CLINICAL INVESTIGATION

STRATOS 1 and 2: considerations in clinical trial design for a fully human monoclonal antibody in severe asthma

New therapies are being developed to target proinflammatory mediators thought to be involved in the pathophysiology of severe asthma. Tralokinumab is an investigational fully human monoclonal antibody that specifically blocks binding of IL-13 to its receptors. Here, we describe the background leading to the design of two Phase III trials, STRATOS 1 and 2 (NCT02161757 and NCT02194699), which aim to provide confirmatory evidence of the efficacy and safety of tralokinumab in patients with asthma that is uncontrolled despite treatment with inhaled corticosteroids and long-acting β2-agonists. These trials will also confirm the validity of periostin and dipeptidyl peptidase-4, identified in a prior Phase IIb study (NCT01402986), as predictors of an enhanced response to tralokinumab.

Keywords: airway disease • anti-IL-13 monoclonal antibody • biomarker • DPP-4 • exacerbation • IL-13 • periostin • severe persistent asthma • tralokinumab • uncontrolled asthma

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Asthma is heterogeneous in its clinical presentation [1,2]. In a mild form, asthma is exercise-induced bronchospasm, with symptoms occurring only with vigorous exercise. Severe asthma involves persistent symptoms and frequent exacerbations, which are both life threatening and chronically disabling. Between these extremes, mild-to-moderate forms of the disease exist, requiring intermittent use of short-acting inhaled β2-agonists or long-acting inhaled β2-agonists (LABAs) for symptom relief alone or with regular inhaled corticosteroids (ICS).

Severe persistent asthma is defined by treatment with medium-to-high dose ICS and LABA or other add-on therapies (e.g., leukotriene inhibitors, theophylline, anti-IgE and oral corticosteroids [OCS]), or asthma that remains uncontrolled despite this treatment [2]. A proportion of patients with asthma, approximately 5–10%, have severe persistent disease [3]. The burden of disease, both on the patients and the healthcare system, increases with asthma severity. Challenges exist in managing severe persistent asthma when exacerbations reoccur despite the use of medium-to-high dose ICS and other controller therapy, including maintenance therapy with systemic OCS [4,5]. More effective therapies are urgently required for these patients.

An expanding understanding of the inflammatory mediators involved in asthma pathophysiology is now leading to the development of new agents, including biologics such as monoclonal antibodies, for the treatment of patients with severe asthma. The first such agent to be approved for therapy in patients with severe asthma was omalizumab, a fully human IgG_1 monoclonal anti-IgE antibody which causes a rapid decrease of free-circulating IgE, and hence is targeted to patients with severe asthma with a known allergen and high levels of IgE [6].

More recently, a wide assortment of other inflammatory mediator targets have been identified as likely to be involved in the pathophysiology of severe asthma [7]. Examples of these targets include IL-5, IL-4, IL-13, IL-9, OX40 ligand, thymic stromal lymphopoietin, IL-17, IL-23 and TNF-α.

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Characterizing the inflammatory cascade involved in the individual patient with asthma will become increasingly crucial in guiding the clinical use of these targeted therapies [8].

Extensive investigation has since shown that IL-13 plays an important role in mediating several of the key pathophysiologic processes characteristic of asthma. Specifically, IL-13 promotes airway hyper-responsiveness in humans and animal models [9,10], smooth-muscle proliferation *in vitro* and in animal *in vivo* models [11–14], as well as increased mucus production in human airway epithelial cells, *in vitro* and in animal models [10,14–16]. IL-13 is thought to be a key mediator of airway inflammation in asthma [17]. There is also some evidence to suggest that IL-13 may promote eosinophil accumulation and reduce migration into the lungs [18,19–21]. IL-13 expression is upregulated in peripheral blood, sputum, bronchial mucosa and airway smooth muscle in patients with asthma [22–27]. Furthermore, upregulation of IL-13 can occur even in patients receiving ICS therapy [28]. Due to these multiple effects, IL-13 has been identified as a promising target for drug therapy in patients with severe asthma [29,30].

