a study designed to relate circulating markers of eosinophil and neutrophil activation to measures of airway function. They found a dose-dependent effect of budesonide on both eosinophil markers and lung function. However, when the budesonide-treated groups were separated into smokers and non-smokers, they discovered that the positive effects of budesonide were attributable primarily to its effects in non-smokers. Even high-dose budesonide (1600 mcg/ day) had no effect on FEV_1, PC_{20}Histamine, beta-agonist use, or serum levels of eosinophils, ECP, or EPX in those asthmatic subjects who smoked.





They concluded that "the smoking asthmatic subjects behaved as steroid-resistant subjects."

In a second study (18), designed to compare the effects of high versus low dose budesonide as initial treatment in newly detected asthma, the authors found that the FEV_1 response was significantly blunted in smokers ($p<0.05$) and that the reduction in methacholine reactivity was less ($p=0.07$). However, this study was not designed to

compare smokers and non-smokers and, as the authors themselves pointed out, “The number of smoking asthmatics was relatively small…making it difficult to draw definite conclusions.”

Thus, while data from retrospective analyses of subgroups of asthmatic subjects who smoke suggest that the response to inhaled corticosteroids in these patients may be less than that observed in non-smoking asthmatics, we could find no study designed to examine this important issue. We therefore propose this randomized, double-blind, crossover study that will compare the effect of an inhaled corticosteroid on FEV_1 in asthmatic subjects who do and do not smoke. Secondarily, we will compare the response in the 2 groups with regard to AM- and PM-Peak Expiratory Flow (PEF), PC_{20}Methacholine, and markers of inflammation in induced sputum (total cell count and differential, eosinophil cationic protein, and tryptase), as well as the effects on these variables of treatment with a leukotriene receptor antagonist.

C. Airway Inflammation in Asthma and in Smoking-Related Airway Disease

It is now generally accepted that asthma is an inflammatory disease, characterized by infiltration of the airway mucosa and submucosa with eosinophils, lymphocytes, and other cell types (19). At times, for example during asthma exacerbations, expectorated sputum from asthmatics reveals a shift in this inflammatory cell profile to a polymorphonuclear predominance (20). The exact mechanism of airway injury by tobacco smoke is not well understood, but inflammation and inflammatory mediators again seem to be involved. Exposure to tobacco smoke causes both bronchoconstriction and increased airway responsiveness in asthmatics (21), and airway hyperresponsiveness is a marker of airway inflammation. Analysis of induced sputum obtained from patients with COPD has demonstrated increased polymorphonuclear leukocytes, but rare eosinophils (22). A single study comparing the constituents of induced sputum obtained from smoking and non-smoking asthmatics found that the asthmatics who smoked had significantly more neutrophils and higher levels of IL-8 than did the non-smokers (23). These differences in the inflammatory cell profile might be reflected as differences in the way smoking and non-smoking asthmatics respond to various pharmacologic therapies.

Although inhaled corticosteroids are the most widely-prescribed anti-inflammatory (controller) medication for asthma (24), there are now alternatives to consider, including the leukotriene receptor antagonists. In addition to causing airway narrowing through activation of airway smooth muscle, the leukotrienes are known to produce airway edema, inflammatory cell influx, and possibly increased mucus production (25-29). Use of the leukotriene modifiers has been shown to decrease asthma exacerbations (30-35). Further, they have been shown to have effects similar to inhaled corticosteroids on inflammatory mediators that we associated with discontinuation of inhaled corticosteroids in the SOCS trial. These therapeutic agents have been shown to reduce exhaled nitric oxide levels (36, 37) and they reduce circulating and sputum eosinophils (36, 38, 39) and other mediators of airway inflammation (40, 41). They may also reduce levels of sputum tryptase (42), a marker that increased with inhaled corticosteroid withdrawal in the SOCS/SLIC trial and that may predict those patients who deteriorate upon withdrawal of inhaled corticosteroids.

Several studies have now demonstrated that leukotriene receptor antagonists provide additional physiologic and clinical improvement when added to the regimen of patients whose asthma is inadequately controlled on inhaled corticosteroids alone (43-46). This additional benefit of leukotriene receptor antagonists suggests the presence of leukotrienes in these subjects treated with inhaled corticosteroids. In fact, there are substantial data both from *in vitro* (47) and *in vivo* (48, 49) studies, including those of patients with asthma (47, 50), demonstrating that corticosteroids may not inhibit generation of leukotrienes. Thus, if the response to inhaled corticosteroids is blunted in asthmatics who smoke, the reponse to a leukotriene receptor antagonist will be interesting. If leukotrienes play an important role in this patient population, the difference in response between smokers and non-smokers seen when these subjects are treated with an inhaled corticosteroid may not be seen when the two groups are treated with a leukotriene receptor antagonist.

III. SPECIFIC AIMS

1. To determine whether treatment with an inhaled corticosteroid is less effective in asthmatic subjects who smoke cigarettes than in asthmatic subjects who do not smoke with regard to:

- Improvement in FEV_1
- Improvement in other measures of airway function (AM-PEF, PM-PEF, PC_{20}Methacholine)
- Reduction in the number of inflammatory cells and inflammatory mediators recovered from the airways
- Improvement in clinical outcomes such as asthma symptom scores, quality of life scores, use of ß-agonist rescue, asthma exacerbations

2. To determine whether treatment with a leukotriene receptor antagonist is less effective in asthmatic subjects who smoke cigarettes than in asthmatic subjects who do not smoke with regard to:

- Improvement in airway function
- Reduction in the number of inflammatory cells and inflammatory mediators recovered from the airways
- Improvement in clinical outcomes such as asthma symptom scores, quality of life scores, use of ß-agonist rescue, asthma exacerbations

3. To compare the response to an inhaled corticosteroid with the response to a leukotriene receptor antagonist with regard to:

- Improvement in FEV_1
- Improvement in other measures of airway function (AM-PEF, PM-PEF, PC_{20}Methacholine)
- Reduction in the number of inflammatory cells and inflammatory mediators recovered from the airways
- Improvement in clinical outcomes such as asthma symptom scores, quality of life scores, use of ß-agonist rescue, asthma exacerbations

IV. RATIONALE FOR CHOOSING THESE QUESTIONS

Recognition of the critical importance of inflammation in the pathogenesis of asthma has led to recommendations that primary therapy for asthma include an agent effective at reducing airway inflammation. Corticosteroids are the most effective agents for the treatment of asthma, and currently, inhaled corticosteroids are the most widely-prescribed controller medication in the United States (24). Studies such as those by Haahtela (51, 52), Agertoft (53), and Overbeek (54) have suggested that delay in instituting inhaled corticosteroid therapy in newly-diagnosed asthma may result in loss of lung function. This has led national (55) and international (56) expert panels to recommend that all patients with persistent asthma (including many that previously would have been treated only on an "as needed" basis) receive inhaled corticosteroids. Because of the large number of patients affected by this recommendation, its economic impact is enormous. The prevalence of smoking amongst patients with asthma is disappointing, approximately the same as that in the population at large, i.e., 20-30% (9-13). Few studies have compared the effect of asthma medications in smokers versus non-smokers. In fact, most studies of asthma therapy have deliberately and meticulously excluded patients who smoke, to avoid the confounding effects of enrolling patients with COPD. There are, however, at least 2 studies in which a *post hoc* analysis suggests that smokers have a blunted response to inhaled corticosteroids. If true, there are important implications for how we treat asthmatics who smoke. If we assume that 20-30% of the 17 million Americans with asthma smoke, and that approximately 60-70% of these have mild, moderate or severe persistent asthma requiring 1 canister of inhaled corticosteroid/month at approximately $45/canister, then the cost of administering inhaled corticosteroids to this population would be more than $900 million/year. If asthmatics who smoke do not respond to inhaled corticosteroids (or if they have a significantly blunted response) this may not be an appropriate form of therapy, and the risk/benefit ratio shifts significantly. The purpose of this study is to investigate this important issue in a double-blind randomized controlled trial.

