PrecISE: Precision Interventions for Severe and/or Exacerbation-Prone Asthma Network

National Clinical Trial (NCT) Identified Number: NCT04129931

IND Sponsor: University of North Carolina at Chapel Hill

Funded by: National Heart, Lung, and Blood Institute

Version Number: v 3.3

27 September 2024

Summary of Changes from Previous Version:

Affected Section(s)	**Summary of Revisions Made**	**Rationale**
9.2 Sample Size Determination	Indicate that 93 participants (2:1 ratio of biomarker positive to negative participants) is needed when futility analysis is not performed for an intervention.	Clarify the sample size needed when futility analysis is not performed.
1.3 Schedule of Activities	Indicate that some procedures and assessments are not required at the participant's last Visit X.6	Since the participant will not be participating in another treatment period, some assessments and procedures are not necessary.
9.4 Populations for Analyses	Revise population definitions and move details to the Statistical Analysis Plan	Clarification.

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STATEMENT OF COMPLIANCE

The PrecISE study will be conducted in accordance with International Council on Harmonization E6 Guideline for Good Clinical Practice (ICH E6 GCP), applicable United States (US) Code of Federal Regulations (CFR), and the NHLBI Terms and Conditions of Award. The study is being conducted under an Investigational New Drug application (IND) from the US Food and Drug Administration (FDA), and Dr. David B. Peden, MD, MS of the University of North Carolina at Chapel Hill is the IND sponsor. The PrecISE Principal Investigators will assure that no deviation from or changes to the protocol will take place without prior agreement from Dr. Peden (as IND sponsor) and the NHLBI (as funding agency), FDA review, and documented approval from the Institutional Review Board (IRB), except where necessary to eliminate an immediate hazard(s) to the trial participants. All personnel involved in the conduct of this study have completed Human Subjects Protection and ICH GCP Training.

The protocol, informed consent form(s), recruitment materials, and all participant materials will be submitted to the central IRB for review and approval. Approval of both the protocol and the consent form must be obtained before any participant is enrolled. Any amendment to the protocol will require review and approval by the IRB before the changes are implemented to the study. When changes are made to the consent form; a determination will be made regarding whether a new consent needs to be obtained from participants who had provided consent earlier using a previously approved consent form.

1 PROTOCOL SUMMARY

1.1 SYNOPSIS

Title:	The PrecISE (Precision Interventions for Severe and/or Exacerbation-Prone Asthma Network) Study
Background and Rationale	PrecISE is an NHLBI-sponsored network established to conduct a phase 2 proof-of-concept study to test interventions in biomarker-defined subgroups of patients with severe asthma. Severe asthma affects 5 - 10% of all patients with asthma (~2.5 million Americans), and patients with severe asthma experience substantial morbidity and require extensive use of healthcare resources.[1] Despite the use of high dose inhaled corticosteroids and a second controller (and/or systemic corticosteroids), severe asthma patients continue to have poor control, low lung function and/or increased risk of severe exacerbations. Consequently, there is a need to investigate novel interventions for utility as targeted therapies in this patient population. The PrecISE Investigators proposed novel interventions in biomarker-based subgroups, with the goals of gaining information about the efficacy of the interventions in the severe asthma patient population and additional information on the utility of the biomarkers in identifying patients for treatment. Over 30 interventions were proposed, and six were selected by the PrecISE Steering Committee for initial study based on feasibility, scientific enthusiasm about preliminary evidence of efficacy, safety, and potentially useful biomarkers. Additional interventions were identified for consideration as the study progresses, particularly if any of the initial interventions are discontinued during the study for low

likelihood of success or safety issues. Due to delays incurred by the Covid-19 pandemic, the number of interventions to be initially investigated was reduced to five.

All interventions will be studied under one master protocol using an adaptive platform trial design[2,3] to assess multiple interventions simultaneously, while also obtaining information on biomarker utility in predicting responsiveness to each treatment. Interventions may be discontinued and new interventions entered into the study, as data accrue on the safety and efficacy of the various interventions. Definitions of patient subgroups targeted by each intervention and the biomarkers used to define them are adapted prospectively during the study. By prospectively enriching patient population we increase the chance of demonstrating a treatment effect at the end of the trial, as testing the efficacy of an intervention in an unenriched population of all participants may results in failure to detect the treatment effect.

The master protocol, described in the following sections (1.2 – 10.4), contains all aspects of the study design and all study elements and procedures common across the various interventions. Details of each of the interventions planned for study, including the rationale for their selection into the study and intervention-specific study procedures required for each, are provided in Appendices I-V. As additional interventions are selected for study, appendices for each will be added to the master protocol as protocol amendments and submitted to the DSMB and IRB for their approval and to the FDA for its review prior to implementation.

Our approach to designing PrecISE provides flexibility in studying multiple novel interventions at the same time and adaptability in defining patient subgroups targeted for therapy and in stopping some interventions and starting others through interim decision-making utilizing accruing data. The anticipated result is a wealth of information about the efficacy and safety of new treatments for severe asthma and about which patients might benefit from those treatments in the future, providing a precision medicine solution in an area of significant unmet medical need.

Overall Study Description:

The PrecISE Study is an adaptive platform trial conducted under a single Master Protocol to identify new therapies for severe asthma that are effective in biomarker-defined subgroups of participants. Five novel therapies will initially be investigated. The trial is designed to meet our primary objectives, namely, to: (1) identify novel therapies that work in biomarker-defined subgroups of participants with severe asthma, and (2) optimize the subgroups targeted for treatment by refining the biomarker and subgroup definitions.

For each participant, the study consists of three phases: (i) an initial screening phase, (ii) a 2-period crossover phase, and (iii) a single-period

crossover phase with successive re-randomizations. Common platforms for biomarker screening will be used during the initial screening period to determine a participant's biomarker profile. Throughout the study, interventions will be randomly assigned based on these biomarker profiles. Participants will have a higher likelihood of assignment to an intervention that targets their particular profile. Subgroup definitions and associated treatment assignment probabilities are adapted as data accumulate throughout the study.

Following the initial screening period, participants will be randomly assigned to interventions based on their biomarker profile and enter a 2-period, double-blind crossover phase consisting of two 16-week treatment periods separated by an 8-week washout period. Note that longer washout periods will be considered for therapies with a half-life longer than 11 days. Treatment sequence will be randomly assigned as either test treatment followed by matching placebo or vice-versa.

At the conclusion of the 2-period crossover phase and washout period, participants enter the single-period crossover phase of the study consisting of successive single 16-week treatment periods followed by 8-week (or longer) washout periods. For each single-period crossover treatment, participants are randomly assigned to interventions based on their biomarker profiles that were determined during the initial screening period. The biomarker-based treatment assignment probabilities will be adapted during the study as data on the utility of the biomarkers in predicting treatment response accumulate.

A small percentage of participants will receive placebo in this phase (periods 3-6) to assess seasonal and sequence effects throughout the study (no participant will receive more than three placebos during the study). Masking of treatment assignments will be maintained by matching a participant's placebo to one of the available interventions for which the participant is eligible and has not yet received.

Periodic reviews of safety data will be performed by the Data and Safety Monitoring Board (DSMB) at pre-specified intervals throughout the study, and interventions found to pose significant safety risks in the severe asthma population will be dropped from the study.

Interventions may enter the study at different times, depending on their availability, and may be discontinued, as noted above. Additional interventions may be proposed depending on the progress of the study and innovations in potential treatments for severe asthma.

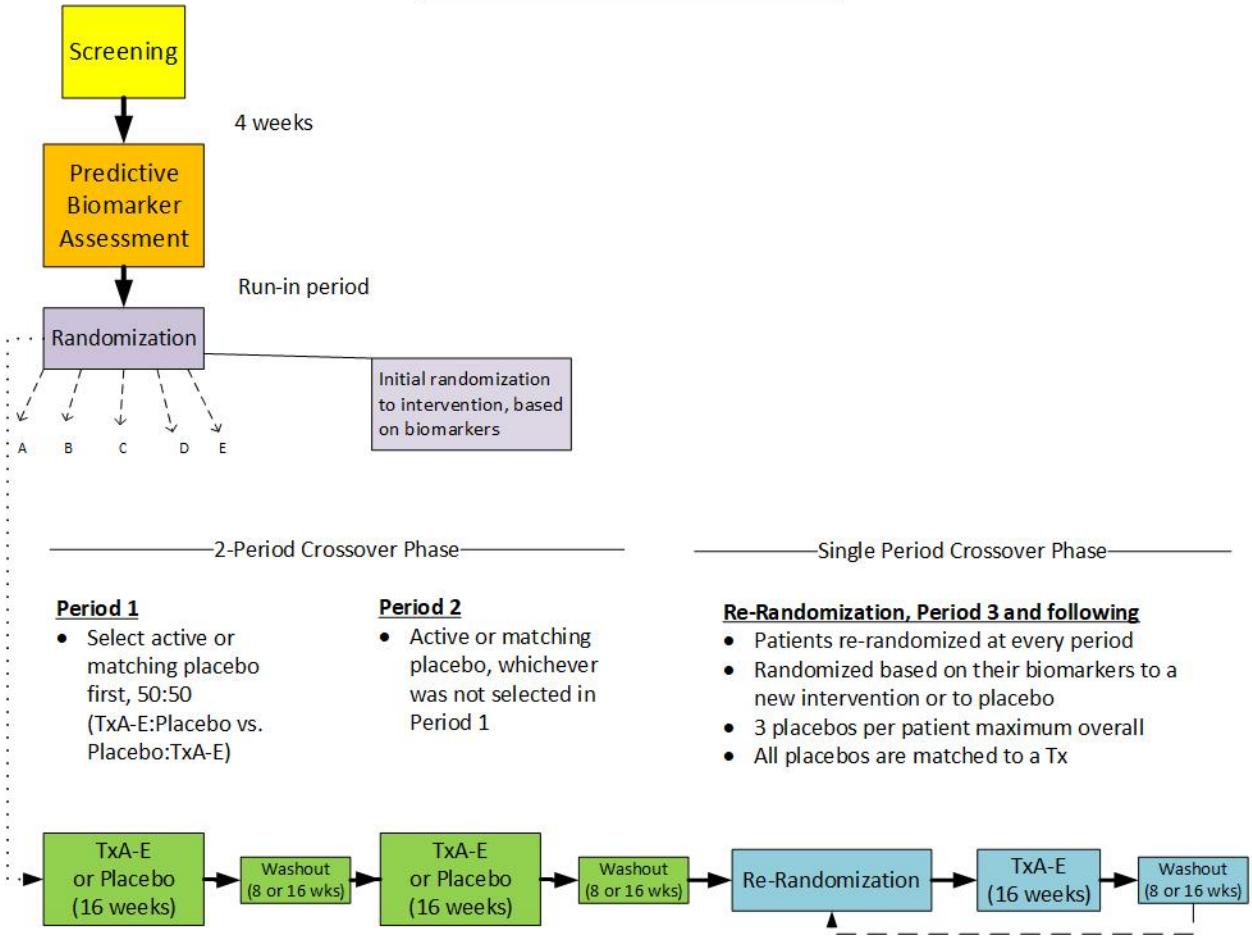
Our study timelines and enrollment goals are such that, depending on the time of enrollment, each participant will receive up to five test treatments

Objectives:	**Primary Objectives:** (1) Identify novel therapies that work in biomarker-defined subgroups of severe asthmatic participants; (2) Optimize the subgroups targeted for treatment by refining the biomarkers and subgroup definitions. **Secondary Objectives:** (1) Gain information about potential response/pharmacodynamic biomarkers for selected therapies; (2) Explore the safety and effectiveness of selected therapies in adolescent participants with severe asthma.
Endpoints:	**There are 2 primary efficacy endpoints:** **1) FEV_1 percent predicted,** assessed prior to bronchodilator administration **2) Asthma symptom control,** assessed via the Juniper Asthma Control Questionnaire (ACQ-6)
Study Population:	The sample will include 395 adult and pediatric (12-18 years of age) participants who meet modified guideline criteria for severe asthma and who are currently uncontrolled or continue to have exacerbations. Participants must be at least 12 years of age, on a stable regimen of asthma medications prior to enrollment, and satisfy other inclusion/exclusion criteria. Not all interventions will be studied in adolescents (see Appendices I-V). We anticipate that at least two of the interventions will be studied in both adults and adolescents.
Sample Size:	A sample size of 111 participants is required to achieve 80% power to detect a treatment effect with respect to at least one of the two primary endpoints (FEV_1 or asthma symptom control) equal to 0.3 times the standard deviation of that endpoint, taking into account the possibility to stop for futility and assuming a treatment period discontinuation rate (active or placebo) of 10%. The Type I error probability will be controlled under the global null hypothesis (no effect on either endpoint) at $\alpha \leq 0.10$ (see Section 9.2). A sample size of 111 participants, at least 74 targeted by the intervention, is required to support the final precision medicine analysis. Secondary efficacy analyses will be conducted in pediatric participants, but power to support hypothesis testing will be limited for these analyses.
Interim Analysis:	Interim analyses will be conducted for the following purposes. (1) A single interim analysis for futility may be conducted for each test treatment based on data from the a priori best subgroup, biomarker positive, patents. Test treatments demonstrating futility will be dropped from the trial. A futility analysis will be also performed in biomarker negative patents. Further enrollment to test treatments demonstrating futility in biomarker negative subgroup will be limited to biomarker positive participants. (2) An interim analysis to estimate the within-participant, between-period correlation might be conducted pooling data across test and placebo treatment periods, maintaining the masking of treatment assignments. If the correlation is much higher than we hypothesized, we

Final Analysis:	will reduce the required sample size prior to unmasking of treatment assignments and modify the final analysis plan to reflect this change. The timing of these interim analyses, will depend on the time the treatment is available to enter the Master Protocol, as well as participant accrual. An intervention will be considered effective if it is effective on at least one of the two primary outcomes. The analysis model for the final efficacy analysis will take into account the correlations on outcomes assessed for the same patient across the different treatment periods. The final analysis will include pairs of the treatment and matching placebo from the first two periods of the trial, as well as unmatched observations on the same patient. Mixed model repeated measures methods will be used for the primary analysis (see Section 9.5).
Phase:	Phase 2 proof-of-concept study
Description of Sites/Facilities Enrolling Participants:	Approximately 35 U.S. sites; all sites are clinic settings.
Description of Study Intervention:	Five novel treatments for severe asthma will be studied, each targeting a specific, biomarker-defined subgroup of participants. Candidates currently include novel medications and dietary supplements that target mechanisms relevant to asthma pathology.
Study Duration:	Approximately 62 months from first participant screened until last participant follow-up
Participant Duration:	**Treatment Supply:** Participant duration will be variable based on the time they enter the study and the number of treatments for which they are eligible (see Section 4.4). We anticipate needing approximately 850 months of treatment (active or placebo) for each intervention.