Tralokinumab, a fully human monoclonal antibody of the IgG4λ subclass, was specifically engineered to have high affinity, specificity and potency for neutralizing human IL-13, by preventing binding of IL-13 to its receptors (IL-13Rα1 and IL-13Rα2) [31]. In a randomized Phase IIa study (NCT00873860), different doses of tralokinumab (150, 300 and 600 mg) were administered subcutaneously every 2 weeks (Q2W) for 12 weeks to 194 patients with moderate-to-severe asthma whose disease was uncontrolled despite the use of medium-to-high dose ICS and usually a LABA [32]. The primary end point, improvement in asthma control, measured by the Asthma Control Questionnaire (ACQ-6), was not met. There was a dose-related improvement in the forced expiratory volume in 1 s(FEV_1) with tralokinumab treatment, with significant benefit found with the highest dose (600 mg). Furthermore, while the majority of patients in this trial had undetectable sputum IL-13 levels, those with measurable levels of IL-13 in sputum had greater improvements in FEV_1 with tralokinumab treatment compared with those without measurable IL-13. Detection of quantifiable IL-13 protein or mRNA in the sputum and blood is challenging and, therefore, highly sensitive assays are required [23,26,28,33,34]. Further development of tralokinumab for severe asthma requires identification of predictive biomarkers for IL-13 pathway activation. Fractional exhaled nitric oxide (Fe_{NO}) and inducible nitric oxide synthase (iNOS) have been investigated as potential biomarkers. Whether variability in Fe_{NO} levels impact its utility as a potential predictive biomarker at patient level needs to be studied further [35,36]. Similarly, iNOS mRNA and protein levels are upregulated in response to IL-13, but there is no correlation between the mRNA and the protein levels, and iNOS is not secreted precluding its use as an easily measured serum biomarker [37].

In a randomized Phase IIb study (NCT01402986), tralokinumab treatment (300 mg, dosed Q2W for 52 weeks, or Q2W for 12 weeks and then every 4 weeks (Q4W) for 40 weeks) or placebo, administered subcutaneously was added to high-dose ICS-LABA in 452 patients. A significant reduction in exacerbations was not found in the overall population with any of the dosing regimens, but there was a significant improvement in FEV_1 in the Q2W treatment group. Exploratory *post-hoc* analyses showed that patients with post-bronchodilator reversibility at baseline, not receiving chronic OCS, and with a baseline periostin level above median at baseline, had significant improvements in exacerbations, FEV_1 and ACQ-6 with tralokinumab 300 mg Q2W. It was found in exploratory *post-hoc* analyses that dipeptidyl peptidase-4 (DPP-4) levels above median at baseline in certain patient subgroups was associated with significant improvements in exacerbations, FEV_1 and ACQ-6 [38]. Therefore, periostin and DPP-4 have emerged as promising potential biomarkers for IL-13 pathway activation. Periostin is a matricellular protein that belongs to the fasciclin family and is secreted by airway epithelial cells in response to IL-13 stimulation. Periostin may have a key role in the pathophysiology of asthma by promoting airway pathological remodeling in response to Th2 cytokines [39]. Although in a long-term multicenter cohort study, periostin alone was not predictive of either decline in FEV_1 over time or an increased risk of exacerbations, the combination of high periostin and high Fe_{NO} was a useful predictor of these poor outcomes [40]. DPP-4 is a multifunctional enzyme, found on nonciliated bronchial epithelial cells and T-lymphocytes. It is expressed at high levels on Th1 cells and may counter-regulate Th1-cytokine activity via its peptidase activity [41–44]. Both plasma levels and lymphocyte surface expression of DPP-4 were found to be elevated in patients with atopic asthma [41]. Both potential biomarkers, periostin and DPP-4, can be measured using a blood test [38].

The Phase IIa and IIb studies have demonstrated that tralokinumab treatment significantly improves lung function in patients with severe uncontrolled asthma already treated with ICS-LABA. In a Phase I study, pharmacokinetic evaluations of tralokinumab (administered as a single 300 mg subcutaneous dose) in 20 adolescents (aged 12–17 years) with asthma requiring daily use of controller medications confirmed that the same dose of tralokinumab is applicable as in adults

[45]. The ongoing tralokinumab Phase III studies, STRATOS 1 (NCT02161757) [46] and STRATOS 2 (NCT02194699) [47], aim to provide confirmatory evidence of the efficacy and safety of tralokinumab in patients with severe uncontrolled asthma, despite treatment with ICS-LABA. As adolescent patients are expected to respond similarly to adults, they will be included as part of the study population. The primary objective of these studies is to evaluate the effect of tralokinumab compared with placebo on annual asthma exacerbation rate (AER) in adults and adolescents with asthma that is inadequately controlled with ICS-LABA. The results from STRATOS 1 will be used to identify a target population with an enhanced response rate based on the presence of high serum periostin or DPP-4. STRATOS 2 aims to confirm that this biomarker-positive subpopulation demonstrates an enhanced response to tralokinumab.