V. PROTOCOL

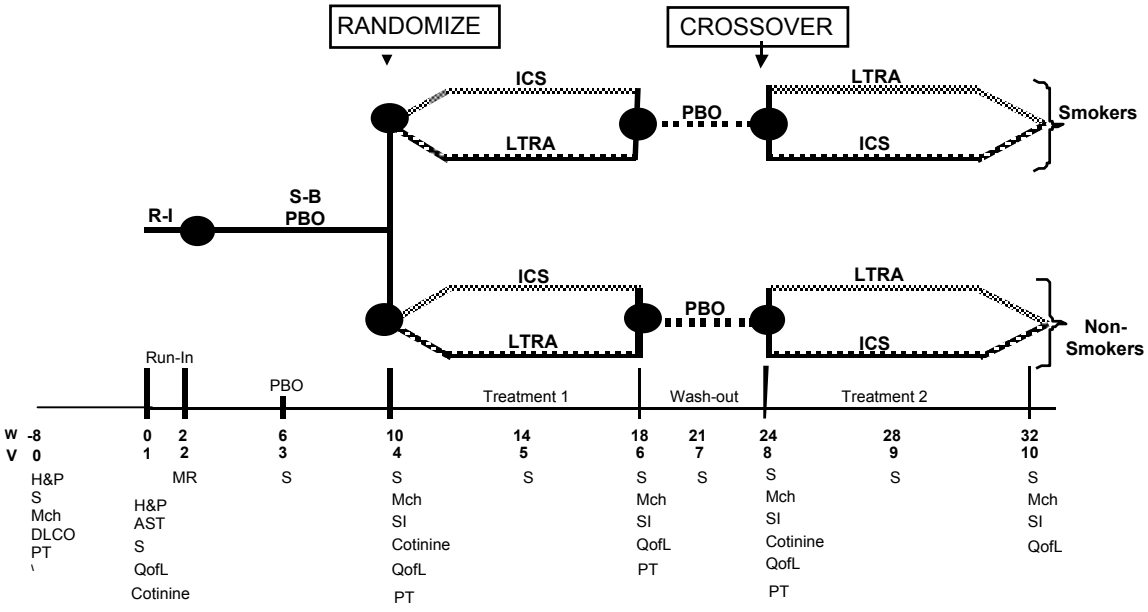
A. Overview

This randomized, double-blind, crossover trial, will compare the effect of inhaled corticosteroid treatment delivered twice daily for 8 weeks in two groups of subjects with persistent asthma: one group will be comprised of smokers; the other will be non-smokers. Smokers and non-smokers will be matched into pairs according to gender and FEV_1 status (70-80% predicted or 80-90% predicted) prior to the run-in period. Subjects will also be matched within a ten year age window. Each member of the matched pair will be randomized together to the same crossover sequence. All subjects will be 18-50 years old. The primary outcome will be change in pre-bronchodilator FEV_1 over the 8-week treatment period in smokers compared with non-smokers. Secondary outcomes are AM- and PM-PEF, PC_{20} Methacholine, and markers of inflammation in induced sputum. A secondary comparison will examine the effect of 8 weeks of treatment with a leukotriene receptor antagonist in asthmatics who smoke versus asthmatics who do not smoke. An additional

analysis will compare the response to inhaled corticosteroid with the response to leukotriene receptor antagonist.

B. Protocol Detail and Schematic

Subjects will come to the clinical centers for 11 visits over 32 - 40 weeks. After a 2-week Run-In Period, to establish eligibility as well as familiarity and adherence with study protocols and forms, subjects will enter an 8-week Single-Blind Placebo Treatment Period. Following this period, asthmatics who smoke and asthmatics who do not smoke will be randomly assigned in parallel to receive an inhaled corticosteroid [beclomethasone dipropionate HFA, QVAR, 320 mcg B1D] or a leukotriene receptor antagonist (e.g., montelukast, 10 mg q evening), for an 8-week Double-Blind Treatment Period, using a double-dummy blinding technique. After a 6-week Placebo Wash-Out Period, subjects will "cross-over" to receive the alternative treatment for an additional 8 weeks.



W=Week; V=Visit number; H&P=History and Physical; Mch=Methacholine reactivity; AST=Allergy skin testing; S=Spirometry; DLCO=Diffusing capacity for carbon monoxide; QofL=Juniper Quality of Life assessment; MR=Maximum Reversibility; SI=Sputum induction; Cotinine=urinary cotinine; PT=Pregnancy Test; LTRA=Leukotreine receptor antagonist; ICS= Inhaled corticosteroid; PBO=Placebo.

C. Visit Structure

Visit 0, Prescreening ≤ 8 weeks prior to Visit 1

Subjects will be interviewed prior to protocol entry (either by phone or in person) regarding their asthma and medical history. Specifically, the current status of asthma control, use of asthma and non-asthma medications, health status in the previous 6 weeks, and smoking status will be determined. An overview of the goals of the study and the visit structure and procedures involved will be presented. If the patient appears to fulfill entry criteria, is interested in study participation, and is not taking oral corticosteroids, a leukotriene modifying drug, salmeterol, inhaled nedocromil or cromolyn, Visit 0 may be scheduled. If the patient is taking one of these asthma medications regularly, a pre-study visit must be scheduled, informed consent obtained, and the patient evaluated by the study investigator as to the appropriateness of drug withdrawal for the 6 weeks prior to Visit 1. If warranted, the investigator may request additional pre-study visits for evaluation of asthma stability during this 6-week period. If, in the investigator's opinion, the subject is likely to be clinically stable, the subject may enroll.

On this first visit, written Informed Consent will be obtained, using a document that has been approved by the ACRN as well as by the local IRB. Vital signs, a brief physical examination, spirometry, diffusing capacity measurement, and reversibility or methacholine challenge (if not performed within 6 months and if permissible by spirometric criteria) will be obtained. Urine will be obtained for a pregnancy test in females. Blood will be drawn for genetic analysis related to allergy, asthma, and inflammation.

Spirometry, measurement of diffusing capacity, and methacholine challenge will be performed according to protocols outlined in the ACRN Manual of Procedures. All data will be recorded electronically and on forms supplied by the ACRN.

At Visit 0, smokers will be questioned regarding their interest in smoking cessation as an alternative to study participation. Those who are interested will be referred to an appropriate program. For those who decline, a similar offer will be made at the end of the study.

Visit 1, Week 0

Subjects will return to the clinical center within 8 weeks of Visit 0. Medical history, vital signs, physical examination, and spirometry will be performed. A quality-of-life questionnaire will be administered and allergy skin tests will be performed.

If, based on the information gathered to this point, the subject meets the specific entry criteria, he/she will be entered into the run-in phase of the trial. Subjects will be given an "open label" albuterol inhaler, to be used for rescue treatment, and an electronic peak flow meter. Prior to distribution the peak flow meter readings will be checked using a Jones Flow-Volume Calibrator. Only peak flow meters whose readings are within a specific range will be distributed. Subjects will be taught how to use their peak flow meters and albuterol

MDIs. They will be instructed to measure and record peak flow on diary cards immediately upon arising and at bedtime (between 2000 and 0100 hrs). Subjects will be instructed to circle on their diary cards those peak flow values obtained less than 2 hours after use of inhaled albuterol. Subjects will also be instructed to record daily asthma symptom severity on diary cards. The use of diary cards will be explained and subjects will be given an appropriate supply. Subjects will be instructed to return to the clinical center in 2 weeks.

Visit 2, Week 2

Subjects will return to the clinical center at the same time of day as on Week 0 ±2 hours. If scheduling permits, all subsequent visits will occur within a ± 2-hour window on the study day. A short physical exam will be administered. Spirometry will be performed, along with the Maximum Reversibility Test, to determine the best achievable post-bronchodilator FEV_1. The subject's peak flow meter will be tested against the Jones Flow-Volume calibrator and replaced if it does not meet defined quality control standards. Adverse events will be noted, using the protocol defined by the ACRN. Diary cards will be reviewed and new ones dispensed. Clinical center personnel will assess adherence by reviewing the appropriateness and timing of peak flow recording and symptom recording. Guidelines will be reviewed. The single-blind placebo-inhaled steroid MDI and placebo-leukotriene receptor antagonist tablets will be issued. Subjects will be instructed as to how and when to take these medications. Open label albuterol will be issued as needed. Subjects will be instructed to return to the clinical center in 4 weeks.

Visit 3, Week 6

Subjects will return to the clinical center at the same time of day as on Week 0 ± 2 hours. If scheduling permits, all subsequent visits will occur within a ± 2-hour window on the study day. A short physical exam will be administered and spirometry will be performed. The subject's peak flow meter will be tested against the Jones Flow-Volume calibrator and will be replaced if it does not meet defined quality control standards. Adverse events will be noted using the protocol outlined by the ACRN. Diary cards will be reviewed and new ones dispensed; peak flow data will be uploaded. Clinical center personnel will assess adherence by reviewing the appropriateness of the timing of medication use, peak flow recording and symptom recording. Guidelines will be reviewed. Single-blind medications and open label albuterol will be issued as needed. Subjects will be instructed to return to the clinical center in 4 weeks.

Visit 4, Week 10 (Randomization)

Subjects will return to the clinical center at the same time of day as on Week 0 ± 2 hours. If scheduling permits, all subsequent visits will occur within a ± 2-hour window on the study day. The subject's peak flow meter will be tested against the Jones Flow-Volume calibrator and will be replaced if it does not meet defined quality control standards. Adverse events will be sought and noted using the protocol outlined by the ACRN. Diary cards will be reviewed and new ones dispensed; peak flow data will be uploaded. Clinical center personnel will assess adherence by reviewing the appropriateness of the timing of

medication use, peak flow recording and symptom recording. Guidelines will be reviewed as needed. Subjects will complete the asthma quality of life questionnaire. Urine will be obtained for a pregnancy test in females.

If adherence criteria and study entry criteria are met, spirometry, methacholine challenge and sputum induction will be performed in that order as per protocols outlined in the ACRN MOP. A short physical exam will be administered and a urine sample will be obtained for cotinine determination.