1.2 SCHEMA

PrecISE Protocol Schema



1.3 SCHEDULE OF ACTIVITIES (SOA)

PrecISE Visit Structure **Procedures**	**Visit SA: Screening**	**Visit SB: Optional as needed to qualify**	**Visit SC: Compliance check***	**Visit 0: Run-In Visit, Safety Labs & Biomarkers**	**Visit X.1: Treatment Visit 1**	**Visit X.2: Treatment Visit 2**	**Visit X.3: Treatment Visit 3**	**Visit X.4: Treatment Visit 4**	**Visit X.5: End of Treatment Visit**	**Visit X.6: Washout**
Consent, Demographics, Contact Info, Registry[1]	X									
Complete Physical Exam	X	X			X					
Brief Physical Exam by Coordinator				X		X[4]	X	X[4]	X	X[11]
CompEx Diary Assessment[1]			X	X	X	X	X	X	X	X[11]
Compliance Assessment[1]			X	X	X	X	X	X	X	X[11]
Dispense Controller Medication[1]	X	X		X	X	X	X	X	X	X[11]
Dispense and train on e-diary and home spirometers[1]	X	X		X						
Assess that participant meets or continues to meet inclusion criteria[1]				X	X					X[11]
Intervention Assignment (occurs between visits 0 and 1.1 for the first period) [1]				X						X[11]
Treatment Assignment (active or placebo) [1]					X					
Dispense Study Treatment[1]					X	X	X	X		
Medical, Asthma and Allergy History questionnaires[1]	X	X								X
Adolescent School questionnaire (participants in school only) [1]	X									
Validated questionnaires (to determine sleep apnea, GERD, depression, sinus disease, vocal cord dysfunction) [1]	X	X								

PrecISE Visit Structure **Procedures**	**Visit SA: Screening**	**Visit SB: Optional as needed to qualify**	**Visit SC: Compliance check***	**Visit 0: Run-In Visit, Safety Labs & Biomarkers**	**Visit X.1: Treatment Visit 1**	**Visit X.2: Treatment Visit 2**	**Visit X.3: Treatment Visit 3**	**Visit X.4: Treatment Visit 4**	**Visit X.5: End of Treatment Visit**	**Visit X.6: Washout**
Validated questionnaires for Quality of Life and Work Productivity[1]	X	X			X	X	X	X	X	
Socioeconomic information/Household[1]	X									
Asthma and non-asthma medication (concomitant medications) status[1]	X	X		X	X	X	X	X	X	X
Blinding Index questionnaire									X	
Pulmonary Function, Asthma Control and Exacerbations										
Spiro PRE with specified med withholds[2]	X	X		X	X	X	X	X	X	X[11]
Spiro POST 4 puffs albuterol[2]				X	X				X	
Maximum Bronchodilator (4 + 2 + 2 puffs albuterol)	X			X[3]						
Methacholine challenge (if needed)		X								
DLCO (if needed bc +smoking Hx)		X								
Cotinine (if needed bc +smoking Hx)		X								
ACQ-6[1]	X			X	X	X	X	X	X	X
Exacerbation History/Health care utilization questionnaire[1]	X			X	X	X	X	X	X	X
Safety Assessments										
Urine: Pregnancy test (females of childbearing potential)	X	X		X	X	X[4]	X	X[4]	X	X[11]
Blood: CBC with Differential Count				X	X	X[4]	X	X[4]	X	X[11]
Blood: Chemistry Panel (including liver function tests)				X	X	X[4]	X	X[4]	X	X[11]
Blood: Triglycerides (non-fasting)				X						

PrecISE Visit Structure **Procedures**	**Visit SA: Screening**	**Visit SB: Optional as needed to qualify**	**Visit SC: Compliance check***	**Visit 0: Run-In Visit, Safety Labs & Biomarkers**	**Visit X.1: Treatment Visit 1**	**Visit X.2: Treatment Visit 2**	**Visit X.3: Treatment Visit 3**	**Visit X.4: Treatment Visit 4**	**Visit X.5: End of Treatment Visit**	**Visit X.6: Washout**
Blood: Viral screens – Hep B and C, HIV Types 1 and 2				X						
Blood: TB via QuantiFERON[5]				X						
Radiology (CXR), not required if the participant has a negative QuantiFERON[5]				X						
Cardiology (EKG)				X						
Adverse event assessment[1]	X	X	X	X	X	X	X	X	X	X
Primary Predictive Biomarkers										
Blood eosinophils (CBC with diff)				X	X					
Plasma IL-6				X						
FeNO				X	X					
Genotyping				X						
Response/pharmacodynamic biomarkers										
Blood eosinophils (CBC with diff)						X[4]	X	X[4]	X	X[11]
Plasma IL-6										
FeNO						X[4]	X		X	
Additional Phenotyping Procedures										
Sputum induction[6]				X						
CT scan for lung phenotyping[6]				X						
Blood: PBMCs[7]					X[8]				X	
Urine for biorepository					X		X		X	
Blood for RNA[9]				X						
Other blood (plasma, serum) for biorepository					X	X[4]	X	X[4]	X	
Nasal EPX[10]				X	X		X		X	

*Visit SC is optional. All other visits are approximately 4 weeks apart. Treatment visits X.1-X.6 are repeated each treatment period.

[1] These procedures and assessments may be conducted remotely via phone, videoconference or electronic survey methods.

[2] Spirometry may be conducted in the home and/or in the clinic.

[3] Optional at site discretion to demonstrate reversibility

[4] Not required for adolescents at VX.2 and VX.4.

[5]Blood for TB via QuantiFERON is collected on adults only

[6] If not collected before V1.1, the Sputum and CT can be collected before or at V2.1 or before or at V3.1. Sputum induction and CT scans may not be conducted at all sites.

[7] PBMCs may not be collected at all sites.

[8] PBMC is only collected at V1.1.

[9] Blood for RNA if not collected before V1.1, should be collected at V2.1, and, again, at V3.1.

[10] Nasal EPX can be skipped if the institution requires a COVID test before the procedure. Nasal EPX collection is optional at the site level.

[11] Not required at the final VX.6.

Note: Due to the COVID-19 pandemic, not all procedures will be conducted at all clinic visits.

2 INTRODUCTION

2.1 STUDY BACKGROUND AND RATIONALE

Severe asthma affects 5 - 10% of all patients with asthma (~2.5 million Americans) and patients with severe asthma experience substantial morbidity and require extensive use of healthcare resources.[1] Despite the use of high dose inhaled corticosteroids and a second controller (and/or systemic corticosteroids), severe asthma patients continue to have poor control, low lung function and/or increased risk of severe exacerbations.

While most asthma is controlled by currently available therapy including beta-agonists and inhaled corticosteroids, severe asthma continues to produce symptoms and/or exacerbations despite these currently available therapies.[1] Persistent symptoms impair quality of life, lead to loss of days from work and school and impose a high personal and societal economic burden. Recurrent exacerbations lead to loss of days from school and work and are associated with accelerated loss of lung function. In addition, recurrent exacerbations are treated with oral corticosteroids; 52% of adults enrolled in the NHLBI Severe Asthma Research Program 3 (SARP3) had received at least two courses of systemic corticosteroids in the prior year.[5] Many patients experience substantial side effects from these short courses, including sleep problems, mood changes, increases in appetite and weight gain, challenges in maintaining glucose control in patients with diabetes, and hypertension. Furthermore, a recent study of over 1.5 million commercially-insured Americans showed that short term use of oral corticosteroids for any condition (including asthma) was associated with significantly increased risks of developing sepsis, venous thromboembolism, and bone fracture within 30 days of corticosteroid use.[6] Thus, identification of therapeutic strategies that reduce asthma symptoms or reduce the risk of exacerbations among patients with severe asthma is essential.

Optimal treatment for patients with severe asthma is uncertain, because the pathophysiologic underpinnings of severe asthma are heterogeneous.[1] For example, in the cluster analyses performed using SARP data, five clusters of asthma were identified. Three of them were characterized by early onset of disease and aeroallergen sensitivity (cluster 1: mild, 2: moderate and 4: severe).[7] Other patients with severe asthma may have fixed or progressive reductions in lung function (cluster 5) or a later age of

asthma onset (cluster 3). Finally, a subgroup of patients with severe asthma have persistent eosinophilic or neutrophilic airway inflammation (clusters 4 and 5).

Other factors that may influence asthma severity include disease duration, sex, obesity, and respiratory infections. A treatment may be effective for only a subset of patients with particular phenotypes/endotypes of severe asthma or other factors. The newest therapies for severe asthma, those targeting eosinophils, appear to be primarily effective in patients with Type 2 inflammation. In the ACRN TALC study,[8] African-Americans were less responsive to each of three add-on therapeutic regimens than Caucasians. Traditional biomarker-stratified parallel arm trial designs are inefficient and could be considered as a barrier to drug discovery for newly recognized inflammatory pathways and/or uncommon patient subsets. Therefore, a need exists to take a personalized medicine approach to severe asthma, with treatments that are tailored to an individual participant's clinical or biomarker profile.

Precision medicine allows more patients to receive the specific treatment that is likely to be best for them. Severe asthma is a disease that stands to benefit from the use of novel trial designs and statistical methods to identify effective targeted therapies. The goal of this trial design is to efficiently use resources to test treatments in specific subgroups of patients. The adaptive platform design proposed here has multiple advantages. It allows rapid evaluation of innovative therapies while holding out the potential of providing information on informative phenotypic biomarkers and possibly early-response/ pharmacodynamic biomarkers. It permits patients to receive multiple interventions and will ultimately provide information on therapies that might be most suited to them. Design features that allow for this flexibility include shortening the required time to follow up participants, reducing the number of participants in different treatments, and initiating novel treatments more quickly.

The background information for individual test treatments is found in Appendices I–V, Section 2.2.

2.2 RISK/BENEFIT ASSESSMENT

The overall burden of severe asthma is much greater than that associated with mild-to-moderate disease. In addition to the side effects and decreased efficacy of systemic corticosteroids in these patients (reviewed in Section 2.1), many patients with severe asthma have daily symptoms, functional impairment and frequent health care utilization. These indicators of morbidity persist despite introduction of newer therapeutic strategies which are not uniformly effective across the spectrum of severe asthma. Discussions with patients and advocacy organizations indicate that patients with severe asthma are often also frustrated by their current clinical care, particularly if they do not have access to an asthma specialist.

Participants in these forums have expressed a general interest in learning more about the subsets of severe asthma and a willingness to try new medications, even those associated with side effects.

The PrecISE Network convened three Participant Advisory Councils (PAC, see Section 5.5) in the Midwest, Southeast, and Southwest. These PACs consisted of twenty adults with severe asthma who completed a half-day workshop with PrecISE investigators. Input of the PAC was incorporated into the creation of this master protocol and will be briefly summarized.

The PAC expressed the opinion that they would recommend participating in a PrecISE protocol to others with severe asthma, and specifically commented that use of the placebo, multiple treatment periods, and a washout period are acceptable features of a clinical trial. The PAC also expressed a willingness to undergo multiple sequential interventions followed by washout periods with the possibility of placebos during treatment periods. Finally, most patient advisors felt that the study procedure and visit burden was acceptable for the potential benefit of advancing the state of knowledge in the treatment of severe asthma. However, there was concern among patients about the length and number of visits and the

number of PFTs to be performed. Therefore, the investigators reduced the number of PFTs and visits in consideration of this feedback. The PrecISE Network also conducted one-on-one interviews with five adolescents with severe asthma and their caregivers. Overall, parents were very protective of their children and less enthusiastic about the trial. They were concerned about use of placebos but expressed that they would be willing to consider the trial if their physician recommended it. The pediatric committee considered the concerns of the caregivers and understands how important physician recommendation for the trial will be in recruitment of these patients.

The risks of study participation and the specific interventions in PrecISE are reviewed in Appendices I-V, Section 2.3, but in general, include risks of worsening asthma control, major infection, diarrhea and electrolyte abnormalities. In addition to safety lab monitoring, the overall risk of these side effects is mitigated by the short treatment periods (16 weeks) in this protocol, followed by washout periods of the study treatments.