Study designs

STRATOS 1 and STRATOS 2 are Phase III, multicenter, randomized, double-blind, parallel-group, placebo-controlled studies that will run concurrently and will be analyzed sequentially (Figure 1). For STRATOS 1, approximately 1140 patients will be randomized at more than 320 sites at centers in the USA, Argentina, Belgium, Brazil, Bulgaria, Colombia, Germany, Hungary, Republic of Korea, Peru, Poland, Slovakia and Spain. For STRATOS 2, approximately 770 patients will be randomized at more than 290 sites at centers in the USA, Canada, Chile, Italy, Japan, Mexico, Philippines, Russia, the Czech Republic, South Africa and the UK. The studies comprise a 4–6 week screening/run-in period, followed by a 52-week treatment period with study visits every 2 weeks. Follow-up visits at weeks 56 and 72 are intended to provide safety and immunogenicity information.

These studies are sponsored by AstraZeneca. Written informed consent will be obtained from all patients before initiation into the studies.

The studies will comply with the Declaration of Helsinki, the International Conference on Harmonization guidelines, applicable regulatory requirements and the AstraZeneca policy on Bioethics and Human Biological Samples.

Treatment/dosing

In STRATOS 1, 1140 patients will be randomized in a 2:1:2:1 ratio to receive tralokinumab 300 mg or placebo Q2W, or tralokinumab 300 mg or placebo Q4W (Figure 1A). Although efficacy was not established with the Q4W dosing regimen in the Phase IIb study, the Q4W dosing arm will be included in STRATOS 1 to confirm the dose–response relationship. In STRATOS 2, 770 patients will be randomized in a 1:1 ratio to receive tralokinumab 300 mg or placebo Q2W (Figure 1B). In both studies, tralokinumab 300 mg (150 mg/ml solution) or placebo will be administered via subcutaneous injections. Since tralokinumab and placebo are visually distinct, treatments will be handled and administered by unblinded team members who will not be involved in the management of study subjects. The first dose of tralokinumab or placebo will be administered at week 0. Patients randomized to the Q2W regimen will receive this dosing regimen up until week 50 (26 doses in total) with an end of treatment visit at week 52. Patients randomized to the Q4W regimen, will receive this dosing regimen up until week 48 (13 doses in total) with an end of treatment visit at week 52. Patients will be stratified at randomization by serum periostin level sampled during the run-in period (<16.44 ng/ml or ≥16.44 ng/ml; corresponding to the median value observed in previous studies), geographical region and age group (adults versus adolescents). All patients will continue to receive their prescribed ICS-LABA and any additional asthma controller medication, without change, throughout the study. Inclusion criteria allow additional maintenance asthma controller therapies (such as theophylline, long-acting inhaled anticholinergics and leukotriene blocking agents), according to standard practice of care, provided that the dose had been stable for 3 months prior to enrollment and that the dose will be maintained at a stable level throughout the study. Salbutamol, albuterol or levalbuterol will be used as rescue medications during the study period.

End points

Efficacy end points

The primary end point of both STRATOS 1 and 2 is the annual AER up to week 52. An asthma exacerbation is defined by either a worsening of asthma requiring the use of systemic corticosteroids for ≥3 days (or a single depo-injectable dose of corticosteroids), or an emergency room/urgent care visit due to asthma requiring systemic corticosteroids, or an inpatient hospitalization due to asthma. Additional systemic corticosteroid treatments, emergency room or urgent care visits requiring use of systemic corticosteroids, or inpatient hospitalization due to asthma occurring during an exacerbation will not be regarded as a new exacerbation unless it is preceded by ≥7 days in which neither criterion is fulfilled. For the purpose of the protocol, worsening of asthma is defined as new or increased symptoms and/or signs (i.e., physical examination or lung function) that can be either concerning to the subject (subject driven) or related to an asthma daily diary alert (diary driven). If an exacerbation event is