If the subject continues to meet the inclusion criteria for the study, and fulfills the criteria for randomization, the ACRN DCC will be contacted and the subject will be randomized to one of the 2 double-blind treatment arms (inhaled corticosteroid or oral LTRA). Based upon this randomization, all subjects will receive one coded, double-blind "regular use" metered-dose inhaler to be taken twice a day. This inhaler will contain either the inhaled steroid or its placebo. Subjects will be instructed to take their "regular use" inhalers at the same time each evening and morning (approximately 0500 – 1000 and 2000 – 0100 hrs), and to measure their peak flow at the same time each morning. Subjects will also receive a five weeks' supply of LTRA or its placebo, and will be instructed to take the LTRA as appropriate. All subjects will receive open label albuterol inhalers and prednisone, to be used as needed for rescue treatment. New study specific diary cards will be issued. Subjects will be instructed to return to the clinical center in 4 weeks.

Visit 5, Week 14

Subjects will return to the clinical center at the same time of day as on Week 0 ± 2 hours, having taken their study medication approximately 12 hours before the time of the visit. If scheduling permits, all subsequent visits will occur within a ±2-hour window on the study day. A short physical exam will be administered and spirometry will be performed. The subject's peak flow meter will be tested against the Jones Flow-Volume calibrator and will be replaced if it does not meet defined quality control standards. Adverse events will be noted using the protocol outlined by the ACRN. Diary cards will be reviewed and new ones dispensed; peak flow data will be uploaded. Clinical center personnel will assess adherence by reviewing the appropriateness of the timing of medication use, peak flow recording and symptom recording. Guidelines will be reviewed. Double-blind medications and open label albuterol will be issued as needed. Subjects will be instructed to return to the clinical center in 4 weeks.

Visit 6, Week 18

Subjects will return to the clinical center at the same time of day as on Week 0 ± 2 hours, having taken their study medication approximately 12 hours before the time of the visit. If scheduling permits, all subsequent visits will occur within a ±2-hour window on the study day. A short physical exam will be administered. The subject's peak flow meter will be tested against the Jones Flow-Volume calibrator and will be replaced if it does not meet defined quality control standards. Adverse events will be noted using the protocol outlined by the ACRN. Diary cards will be reviewed and new ones dispensed; peak flow data will be

uploaded. Clinical center personnel will assess adherence by reviewing the appropriateness of the timing of medication use, peak flow recording and symptom recording. Guidelines will be reviewed. Subjects will complete the asthma quality of life questionnaire. Urine will be obtained for a pregnancy test in females.

Spirometry, methacholine challenge and sputum induction will be performed in that order as outlined in the ACRN MOP.

Double-blind medications will be collected and single-blind placebo and open label albuterol will be issued. Subjects will be instructed to return to the clinical center in 3 weeks.

Visit 7, Week 21

Subjects will return to the clinical center at the same time of day as on Week 0 ± 2 hours. If scheduling permits, all subsequent visits will occur within a ± 2-hour window on the study day. A short physical exam will be administered and spirometry will be performed. The subject's peak flow meter will be tested against the Jones Flow-Volume calibrator and will be replaced if it does not meet defined quality control standards. Adverse events will be noted using the protocol outlined by the ACRN. Diary cards will be reviewed and new ones dispensed; peak flow data will be uploaded. Clinical center personnel will assess adherence by reviewing the appropriateness of the timing of medication use, peak flow recording and symptom recording. Guidelines will be reviewed. Single-blind medications and open label albuterol will be issued as needed. Subjects will be instructed to return to the clinical center in 3 weeks.

Visit 8, Week 24

Subjects will return to the clinical center at the same time of day as on Week 0 ± 2 hours. If scheduling permits, all subsequent visits will occur within a ± 2-hour window on the study day. The subject's peak flow meter will be tested against the Jones Flow-Volume calibrator and will be replaced if it does not meet defined quality control standards. Adverse events will be sought and noted using the protocol outlined by the ACRN. Diary cards will be reviewed and new ones dispensed; peak flow data will be uploaded. Clinical center personnel will assess adherence by reviewing the appropriateness of the timing of medication use, peak flow recording and symptom recording. Guidelines will be reviewed as needed. Subjects will complete the asthma quality of life questionnaire. Urine will be obtained for a pregnancy test in females.

If adherence criteria and study entry criteria are met, spirometry, methacholine challenge and sputum induction will be performed in that order as per protocols outlined in the ACRN MOP. A short physical exam will be administered and a urine sample will be obtained for cotinine determination.

Subjects will then enter the second double-blind treatment period, during which they will crossover to receive whichever medication they did not receive during the first double-blind

treatment. All subjects will now receive one coded, double-blind "regular use" metered-dose inhaler to be taken twice a day. This inhaler will contain either the inhaled steroid or its placebo. Subjects will be instructed to take their "regular use" inhalers at the same time each evening and morning (approximately 0500 - 1000 and 2000 - 0100 hrs), and to measure and record their peak flow at the same time each morning. They will also be instructed to record daily asthma symptom severity. Subjects will also receive a five weeks' supply of LTRA or its placebo, and will be instructed to take the LTRA as appropriate. All subjects will receive open label albuterol inhalers and prednisone, to be used as needed for rescue treatment. New study specific diary cards will be issued. Subjects will be instructed to return to the clinical center in 4 weeks.

Visit 9, Week 28

Subjects will return to the clinical center at the same time of day as on Week 0 ± 2 hours, having taken their study medication approximately 12 hours before the time of the visit. If scheduling permits, all subsequent visits will occur within a ±2-hour window on the study day. A short physical exam will be administered and spirometry will be performed. The subject's peak flow meter will be tested against the Jones Flow-Volume calibrator and will be replaced if it does not meet defined quality control standards. Adverse events will be noted using the protocol outlined by the ACRN. Diary cards will be reviewed and new ones dispensed; peak flow data will be uploaded. Clinical center personnel will assess adherence by reviewing the appropriateness of the timing of medication use, peak flow recording and symptom recording. Guidelines will be reviewed. Double-blind medications and open label albuterol will be issued as needed. Subjects will be instructed to return to the clinical center in 4 weeks.

Visit 10, Week 32

Subjects will return to the clinical center at the same time of day as on Week 0 ± 2 hours, having taken their study medication approximately 12 hours before the time of the visit. A short physical exam will be administered. The subject's peak flow meter will be tested against the Jones Flow-Volume calibrator. Adverse events will be noted using the protocol outlined by the ACRN. Diary cards will be reviewed; peak flow data will be uploaded. Clinical center personnel will assess adherence by reviewing the appropriateness of the timing of medication use, peak flow recording and symptom recording. Subjects will complete the asthma quality of life questionnaire. Urine will be obtained for a pregnancy test in females.

Spirometry, methacholine challenge and sputum induction will be performed in that order as outlined in the ACRN MOP.

Double-blind medications will be collected.

Smokers will be questioned regarding their interest in smoking cessation. Those who are interested will be referred to an appropriate program.

D. Protocol in Tabular Form

Variable		Run-in		Single-blind Placebo		Double-blind Treatment #1		Wash-out		Double-blind Treatment #2	
Visit	0	1	2	3	4	5	6	7	8	9	10
Week	-8	0	2	6	10	14	18	21	24	28	32
Window	±8w	±3d	±3d	±3d	±3d	±3d	±3d	±3d	±3d	±3d	±3d
Informed Consent	X										
Randomization					X						
History	X	X									
Long Physical Exam		X									
Short Physical Exam	X		X	X	X	X	X	X	X	X	X
Refer to Smoking Cessation	X										X
Blood for Genetic Analysis	X										
Allergy Skin Test		X									
Spirometry	X	X	X	X	X	X	X	X	X	X	X
Pregnancy Test	X	X			X		X		X		X
Methacholine Challenge	X				X		X		X		X
Maximum Reversibility			X								
DLCO	X										
Sputum Induction					X		X		X		X
Cotinine		X			X				X		
Quality of Life Questionnaire		X			X		X		X		X
Adverse Events Assessment	X	X	X	X	X	X	X	X	X	X	X
Review Peak Flow Data			X	X	X	X	X	X	X	X	X
Peak Flow QC			X	X	X	X	X	X	X	X	X
Dispense/ Review Diary Cards		X	X	X	X	X	X	X	X	X	X
Dispense / Collect Medicines			X	X	X	X	X	X	X	X	X
Dispense Prednisone		X									
Collect Meds											X

VI. SUBJECTS AND INCLUSION AND EXCLUSION CRITERIA

A. Study Population

This study will require a total of 96 adults with persistent asthma, half of whom smoke. Patients will be recruited from the "standing" populations of the participating centers, by advertisement, and by referral from participating physicians. Patients will meet the inclusion criteria specified herein and not possess any of the exclusion criteria. Both heterogeneity of the study group and rapidity of recruitment are greatly facilitated by the involvement of several geographically dispersed study centers in a multi-center collaboration. At randomization the smoking and non-smoking subjects will be matched according to gender and FEV_1. The Data Coordinating Center will distribute monthly accrual reports for each clinical center, categorizing patients entered by gender, age, and ethnicity. This routine monitoring will allow early identification and resolution of problems in achieving demographic goals.