The following sections also discuss the nonpharmacologic risks and benefits of study participation.

2.2.1 KNOWN POTENTIAL RISKS

Participating in the research study has the potential for some risks to the research participant. The risks of our therapeutic interventions are listed for each intervention under its respective section. In addition, we have delineated below the risks of our research procedures, which may include physical, psychological, social, or legal risks.

The risk of disclosure of study participation is a risk to participants in the study.

Questionnaires pertaining to related allergy, asthma and pulmonary symptoms, medication, and history will be conducted as part of the study and may be considered burdensome and repetitive. This may occur but is not considered a serious risk to the research participant.

Venipuncture to collect blood for biomarkers and safety studies is occasionally associated with minor bruising, risk of infection, discomfort, feeling light headed, or faint.

Spirometry measurements are conducted to evaluate a participant's lung function. Spirometry can cause any participant to become light-headed or dizzy. This may make asthmatic participants wheeze or experience shortness of breath or chest tightness, or they may worsen these symptoms if they were already occurring. The chance of these symptoms occurring is not common but treatment (including albuterol) will be available to the participant if any of the symptoms should occur. During the procedure, **albuterol** will also be given to determine **reversibility**. Common risks associated with albuterol include headache, increased pulse rate, and shakiness of hands. Uncommon risks include heart pounding or racing, mouth and throat irritation and muscle cramps. Rare side effects include low blood potassium, irregular heartbeat or heart rhythm, hyperactivity and an immediate increase in wheeze after dosing. The same potential risks associated with spirometry conducted in the clinic exist for spirometry conducted in the home. Participants will have treatment (including albuterol) at home to treat any of the symptoms listed above, should they occur. Moreover, the study coordinator will be video-conferencing with the participant during in-home spirometry assessments to monitor the participant's safety.

As part of the preparation for spirometry testing, a participant may be asked to **withhold using certain medications** prior to certain breathing tests and study visits. An increase in allergy or asthma symptoms may occur as a result.

Methacholine may cause coughing, chest tightness, shortness of breath, and wheezing. On rare occasion, severe bronchospasm can occur.

Induced sputum can be associated with bronchospasm in participants with asthma.

Urine will be collected by clean-catch and there are no foreseeable risks to its collection.

Exhaled nitric oxide is a non-invasive procedure to measure the amount of nitric oxide in exhaled breath and there are no clinically significant risks to its measurement.

Pregnant or nursing women are not permitted to participate in this study. Certain procedures and test treatments could cause harm to the fetus including but not limited to radiation from the CT scan, and methacholine inhalation. Females of childbearing potential will be required to use acceptable methods of birth control and will undergo pregnancy tests regularly throughout the study.

Low-dose chest CT has been used safely in numerous clinical research studies of COPD and asthma. However, x-ray Radiation from CT at clinical exposure levels has been shown to have a nominal cancer risk at the population level. To minimize this risk, CT dose levels are kept to a minimum and not all lung volumes are scanned for all time points. For the proposed CT protocol, participants receive a total effective dose of 3 times the annual background radiation (3.3 mSv) for the total of the 21-month treatment period. This is lower overall than the effective dose in comparable studies such as SARP III despite more frequent monitoring. Additional time points may be specified using a 50% reduced dose protocol developed within QIBA (http://qibawiki.rsna.org/index.php/Lung_Density_Biomarker_Ctte). Noise associated with lower dose is mitigated using median filtering or iterative reconstruction (where available).

Protection against risk

Safety monitoring is multifaceted, including addressing safety of the research participants, processes for minimizing research-associated risk, protection of study data and participant identification, and review and reporting of adverse and unanticipated events. This information is outlined in the Data and Safety Monitoring Plan for the study (see Section 10.1.6). Adverse events will be reviewed by the Data, Modeling and Coordinating Center (DMCC), IRB, PrecISE Data and Safety Monitoring Board (DSMB), and NIH, as required. Any participant may choose to withdraw from the study at any time, and any participant may be electively discontinued from the study by the PI.

Every precaution is taken to ensure participant safety. The measures taken to minimize risk are described in the section below under each specific procedural risk. The staff responsible for this protocol are skilled in the management of severe asthma, allergies, and anaphylaxis. All coordinators administrating study medications will be certified in preparation and administration. Venipuncture and test treatment administration will be performed by trained medical personnel using aseptic technique. Having experienced research staff following established guidelines for all study procedures will help reduce the risk to participants. All PrecISE clinical centers have extensive experience in the performance of these procedures, and safety checks and procedures are in place to minimize risk.

Central training on study procedures and systems will be held for clinical personnel prior to study start and will be recorded for use by new staff members as needed during the study. If new procedures or forms are adopted by the study, additional training will be conducted via webinars. New clinical center staff will be trained by experienced staff at their site in addition to the training modules available via webinars. Each clinical site staff member will be certified on study procedures and systems, and certification records will be maintained at the sites for auditing.

Minimization of research-related risk

The protocol includes information about processes by which procedure related research risk is minimized.

The risks for each of the interventions we propose is described in Appendices I-V, Section 2.3, as are the strategies to minimize risk. For each of the interventions, we will exclude patients at high risk for known side effects of the intervention. Throughout (and after the intervention if appropriate) we will monitor for known side effects as well as unknown and unexpected side effects. Suspension of therapy and discontinuation of therapy thresholds have been specified.

Risk of **disclosure** will be minimized through the use of several procedures. The DMCC operates under FISMA (Federal Information Security Management Act) guidelines, and the data management system used in PrecISE is compliant with FDA 21 CFR Part 11, which establish security policies for the study databases, such as requiring use of strong passwords, limiting access to data based on study role, and the use of secure sockets layer encryption during data transmission. Within the data management system, data elements are persisted in a format which isolates personal identifying information from other data elements.

During the **questionnaires**, a participant can refuse to answer any question that makes them feel uncomfortable.

The risk of **venipuncture** is minimized by allowing the participants to recline during the venipuncture. Aseptic technique will be used to prevent infection.

Risks related to **spirometry** and **reversibility** will be minimized by monitoring participants for development of asthma symptoms or drop in lung function during the procedure. For in-home spirometry, monitoring will be conducted by video-conference.

The risk of **methacholine challenge** is minimized by gradually increasing the agonist dose, while monitoring pulmonary functions after each dose. Participants with a baseline FEV_1 <50% of predicted will not undergo bronchoprovocation. Albuterol is readily available to treat any symptoms. Lung function is monitored throughout the challenge, and methacholine dosing is stopped once lung function declines by 20%. Any acute severe bronchospasm will be treated with an inhaled beta-agonist. Medical and nursing personnel, medication, and equipment will be available at the study sites to treat and manage any bronchoconstriction episodes. If symptoms become intolerable at any time during the procedure, the procedure will be stopped.

Withholding asthma medications can cause chest tightness and wheezing. Participants are informed that if they cannot hold their medications and feel they need to take them, they should do so and call the study coordinator to let them know.

The risk of **sputum induction** is minimized by monitoring participants for development of asthma symptoms or drop in lung function during the procedure. Pre-medication with albuterol will be provided and albuterol treatment will be available for clinically significant symptoms or drop in FEV_1 >20% during or after the induction.

To minimize risks during **pregnancy**, females of childbearing potential must have a negative pregnancy test prior to any testing and prior to any test treatment administration. They must agree to use acceptable forms of birth control throughout the study.

Risks related to administration of **prednisone** to treat significant asthma exacerbations will be minimized by prescribing the shortest course of prednisone that is possible and monitoring for adverse events.

Although there are no proven harmful effects from the doses of radiation received during the **CT scans** proposed in this study, long term effects on the participant's health cannot be ruled out. Special precautions are taken for all of the procedures involving radiation to minimize the amount of radiation. This includes the use of low-dose CT scans when possible and the use of breast shields in the CT scans of women and adolescents.

2.2.2 KNOWN POTENTIAL BENEFITS

Although the goal of testing these add-on therapies is to find new therapies that improve lung function, asthma symptoms, or the risk of loss of asthma control events, there is no guaranteed benefit from participating in the trial. Some participants with asthma may achieve psychological benefit from involvement in an important research study and from interaction with the study staff. There is a possibility that the information obtained from participation in this study or treatments used will also directly benefit the participating participants. All research procedures will be completed free of charge to the research participants, including provision of asthma controller medications (if available). Participation in these studies may ultimately help in future understanding of the treatment and progression of respiratory diseases and learn more about the phenotypes that predict favorable and unfavorable responses to medications. Although some of the study procedures contain risks, the participants will be well monitored not only by the study personnel but also by the DSMB and the IRB. Because the study will be conducted under an IND, the study will provide reports to the FDA as required by federal regulations. Risks will be minimized on site by close monitoring, thorough training prior to and during the study, and appropriate written documentation (Manual of Procedures).

The knowledge to be gained from this study is important. Asthma is characterized by striking heterogeneity that influences its severity and responsiveness to treatment. The Severe Asthma Research Program (SARP) has found severe asthma to include multiple phenotypes that contribute to ongoing impairment and risks for exacerbations. However, precision in predicting treatment responses in severe asthma is limited, and the lack of data to inform treatment guidelines in these high-risk participants remains a major unmet need. As a next step to better identify modifiable treatment targets and more effective treatments, we have focused on establishing underlying molecular causes (endotypes) that drive phenotypic disease expression in severe asthma subpopulations. Although traditional parallel group or cross-over designs for randomized, placebo-controlled clinical trials can provide guidance as to the efficacy of test treatments, adaptive trial designs offer more flexibility and efficiency in selecting personalized treatment options for severe asthma. Our vision is to develop personalized medicine approaches for the treatment of severe asthma phenotypes that are directed at disease pathogenesis, and will facilitate drug development and ultimately impact clinical practice.

The risk/benefit assessment information for individual test treatments is found in Appendices I–V, Section 2.3.

3 OBJECTIVES AND ENDPOINTS

The study has two primary objectives:

1. Identify novel therapies that work in biomarker-defined subgroups of severe asthma patients
2. Optimize the subgroups targeted for treatment by refining the biomarkers and subgroup definitions

To achieve the first objective, we will perform hypothesis tests for comparisons of each intervention to placebo, (independently of the other interventions) for each of the two primary endpoints (defined below). An intervention will be considered efficacious if significant benefit is found with respect to either of the two primary endpoints. To achieve the second objective, we will enroll participants outside the target subgroups and optimize the target subgroup definitions using precision medicine methods performed during the final analysis.

The study also has two secondary objectives:

1. Gain information about potential response/pharmacodynamic biomarkers for selected therapies
2. Explore the safety and effectiveness of selected therapies in adolescent patients with severe asthma

For the first secondary objective, we will identify which response/pharmacodynamic biomarkers could potentially serve as early or intermediate outcomes for an investigation. These biomarkers may prove useful in predicting whether a treatment will be effective with respect to clinical outcomes, which could in turn inform future trials of the treatment. For the second, we will describe trends with respect to the primary efficacy outcomes observed in the adolescent subgroup and determine if the risk profile of an intervention appears to differ between adults and adolescents. We do not expect to have sufficient power for formal hypothesis tests of effects in the adolescent subgroup alone or for a test of age by treatment interactions.

We have defined two primary endpoints:

1) FEV_1 percent predicted, assessed prior to bronchodilator administration
2) Asthma symptom control, assessed via the Juniper Asthma Control Questionnaire (ACQ-6)

An intervention will be considered beneficial if it demonstrates improvement with respect to either of the two primary endpoints relative to placebo, and optimization of the subgroup to be targeted by each intervention will be performed taking both primary endpoints into account. See Section 8.1 for details about assessments for each of these two primary endpoints.

4 STUDY DESIGN

4.1 OVERALL DESIGN

The PrecISE Network is undertaking an adaptive platform trial conducted under a single Master Protocol to identify new therapies for severe asthma that are effective in biomarker-defined subgroups of participants. Five therapies will initially be investigated. The trial is designed to meet our primary objectives, namely, to: (1) Identify novel therapies that work in biomarker-defined subgroups of participants with severe asthma, and (2) Optimize the subgroups targeted for treatment by refining the biomarker and subgroup definitions.

For each participant, the study consists of three successive phases: (i) an initial screening phase, (ii) a 2-period crossover phase, and (iii) a single-period crossover phase with successive re-randomizations. Common platforms for biomarker screening will be used during the initial screening period to determine a participant's biomarker profile. Throughout the study, interventions will be randomly assigned based on these biomarker profiles. Participants will have a higher likelihood of assignment to an intervention that targets their particular profile. Subgroup definitions and associated treatment assignment probabilities are adapted as data accumulate throughout the study.

Following the initial screening period, participants will be randomly assigned to interventions based on their biomarker profile and enter a 2-period, double-blind crossover phase consisting of two 16-week treatment periods separated by a washout period. Treatment sequence will be randomly assigned as either test treatment followed by matching placebo or vice-versa.

At the conclusion of the 2-period crossover phase and a washout period, participants enter the single-period crossover phase of the study consisting of successive single 16-week treatment periods followed by washout periods. Participants are randomly assigned to interventions based on their initial biomarker profiles as ascertained during the screening period to reduce the likelihood of misidentification based on carry over effects from a treatment. The biomarker-based treatment assignment probabilities will be adapted during the study as data on the utility of the biomarkers in predicting treatment response accumulate.