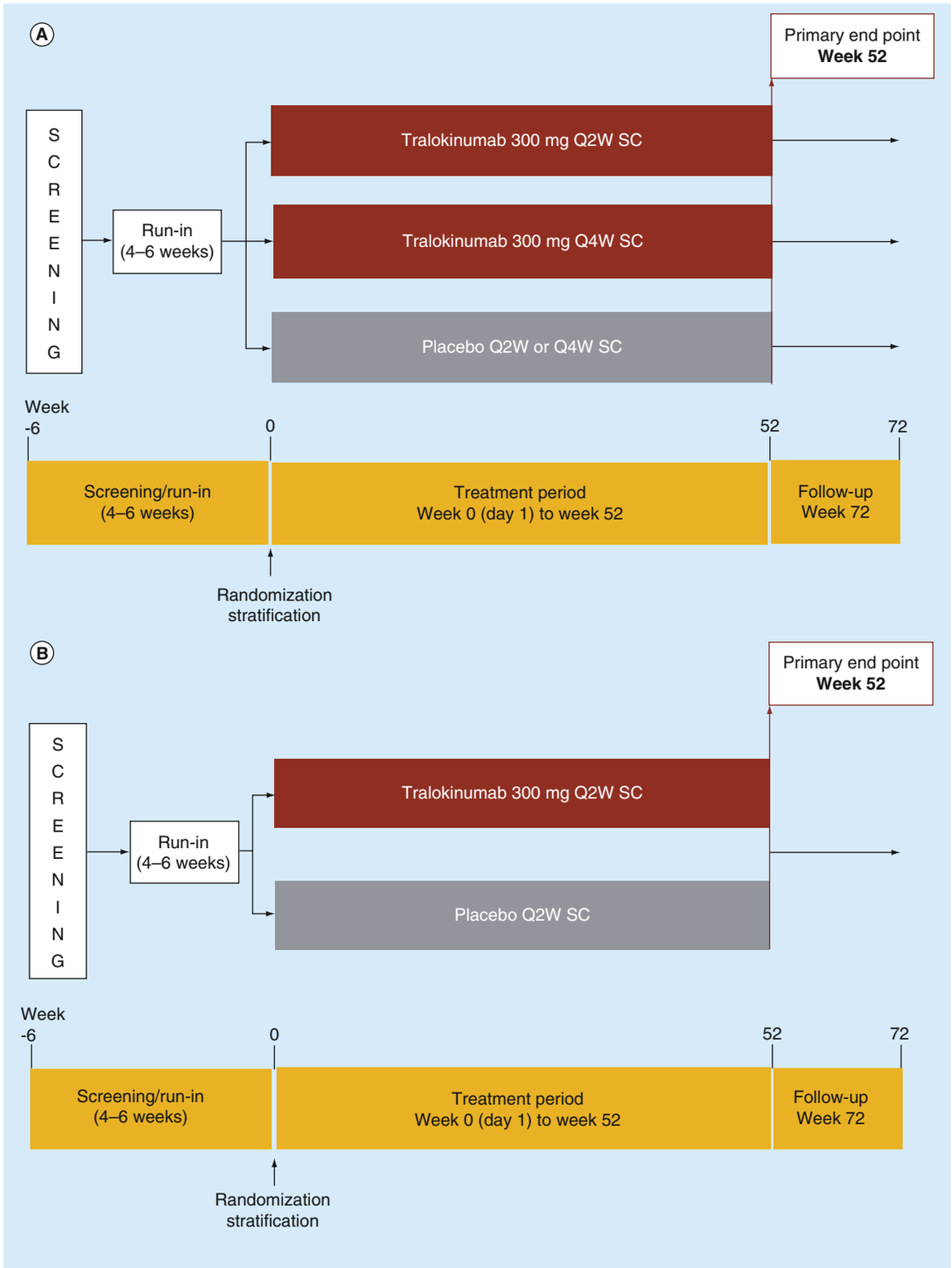

Figure 1. Study design of STRATOS 1† (A) and study design of STRATOS 2† (B).
†Study treatment is in addition to ongoing ICS-LABA therapy.
ICS: Inhaled corticosteroids; LABA: Long-acting inhaled β2-agonists; Q2W: Every 2 weeks; Q4W: Every 4 weeks; SC: Subcutaneous.

not associated with deterioration in at least one of the prespecified subject-driven and/or diary-driven objective measurements, the Investigator will have to justify the decision for defining the event as an exacerbation. Events that are not supported by any objective assessment of worsening of asthma will be deemed not to be a protocol-defined exacerbation.