B. Inclusion Criteria (Visit 0)

1. Male and female subjects, 18 to 50 years of age.
2. History of asthma, as defined by ATS criteria (57)
3. Heightened airway reactivity, shown by reversible airflow obstruction ≥12% after aerosolized albuterol, or by PC_{20}Methacholine ≤ 8mg/ml (documented within 6 months or performed at Visit 0).
4. FEV_1 70-90% of predicted after withholding bronchodilator and restricted medications per Manual of Operations.
5. Ability to provide informed consent, as evidenced by signing a copy of the consent form approved by the Committee on Human Research of the study institution.
6. No smokeless tobacco for at least 1 year.
7. No marijuana for ≥12 months.
8. D_LCO ≥80% of predicted. (Each center will use individual D_LCO predicteds regularly in place.
9. For heterosexual females, use of reliable contraception throughout the study.
10. For Non-Smokers:
  a. Total lifetime smoking < 5 pack-years.
  b. No smoking for at least 1 year.
11. For Smokers
  a. Total lifetime smoking 2-15 pack-years.
  b. Current smoking 10-40 cigarettes/day (1/2 – 2 packs/day).

C. Exclusion Criteria (Visit 1)

1. Use of any drugs listed in Table 1 during the designated washout period prior to Visit 1, or intention to take the drug during the study.

Table 1. Drugs to be withheld throughout the study	Washout prior to Visit 1
Inhaled Steroids, except as provided in study	> 6 weeks
Oral Steroids	> 6 weeks
Cromolyn/Nedocromil	> 2 weeks
Oral beta-adrenergic agonists	> 48 hours
Monoamine oxidase inhibitors	> 4 weeks
Tricyclic antidepressants	> 4 weeks
Beta-adrenergic blockers	≥ 48 hours
ACE inhibitors and Angiotensin II antagonists	> 2 weeks
Inhaled beta-adrenergic agonists (intermediate-acting, e.g., albuterol, terbutaline, metaproterenol, pirbuterol, bitolterol), except provided in study	≥ 8 hours
Salmeterol, formoterol	≥ 48 hours
Anticholinergics	≥ 48 hours
Short-acting theophylline (e.g., Slophyllin tablets)	≥ 48 hours
Long-acting theophylline (e.g., Theo-Dur, Slo-bid)	≥ 48 hours
Ultra long-acting theophylline (e.g., Theo-24, Uniphyl)	≥ 48 hours
Antihistamines (1st generation)	≥ 48 hours
Loratadine, Cetirizine	≥ 7 days*
Zafirlukast (Accolate) and Montelukast (Singulair), except as provided in protocol	> 2 weeks
Zileuton (Zyflo)	> 2 weeks
Drugs withheld prior to pulmonary function and/or methacholine challenge, per MOP	**Specified time period**
Albuterol	≥ 6 hours
Salmeterol	≥ 48 hours
Fexofenadine (Allegra)	≥ 72 hours
Chlorpheniramine (ChlorTrimeton)	> 48 hours
Methylxanthine-containing foods or beverages (e.g., coffee, tea)	≥ 6 hours
Alcohol-containing foods or beverages	≥ 6 hours

*Since these medications may remain in the body for weeks following administration, it is recommended that histamine prick skin testing be performed in patients who are currently on treatment. If a wheal and flare > 5 mm develops following testing, the subject would be eligible for study participation. If not, the subject would need a longer washout period (possibly weeks) prior to re-testing and enrollment.

2. Medication use: Chronic use of any medication other than short-acting ß-agonists, except oral contraceptives and other hormonal forms of contraceptives (i.e., DepoProvera, Norplant), vitamins, any nasal inhaled corticosteroid at a stable dose throughout the entire study (see MOP), acetaminophen, non-steroidal anti-inflammatory medications (e.g., aspirin, naproxen, ibuprofen, cox-2 inhibitors), thyroid replacement medications, lipid-lowering medication, or medium and low potency topical cutaneous steroids.

Allowable medications also include:

(a) calcium supplements
(b) nasal saline spray
(c) topical eye preparations for allergic eye symptoms (e.g., antihistamines, NSAIDs, or antiallergic compounds)
(d) diuretics
(e) specific antihypertensives (e.g., calcium channel blockers, clonidine,etc)
(f) acyclovir
(g) chlorpheniramine and fexofenadine (48 hour washout)
(h) pseudoephedrine (48 hour washout)
(i) antibiotics for acne
(j) stool softeners and bulk laxatives

3. Lung disease other than asthma.

4. Established diagnosis of vocal cord dysfunction.

5. Significant medical illness other than asthma.

6. History of respiratory tract infection within the previous 6 weeks.

7. History of a significant asthma exacerbation in the previous 6 weeks.

8. History of life-threatening asthma requiring treatment with intubation and mechanical ventilation within the past 10 years.

9. Hyposensitization therapy other than an established maintenance regimen.

10. Inability, in the opinion of the investigator or clinical coordinator, to coordinate use of a dry-powder or metered-dose inhaler.

11. Pregnancy or lactation. If potentially able to bear children, not using an acceptable form of birth control (see ACRN MOP).

12. Inability, as evidenced through biological quality control testing, to correctly use an electronic peak flow meter for recording peak flow measurements.

D. Exclusion Criteria for Continuation (Visit 4)

1. Significant exacerbation of asthma during the placebo treatment period (See Section XIII for definition of significant exacerbation and guidelines for treatment).

2. Inability to comply with regular use of study medications (missed >15% of doses in last 2 weeks of placebo treatment period, as reflected by Doser + eDEM).

3. Failure to record peak flow measurements and symptoms in diary on average >15% of required time during the last 2 weeks of the placebo treatment period.

4. Any changes with regard to any of the exclusion criteria for Visit 1 (See Section VI C above).

5. Use of an average of >56 puffs of albuterol per week during last 4 weeks of placebo treatment period (Week 6 -10).

E. Criteria for Assigning Treatment Failure Status during Double-Blind Treatment Periods

1. Requirement for corticosteroids (oral or parenteral) for treatment of asthma deterioration.

2. A pre-bronchodilator FEV_1 < 80% of that established at Visit 4.

3. More than one Emergency Department or Urgent Care Visit for treatment of asthma exacerbation.

4. Hospitalization for treatment of asthma attack.

5. Physician clinical judgment for safety reasons.

F. Criteria for Asthma Exacerbation

Increased cough, chest tightness, or wheezing in association with one or more of the following:

1. Any treatment failure criteria.

2. Rescue ß-agonist use ≥ 8 puffs per 24 hours over baseline use (see section XIIIC) for a period of 48 hours.

3. Rescue ß-agonist use ≥ 16 puffs per 24 hours for a period of 48 hours.

4. PEF < 65% of reference levels despite 60 minutes of rescue treatment.

5. Symptoms despite 60 minutes of rescue treatment.

VII. OUTCOME VARIABLES

The principal outcome to be assessed in this study is the change in pre-bronchodilator FEV_1 over the 8-week double-blind treatment period during which subjects receive inhaled corticosteroid, comparing the change in FEV_1 in the group of smokers with that seen in the non-smokers. Other important secondary outcomes during this period are AM- and PM-PEF, PC_{20}Methacholine, daily symptom scores, and quality of life measures. To determine if differences between the smoking and non-smoking groups reflect differences in the character of inflammation, we will examine induced sputum samples for total and differential cell counts, and for concentrations of eosinophil cationic protein and tryptase, as markers of airway inflammation, eosinophil activation, and mast cell activation, respectively. Changes observed during the treatment period will be compared with changes observed during the placebo run-in period.

A secondary comparison will consist of the leukotriene receptor antagonist on the same variables described above, comparing the non-smoking asthmatics with those who smoke. An additional comparison will consist of the effect of the inhaled corticosteroid versus the effect of the leukotriene receptor antagonist, primarily with respect to the change in FEV_1.

VIII. STATISTICAL DESIGN AND ANALYSIS

A. Sample Size and Effect Size Calculation

A total of 48 smokers and 48 non-smokers (8 smokers and 8 non-smokers per Clinical Center) are necessary for this study. With a two-sided, 0.05 significance level test and a 10% drop-out rate, the sample size of 48 smokers and 48 non-smokers provides (1) greater than 90% statistical power for detecting a 10% improvement in FEV_1 in non-smokers vs. a 5% improvement in FEV_1 in smokers when inhaled corticosteroid is administered, and (2) 80% statistical power for detecting an 8% improvement in FEV_1 in non-smokers vs. a 4% improvement in FEV_1 in smokers when a leukotriene receptor antagonist is administered.

Using the data from our own ACRN DICE (Dose of Inhaled Corticosteroids with Equisystemic Effects) and MICE (Measuring Inhaled Corticosteroid Efficacy) trials, we have reliable estimates of the standard deviation for the improvement of FEV_1 after inhaled corticosteroid administration. The standard deviation for this improvement averaged 0.20 L across the various inhaled corticosteroid treatment groups. A 10% improvement in FEV_1 corresponds approximately to a 0.3L improvement, so the standardized effect in the non-smoking group would be 1.5 standard deviations. A 5% improvement in FEV_1 corresponds approximately to a 0.15L improvement, so the standardized effect in the smoking group would be 0.75 standard deviations.