The duration of the washout period following interventions with half-lives of 11 days or less is eight weeks. If the half-life of an intervention is longer than 11 days, the duration of the washout period will be determined such that the time from last administration of the intervention to the time of the start of the next treatment is equal to or exceeds five half-lives of the intervention (see Appendices I-V).

A small percentage of participants will receive placebo again in periods 3-6 to assess seasonal and sequence effects throughout the study (no participants will receive more than 3 placebos during the study). Masking of treatment assignments will be maintained by matching a participant's second placebo (if assigned) with one of the available interventions for which the participant is eligible but has not yet received (see Section 6.3). Placebo data will be shared across interventions for comparative analyses of efficacy and safety of each intervention, independently of the others in the study, so that the number of placebo periods are minimized (see Section 9.5).

Two of the interventions will be studied in adolescents. The treatment assignment probabilities at each period will be restricted accordingly.

Based on our power calculations and analysis assumptions, a total of 111 participants both within and outside a priori best subgroup are needed for the final efficacy analysis (see Section 9.2) of each intervention.

Reviews of safety data will be performed by the DSMB at periodic intervals throughout the study, and interventions found to pose significant safety risks in the severe asthma population will be dropped from the study. In addition, we plan to perform a single interim analysis for futility for each intervention (independently of the others) to allow for early stopping. Those that have not been eliminated due to either futility or safety will continue to accrue patients.

Interventions may enter the study at different times, depending on their availability, and may be discontinued, as noted above.

Our current study timelines and enrollment goals are designed so that each participant will receive up to five test treatments, depending on their time of enrollment.

4.2 SCIENTIFIC RATIONALE FOR STUDY DESIGN

As described above, participants with severe asthma who remain poorly controlled despite the availability of effective asthma therapies account for a large proportion of the morbidity and mortality associated with the disease. This phase 2 proof-of-concept trial is designed to provide preliminary evidence of effectiveness of up to six novel therapies that target specific phenotypes of severe asthma participants based on their biomarker profiles. Additionally, we believe that each therapy investigated in PrecISE has a differential effect across the biomarker profiles of participants. Consequently, this

platform trial has tremendous advantage over a traditional design in terms of efficiency of participant recruitment. The optimal subgroup of participants, defined by biomarker profiles that should be targeted by each therapy, will be determined through precision medicine analyses.

4.3 JUSTIFICATION FOR DOSE

The justifications for doses for individual test treatments are found in Appendices I-V, Section 4.3.

4.4 END OF STUDY DEFINITION

The study will end approximately 62 months after randomization begins. At the end of the study, participants will have completed varying numbers of treatment periods (test treatments and placebo) through the crossover design, depending on their enrollment dates. The number of periods will range from 2-6, with the majority of participants expected to complete four or fewer periods during the study.

An intervention may be stopped early for safety or futility. Consideration will not be given to stopping treatments based on early evidence of efficacy, to ensure sufficient power is available to support the final efficacy analysis and precision medicine analyses. If not stopped early, an intervention will be stopped when the total required sample size has been reached.

5 STUDY POPULATION

5.1 INCLUSION CRITERIA

To be eligible to enter the screening period, an individual must meet all of the following inclusion criteria:

1. Provision of signed and dated informed consent form
2. Stated willingness to comply with all study procedures and availability for the duration of the study
3. Male or female, age $\geq$ 12 years
4. Stable asthma medications
    a. No change in asthma medications for the past interval of time based on the participant's circumstances:
        i. Participants who have not been on a biologic (Table 1C) in the past 4 months must have all asthma medications stable for the last month
        ii. Participants currently on a biologic to be continued during PrecISE must be stable on that biologic for 2 months, with all other asthma medications stable for one month
        iii. Participant who stopped a biologic listed in Table 1C prior to screening must be free of treatment for 4 months. Other biologics not listed in Table 1C would be evaluated separately based on the half-life of the new medications.

    These rules also apply if the biologic (Table 1C) is prescribed for other conditions, such as atopic dermatitis etc.
    b. Use of medium or high dose inhaled corticosteroids (ICS) (defined by Table 1A) AND
    c. Use of an additional asthma controller/biologic (defined in Tables 1B and 1C) AND

d. For participants who enter into the run-in on medium dose ICS: they will be switched to high dose ICS; the participant's Visit 0 should occur at least 4 weeks after initiation of the high dose ICS; to qualify, the participant must meet one of the first two entry criteria for uncontrolled asthma (low FEV_1 with reversibility or high ACQ-6 score).

5. Verification of the asthma diagnosis at VSA or VSB demonstrated by either bronchodilator reversibility or methacholine responsiveness. This can be verified by direct measurement during the run-in or by historical evidence of either criterion. Historical evidence of testing must have been performed under either the 2017 ERS technical standard[9] or the 1999 ATS Guidelines[10], and full sets of flow volume loops require review and approval by the Spirometry core. These criteria are defined as:
   - An increase in FEV_1 ≥12% AND 200 ml after up to 8 puffs of albuterol OR
   - Positive methacholine defined as PD20 ≤400 mcg (PC20 ≤ 16 mg/ml for historic data).

6. Baseline poor or uncontrolled asthma at VSA and Visit 0, defined as meeting at least one of the following:
   - Poor symptom control – Asthma Control Questionnaire (ACQ-6) Score ≥1.5
   - Frequent exacerbations – Participants on high dose ICS at screening: ≥ 1 exacerbation defined as a documented burst of systemic corticosteroids (>3 days for adults and adolescents or >1 day for adolescents treated with dexamethasone) in prior year for those not receiving chronic OCS or an increase in >50% of baseline corticosteroid dose for ≥3 days in those receiving chronic OCS.
     - For patients on a biologic agent who do not meet criteria for low FEV_1 or ACQ score, at least one asthma exacerbation must have occurred at least 2 months after the initiation of the biologic agent. The definition of acceptable documentation for asthma exacerbations can be found in Section 6.5.3.
   - Low lung function with potential for improvement – During the run-in, the FEV_1 <80% predicted (for adults≥18) or FEV_1<90% (pediatric participants <18).
     - The potential to improve is defined at Visit 0 by an improvement of 12% OR 200 mL after up to eight puffs of bronchodilator has been administered.
7. Agreement to adhere to Lifestyle Considerations (see Section 5.4) throughout study duration
8. Owns or is able to obtain a device compatible with the eDiary system used for CompEx. As of February 2020, the system requirements are an iOS 11+ device such as iPhone, iPad or iPod, or a smartphone or tablet running on Android 5.0+.

Table 1A: Definitions of lower and high dose ICS daily dosing for protocol qualification

Inhaled corticosteroid (ICS)	Lower dose* (mcg)	High dose (mcg)
Beclomethasone (Qvar) – HFA (40,80)	160-320	>320
Budesonide (Symbicort) – HFA (80,160)[1]	320-639	≥640
Budesonide (Pulmicort Flexhaler) – DPI (90,180)[1]	360-720	>720
Budesonide (Pulmicort Respules) – nebulized[2]	500-999	≥1000
Budesonide (Breztri)	320 - 639	≥640
Ciclesonide (Alvesco) – HFA (80,160)	160-320	>320
Fluticasone furoate (BREO, Arnuity Ellipta, Trelegy Ellipta) – DPI	100-199	≥200
Fluticasone propionate (Flovent) – HFA (44,110,220)[3]	220-440	>440
Fluticasone propionate (Diskus, Armonair respiclick) – DPI (50,100,250)	250-500	>500
Fluticasone propionate (Advair) – DPI (100,250,500)	250-500	>500
Fluticasone propionate (Advair) – HFA (45,115,230)	230-460	>460
Fluticasone propionate (AirDuo) – DPI (113)	226-452	>452
Mometasone HFA (Asmanex HFA) (100,200)	200-400	>400
Mometasone (Dulera) – HFA (50,100,200)	200-400	>400
Mometasone (Asmanex Twisthaler) – DPI (110,220)	220-440	>440

HFA= Hydrofluoroalkane inhaler
DPI= Dry Powder Inhaler
*Doses from which patients can be switched to high dose in the run-in to ascertain whether they will meet entry criteria (these doses are also referred to as medium doses)
[1]Not included in GINA table
[2]Not included in GINA for adult dosing
[3]Converted to US product

Table 1B: Medications that qualify as an additional asthma controller

Long Acting Beta Agonists (LABA)
Long Acting Muscarinic Antagonist (LAMA)
Leukotriene Modifiers
Theophylline

Table 1C: Medications that qualify as biologics

Anti IgE : Omalizumab
Anti IL-5: Mepolizumab, Reslizumab, Benralizumab
Anti IL-4R: Dupilumab
Anti TSLP: Tezepelumab

5.2 EXCLUSION CRITERIA

An individual will be excluded from participation in this study if they meet any of the following criteria:

9. Current participation in an interventional trial (e.g. drugs, diets, etc.)
10. Enrollment in a clinical trial where the study medication was administered within the past 60 days or within 5 half-lives (whichever is greater)
11. Physician diagnosis of other chronic pulmonary disorders associated with asthma-like symptoms, including, but not limited to, cystic fibrosis (CF), chronic obstructive pulmonary disease (COPD), chronic bronchitis, emphysema, severe scoliosis or chest wall deformities that affect lung function, or congenital disorders of the lungs or airways.
12. Receiving one or more immune-modulating therapies for diseases other than asthma. This does not include biologics that are also approved for asthma.
13. Receiving methotrexate, mycophenolate (CellCept®), or azathioprine (Imuran®)
14. Receiving aero allergen immunotherapy and not on at least 3 months of maintenance allergen immunotherapy
15. Underwent a bronchial thermoplasty within the last two years
16. Premature birth
    – Born before 32 weeks of gestation
    – For those born between 32 and 35 weeks, we will exclude patients who went home from hospital on oxygen or have ever had a diagnosis of bronchopulmonary dysplasia (BPD) /Chronic lung disease (CLD).
        - Regarding the diagnosis of BPD and CLD, the investigator will discuss what is known about perinatal course, supported by medical record review, if available. If the patient received oxygen for 28 or more days or went home from the hospital on oxygen, then the patient will be excluded.
17. Uncontrolled hypertension, defined as systolic blood pressure > 160 mm/Hg or diastolic blood pressure > 100 mm/Hg
18. History of malignancy except non-melanoma skin cancer within the last five years

19. History of smoking

    Participants with a history of smoking who meet the following criteria may be included in PrecISE:

    - Participant is <30 years old, smoked < 5 years and none in the past year, and has a normal urine cotinine
    - Participant is ≥30 years old, smoked < 10 pack years and none in the past year, and has a normal urine cotinine
    - Participant is ≥ 40 years old, smoked 10-20 pack years and none in the past year, and has a normal urine cotinine and a normal DLCO (>70% predicted)

    If participant meets any of the following criteria, they are excluded:

    - Participant is <30 years old and smoked for ≥5 pack-years

    - Participant is 30-39 years old and smoked for ≥10 pack years
    - Participant is ≥40 years old and smoked ≥ 20 pack years

    * Smoking equivalent pack years. One pack of cigarettes a day for 1 year is equivalent to:
        - 1 cigar or pipe per day for 1 year
        - Smoked hookah or shisha =1 session per day for 1 year
        - Vaped e-cigarettes =0.5 mLs e-liquid per day for 1 year, or =1 cartridge/tank/pod per day for 1 year
        - 1 use of marijuana per day for 1 year

20. Active use of any inhalant >1 time weekly in the past year
    - Active smoking of conventional tobacco, inhaling of marijuana or other drugs, or vaping of e-cigarettes or vape pods >1 time per week in the past year.
    - Any form of tobacco qualifies, such as: 1 cigarette, 1 hookah or shisha sessions, 1 cigar, 1 pipe, etc.
    - Any electronic (e)-device included: e-cigarette e-cig, mod, vape pen, JUUL, e-cigar, e-hookah, e-pipe, vape pods, etc.
    - Any form of inhaled marijuana, including smoking marijuana leaves or inhaling THC via e-cigarette or device
21. Substance abuse within the last year
22. Unwillingness to practice medically acceptable birth control or complete abstinence during the study, current pregnancy, or lactation. Medically acceptable birth control/abstinence is defined as:
    - Career, lifestyle, or sexual orientation precludes intercourse with a male partner
    - For those in a monogamous relationship that precludes sexual activity with other partners, one of the sexual partners has been sterilized by vasectomy (in males) or hysterectomy and/or bilateral salpingo-oophorectomy (in females)
    - Use of highly effective methods of birth control defined as those, alone or in combination, that result in a low failure rate (i.e. less than 1% per year) when used consistently and correctly. Contraception should be used for at least 1 month prior to

screening, throughout study participation and for an additional 16 weeks after the end of the final test treatment.

For females of child-bearing potential, pregnancy tests will be given prior to study enrollment and at each clinic visit.

Each male participant will agree to inform his sexual partner(s) of the potential for harm to an unborn child. If a sexual partner becomes pregnant while he is participating in the study, he will notify study staff within 24 hours of receiving medical confirmation. His partner will be advised to promptly notify her doctor.

Any pregnancy (of a participant or a partner) will be monitored for adverse events with respect to pregnancy outcome until one month after birth.