Key secondary end points include FEV_1; asthma symptom score (total, daytime and nighttime); standardized asthma quality of life questionnaire (AQLQ[S]) for ≥12 years [48]; and ACQ-6 [49,50]. In order to demonstrate increased asthma symptom control compared with placebo, tralokinumab needs to show a significant effect on the percentage change from baseline in predose/prebronchodilator FEV_1 at week 52; and either a significant effect in mean change from baseline in bi-weekly daily asthma symptom score (combined daytime and nighttime score as captured in the asthma daily diary) at week 52 or a significant mean change from baseline in AQLQ(S) for ≥12 years total score at week 52.

Other secondary end points in both STRATOS 1 and 2 include the annual AER associated with an emergency room, or urgent care visit or a hospitalization; time to first asthma exacerbation; proportion of patients with ≥1 asthma exacerbation [51]; European Quality of Life – 5 Dimension 5 Level Daily Living Questionnaire [52]; Work Productivity and Activity Impairment Questionnaire [53] and Classroom Impairment Questionnaire [54]; asthma-specific resource utilization; rescue medication use; home-peak expiratory flow (morning and evening); nighttime awakening due to asthma [51]; and the pharmacokinetic and immunogenicity profile of tralokinumab.

Safety/tolerability end points

The safety and tolerability of tralokinumab will be evaluated by assessing adverse events (AEs), serious adverse events, and laboratory tests, vital signs, electrocardiograms and physical examination. AEs and serious adverse events will be collected from the time the patient signs the informed consent form, throughout the treatment period and the follow-up periods. Unresolved AEs at any follow-up visit will be monitored by the Investigator for as long as it is medically indicated, but without further recording in the case report form. Immunogenicity of tralokinumab will be assessed by the incidence rate of positive antidrug antibodies and characterization of their neutralizing potential.

Secondary objectives

The utility of baseline levels of periostin and DPP-4 as predictive biomarkers for the response to tralokinumab will be assessed. The results of STRATOS 1 will determine the biomarker-positive population to be assessed in STRATOS 2.

Exploratory analysis

Other potential biomarkers, which may be associated with increased IL-13 activation, will be evaluated including Fe_{NO}, blood eosinophils and total serum IgE. Additional exploratory end points relating to onset of treatment effect will be assessed by percent and absolute change from baseline in predose/prebronchodilator FEV_1, change from baseline in mean daily asthma symptom score (combined daytime and nighttime score as captured in the asthma daily diary); change from baseline in AQLQ(S) for ≥12 years score; and percentage change from baseline in forced vital capacity and forced expiratory flow 25–75% (STRATOS 1 only). DNA samples will be collected for future genetic analyses for which patient consent will be obtained separately.

Key inclusion & exclusion criteria

The key inclusion and exclusion criteria for STRATOS 1 and STRATOS 2 are listed in Box 1. After initial enrollment and confirmation of entry criteria, patients will enter a run-in period of 4–6 weeks to allow adequate time for all the eligibility criteria to be evaluated. Following this run-in period, patients who meet the eligibility criteria will be randomized to the assigned treatment over the 52-week period.

Statistical methods

The primary analysis of the efficacy end points will include all subjects randomized and receiving any study treatment, irrespective of their protocol adherence and continued participation in the study. Subjects will be analyzed according to their randomized treatment, irrespective of whether or not they have prematurely discontinued study treatment. For subjects who withdraw consent or assent to participate in the study, all data will be included up to the date of their withdrawal. Efficacy and safety comparisons of tralokinumab versus placebo will be performed using the pooled placebo cohorts (Q2W and Q4W).

A testing strategy will be applied to provide global strong control of the type I family-wise error rate across the primary and key secondary end points, and across dosing regimens in STRATOS 1 and the co-primary overall and biomarker-positive populations in STRATOS 2.

The primary efficacy objective will be evaluated based on a negative binomial model with the number of asthma exacerbations experienced by a subject during the 52-week double-blind treatment period as response variable, and the logarithm of the subject's corresponding follow-up time as an offset in the anal-

Box 1. Key inclusion and exclusion criteria in STRATOS 1 and 2 studies.