However, the expected improvements due to the leukotriene receptor antagonist are smaller, namely 0.24L in the non-smoking group and 0.12L in the smoking group. Assuming that the standard deviation for the change in FEV_1 from leukotriene receptor antagonist administration is similar to that seen with inhaled corticosteroid administration, the improvements due to leukotriene receptor antagonist administration correspond to 1.2 standard deviations in the non-smoking group and 0.6 standard deviations in the smoking group. The expected mean improvements in FEV_1 are illustrated in the following table:

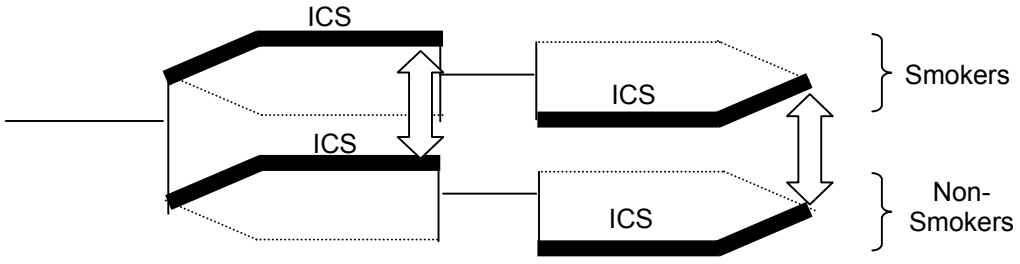
	ICS	**LTRA**
Smokers	0.15 L	0.12 L
Non-smokers	0.30 L	0.24 L

Applying the usual sample size formula for a two-sample t test with 80% statistical power, the required sample size for the primary comparison of smokers and non-smokers with respect to FEV_1 improvement from ICS administration is 32 subjects per group. The sample size of 48 smokers and 48 non-smokers actually yields statistical power greater than 90% for this primary comparison. The required sample size with 80% statistical power for the secondary comparison of smokers and non-smokers, with respect to FEV_1 improvement from LTRA administration, is 48 subjects per group. Thus, the target sample size is 48 smokers and 48 non-smokers for this trial.

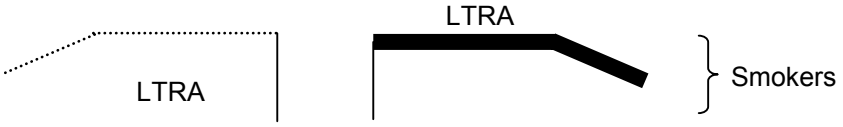
Given this target sample size of 48 smokers and 48 non-smokers, there is 80% statistical power for detecting a difference of 0.09L in FEV_1 between the inhaled corticosteroid and the leukotriene receptor antagonist. This effect size of 0.09L holds for the non-smoking group as well as the smoking group.

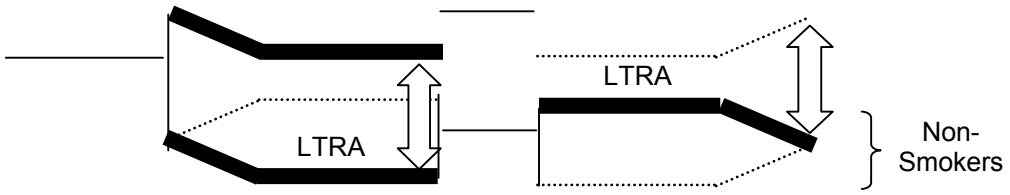
B. Statistical Analysis

The primary question to be addressed by this study is whether the change in pre-bronchodilator FEV_1 between the beginning and the end of the 8-week double-blind treatment period, during which subjects receive inhaled corticosteroid, differs between the group of asthmatic subjects who smoke and those who do not smoke.



A secondary question is whether the two groups differ in their response to a leukotriene receptor antagonist.





Although we are interested in whether the response to the leukotriene receptor antagonist differs from the response to the inhaled corticosteroid, this is really a third-order question, that becomes interesting only if there are significant differences between groups in the response to ICS and LTRA.

Other variables to be analyzed include AM- and PM-PEF, PC_{20}Methacholine, markers of inflammation from induced sputum, asthma symptom scores, quality of life measures, the number of occasions and actuations of rescue albuterol, and episodes of adverse asthma control.

The baseline information will be obtained from the end of the run-in. All outcome measures and demographic measures will be summarized at baseline as means and standard deviations for continuous variables (e.g., FEV_1, PC_{20}) or frequencies and percents for categorical variables (e.g., gender, race).

The primary outcome variable, FEV_1, will be measured at the beginning, middle, and end of each 8-week treatment period. The change over the treatment period will be analyzed using a mixed effects linear model.

The expected value for the change from baseline in FEV_1 for each period within each sequence and smoking category can be expressed as:

	Sequence	**Period 1**	**Period 2**
Smokers	ICS-LTRA	$\mu_I^{(s)} + \delta^{(s)} + \gamma^{(s)}$	$\mu_L^{(s)} + \delta^{(s)} - \gamma^{(s)}$
	LTRA-ICS	$\mu_L^{(s)} - \delta^{(s)} + \gamma^{(s)}$	$\mu_I^{(s)} - \delta^{(s)} - \gamma^{(s)}$
Non-smokers	ICS-LTRA	$\mu_I^{(ns)} + \delta^{(ns)} + \gamma^{(ns)}$	$\mu_L^{(ns)} + \delta^{(ns)} - \gamma^{(ns)}$
	LTRA-ICS	$\mu_L^{(ns)} - \delta^{(ns)} + \gamma^{(ns)}$	$\mu_I^{(ns)} - \delta^{(ns)} - \gamma^{(ns)}$

where
μ_I is the mean effect for ICS
μ_L is the mean effect for LTRA
δ is the nuisance parameter for the sequence effect
γ is the nuisance parameter for the period effect.

The comparison between smokers and non-smokers for each of the treatments ($\mu_I^{(ns)} - \mu_I^{(s)}$ and $\mu_L^{(ns)} - \mu_L^{(s)}$) will be obtained by testing the appropriate contrast of the model parameters. The model parameters will be estimated using PROC MIXED in SAS version 8.1. The model can easily be adjusted for other covariates, such as age or clinical center.

Outcome variables that are measured daily from the patient diary cards (e.g., PEF, symptom scores, ß-agonist rescue) will be averaged on a weekly basis, and analyzed using the same model framework.

Although urinary cotinine levels will be determined, these will be used to characterize smokers and non-smokers, and not as inclusion criteria. Subjects will be classified as "smokers" and "non-smokers" based on their self-identification, rather than on cotinine levels, and data will be analyzed by intent-to-treat based on this classification. However, if there are significant discrepancies between self-reported smoking status and cotinine levels, a secondary analysis will be performed with data from these "outliers" excluded.

In summary, the mixed-effects linear model will be applied to the following response variables and comparisons (in the order of priority):

1. Improvement in pre-bronchodilator FEV_1 due to inhaled corticosteroid between smokers and non-smokers
2. Improvement in pre-bronchodilator FEV_1 due to leukotriene receptor antagonist between smokers and non-smokers
3. Improvement in AM-PEF, PM-PEF, PC_{20} methacholine, markers of inflammation from induced sputum, asthma symptom scores, quality-of-life scores, and rescue albuterol use due to inhaled corticosteroid between smokers and non-smokers
4. Improvement in AM-PEF, PM-PEF, PC_{20} methacholine, markers of inflammation from induced sputum, asthma symptom scores, quality-of-life scores, and rescue albuterol use due to leukotriene receptor antagonist between smokers and non-smokers
5. Improvement in pre-bronchodilator FEV_1 between inhaled corticosteroid and leukotriene receptor antagonist in smokers and in non-smokers
6. Improvement in AM-PEF, PM-PEF, PC_{20} methacholine, markers of inflammation from induced sputum, asthma symptom scores, quality-of-life scores, and rescue albuterol use between inhaled corticosteroid and leukotriene receptor antagonist in smokers and in non-smokers

IX. DATA COLLECTION AND DATA MANAGEMENT

A. Data Recording

Each center will have a computer configuration that includes an X-terminal, a postscript printer, and a modem. This will give each center the capability of logging directly into the data coordinating center (DCC) computing system over the Internet with the modem as a backup if the connection is not possible. Though this set-up is installed primarily to allow for distributed data entry into a centralized database on the ACRN project server at the DCC, menu options will also include sending e-mail, downloading study documents such as forms and reports, and viewing a calendar of ACRN events. A sophisticated security system will limit access to qualified personnel and prevent corruption of the study database.

All data including the informed consent, history, physical examination, results of allergy skin testing, vital signs, results of pregnancy tests, adverse events, confirmation of medication dispensation, methacholine challenge testing, and quality of life testing will be recorded on forms prepared by the ACRN DCC. Once the data collection forms have been filled out and reviewed, the Clinic Coordinator will log into the DCC computer system and enter the data. The advantage of this distributed data entry system is that the Clinic Coordinators will review the data a second time as they are entering it, which serves as another level of quality control. Forms will then be forwarded to the DCC for the second data entry and filing. The DCC will be responsible for identifying problem data and resolving inconsistencies.

Results from pulmonary function tests and the electronic peak flow meter will be transmitted electronically to the DCC.

All data will be stored and analyzed at the DCC.