23. Requirement for daily systemic corticosteroids above 10 mg of prednisone (or equivalent) per day for the past 2 months.
24. Respiratory infection within 1 month of screening
25. Intubation for asthma in the last 12 months
26. Use of warfarin, current or last 30 days
27. Any clinically significant abnormal findings in the history, physical examination, vital signs, electrocardiogram, hematology or clinical chemistry during run-in period, which in the opinion of the site investigator, may put the participant at risk because of his/her participation in the study, or may influence the results of the study, or the participant's ability to complete the entire duration of the study.
28. Additional exclusions for specific interventions (and not for others) are listed in the Appendices I-V, Section 5.2.

Safety Exclusion Criteria

Participants who meet the following criteria will be excluded from the study:

- Hemoglobin < 10 g/dL
- ANC <1000/µl for black participants, <1500/µl for other participants
- Lymphocytes <500/µl
- Platelet count <100,000/µl
- ALT/AST >2x ULN
- Bilirubin ≥2x ULN
- Estimated glomerular filtration rate (eGFR) <54 ml/min/1.73 sq m on two sequential tests performed within 2-4 weeks OR eGFR 54-60 ml/min/1.73 sq m and proteinuria detected on urine dipstick
- Positive Human Immunodeficiency Virus, Types 1 & 2 (HIV 1&2) Ab/Ag immunoassay followed by a confirmatory positive test (Geenius™ HIV-1/HIV-2 antibody differentiation immunoassay)
- Positive Hepatitis B surface Ag (active infection) or Hepatitis B core total antibody (marker of past infection that could reactivate)
- Positive Hepatitis C RNA test following positive Hepatitis C Antibody
- EKG with significant clinical findings

Adult participants (age≥18) will be screened for TB. A positive QuantiFERON-TB Gold test requires further screening. A participant may be included in PrecISE if at least one of the following criteria are met:

- A chest radiograph (CXR) done within the last six months of the test that shows no evidence of active TB

- A chest CT scan done within the last six months of the test showing no evidence of active TB
- Documentation of adequate treatment for latent TB.

In cases of an indeterminate QuantiFERON-TB test result, a second blood specimen must be drawn. A chest x-ray is not required if the participant has a negative QuantiFERON-TB Gold test.
Adolescent participants (age<18) will not be screened for TB.

Comorbid Conditions
Comorbidities are commonly present in severe asthma. Specific questionnaires will be used to identify common comorbidities as follows:

- Sleep apnea: STOP-BANG
- GERD (GERD- Questionnaire)
- VCD (Pittsburgh vocal cord dysfunction index)
- Chronic Rhinitis Sinusitis (Sinonasal questionnaire-SNQ5)
- Depression- Anxiety (Hospital anxiety and depression Scale: HADS)

These questionnaires are best used as screening tools. As such they typically have high sensitivity but relatively low specificity. Many of their symptoms overlap with the symptoms reported by participants with asthma who do not suffer from these conditions. Therefore, participants who meet the established cut offs for these questionnaires will need to be evaluated by the investigator to consider the clinical significance of the positive questionnaire based on history and physical and available testing. The investigator will need to judge the presence, severity and control of a specific condition and determine if it is sufficiently controlled to keep the participant in the PrecISE protocol. If the comorbid condition(s) is/are not adequately controlled, the investigator may refer the participant for further evaluation/treatment, prior to enrollment in PrecISE. Rescreening is permitted (after at least four weeks) to determine if the participant is able to move forward in PrecISE once the comorbid condition(s) is/are under adequate control. It is expected that some of the participants may also have other conditions such as cardiovascular disease, diabetes and obesity. These should be evaluated clinically as part of the complete history and physical done at initial evaluation. Their inclusions should be based on the investigator clinical judgement in line with good clinical practice principles.

5.3 RANDOMIZATION CRITERIA

In order to be eligible to enter the study following run-in period, an individual must meet all of the following additional randomization criteria:

- Must have stable asthma medications in the 4 weeks prior to visit 1.1 and no major change in therapy during the run in except for switching from medium to high dose ICS
- Must still meet criteria for poor baseline asthma control as defined above. For asthma exacerbation criteria, the exacerbation must have occurred within the past year from the start of the run-in period
- To be eligible for randomization, individuals must demonstrate adherence by:
    - taking controller medications ≥70% of scheduled doses and completing e-diaries and PEFs ≥70% [20/28 possible entries] during any 14 consecutive days among the 21 days prior to visit 0.
    - taking controller medications ≥70% of scheduled doses 28 days prior to visit 1.1
    - completing e-diaries and PEFs ≥70% [40/56 possible entries] 28 days prior to visit 1.1
    - completing e-diaries and PEFs ≥70% [20/28 possible entries] 14 days prior to Visit 1.1 for establishment of the CompEx baseline.

  All criteria assessed 3 days prior to visit 0 or visit 1.1

Electronic surveys, and occasionally paper diaries, may be used as a backup data source for recording e-diary and peak flow measurements if data are missing due to a technical issue with the electronic data system.

Individuals who do not meet the adherence criteria prior to target dates for visits 0 and 1.1 will be retrained, either in-person or by telephone. The scheduled visit will be postponed to allow the participant to bring adherence up to the 70% targets. If the participant has already been allowed 4 additional weeks in the screening period and does not meet adherence requirements, then he or she will be considered a screen failure. Depending on the circumstances, these individuals may be allowed to re-enroll at a later time. Participants may be contacted via phone to ensure safety and to assess any issues that may be ongoing during screening.

5.4 LIFESTYLE CONSIDERATIONS

Throughout the duration of this study, participants are asked to:

- Abstain from use of all tobacco products and nicotine containing products (including e-cigarettes, patches)
- Abstain from use of inhaled marijuana
- Abstain from use of any illegal drugs including abuse of prescription drugs

5.5 SCREEN FAILURES

Screen failures are defined as participants who consent to participate in the clinical trial but are not subsequently randomized to a test treatment. A minimal set of screen failure information is required to ensure transparent reporting of screen failure participants, to meet the Consolidated Standards of Reporting Trials (CONSORT) publishing requirements and to respond to queries from regulatory authorities. Minimal information includes demography, screen failure details, eligibility criteria, and any serious adverse event (SAE).

Screening visits (see Section 8.2) are designed to ensure that participants meet inclusion criteria for lung function and criteria for bronchodilator reversibility or methacholine reactivity.

Individuals must wait 4 weeks after screen failure to undergo rescreening. Participant's visit will be delayed rather than a rescreen, if he/she fails to meet the criteria for participation or randomization in the trial during the initial screening for reasons such as:

- An asthma exacerbation within two months of initiation screening/during run-in
- A respiratory infection within one month of screening
- Inability to demonstrate adherence to daily diaries, and PEFs of at least 70% for the two weeks prior to Visit 1.1

5.6 STRATEGIES FOR RECRUITMENT AND RETENTION

Asthma patients will be recruited largely from a variety of settings, via direct contact in clinical settings, advertisement, word of mouth, social media, mass mail and mass email; however, some severe asthma patients may come from hospitals and emergency departments, urgent cares and primary care practices A conscious effort will be made to have a balanced representation of males and females and various

ethnic groups. Participants who have already participated in studies focused on severe asthma, and have expressed interest in clinical trials would be an initial target for recruitment. Study goals, risks and benefits will be explained as part of the informed consent process. Various approaches will be considered to enhance participant retention including: scheduling flexibility, offering time with a physician investigator to examine the participant and answer their questions at each visit, thus building a connection and trust, regular phone call from the coordinators (or electronic messaging), postcard reminders, birthday cards and Thank You notes, updates on study progress (such as regular study newsletter targeted at the participants, celebrating milestones), participant support groups and invitation to provide input into future studies.

The PrecISE Network convened a Participant Advisory Council (PAC) which provided patient-level input about the potential experience of enrolling and participating in this trial. The insights gained from the PAC will be deployed in real-time to assist with recruitment and retention. As part of this effort, the network has also incorporated investigators with expertise in leveraging human-centered design methods in clinical trials to promote effective recruitment and retention strategies fit-for-purpose. Three PAC meetings were convened in Fall of 2018 and Winter of 2019 (also discussed in Section 2.2 Risk/Benefit Assessment). These meetings occurred in different regions of the US including the Southwest, Southeast, and Midwest and were each four hours long. A total of twenty adults participated and provided in-depth feedback on the length of study, visit schedule, randomizations and procedures, study medications, monitoring, compensation, recruitment and retention. In addition, one-on-one interviews were conducted with six adolescents and their primary caregiver to understand their perspective on these important aspects trial.

Recruitment and retention throughout the study will be monitored by the Recruitment and Retention Committee. This committee will oversee development of materials to facilitate recruitment and retention, monitor progress with respect to both as the study progresses, and develop strategies for addressing recruitment and retention issues as they arise. The DSMB will monitor accrual at each clinical center based on periodic summary reports provided by the DMCC for their review (see Section 10.1.6) and make recommendations on proposals to address recruitment or retention challenges, such as adding more clinical centers or extending enrollment, if needed.

6 STUDY INTERVENTION

6.1 STUDY INTERVENTION(S) ADMINISTRATION

6.1.1 STUDY INTERVENTION DESCRIPTION

Details about each of the interventions being considered for initial study are provided in Appendices I-V, Section 6.1.1. As new interventions are identified for study under the master protocol, additional appendices describing each will be added via protocol amendment.

6.1.2 DOSING AND ADMINISTRATION

The dosing and administration information for the initial five individual interventions is found in Appendices I-V, Section 6.1.2.

6.2 PREPARATION/HANDLING/STORAGE/ACCOUNTABILITY

The preparation/handling/storage/accountability information for the individual interventions is found in Appendices I-V, Section 6.2.

6.3 MEASURES TO MINIMIZE BIAS: RANDOMIZATION AND BLINDING

Placebo administration and masking

Each test treatment will have its own placebo that matches the test treatment in mode of administration, color, size, and/or taste. Each participant will receive at least one placebo during their participation in the network.

In the first two periods, each participant will receive a test treatment and its matching placebo, with the order being randomly determined (a two-period crossover design). After the first two periods, a test treatment will be randomly selected for each participant in each period. Participants may receive a second placebo during periods 3-6. The number of participants receiving a second placebo will be monitored throughout the study, and the probability of a second placebo assignment adjusted so that approximately 25% of participants receive a second placebo. Masking of treatment assignments will be maintained by matching a participant's additional placebo(s) (if assigned) with one of the available interventions for which the participant is eligible but has not yet received. Note that a participant may discontinue treatment early in their initial placebo period (period 1 or 2) due to safety or other reasons. In this case, the probability of additional placebo assignment(s) may be adjusted such that the participant may be assigned to placebo for at most three periods. Further details about randomization are provided in the Statistical Analysis Plan.

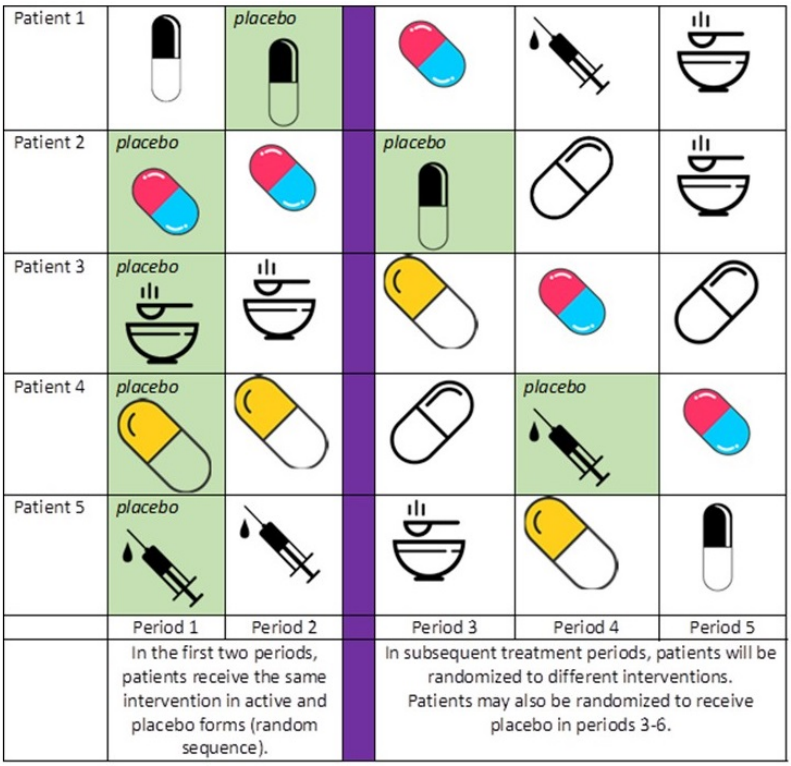
Examples of possible treatment sequences are shown in Figure 1. No participant will receive placebo in more than three periods unless the participant experiences a major change in therapy and is re-enrolled (Section 6.5.3). This strategy results in participants enrolled earlier, and therefore eligible for more treatment periods, having a higher chance of receiving a second placebo than participants enrolled later in the trial, when fewer treatment periods are available before the study ends.

The initial randomization to a complete two-period crossover design, with all participants receiving placebo during one of these two periods, allows for efficient estimation of treatment effects to support futility analysis of each test treatment early in the study. The addition of a randomly assigned additional placebo(s) allows for the estimation of time differences in placebo response and the potential to adjust the final efficacy analysis for any time trends observed. Because some participants will receive two placebos with different modes of administration (e.g., oral and injection or dietary and oral, etc.), we will also be able to estimate differences in placebo response between different modes of placebo administration.