Key inclusion criteria

- Age 12–75 years
- Documented physician-diagnosed asthma for ≥12 months prior to enrollment, with the patient having received an asthma controller regimen requiring treatment with medium-to-high dose inhaled corticosteroids for at least 6 of the 12 months prior to enrollment
- Documented treatment with inhaled corticosteroids at a total daily dose corresponding to ≥500 μg fluticasone propionate dry powder formulation equivalents and a long-acting β2-agonist for ≥3 months prior to visit 1
- Morning prebronchodilator forced expiratory volume in 1 s value of ≥40 and <80% value (<90% for patients 12–17 years of age) of their predicted normal value
- Post-bronchodilator reversibility of ≥12% and ≥200 ml in forced expiratory volume in 1 s
- ≥2 documented asthma exacerbations in the 12 months prior to the date informed consent is obtained that required use of a systemic corticosteroid
- Asthma Control Questionnaire-6 score ≥1.5 at visit 1

Key exclusion criteria

- Clinically important pulmonary disease other than asthma
- History of anaphylaxis following any biologic therapy
- Clinically significant asthma exacerbation, in the opinion of the Investigator, including those requiring use of oral corticosteroids 30 days prior to the date of enrollment (visit 1), or during the screening/run-in period
- Use of immunosuppressive medication (including but not limited to methotrexate, troleandomycin, cyclosporine, azathioprine, systemic corticosteroids, such as regular treatment with oral corticosteroids and intramuscular long-acting depot corticosteroids, or any experimental anti-inflammatory therapy) within 3 months prior to enrollment (STRATOS 1 and 2), or to the date informed consent or assent is obtained (STRATOS 2)
- Treatment with any marketed or investigational biologic agent within 4 months or five half-lives prior to the enrollment visit, whichever is longer
- Current tobacco smoking or a history of tobacco smoking for ≥10 pack-years
- Previous treatment with tralokinumab

ysis to adjust for the subject-specific exposure times. Model covariates and factors will include at least treatment group and stratifying variables. Key secondary outcome variables will be analyzed using a repeated measures analysis approach including at least treatment and stratifying variables as the explanatory variables.

The interpretation of exacerbation data post-discontinuation of treatment is likely to be confounded by reduced quality of objective confirmation of deterioration, and by the use of subsequent therapies. Sensitivity analyses for the primary end point will be carried out to explore the impact of this, for example, exclusion of data post-discontinuation of treatment. Sensitivity analyses for the primary end point and the key secondary end points based on different missing data mechanism assumptions, including those expected to be more conservative such as 'missing not at random,' will be used to explore the robustness of any treatment effect, including multiple imputation approaches.

In order to evaluate the prognostic and predictive value of biomarkers, and identify a biomarker-positive subpopulation, a structured exploration of the relationship between outcome, biomarkers and treatment group will be performed in STRATOS 1. The biomarker positive subpopulation will be further studied in STRATOS 2 to confirm the effect of tralokinumab on the primary and key secondary end points in this population. The relative and absolute benefit of treatment in relation to the size of the potential subpopulation will be considered in selecting the final biomarker-positive subpopulation. The focus will be on the Q2W treatment regimen compared with placebo, with baseline periostin as the primary biomarker of interest, but also considering baseline DPP-4 values.

Sample size estimates

In STRATOS 1, a total study sample of 1140 patients (380 patients in each active dosing regimen and 190 in each of the Q2W and Q4W placebo treatment regimens) provides at least 90% power to show a reduction of 32% in AER for the Q2W dosing regimen of tralokinumab, compared with the pooled placebo cohorts. In STRATOS 2, a total study sample of 770 patients (385 patients per group) provides at least 90% power to show a reduction of 37% in AER for tralokinumab compared with placebo in the overall study population and 44% in the biomarker-positive population, if this includes approximately 50% of the overall population. The sample size calculations are made in terms of patient years at risk, and are based on an assumed AER of 0.8 in the placebo group and shape parameter of 0.95 (overdispersion) and a uniform loss to follow-up of 15% during the study in both the overall population and the biomarker-positive population.