B. Matching

The randomization scheme will require that smoking and non-smoking individuals be "matched" prior to the run-in period. A smoking and non-smoking individual are matched if they are the same gender and in the same FEV_1 category (70-80% predicted or 80-90% predicted). All subjects will be matched within a 10 year age window; that is, a 28 year old subject will be matched to a subject between age 18 and 38 years. A 50 year old subject must be matched with a subject 40 years and above. Every attempt will be made to match subjects within a center, but if no match occurs within 4 weeks of Visit 0, subjects will be eligible to match across centers. To the extent possible, clinical personnel will be blinded with respect to information regarding which subjects are paired.

C. Randomization

When a subject at a particular center is deemed eligible for the study, the Clinic Coordinator will log into the ACRN network server and indicate to the system that a subject requires randomization. If the subject's paired member already has been randomized to a sequence, then this subject will be assigned to the same sequence as its paired partner. Otherwise, this subject will be randomized to one of the two sequences. After entering the pertinent information with respect to clinical center and eligibility criteria, the Clinic Coordinator will be asked to verify that all of the information has been reviewed carefully and the subject is eligible. If so, the Clinic Coordinator will be given a packet number, from which all medication for that subject will be dispensed. In order to maintain security of the randomization schedules, the data manager of the DCC will automatically receive a notice from the ACRN network server that a subject has been randomized. If no follow-up information is forthcoming on such a subject, the data manager will contact the Clinic Coordinators concerning the status of the subject. The randomization scheme will be stratified according to center because differences among clinical centers typically yield a large amount of variability.

D. Drug Supplies

Drug supplies for this study will include an inhaled corticosteroid (beclomethasone dipropionate HFA), a leukotriene modifier (e.g. montelukast or zafirlukast), albuterol sulfate, and placebos (tablets and dry powder inhalers). The ACRN will work with contractors and coordinate with the DCC to ensure proper blinding and coding of drugs.

E. Masking

Careful procedures are required in order to maintain the triple-masking of the study participants, clinical center personnel, and DCC personnel as to whether individual subjects are taking inhaled corticosteroid, leukotriene receptor antagonist, or placebo. Treatment medication for each subject will be packaged together and labeled with a unique number. The contents of the packages will be known only to limited personnel at the DCC. These packages, and MDIs for the run-in and wash-out will be delivered to the Clinic Coordinators. Triple-masking, i.e., masking of the DCC personnel in addition to the study participants and clinical center personnel, will be employed so that the statistical analyses are not biased by preconceived notions. Until the time of manuscript preparation, DCC personnel will identify the randomized groups as A and B, and only limited personnel within the DCC will know the identity of A and B.

In order to decrease the likelihood of incorrect drug distribution, each coded package designated for a study participant will have a sheet of removable labels attached to it. When the Clinic Coordinator retrieves a canister for the study participant, he/she will remove one of the labels and attach it to the data collection form prior to mailing the form to the DCC. The Clinic Coordinator will initial across the label to indicate that he/she checked to make sure the appropriate canister was distributed to the participant.

F. Adherence and Monitoring

In order to determine subject adherence with the inhaled corticosteroid placebo and the leukotriene receptor antagonist placebo during the single-blind placebo treatment period, information recorded on subjects' diary cards regarding number of puffs and the number of pills taken each day will be reviewed at designated visits. This information will be compared against PEF measurements obtained from the electronic peak flow device. Limitations of this mechanism for monitoring adherence are accuracy of the subject's recall and honesty since the timing of dosing cannot be determined.

During the double-blind treatment periods, adherence will be monitored using the Doser™ for the MDI, and the eDEM Electronic Drug Exposure Monitor™ for the LTRA or placebo.

G. Special Study Techniques

No techniques or procedures new to the ACRN are proposed for this study. Standard methods have been developed and described in the Manual of Procedures for spirometry,

methacholine challenge, measurement of maximum bronchodilator response, sputum induction and analysis, asthma diary instruction, skin testing, and quality of life assessment. The ACRN also has experience with the methods intended to monitor and insure adherence, and for measuring peak flow. All personnel are certified in each procedure, and new personnel will require certification before participating.

X. RECRUITMENT

Harvard Clinical Center/Boston

The Asthma Clinical Research Center at the Brigham & Women's Hospital utilizes three primary resources for identifying and recruiting potential subjects as described below.

Research Patient Database: The Asthma Clinical Research Center at the Brigham and Women's Hospital has a database of over 1,500 asthmatics who have expressed interest in participating in research. All of these subjects have completed questionnaires regarding their asthma and medication use. In addition, many have undergone physiological screening. The database is screened based on entry criteria, and subjects are contacted in a manner approved by the IRB to ascertain their interest in participation.

Advertisements: We utilize IRB-approved radio and newspaper advertisements to inform potential subjects of our studies and solicit participation. In addition, we use posters in selected locations.

Asthma Patient Lists: Following IRB guidelines, the center has permission to contact patients with a diagnosis of asthma to ascertain these patients' possible interest in participating in asthma studies. Lists generated at the Brigham & Women's Hospital contain over 5,000 such patients. In the past, we have also used patient lists from the Harvard Pilgrim Health Care HMO. The latter list can be screened by medication use to preliminarily identify patients with specific patterns of medication use.

National Jewish Medical and Research Center/Denver

Research subject recruitment has been very successful for all types of asthma patients at the National Jewish Medical and Research Center. The total number of subjects, with one-half being female and one-third minority population, will come from the following areas.

NATIONAL JEWISH OUTPATIENT CLINIC

The adult clinic saw 1,079 new asthmatic patients over the last year with 503 being from the Denver metropolitan area. Another 335 from the Denver area were seen in follow-up. The severity of asthma varies among these patients, but at least 15% are in the mild category. The pediatric clinic saw 490 new asthmatic children with 352 being from the Denver metropolitan area. Again these patients were of varying severity, but about 10-15% are in the mild category. Ninety-seven additional children were seen in follow-up. National Jewish Center changed markedly over the last decade. We have evolved from a primary inpatient facility with a small clinic to a very active outpatient service. Thus, we are seeing many more asthmatic patients of all degrees of severity.

NATIONAL JEWISH ASTHMA RESEARCH POOL

There are over 600 asthma patients (not followed in the NJC outpatient clinic) who have participated in our research studies. Many of these subjects have been through various medication studies and bronchoscopies with lavage/biopsies. Their FEV_1 range from 30-110% of predicted.

DENVER HEALTH MEDICAL CENTER

Dr. James Fisher, Head of Pulmonary Medicine, is supporting our efforts by helping us to recruit from the asthmatic patient population at Denver General. This is a large county hospital with a patient population comprised mainly of Hispanic and African-American people.

DENVER VETERANS ADMINISTRATION HOSPITAL

Dr. Clifford W. Zwillich, Head of Medicine, will support this grant. The V.A. hospital has a large outpatient clinic of patients with asthma.

DENVER KAISER PERMANENTE HMO

Dr. Timothy Collins is the Director of Pulmonary Medicine and Dr. John Williams is the Director of Allergy at Kaiser. Drs. Collins and Williams have been actively involved in supporting research at NJ in the past by referring patients to us. Their groups will continue to play an active role.

University of Wisconsin/Madison

The Asthma/Allergy Clinical Research Program of the University of Wisconsin maintains an ongoing computer database of potential subjects with asthma and allergic diseases who are interested in future research participation. These individuals have been screened and/or participated in previous clinical studies with our unit. Their names have been generated in response to extensive newspaper advertisements, physician referrals, radio advertisement campaigns, community health screening events, and by email communications to the entire student enrollment of the University of Wisconsin (approximately 40,000 students); all advertisement modalities have been approved by the Human Subjects Committee. Approximately 85% of the subjects in this database have "mild to moderate persistent asthma" and are, therefore, eligible for ACRN protocols. The following patient data is maintained: birth date, gender, ethnic background, duration of asthma, childbearing and contraceptive use status, smoking history, atopic status (including allergy skin testing results if previously performed), pulmonary function tests, concurrent medical history, asthma and non-asthma medication use, methacholine test results, and exercise challenge test results (if previously performed). When additional subjects are needed, referrals from physicians in the University of Wisconsin Clinics and Physicians Plus network are solicited.

Even though this database serves as the foundation of our recruitment efforts, the Madison ACRN site has utilized some additional approaches to target minority recruitment. We have utilized a marketing expert to coordinate and oversee our overall efforts in recruiting and retaining minorities. He is uniquely qualified for this task due to his combined professional and personal background (he is an ethnic minority, has a long history of asthma, and has participated in previous asthma studies with our unit). As a result of his efforts, we have

advertised widely in newspapers and other publications that target ethnic minorities, established contacts with various ethnic community, university, church, and business groups, and conducted community-based asthma programs. We will continue our annual asthma screening services for all of the incoming University of Wisconsin freshmen athletic teams, which has been highly successful. Historically, retention of students in our asthma studies has been excellent, especially if contacted early upon arrival to the campus. These individuals discover that study participation serves as an ongoing source of quality medical management for their asthma. In addition, we will utilize published examples of successful retention strategies such as frequent payment of subject honoraria as study landmarks are achieved and study participant group social events. Study visits will be carefully planned and scheduled to avoid exam-time and university calendar breaks.