The additional placebo(s) also provides some protection against expectation bias (relative to outcome assessments and other reporting behaviors) on the part of both the participants and the investigators/clinical staff, because there is a positive probability of placebo assignment in each treatment period for periods 3-6.

In addition to the safety assessments conducted on all participants throughout the trial (see Section 8), one or more interventions may require additional safety monitoring specific to that intervention (see Appendices I-V for additional safety requirements for each intervention). Participants randomly assigned to an intervention or its matching placebo will receive the treatment specific safety assessments required during the assigned periods. Performing the additional safety assessments on the matching placebo participants will help maintain the masking of treatment assignments for the study. Note also that to further maintain masking in periods 3-6, a participant can get a placebo matched to intervention A only if the participant has not yet received intervention A, and a participant who has completed a period with placebo matched to intervention A is not eligible to receive intervention A for the remainder of the study.

Figure 1: Example Treatment/Placebo Sequences



Treatment assignment

Predictive biomarkers will be used to define an initial subgroup to target for each intervention based on a priori information available at the time the intervention enters the trial. During the screening period, biomarkers will be assessed using common screening platforms to determine the biomarker profile for each participant. Based on these biomarker profiles, a determination will be made as to which participants should be targeted by which interventions. The biomarker profile determined at screening will be used to inform the randomizations at all treatment periods.

For the initial two-period crossover phase, participants will first be assigned to an intervention based on their biomarker profile using a biased-coin type design that favors treatments targeting the particular profile of most participants. A second randomization then occurs to determine the crossover sequence, i.e., whether participant receives test treatment followed by placebo (T:P) or placebo followed by test treatment (P:T). In subsequent treatment periods (periods 3-6), the same biased-coin design will be repeated to assign interventions, but restricted to interventions not yet received by a participant in prior periods.

The target subgroups initially defined for each intervention will have some degree of overlap, such that some participants will be targeted by more than one intervention. The randomization probabilities at any point in the trial will reflect the priorities in place at that time. For example, interventions that have not yet reached the required sample size to test for futility will be favored over interventions that have already enrolled enough participants to conduct the futility analysis.

Apart from intervention-specific predictive biomarkers, treatment assignment will take into account three factors thought to be prognostic for asthma outcomes: past-year exacerbations at the study's baseline (assessed during the initial screening period), blood eosinophils, and plasma IL-6 levels. The

goal is to ensure that participants contributing to the efficacy analysis (interim and final) of each test treatment resemble the severe asthma subpopulation with respect to these three factors. Initial targets for balancing with respect to these three factors will be based on the factor distributions in the SARP database. Randomization will be implemented in such a way that biomarker positive participants assigned to an intervention are likely to include at least 50% of the SARP distribution as far as baseline past year exacerbations ≥ 2. For example, if a random sample from the biomarker positive subgroup for a particular intervention would be expected to have 35% of participants with baseline past year exacerbations ≥ 2 based on SARP data, where possible, randomization will favor treatments that have less than 0.5*35% = 13% of participants with baseline past year exacerbations ≥ 2. Similarly, we will balance assignments with respect to blood eosinophils ≥300 µl and plasma IL-6 level ≥ 3.1. As data accumulate in the trial, the initial targets based on SARP distributions will be refined reflecting the PrecISE study population.

6.4 STUDY INTERVENTION COMPLIANCE

The following mechanisms will be employed to determine protocol and medication adherence and measure outcomes:

1. An electronic peak flow meter/e-diary device will be used to measure peak expiratory flows (PEF). The device will serve to obtain PEF and will function as a general adherence check (date and time are electronically recorded). Electronic measurements will be transmitted to a central platform and a compliance report will be reviewed with the participants. PrecISE coordinators will provide positive feedback to participants who demonstrate good adherence, and ongoing encouragement when warranted.

2. The PrecISE network will leverage the e-diary platform from Propeller Health (or other provider) to administer a daily question for self-report of intervention compliance. Qualtrics e-diaries have been developed as a backup to the e-diaries provided by Propeller Health. In addition, adherence with the e-diary symptom questions for use in the CompEx serves as a proxy for study procedure adherence in general. A financial compensation structure is planned to promote e-diary adherence, which will in turn promote intervention compliance. Recognizing that self-report may not be fully adequate in assessing intervention adherence, we will use the following procedures/measures to assess adherence as appropriate for each intervention (see appendices): counts of returned pills or capsules for oral formulations, counts of returned packets for powder formulations and counts of in-clinic injections.

3. Participants who take fewer than 60% of doses (active or placebo) required in a 16-week treatment period will be considered in violation of the protocol for that treatment period. The treatment period will be excluded from the per protocol analysis population (see Section 9.4).

6.5 CONCOMITANT THERAPY

Concomitant medications will be reviewed at each study visit. Participants who are receiving immune-modulating therapies for diseases other than asthma will not be permitted to enroll. Participants who are taking biological agents for asthma will be permitted to enroll in the study and will be asked to continue their current biologic throughout the study. Intervention-specific lists of excluded medications are included in Appendices I-V. Table 2 shows the required washout time prior to Screen Visit and visits with spirometry, BDR or methacholine. These withholds may also be used for home spirometry.

Table 2

	Washout prior to Screen Visit and visits with Spirometry, BDR or methacholine
Inhaled beta-adrenergic agonists (intermediate-acting, e.g., albuterol, terbutaline, metaproterenol, pirbuterol, bitolterol)	**≥ 6 hours**
Inhaled long-acting beta-agonists (e.g., Salmeterol/formoterol) Taken twice daily Taken once daily	**≥ 24 hrs** **≥ 36 hrs**
Inhaled anticholinergics Long acting taken twice daily Long acting taken once daily	**Short-acting: ≥6 hours** **Long acting BID: ≥24 hours** **Long acting QD: ≥36 hours**
Short-acting theophylline (e.g., Slophyllin tablets)	**≥ 12 hours**
Long-acting theophylline (e.g., Theo-Dur, Slo-bid, Theo24, Uniphyl)	**≥ 24 hours**
Leukotriene modifiers (Singulair, Accolate, Zyflo)	**≥ 24 hours**
Methylxanthine-containing foods or beverages (e.g., coffee, tea) or medications	**≥ 4 hours**

Chronic daily or intermittent use of the following medications is allowed (some exclusions may apply for specific interventions – see appendices):

- oral contraceptives and other hormonal forms of contraceptives (e.g., DepoProvera-7, Norplant-7)
- estrogen / progesterone replacement therapy for post-menopausal women
- vitamins and calcium supplements
- any nasal inhaled corticosteroid used at a stable dose throughout the entire study
- acetaminophen
- non-steroidal anti-inflammatory medications (e.g., aspirin, naproxen, ibuprofen, Cox-2 inhibitors)
- thyroid replacement medications
- lipid-lowering medication
- stable dose medical therapy for well-controlled hypertension and well-controlled diabetes, except those meds specifically excluded due to specific intervention exclusions
- medium and low potency topical cutaneous steroids
- nasal saline spray
- topical eye preparations for allergic eye symptoms (e.g. antihistamines, NSAIDs, or antiallergic compounds)
- diuretics and specific antihypertensives (e.g. calcium channel blockers, clonidine, etc.)
- acyclovir
- oral and topical antihistamines
- pseudoephedrine and oxymetazoline and other decongestants
- antibiotics for acne
- non-macrolide antibiotics
- macrolide antibiotics (both chronic and acute use)

- stool softeners and bulk laxatives
- H_2 blockers and proton pump inhibitors for GERD
- Imitrex for migraines
- Propecia (finasteride)
- SSRI class antidepressants
- non-SSRI antidepressants
- migraine analgesics (e.g., butalbital)
- antianxiety agents
- ACE inhibitors
- Librax
- CNS stimulants/appetite suppressants

Chronic use of the following medications is not allowed:

- Methotrexate
- Mycophenolate (CellCept®)
- Azathioprine (Imuran®)
- Immune-modulating therapies for diseases other than asthma

6.5.1 CONTROLLER MEDICATION

Participants must be taking an additional asthma controller/biologic (defined in Tables 1B and 1C in Section 5.1). Study sites provide controller medication from our supplier to participants beginning at the Screening Visit and at every subsequent visit while the participant is in the study. If a participant prefers to continue the controller medication, they regularly use rather than switch to the controller medication provided by the study, they will be allowed to do so. This information will be documented.

Participants will be encouraged to continue on the provided controller medication for the duration of the trial. If a subject decides to switch controller medication during the trial, this may be allowed, but the cost will not be covered by the trial. Participants will be encouraged to wait until the end of their current 4-month treatment period before changing medications. See Section 6.5.4 for more information on allowed changes in controller medication during the trial.

6.5.2 RESCUE MEDICINE

Study sites provide inhaled rescue medication from our supplier to participants beginning at the Screening Visit and at every subsequent visit while the participant is in the study. Participants may use their personal rescue medication as needed as prescribed by their treating physician if they prefer it to the study supplied rescue medication.

- Rescue medications may include: albuterol, levalbuterol, ipratroprium and albuterol/Ipratropium. Participants may not use ICS/LABA combination drugs for rescue – i.e. no SMART therapy.
- The use of rescue medications is allowable at any time during the study.
- The study will record the use of rescue medications.

6.5.3 EPISODES OF WORSENING ASTHMA/EXACERBATIONS

An asthma exacerbation in PrecISE is defined as a worsening of asthma requiring the use of a systemic corticosteroid (at least 3 days of treatment) to prevent a serious outcome. This definition is consistent with recommendations from the NIH Outcomes Workshop.[11] Events meeting one or more of the following criteria will be recorded as asthma exacerbations during the study:

- All worsening asthma events in which systemic corticosteroids were initiated to prevent a serious outcome, including use of systemic corticosteroids in association with any form of healthcare provider encounter
- All asthma-specific emergency department or urgent care visits that involved treatment with systemic corticosteroids (defined as evaluation and treatment for <24 hours in an ED or urgent care center)
- All asthma-specific hospitalizations that involved treatment with systemic corticosteroids (defined as admission to an inpatient facility and/or evaluation and treatment in a healthcare facility for ≥24 hours) (also reported as a serious adverse event)
- All asthma-specific intensive care unit admissions or intubations (also reported as a serious adverse event)
- All asthma-related deaths (also reported as a serious adverse event)

For the purposes of this study, a course of oral systemic corticosteroids (OCS) is defined as any increase of at least 20 mg prednisone or double the daily maintenance dose for a minimum of 3 days. Note that a single depo-injectable dose of corticosteroids or oral dexamethasone will be considered equivalent to a 3-day course of OCS. If the maintenance dose is <5 mg, an increase must be both a doubling of that dose and at least a 5 mg increase (e.g., 2.5 mg would need to increase to at least 7.5 mg to count as a course).

The recommended course of OCS for treatment of an asthma exacerbation that occurs during the study is prednisone 60 mg x 3 days, then tapering by 10 mg each day (60-60-60-50-40-30-20-10-0). If the patient is receiving OCS on a daily basis, the taper ends at that dose. A patient receiving a course of OCS that differs from this recommendation will still be considered as having had an exacerbation, provided one of the defining criteria for an asthma exacerbation listed above is met.

Two courses of systemic corticosteroids must be separated by at least two weeks to count as two exacerbations. The two-week clock starts when the corticosteroid course for the first exacerbation ends.

Exacerbations may result in treatment discontinuation and re-randomization (see Section 7.1) or withdrawal from the study (see Section 7.2).

If no asthma symptoms are present when a course of OCS is started during the study, the OCS use should be reported as a concomitant medication, but the event would not qualify as an asthma exacerbation. For example, if an adult participant starts an OCS course because a family member developed a viral respiratory infection, this OCS course would not be considered a steroid course for asthma, but it would be recorded as a concomitant medication.

Acceptable documentation to support an exacerbation, in order of preference, is as follows:

- Discharge summaries from a hospital, emergency room, or an urgent care facility indicating that a subject was hospitalized/treated with systemic corticosteroids for an asthma exacerbation

- Signed and dated notes from a referring physician, including information regarding diagnosis and treatment of an exacerbation with systemic steroids
- Evidence of prescriptions provided by the participant for systemic corticosteroids used during an exacerbation
- A documented conversation that is recorded in a timely manner between the investigator/nurse or nurse practitioner and a subject who is already on an OCS action plan, detailing the diagnosis and treatment of an asthma exacerbation.
- A documented conversation between the treating/referral physician, health care provider, or nurse/nurse practitioner certifying that a participant was treated for an exacerbation with corticosteroids at their clinic or under their supervision. The dates (month/year) of the exacerbations and verbal confirmation that appropriate prescriptions were provided is necessary. This option should be used only if reasonable attempts to procure participant records have been unsuccessful.

Participants may receive treatment for asthma exacerbations during the study outside of the PrecISE study clinics. We recognize that participants may live some distance from a study site; may wish to have exacerbations managed by their usual provider; may be at work, traveling, or away from home when the exacerbation occurs; and/or may have health insurance that precludes treatment at the PrecISE study site or affiliated hospital. The ethical management of asthma exacerbations in study participants requires that 1) we are prepared to help participants manage these exacerbations, 2) we work to ensure the safety of participants when they seek assistance for an exacerbation, and 3) we work collegially with other health care providers who may be called upon to treat a PrecISE participant.

An informational pamphlet will be provided to each study participant that they can give to a provider in the event of an asthma exacerbation. This pamphlet will inform the provider of: a) the general nature of the PrecISE study, b) the types of interventions in the study, and c) contact information for the study sites.