Discussion & conclusion

The development of tralokinumab as a potential precision therapy in the treatment of patients with severe asthma can be viewed as a classic example of the progression of a biologic from the observation of an unmet need to the bedside via observations in the laboratory. The first step was the clinical recognition of an important need. Patients with severe asthma could not be managed adequately with currently available therapies, specifically ICS, LABA and other possible add-on controller medications. The second step was the identification of IL-13 as an important mediator of asthma pathophysiology, and the third step was the evaluation of tralokinumab, an engineered biologic that interferes with IL-13 signaling, in Phase I and II studies. The efficacy data obtained in these studies support further development of tralokinumab and, since no serious safety concerns that would interfere with the progression were identified in the earlier phases, confirmatory Phase III studies have been initiated.

There are clear limitations with current therapies for patients with severe asthma. International guidelines recommend adding other controllers, usually LABAs, to medium-to-high dose ICS when severe asthma is not controlled [2]. In combination with ICS, LABAs have been shown to further improve symptoms and lung function and reduce the risk of exacerbations [55]. Nevertheless, a subset of patients continues to have exacerbations, despite being compliant with the use of ICS-LABA combination therapy [4,5]. Other controllers have been studied in combination with either ICS alone or the ICS-LABA combination, but the benefits in terms of exacerbation reduction is limited. When added to ICS, leukotriene receptor antagonists, including montelukast, improve symptoms and lung function [56], providing similar benefits to those found with the addition of LABA to ICS therapy [57]. In an open-label observational trial in Europe, 12 months of treatment with montelukast added to ICS-LABA combination therapy did improve asthma control and reduce exacerbations, but these patients continued to have 'asthma attacks' [58]. In a three-way cross-over trial, Peters *et al.* showed that tiotropium added to ICS provided similar benefits in improving lung function and symptoms as LABA [59]. Addition of tiotropium to ICS-LABA does increase lung function and decrease exacerbations in patients with difficult to control asthma, but the effect is limited and a substantial percentage of patients continue to have difficulties in managing their symptoms [60]. Omalizumab, an anti-IgE antibody, is effective in patients with a known allergen and an increase in serum IgE, but has shown a nonuniform response and high discontinuation rates across patients [8,61]. The addition of low-dose OCS to ICS-LABA may be effective in some patients with severe asthma; however, OCS are associated with substantial side effects, and their use requires ongoing monitoring [62].

The identification of IL-13 as one of the central mediators of asthma pathophysiology has enabled the development of new therapeutic approaches that target this cytokine in patients with severe asthma [29,30]. Tralokinumab, an investigational fully human monoclonal antibody, specifically binds to IL-13 to prevent its interaction with both IL-13Rα1 and IL-13Rα2 receptors [31]. Preclinical studies suggest that tralokinumab prevents eotaxin release from normal human lung fibroblasts, CD23 upregulation in human monocytes and isotype switching in B cells (IgE production), as well as the development of airway hyper-responsiveness in animal models of allergen challenge [31]. These benefits in preclinical models suggested that tralokinumab, by blocking IL-13, may play an important role in treating patients with asthma.

The Phase II studies demonstrated a consistent beneficial effect on FEV_1 with tralokinumab treatment in patients with severe uncontrolled asthma and identified an exploratory, *post-hoc* subgroup of patients who may be most likely to respond to this biologic. In the Phase IIa study (NCT00873860) in patients with moderate-to-severe asthma, tralokinumab (150, 300 or 600 mg Q2W) did not improve ACQ-6 score, but showed a clear dose–response relationship for FEV_1, and *post-hoc* analyses suggested a link between the presence of IL-13 in sputum and a response to tralokinumab [32]. The following Phase IIb study (NCT01402986) in patients with uncontrolled severe asthma, showed that tralokinumab (300 mg Q2W) added to high-dose ICS-LABA did not reduce exacerbations, but did significantly improve lung function in the overall population [38]. Moreover, exploratory *post-hoc* analyses identified reversible, nonchronic OCS-requiring, biomarker-high (periostin or DPP-4) patient subgroups that demonstrated improvements in lung function, exacerbations and ACQ-6 [38]. These data suggested that periostin and/or DPP-4 could serve as predictive biomarkers in order to identify patients who may show an enhanced response to tralokinumab. Similarly, with lebrikizumab, a monoclonal antibody that also targets IL-13, marked clinical benefits have been observed in patients with high pretreatment periostin levels [63,64].