Harlem Hospital Center, Columbia University, New York City

The ACRN clinical center at Harlem Hospital Center draws study participants from four sources, including the Chest Clinic, the Emergency Department, the general community, and through advertising and outreach efforts. We advertise through local radio stations, newspapers, and newsletters of local churches and other community based organizations. In addition, we disseminate information about inclusion criteria for specific studies through ongoing outreach activities with volunteers in the AHA! (Asthmatics Helping Asthmatics) support and advocacy group, and through educational efforts in the community, including a series of asthma educational workshops.

The Chest Clinic, an outpatient pulmonary clinic in Harlem Hospital Center, sees a diverse group of patients with asthma. Patients learn about research at the Lung Center and about opportunities for participation in clinical trials, during their clinic visits.

The Harlem Hospital Center Emergency Department (ED) sees an average of eight adult patients per day for asthma. Through the REACH (Reducing Emergency Asthma Care in Harlem) project, we have been recruiting study participants at the ED. We have successfully recruited and interviewed 380 patients from the ED for that project, and most are currently being followed. One-third to one-half of REACH participants may be classified with mild intermittent or mild persistent asthma (self-reported symptoms, by NAEPP guidelines criteria).

Responses to inquiries about participation in research studies are answered by a dedicated phone line that is manned during business hours and answered by voicemail at all other times. A research assistant responds to each inquiry immediately, using a screening instrument that inquires about potential respondents' contact information, demographics, smoking history, and medical history. Our database includes 1,600 individuals with physician-diagnosed asthma.

Thomas Jefferson Medical College/Philadelphia

Patients are recruited for clinical trials at the Jefferson Center through two primary mechanisms: (1) local advertising and (2) identification in the asthma patient registry (database). Local advertising takes advantage of the printed as well as the audio-visual media. Printed media include posters placed in public information centers of local colleges

and universities as well as brochures sent to selected physicians in the Philadelphia area. Printed advertising is placed in local neighborhood newspapers and occasionally in the *Philadelphia Inquirer*. Audio-visual media advertisements are also placed in public service announcements on television and radio. All advertising in the printed and audio-visual media has prior approval of the Institutional Review Board.

The Jefferson patient registry (database) has been maintained since 1992 and currently contains 3,100 patients. The patient registry infrastructure includes a computer network linking those divisions of the institution that serve significant numbers of asthmatic patients (pulmonary medicine, family medicine, pediatric and adult allergy, and general internal medicine). Personal computers in each outpatient clinic site are linked to a dedicated file server located in the clinical research offices of the Pulmonary Division. The network operates on Novell Netware 3.22, and the database application is a customized version of Approach for Windows. The database provides a graphic interface for data entry. Fields for demographic information, smoking history, allergic history, medication used, pulmonary function tests, other laboratory tests, and other diagnoses are provided. Designated personnel are able to access the database and perform searches based on any field or combination of fields to define subsets of patients who qualify for particular research studies. The data coordinator is responsible for maintaining the database, assuring its accuracy, and keeping it current. It is estimated that 300-400 new asthmatic patients are seen each year, while a smaller number become inactive due to relocation, change of health care provider, etc. Once identified in the database, patients potentially eligible for a specific study are contacted by the nurse coordinator who explains the study and ascertains the patient's interest. If interested, the patient is seen in the clinical research laboratories where more detailed evaluations are made.

University of California/San Francisco

The approach to recruiting subjects with mild asthma for research studies at the San Francisco Center relies heavily on community advertising and on maintaining a database of subjects who have participated in previous studies, who have come for a "characterization visit", or who have expressed interest in participating. Advertisements are placed in editions of the *San Francisco Chronicle*, the *San Francisco Examiner*, the *Bay Guardian*, and in small neighborhood and college campus newspapers. We post numerous fliers on bulletin boards on the UCSF campus, in community health centers, at campuses of local colleges and universities in the Bay Area, and we broadcast advertisements on local radio stations. We make frequent presentations to different physician groups on and off campus describing our research studies and the enrollment criteria for future studies. Responses to these advertisements are made to a dedicated telephone number equipped with voice mail. A dedicated recruiter, Lila Glogowsky, either responds herself or directs other staff (technicians and clinical coordinators) to respond to each inquiry to obtain basic information about the subject's demographics and about the severity, duration, required treatment, and frequency of symptoms of asthma. Subjects who appear to meet entry criteria for a study are then referred to a study coordinator, who then contacts the subject to schedule a "characterization visit" in which details of the medical history and medication use are obtained, and spirometry (before and after albuterol administration), and skin testing is performed. To date, over 3,000 subjects have been screened for the database. We have

met the goals for recruitment of women and of members of ethnic minorities in all studies so far.

XI. RISKS/BENEFITS

This study compares two approved usage strategies for 2 currently approved pharmaceutical products. Risks of short-term inhaled corticosteroid use include oral candidiasis and dyphonia. An extremely remote risk of treatment with a leukotriene receptor antagonist is the development of Churg-Strauss syndrome. Most cases have occured in patients with moderate or severe asthma requiring inhaled or oral corticosteroids. Subjects for this study will have less-severe asthma, and will have been on ß-agonists only at entry. There are two periods during the study when subjects will receive placebo as their study drug: during the run-in and single blind placebo periods (Weeks 0-10), and during the wash-out period (Weeks 18-24). To ensure the safety of individuals whose asthma worsens during this period, specific criteria have been developed for assigning “asthma exacerbation” or "treatment failure" status, and for initiating appropriate asthma therapy. All subjects may continue to use rescue albuterol throughout the study. There will be no direct benefit to individual subjects participating in this study. The results of this study may be of potential benefit to the entire group of patients with asthma, especially those who smoke, as it may lead to a better definition of guidelines for asthma therapy.

XII. ANTICIPATED RESULTS AND SIGNIFICANCE

We anticipate that this study will help to guide the development of a treatment algorithm for people with asthma who smoke cigarettes. Assuming that we are successful at excluding those subjects who have COPD (by limiting enrollment to those ≤ 50 years old, with a 2-12 pack-year smoking history, reversibility, and normal D_LCO), we will attempt to confirm or disprove the *post hoc* observations from 2 studies suggesting that the response to inhaled corticosteroids is significantly blunted in asthmatics who smoke. Many people with asthma continue to smoke despite universally-accepted recommendations against smoking. If smoking interferes with asthma therapy, that information will be important for these patients, and for the clinicians who care for them.

If, in this carefully controlled, prospective study there is no difference in the response to inhaled corticosteroid between the group of asthmatic subjects who smoke and those who do not, then the only major treatment specified uniquely for asthmatics who smoke would be that directed at facilitating smoking cessation.

It is possible that smokers will have a better response than non-smokers. However, this is unlikely, and will not be considered further.

If, on the other hand, the improvement in FEV_1 that we expect to see in non-smokers after 8 weeks of inhaled corticosteroids is either absent or attenuated in smokers, this will be very important information. We would interpret this as indicating either that smoking modulates the response to corticosteroids, or that smokers with "reversible airway disease" have something other than asthma. The latter seems unlikely, given the fairly rigid entry criteria designed to exclude those with COPD. It is possible that other outcomes may provide clues regarding the mechanism underlying the differences in response. For example, we may find different patterns of inflammatory cells and mediators at baseline, or different patterns of response of these markers of inflammation to treatment. Smoking may induce specific enzymes (e.g., P450 system) or mutations, and thereby change the absorption or metabolism of corticosteroids, contributing to a blunted corticosteroid response. This study will not identify such biochemical events, but would provide the basis for designing such follow-up studies. Finally, it is possible that smokers as a group may adhere less well to a regimen of daily inhaled corticosteroids. Thus, measures to maximize and monitor adherence will be important, to provide some reassurance that a difference in outcomes does not reflect a difference in medication use.

If the study results support our primary hypothesis, i.e., that subjects who smoke demonstrate a lesser response to inhaled corticosteroids than do subjects who do not smoke, it will be important to examine the responses to a leukotriene receptor antagonist. A blunted response to this class of therapy also will suggest a fundamental difference in the behavior of the airway in asthmatics who smoke: acute reversibility in response to albuterol, but failure to respond to two classes of agents that act by very different mechanisms. If there is no difference between smokers and non-smokers in the response to a leukotriene receptor antagonist this will suggest that the leukotriene pathway is involved, and that smoking does not impact either the synthesis of leukotrienes or the action of leukotrienes at their receptor. Failure of these subjects to respond to corticosteroids, which have little or no ability to reduce synthesis of leukotrienes, would suggest that a leukotriene-predominant mechanism might be involved, though we will not be able to determine if smoking alters synthesis or metabolism of leukotrienes.

XIII. ADVERSE EVENTS AND TREATMENT FAILURES

A. Definition

An adverse event shall be defined as any detrimental change in the subject's condition, whether or not it is related to an exacerbation of asthma or to another unrelated illness. Adverse events related to asthma exacerbations will be managed according to rescue algorithms outlined below in Section C.