6.5.4 MAJOR CHANGES IN THERAPY

The PrecISE study has special challenges, as participants have severe and/or exacerbation prone asthma not adequately controlled on high-dose ICS plus additional therapy such as a LABA, LAMA, oral CSs, or all three, they may already be on a biologic for their asthma due to disease severity, and they may be in the study for as long as 40 months. As a result, some participants may have their treatment changed while in the study. The number of participants in whom this occurs is difficult to predict, but as more treatments, especially biologics, for severe asthma become available, it is likely to increase.

A *major change* is:

- the addition, dose adjustment (see below) or discontinuation of an oral corticosteroid for a period of time greater than that specified under treatment of exacerbations (Section 6.5.3) with the intention of continuing such therapy.
- the addition, dose adjustment, or period adjustment of a parenteral corticosteroid administered during the study;
- the addition, dose adjustment, or discontinuation of an additional asthma controller (note that changes within a class of therapy that do not change the dose of that type of therapy are not major changes)
- the addition, dose adjustment, or discontinuation of a biologic therapy believed to affect asthma,
- the addition, dose adjustment, or discontinuation of a beta blocker,

- the addition or discontinuation of azithromycin to a participant's baseline (chronic) regimen for the treatment of asthma.

A major change in asthma therapy typically occurs due to worsening (or dramatic improvement of) asthma, or a perception of worsening asthma by a treating physician, or the availability of a new therapy the treating physician wants to prescribe in the absence of a change in the participant's overall status. Such a change generally occurs outside the period of an asthma exacerbation.

A 5 mg/d change in prednisone (or equivalent) therapy for patients on chronic therapy, outside the strict definition of treatment for an asthma exacerbation in Section 6.5.3, is considered a major change in therapy. Additionally, since the maximum dose of oral prednisone allowed for entry into the study is 10 mg, an increase in oral prednisone therapy to greater than 10 mg daily, outside the strict definition of treatment for an asthma exacerbation in Section 6.5.3, is considered a major change in therapy.

The addition, dose modification, or discontinuation of any immune-modulating therapy, thought to potentially affect asthma pathobiology, for non-asthmatic disease will also be considered a major change in therapy. These treatment changes make it difficult to assess the specific effect of the asthma intervention.

It is expressly noted that a change within a class of therapy that does not change the total dose of that type of therapy is not considered a major change. For example, the patient's insurer may decide to cover one brand of combination ICS/LABA therapy, and the patient may then want or need to switch from the brand he/she is currently using. As another example, a patient may change (for insurance reasons) from one anti-IL-5 biologic to another. These are not considered major changes, whereas a change from one biologic to another for purposes of achieving increased therapeutic efficacy would be considered a major change.

A major change in therapy can occur during a treatment period (test treatment or placebo) or a washout period. In most asthma studies with an intervention lasting up to 12 months, major therapy changes (e.g., addition of a biologic) are not permitted. Consistent with this practice, major changes in therapy will be discouraged during the 4-month treatment periods in PrecISE.

If a participant has a major change in therapy during a treatment period, he/she will discontinue their current assigned treatment, and then discontinue the study. The following guidelines apply:

- Changes that require removal of a therapy, or a decreased dose of therapy, should include the reason for the removal (e.g., adverse event from the medication).
- Likewise, changes that require a new medication for asthma control (e.g., introduction of an anti-IL-5 directed biologic) should include the reason for the addition. When feasible, it is recommended that such changes be done at the end of, rather than in the middle of, an intervention. However, patient safety is paramount in such additions of therapy, and the PrecISE study will not override the judgment of the treating physician.
- Following discontinuation from the study due to a major change in therapy, the participant has the option of re-entering the study as a new participant. Inclusion criteria will be re-assessed to see if the patient still meets the criteria for uncontrolled severe asthma after the change in therapy. The patient will undergo the usual screening and run-in process and additional specimens will be collected. The participant's biomarker profiled will be reassessed to determine randomization probabilities. The participant will not receive any interventions already received when previously enrolled.

7 STUDY INTERVENTION DISCONTINUATION AND PARTICIPANT DISCONTINUATION/WITHDRAWAL

7.1 DISCONTINUATION OF STUDY INTERVENTION

7.1.1 DISCONTINUATION OF A PARTICIPANT'S ASSIGNED INTERVENTION

Participants will be monitored for safety during the trial and the occurrence of certain adverse events or laboratory abnormalities will result in discontinuation from the participant's current treatment; that is, participation in the current treatment period will stop, and the participant will enter the washout period. The following adverse events or lab abnormalities will result in discontinuation from the participant's current treatment:

1. Serious infection (including pneumonia, cellulitis, kidney or neurologic infections, and any bacteremia/sepsis).
2. Any bleeding event thought to be associated with a platelet count reduction per the judgment of the center investigator.
3. Any other adverse event, per judgment of the center investigator, requiring discontinuation from treatment.
4. Laboratory values are monitored per the schedule of activities in Section 1.3. For adults, the below lab tests will be assessed at every 4-week visit during the treatment period. Abnormalities in the following laboratory values require prompt retesting within 3 to 5 days:

    - ANC <1000/µl for black participants, <1500/µl for other participants.
    - Lymphocyte count <500/mm3.
    - Platelet count <50,000/mm3.
    - Hemoglobin <9.0 g/dL.
    - AST and/or ALT elevation >3 times the upper limit of normal.
    - Bilirubin ≥2 times the upper limit of normal.
    - Calculated eGFR < 54 mL/min/1.73 m^2 and baseline eGFR was >60 mL/min/1.73 m^2 OR calculated eGFR < 50 mL/min/1.73 m^2 and baseline eGFR was 54-60 mL/min/1.73 m^2

    If both tests violate the following cut-off values, the participant is removed from treatment:

    a. Two sequential platelet counts <50,000/mm3.
    b. One platelet count <25,000/mm3 unless the confirmatory test demonstrates a count >50,000/mm3. Treatment must be withheld pending the confirmatory retest.
    c. Two sequential neutrophil counts <500/mm3.
    d. Two sequential lymphocyte counts <500/mm3.
    e. Two sequential hemoglobin assessments <8.0 g/dL or >30% below the participant's initial baseline value or the baseline value for the current treatment period.
    f. Two sequential AST or ALT elevations >3 times the upper limit of normal with at least one total bilirubin value ≥2 times the upper limit of normal.
    g. Two sequential AST or ALT elevations >3 times the upper limit of normal accompanied by symptoms consistent with hepatic injury, per the judgment of the center investigator.

h. Two sequential AST or ALT elevations >5 times the upper limit of normal, regardless of total bilirubin or accompanying symptoms.
i. Two sequential decreases in eGFR < 54 mL/min/1.73 m^2 and baseline eGFR was >60 mL/min/1.73 m^2 OR eGFR < 50 mL/min/1.73 m^2 and baseline eGFR was 54-60 mL/min/1.73 m^2

5. Any safety issue that has required or will require (per the investigator's judgment) interruption to treatment for >28 days.
6. Any event (i.e. asthma exacerbation, asthma treatment, or any other medical condition) requiring 21 or more contiguous days of prednisone above the participant's usual baseline dose.

Specific interventions may have additional safety criteria for discontinuing treatment (see appendices).

A participant will be discontinued from treatment if, during an exacerbation, he/she

- Is admitted to an ICU and requires non-invasive positive pressure ventilation or
- Experiences three exacerbations during a treatment period.

If, following treatment discontinuation for any of the criteria listed above, participants are deemed eligible (by safety criteria) to continue in the study, they will enter the 8-week (or longer) washout period preceding the next intervention. Entry into the washout period will begin at least 7 days after completing treatment of the exacerbation with oral or parenteral corticosteroids.

Participants who respond to treatment with systemic corticosteroids or who receive a single IM injection of steroid at the beginning of an exacerbation will not be discontinued from treatment, unless this is their 3rd exacerbation within the same treatment period, in which case, the above treatment discontinuation criteria apply.

7.1.2 DISCONTINUATION OF AN INTERVENTION FROM THE MASTER PROTOCOL

The DSMB will review accumulating data throughout the study to determine if an intervention should be terminated from the PrecISE study for safety reasons. Interventions may also be discontinued from the study based on the interim futility analyses planned for the trial. Rules for termination for specific interventions due to safety may be required and, if so, will be specified in the appendix to this protocol for the intervention. If an intervention is discontinued from the study for either futility or safety reasons, participants who are receiving the intervention or its matching placebo will immediately begin an 8-week (or longer) washout phase and be re-randomized to another treatment.

7.1.3 DISCONTINUATION OF THE STUDY

At this time, we do not anticipate there will be a reason to stop the entire PrecISE study, with the exceptions of termination of funding by NHLBI or poor recruitment to the point where the study is highly unlikely to meet its goals.

7.2 PARTICIPANT DISCONTINUATION/WITHDRAWAL FROM THE STUDY

A participant may be discontinued from the study if he/she is shown to not meet the inclusion criteria based on new information that was not available at the time of initial enrollment (e.g. uncovering a diagnosis of another lung disease), and continuing in the study poses a safety risk. Participants should not be discontinued for reasons other than new information that constitutes a safety risk. For example, if new information indicates that a participant meets one of the exclusion criteria for a test treatment, but the criteria does not pose a safety risk to the participant, then the participant should not be

discontinued. In this case, the participant will be considered in violation of the protocol for that period and excluded from the per protocol analysis (see Section 9.4). In general, participants with poor compliance but at no increased risk will continue in the study to complete all study assessments and minimize attrition.

Other criteria for participant discontinuation at any point in the study:

1. Safety laboratory exclusions as defined in Sections 5.2 and 7.1 that do not resolve in the washout phase that in the judgment of the site physician or Safety Committee constitutes an on-going safety risk for the participant.
2. Participant or parent of participant (for children) withdraws consent or child withdraws assent.
3. Participant becomes pregnant.
4. Study physician determines that continuation in the study is not in the best interest of the participant.
5. Participant suffers life-threatening asthma exacerbation. A life-threatening asthma exacerbation is defined as an ICU admission requiring intubation, hypoxic seizure or loss of consciousness.
6. Frequent asthma exacerbations
    a. For participants with 3 or fewer asthma exacerbations in the prior year at screening, the participant has five or more exacerbations in one year of the study.
    b. For participants with 4 or more asthma exacerbations in the prior year at screening, the participant has a 50 percent or greater increase in exacerbations in one year of the study, compared to the prior year at screening.
7. Frequent hospitalizations
    a. For participants who have 3 or fewer hospitalizations in the prior year at screening, the participant has four or more hospitalizations (greater than 24 hours) in one year of the study.
    b. For participants who have 4 or more hospitalizations in the prior year at screening, the participant has a 50 percent or greater increase in hospitalizations in one year of the study, compared to the prior year at screening.
8. Participant has a major change in therapy (see Section 6.5.4).

As always, the participant can withdraw consent from the master protocol at any point, and that will lead to discontinuation from the study. At that point, the investigator will make reasonable effort to ensure participant's well-being and safety and provide a termination visit with appropriate documentation.

The site investigator may decide to stop a participant's treatment due to side effects or other safety concerns. In that case, the participant would immediately begin the washout phase and then receive the treatment assigned according to their initial crossover sequence (period 2), or be re-randomized for the next treatment (period 3-6). If a participant elects to withdraw their consent from a treatment, this will result in discontinuation from the entire study.

7.3 LOST TO FOLLOW-UP

Participants who do not show up for a scheduled visit will be contacted by available means (calling, email, text). If an emergency number is provided, it can be used to contact the participant, at least to ensure they are doing well. The study coordinator will also contact the patient's referring physician. After multiple attempts during the 16-week treatment period, if the participant fails to respond to voice

and written requests to contact the clinical site, he or she can be considered to have been lost to follow-up. A termination form should be completed by the investigator to record the exit of the participant from the study.

8 STUDY ASSESSMENTS AND PROCEDURES

8.1 EFFICACY ASSESSMENTS

Primary Efficacy Assessments: Efficacy of each intervention will be evaluated with respect to lung function and asthma symptom control. Lung function will be measured prior to and post bronchodilator administration (pre-BD and post-BD). The American Thoracic Society (ATS) criteria for assessing lung function will be applied for spirometric maneuvers. A central spirometry reading center will provide quality control over-reads of the measurements and identify any issues requiring retraining of clinical staff. The primary efficacy outcome with respect to lung function is FEV_1 percent predicted pre-BD.

Asthma symptom control will be measured using six items from the Juniper Asthma Control Questionnaire (ACQ-6) corresponding to five self-reported symptoms and self-reported rescue BD use. The primary efficacy outcome with respect to asthma control symptoms is the average score of these six items (range 0-6).

Secondary Efficacy Assessments: The use of CompEx events as a secondary endpoint provides a measure of loss of asthma control with statistical properties that approximate exacerbations but with a shorter follow-up time.[4] CompEx is a composite outcome specific to asthma that combines clinically-relevant deteriorations captured by diary events with exacerbations, thereby providing an increase in power compared to using exacerbations alone (see Section 9.2). Exacerbations will be analyzed as a secondary efficacy endpoint.