Thus far, tralokinumab has demonstrated an acceptable safety profile with no serious drug-related AEs of concern. Most treatment-emergent AEs were mild or moderate in severity and were not considered related to treatment [32,38]. In STRATOS 1 and 2, tralokinumab will be delivered via subcutaneous injections; safety information from the Phase II studies have shown that injection site reactions after tralokinumab administra-

tion are generally mild, and well tolerated, and discontinuation rates due to these reactions are low [32,38]. In addition, laboratory tests have demonstrated the low immunogenicity of tralokinumab, with no safety concerns raised [32,38].

The Phase III STRATOS 1 and 2 trials, which will run in parallel, aim not only to confirm the efficacy of tralokinumab, but also to identify a biomarker-positive subpopulation of patients that might show enhanced clinical response to the drug. This is in contrast to conventional strategies that assess the efficacy of investigational agents in an unselected patient population. Although, recent Phase II and III trials of the anti-IL-5 agents benralizumab [65], reslizumab [66] and mepolizumab (Phase III) [67] have used blood elevated eosinophil counts as biomarkers to identify patients with eosinophilic severe asthma who may have an enhanced response to anti-IL-5 therapy. Such an approach is reasonable because the mechanism of action of these anti-IL-5 biologics is based on suppressing eosinophil production. However, unlike anti-IL-5 agents that lower blood eosinophil counts, biologics targeting IL-13, like lebrikizumab and tralokinumab, increase blood eosinophil levels [32,38,63]. The mechanism of this effect is not understood and perhaps may be related to the effects of IL-13 on eosinophil trafficking. Further studies are planned to fully understand the effect of tralokinumab on blood and airway eosinophil movement (see Busse *et al.* in this issue for a description of the MESOS study).

Future perspective

For a complex disease such as severe asthma, precision therapies based on the inflammatory profile of the individual patient may hold the key to more effective treatments. Phenotypes can be linked to endotypes by combining an understanding of clinical characteristics with knowledge of underlying molecular pathways, and, consequently, precisely targeted therapies can be designed for these specific asthma endotypes [68]. Therefore, clinically relevant definitions of phenotypes and endotypes are required and the use of reliable biomarkers will be indispensable in predicting subsequent clinical responses. In the future, integrating systems biology (genomics and proteomics approaches) and computational modeling (structural and functional approaches) with pharmacology will enhance our efforts to identify suitable biomarkers, and will contribute to personalized healthcare in severe asthma [69,70].

Executive summary

Background

- Patients with severe asthma that is uncontrolled despite the use of medium-to-high dose inhaled corticosteroids and other controller therapy, or long-term oral corticosteroids, represent a major unmet need.
- New-generation biologic therapies are under development that target various underlying molecular pathways in asthma.
- IL-13 is one of the central mediators of airway inflammation in asthma, and is upregulated in a subset of patients.
- Tralokinumab, an investigational fully human monoclonal antibody, has been designed to specifically target and neutralize human IL-13.
- Measurement of IL-13 in blood and sputum has proved inconsistent and, therefore, identification of surrogate IL-13 biomarkers will be important to identify patients with an activated IL-13 pathway, who are likely to have an enhanced response to anti-IL-13 therapy.

Study rationale

- Phase IIa and IIb studies have demonstrated that tralokinumab improves lung function in patients with moderate-to-severe asthma.
- A previous Phase IIb study of tralokinumab has identified an exploratory, *post-hoc* subgroup of patients with serum periostin or dipeptidyl peptidase-4 (DPP-4) levels above median at baseline, which demonstrated improvements with tralokinumab versus placebo in exacerbations, lung function, asthma symptoms and quality of life.
- Periostin and/or DPP-4 could be used as biomarkers for phenotyping patients who are likely to have an enhanced response to tralokinumab.

Study design

- STRATOS 1 and 2 (NCT02161757 and NCT02194699) are Phase III multicenter, randomized, double-blind, parallel group, placebo-controlled studies conducted globally.
- The studies aim to confirm the efficacy and safety/tolerability of tralokinumab in patients with severe uncontrolled asthma despite treatment with inhaled corticosteroids and long-acting inhaled β2-agonists. STRATOS 1 will also address the potential identification of a target population with an enhanced response rate based on the presence of high serum periostin, or DPP-4 as a biomarker of IL-13-driven asthma. STRATOS 2 also aims to confirm that the biomarker-positive target population, identified in STRATOS 1, demonstrates an enhanced response to tralokinumab.

Financial & competing interests disclosure

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Papers of special note have been highlighted as:
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