B. Adverse Events Unrelated to Asthma

Adverse events due to intercurrent illnesses other than asthma may be grounds for withdrawal if the illness is considered significant by the investigator or if the subject is no longer able to effectively participate in the study. Subjects experiencing minor intercurrent

illnesses may continue in the study provided that the nature, severity, and duration of the illness are recorded and that any unscheduled medications required to treat the illness are also recorded. Examples of minor intercurrent illnesses include acute rhinitis, sinusitis, upper respiratory infections, urinary tract infections, and gastroenteritis. Medications are allowed for treatment of these conditions in accordance with the judgment of the responsible physician. Documentation of an adverse event unrelated to asthma will be recorded on an Adverse Event Report Form and will include the following information:

- Description of the illness
- Dates of illness
- Treatment of illness (medications, doses, dates)
- Whether hospitalization or emergency treatment was required
- Treatment outcome

C. Adverse Events Related to Asthma

1. Definition

During the course of the study, subjects may experience an exacerbation of asthma. An exacerbation of asthma is characterized by an increase in symptoms of cough, chest tightness, and wheezing and it is generally associated with a fall in PEF. It is recognized that the PEF may be improved by use of a bronchodilator and that increased bronchodilator use may, in this case, be more reflective of the exacerbation than PEF. An increase in symptoms may be brief and self-limited, or it may be of sufficient severity as to warrant documentation as an asthma exacerbation. Although <u>any</u> increase in symptoms or changes in PEF should be carefully monitored by the subject, the clinic coordinator, and the physician, alterations in asthma stability will be considered as constituting an asthma exacerbation when PEF does not increase to > 65% of reference levels, or symptoms are not satisfactorily relieved, after the first 60 minutes of rescue beta agonist (albuterol) use. Albuterol may be used at a dose of 2-4 puffs every 20 minutes during this one hour time period. The reference point for PEF comparisons will be as follows:

During run-in period (Weeks 1-2):
Predicted PEF obtained from spirometry software at Visit 1

During single-blind placebo period (Weeks 2-10):
Mean value of AM prebronchodilator PEF recorded during the last week of the run-in (Week 2)

During blinded treatment and wash-out (Weeks 10-18, 18-24, 24-32):
Mean value of AM prebronchodilator PEF recorded during the last 2 weeks of the placebo period (Weeks 9 and 10)

In addition, an asthma exacerbation will be identified if subjects have a significant increase in symptoms associated with either:

An increase in "as needed" ß-agonist use of ≥ 8 puffs per 24 hours over baseline use for a period of 48 hours,

Baseline defined during run-in period: Average daily use during the first week of the run-in period (Week 1).

Baseline defined during placebo period: Average daily use during last two weeks of the run-in period (Weeks 1 and 2).

Baseline defined during double-blind period: Average daily use during the last two weeks of the placebo period (Weeks 9 and 10).

or

Use of ≥16 puffs of "as needed" ß-agonist per 24 hours for a period of 48 hours.

Once an asthma exacerbation has occurred, the subject should contact the clinic coordinator and/or be evaluated at the study site or the nearest medical emergency facility as quickly as possible.

Since less-significant changes in symptoms and/or PEF may precede more severe alterations in asthma stability, a series of rescue algorithms has been developed to address the various clinical presentations that may occur. Once any of these rescue interventions leads to the administration of corticosteroids, the subject will also be considered to have developed an asthma exacerbation. In addition, if in the opinion of the treating physician, corticosteroid therapy is warranted regardless of any antecedent measurements of pulmonary function (PEF, FEV_1, etc.), value for symptom score, or frequency of rescue beta agonist use, the subject will be considered to have developed a asthma exacerbation.

Any subjects who attain treatment failure status will be considered to have had an asthma exacerbation.

The time at which an asthma exacerbation develops in relationship to the schedule of the SMOG protocol will affect the manner in which future clinic visits, medication adjustments, and diagnostic studies are scheduled or performed.

Subjects developing asthma exacerbations during the run-in or single-blind treatment periods will be removed from the study. Once the exacerbation has resolved, the subject may be considered for re-enrollment, starting again at Visit 1 once they meet all Visit 1 entry criteria.

Asthma exacerbations which occur following randomization (double-blind treatment phase) will be managed according to the following rescue algorithms. During medical management of the exacerbation, trial medication will be continued, unless the treating physician considers it appropriate to suspend such therapy until the exacerbation resolves. Reinstitution of trial medications will occur when the exacerbation has resolved at the

discretion of the investigator. A record of all medications, dosages, and frequency of occurrence will be kept during exacerbations. Additional visits and procedures will be scheduled as needed.

2. Rescue Algorithms

Rescue algorithms will be applied in cases where an exacerbation, as defined in Section XIIIC, fails to resolve or PEF is not improved to > 65% of reference level within 48 hours after increasing as needed albuterol use. Rescue algorithms are based on recommendations from the NAEP Guidelines for the Diagnosis and Management of Asthma (55). Albuterol and oral prednisone are the principal medications for rescue management. Subjects will be instructed in their use for home management and supplies of both will be provided throughout the study. For severe acute episodes of asthma, treatment will be administered according to the best medical judgment of the treating physician.

a) Home Care

- Asthma exacerbations will be recognized by an increase in symptoms and by a corresponding drop in PEF below reference level. Subjects will be educated to recognize exacerbations as early as possible to facilitate prompt treatment and to lessen morbidity.

- Subjects who recognize increased symptoms and/or a fall in PEF ≤65% reference level will use albuterol by MDI, 2-4 puffs every 20 minutes up to 60 minutes if needed, and then every 4 hours, or less, if needed. Subjects will be instructed to use the as needed inhaler for treatment.

- If the PEF does not increase to > 65% reference level or if symptoms are not improved after the first 60 minutes of albuterol therapy, the subject should contact the investigator, their primary physician or seek care in the emergency department.

- Failure of albuterol to control or maintain PEF > 65% reference level may necessitate the use of corticosteroids (see below).

b) Physician's Office or Emergency Room Treatment

- Subjects will be assessed by history, physical examination, and by physiological monitoring including spirometry or PEF. If the subject's PEF or FEV_1 are less than 25% predicted or if the subject shows evidence of altered mental status, cyanosis, labored breathing, or use of accessory muscles, sampling of arterial blood for respiratory gas analysis is indicated, with appropriate action taken depending on the results obtained.

- When treated in the physician's office or the hospital emergency room, subjects should initially be given albuterol by nebulization (0.5 cc of 0.5% solution) every 20 minutes over the first 60 minutes.

- If the PEF increases to > 65% reference level after the first 60 min, the subject can be discharged to continue treatment at home. Prednisone or open-label inhaled corticosteroids (fluticasone 200 μg/day) may be administered at the discretion of the physician to augment therapy.

- If symptoms persist and PEF remains < 65% reference level, nebulized albuterol should be continued as often as every hour and further treatment with oral or parenteral corticosteroids should be considered (prednisone, 60 mg orally; methylprednisolone, 60 mg iv bolus). Monitoring of PEF or spirometry should continue every hour. Within 4 hours of treatment, a decision should be made regarding subject disposition.

- If PEF increases to > 65% reference level within 4 hours, the subject can be discharged to continue treatment at home. Home treatment should include an 8-day course of prednisone followed by open-label inhaled corticosteroid treatment (see above).

- If PEF remains > 40% but < 65% reference level, an individualized decision should be made to hospitalize the subject for more aggressive therapy or to continue therapy at home with a course of prednisone followed by inhaled corticosteroids.

- If PEF is < 40% reference level after repeated albuterol treatments, the subject should be admitted to the hospital unless, in the physician's best judgment, alternative treatment could suffice.

b) Prednisone Treatment

In this protocol, prednisone will be used when, in the judgement of the investigator, acute exacerbations cannot be controlled by albuterol and inhaled corticosteroid therapy. Indications for prednisone therapy include the following:

- To achieve stable control of symptoms and optimize pulmonary function once treatment failure status is achieved.

- For follow-up management after discharge from the physician's office, emergency room, or hospital for an acute exacerbation.

The dose of prednisone used during an acute exacerbation shall consist of 60 mg as a single dose every day for 3 days, followed by a 10 mg/day taper over the next 5 days. The decision to initiate or to continue a course of prednisone beyond 8 days is left to the discretion of the physician.

D. Adjustment of Trial Medications During Asthma Exacerbations

Trial drugs will be continued during exacerbations unless the treating physician considers it appropriate to suspend such therapy until the exacerbation resolves. Reinstitution of trial medications may occur when the exacerbation has resolved at the discretion of the

investigator. A record of all medications, dosages, and frequency of occurrence will be kept during exacerbations.

E. Study Visits Following Asthma Exacerbations

If the subject receives steroids for an exacerbation, regular follow-up evaluations will continue according to the original protocol.

F. Criteria for Discontinuing Subjects Due to Asthma Exacerbations or Treatment Failure

Criteria for assigning Asthma Exacerbation or Treatment Failure Status are described in Section VI E,F**.** In either case, subjects will continue to participate in the data gathering aspects of the protocol until the time they would have completed the trial.

G. Adverse Events as Outcome Variables

During exacerbations, the following variables will be recorded and used as outcome variable measures:

- Hospitalization for asthma
- Emergency Department Visits for asthma
- Unscheduled physician/clinic visits for asthma
- Number of subjects having an exacerbation as defined by corticosteroid use
- Treatment failure

XIV. COST AND PAYMENT

Each subject will be paid an amount determined by his/her local center. For subjects who withdraw, payments will be pro-rated for the length of time they stayed in the study, but payments will not be made until the study would have been completed had the subject not withdrawn.

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