CompEx events include exacerbations and deterioration events defined based on

1) Daily recordings of peak expiratory flow (PEF) morning/evening (L/min)
2) Reliever use morning/evening (doses)
3) Symptoms morning/evening (score 0–3) assessed from twice-daily diary recordings

Participants will be asked to describe their morning symptoms using the following scale: 0-No symptoms to report, 1-I was aware of my symptoms but they were easily tolerated, 2- I had problems sleeping due to my asthma, 3-I could not sleep because of my asthma. Participants will be asked to describe their evening symptoms using the following scale: 0-No symptoms to report, 1-I was aware of my symptoms but they were easily tolerated, 2- I had problems performing my normal activities due to my symptoms. 3-I could not perform my normal activities because of my asthma.

The diary event can occur as defined by threshold and slope criteria within a moving window of 5-day length. Evening and morning recordings are treated as separate variables. The baseline levels of deterioration are calculated for each individual as the mean over the 5-10 days ending just before the day of randomization for each of the diary variables. No imputation of missing diary data after the randomization is performed. Deterioration criteria is assessed for each (single) diary variable for thresholds and slopes as follows:

Thresholds: The change from baseline is calculated. If two consecutive days fulfil the chosen threshold limit as defined in Table 3, the deterioration criterion is met.

Slopes: A slope is calculated via linear regression over 5 days. If the slope fulfills the chosen cut-point as defined in Table 3, the deterioration criterion is met.

Diary event start definition: A diary event can occur when (i) the threshold deterioration criterion is met for at least two diary variables, or when (ii) the threshold deterioration criterion is met for one diary variable, and the slope criterion is fulfilled for all included variables. In case of (i), the diary event is defined to start on the first day of the two consecutive deterioration days (event days 0–1). Any missing data in this two-day window will make the event missing. In case of (ii), the event is defined to start on the first of the two days fulfilling the threshold criterion. This means that the slopes are calculated for days -4 to 0 of an event. At least two days with data are needed to calculate slopes in order to qualify as an event. The end of a diary event is the last day that the criteria for a diary event is fulfilled. In order to be counted as a new diary event it must be preceded by at least 7 days in which neither criterion for a diary event is fulfilled.

Table 3

Diary variable	**PEF (P) morning/evening**	**Reliever use (R) morning/evening**	**Symptoms (S) morning/evening**
Threshold type	Decrease from baseline (%)	Increase from baseline (doses)	Increase from baseline (scores) or absolute maximum score
Threshold	15	1.5	1
Slope type	Decrease rate (% per day)	Increase rate (doses per day)	Increase rate (scores per day)
Slope	3	0.3	0.2

Other secondary efficacy assessments

1. FEV_1, post-BD
2. FVC, pre-BD
3. Exacerbations, time to first (as defined in Section 6.5.3)
4. Symptom Free days (defined using diary/CompEx data)
5. Healthcare utilization
    a. Asthma-specific ED visits
    b. Asthma-specific hospital admissions
    c. Asthma-specific ICU admissions
6. Asthma free days using the AsthmaNet definition[12,13]

Exploratory efficacy assessments

1. Rate of recovery from exacerbation, time to improvement
2. Economic burden of disease assessment (Days missed from work/school, cost of hospitalizations)
3. AQLQ (Asthma Quality of Life Questionnaire)
4. WPAI (Work Productivity and Activity Impairment Questionnaire)[14]
5. Modified ASSESS (Asthma Severity Scoring System)[15]

8.2 SAFETY AND OTHER ASSESSMENTS

All participants who enter PrecISE will undergo a series of visits, each with different intent and visit-specific assessments. Some of these assessments and procedures may be conducted remotely via phone, videoconference or electronic survey methods. Please refer to the visit structure (Section 1.3) for specific details related to these assessments.

1. Discussion and signing of informed consent and registration into the study. Informed consent will be obtained from the participant or the legal guardian if the participant is a child less than 18 years of age. Assent will also be obtained from adolescent participants. The informed consent will include the core characterization and longitudinal procedures to be conducted at all sites, including the sharing of data and the storage and distribution of samples.
2. Assessments for inclusion/exclusion related to asthma status and overall general health as directed by the clinical coordinators and/or attending medical personnel. These assessments will include the following:
    a. Full medical history obtained by the clinical coordinator. This will include vital signs, height, and weight. Further, the medical history will collect data related to family history, recent asthma symptoms and acute episodes of asthma, asthma triggers, allergies, and smoking history. Details will be collected on prior disease, illnesses, and surgeries the participant has had. Mediation history will be collected. Smoking history will be collected, including use of inhaled nicotine products.
    b. Information on the use of asthma treatments at school will be obtained from middle and high school participants. Information will be collected about the location of the participant's rescue medication while at school (self-carry versus nurse's office), prior experience with the most recent asthma attack at school and whether the school allows the participant to receive medications during the school day.
    c. Physical examination by qualified medical professional. This examination will take place at the beginning of the study and before each new treatment arm. Additional, non-scheduled physical examinations will take place if needed during the study to support patient safety and exacerbation management.
    d. Comorbid condition surveys/questionnaires. These surveys are conducted with the purpose of determining the general health of the potential participants and their feasibility for inclusion as determined by the attending physicians. Based upon these surveys and questionnaire, the attending physician may decide to treat the comorbidity and invite the participant back for reevaluation or the attending physician may decide that the comorbidity is too severe for the participant to enter the study. These surveys/questionnaires include:
        i. Sleep (STOP-BANG)
        ii. Sinus disease (SNQ5)
        iii. Depression/Anxiety (HADS)
        iv. Vocal cord dysfunction (Pittsburgh vocal dysfunction survey)
        v. Gastroesophageal reflux disease (using the GERDQ to establish uncontrolled GERD)
    e. Pulmonary function tests (PFT). Spirometry before and after bronchodilator (up to 8 puffs of albuterol during screening for qualification for reversibility and 4 puffs of albuterol subsequently for standardized degree of reversibility) will be used during the

screening visits and throughout the study to capture key lung function parameters, such as pre- and post-BD forced expiratory volume, which is used to monitor both asthma control and treatment outcomes. Spirometry is a maneuver whereby a participant will inhale as big a breath as possible and then blow it out as fast and as long as possible until no more air can leave the lungs. The volume of air leaving the lungs is recorded continuously. Spirometry will be conducted according to the SOPs defined in the MOPs. As a result of the COVID-19 pandemic, spirometry may be conducted at home or in the clinic. For home spirometry assessments, a portable spirometer (see 3.c below) is used by participants in conjunction with application software to record the maneuvers.

f. Methacholine challenge studies will be used to determine asthma status for a subset of participants who fail to bronchodilate during standard pulmonary function testing, yet who are suspected to have severe asthma based upon medical history. Positive methacholine challenge will be needed for those participants to enter the study. During this maneuver, the participant will be asked to inhale a small dose of methacholine, followed by spirometry. If there is no change in baseline after the challenge, additional methacholine is delivered until a significant drop (≥ 20% of baseline) in lung function or symptoms occurs. Bronchodilators are then given to reverse the effects.
g. DLCO (if needed based on smoking history). This assessment will be conducted to establish that potential participants with a smoking history, who say they no longer smoke, do not have evidence of underlying smoking-related disease, such as emphysema related to chronic obstructive pulmonary disease. Evidence of underlying lung disease associated with previous smoking history will be an exclusion criterion.
h. Urine cotinine (if needed based on smoking history). The assessment will be conducted to establish that potential participants with a smoking history, who say they no longer smoke, are indeed not smoking. A positive cotinine for smoking will be an exclusion criterion.
i. Socioeconomic information for the household. This information will be collected to determine in post hoc analyses if treatment effects are related to important socioeconomic factors.
j. Immunoglobulin E. This test will be used to provide basic, necessary phenotyping of the severe asthma population in PrecISE. This evaluation will occur from a blood sample and only once during the recruitment visits.

3. Additional surveys/questionnaires will be used throughout the study to capture other relevant features. These include:

    a. Asthma Control Questionnaire (ACQ-6) will be used throughout the study to capture asthma symptoms.
    b. The Asthma Quality of Life Questionnaire (AQLQ) and the WPAI (work productivity) questionnaires will be used throughout the study to capture the quality of life endpoints.
    c. Issuing of E-Diary (and E-Diary training) will occur throughout the study in order to capture the participants' day-to-day medication use, asthma symptoms, and home spirometry. Participants will be provided with an e-diary, which they will be asked to maintain at home throughout their participation in the study. Electronic surveys, and occasionally paper forms, will be available as a back-up.
        i. If adherence < 70% to controller medications and/or E-Diary is determined to be occurring throughout the course of the study, in-person visits or telephone calls may be scheduled for adherence training.

ii. The E-Diary will also capture the questions related to the CompEx events end-point.
iii. Participants will also be asked to utilize a home spirometer to capture daily peak flow, which is a component of the CompEx events end-point.

d. A second home spirometer will be issued to collect FEV1 data by the ZEPHYRx system at study visits or at home.
e. Asthma medication and concurrent medication assessment
f. Exacerbation and adverse event evaluation
g. Blinding Index questionnaire to capture participant's perception of whether they received an active treatment or a matching placebo in the current period

4. Safety labs (primarily used to establish the health of the participant and potentially to exclude participant from certain treatments). All safety labs will be conducted in a CLIA certified Core Laboratory selected by the PrecISE Network to ensure standardization of test reads across clinical sites. Most safety labs will be performed at a primary laboratory to ensure standardization of test reads across clinical sites. In this scenario, biological specimens are collected from the participants and the specimens are immediately sent to the Core Laboratory. Some safety labs may be performed at patient service centers. In all cases, results will be communicated back to the clinical centers and/or the DMCC where they will be analyzed and used for randomization/exclusion decisions. Further, safety labs will be collected throughout treatment periods and before each new treatment arm.
    a. Urine Pregnancy test for all women of childbearing potential will be utilized in order to exclude pregnant individuals from the study. This test will be conducted at the sites using a urine dipstick.
    b. Complete blood cell count with differentials
    c. Comprehensive metabolic panel
    d. Additional safety labs will be performed on all participants that are to be randomized and these may be further evaluated during the treatment in an intervention-specific manner. Based upon the values of these test, some participants may be excluded from certain treatments. Details of these intervention-specific safety laboratory tests are outlined in the Appendices. These include
        i. Triglycerides
        ii. Ketonuria and/or blood glucose
        iii. Serum phosphate
        iv. Hepatitis B and Hepatitis C testing
        v. HIV testing
        vi. QuantiFERON test for tuberculosis
5. To obtain information needed for randomization, the following biomarkers will be obtained (refer to the visit structure for details of timing).
    a. Blood eosinophils (will come from complete blood count; in most cases the blood for this is also drawn for safety reasons) will be taken utilizing standard phlebotomy.
    b. Single nucleotide polymorphism (SNP) genotypes. Limited genotypes (intervention-specific) will be determined from a blood sample used to isolate DNA. The SNP genotype analysis will be conducted in a CLIA-certified Core Laboratory.
    c. Plasma IL-6 will be obtained from EDTA treated plasma
    d. Exhaled nitric oxide level will be determined using an instrument to measure fractional exhaled nitric oxide (FeNO). This instrument will be used during the visits outlined in the visit structure. The test is easy to conduct and requires that the participant exhale

into the mouthpiece of the FeNO measuring instrument for approximately 10 seconds. SOPs and MOPs will be developed to ensure standardization of the test.

6. Specimen collection for biomarker and subsequent exploratory studies. In general, the samples collected for this purpose will be processed after collection at the collection site and aliquoted according to standardized MOPs and SOPs, and shipped quarterly to the PrecISE Network Biorepository Core. The Biorepository Core will store de-identified barcoded samples. When requested by the PrecISE study investigators according to study needs and priorities, the samples will be sent to appointed laboratories for specific-analyses testing. The following provides a general guideline of possible samples collected and procedures performed.
    a. Blood: At various times throughout the study (refer to visit structure) blood will be drawn for processing and subsequent biomarker exploration. Standardized phlebotomy techniques outlined in SOPs and MOPs will be conducted. At no time will maximal blood draw volumes be exceeded. Blood samples will provide the opportunity to explore a variety of blood-specific biomarkers as well as evaluation of white blood cell characterization (peripheral blood mononuclear cells via flow cytometry or RNA expression studies) and DNA samples for genotyping.
    b. Urine will be collected and processed according to standardized MOPs and SOPs.
    c. Stool will be collected using standardized stool collection kits. Stool will be used for microbiome studies.
    d. Induced Sputum. Sputum induction is a relatively simple, repeatable, and non-invasive method to collect airway secretions, thus, they are a highly asthma-relevant biological sample type. Sputum samples will provide an opportunity to establish the inflammatory cell differential and counts in the patient's airways, while providing an opportunity for extended studies (e.g., gene expression, microbiome, and sputum biomarkers such as tryptase). Sputum cell counts will be determined in a central core laboratory. Other sputum biospecimens will be stored in the Biorepository after processing.
    The process of sputum collection involves nebulization of 3% saline to induce cough and sputum production. Hypertonic aerosols such as 3% can induce bronchoconstriction in patients with asthma, and pre-treatment with albuterol will be provided to guard against such bronchoconstriction. In addition, participants with low FEV_1 will undergo sputum induction with an isotonic aerosol (0.9% saline). Specific and standardized procedures for sputum induction are described in SOPs and MOPs. The procedures for sputum induction differ in adults and in adolescents, as follows, with differences designed to have a more conservative protocol in the adolescents:

        - *Adolescents*: For participants with a post bronchodilator $FEV_1\% \geq 70\%$, sputum will be induced using 3% saline; for participants with a post bronchodilator $FEV_1\% < 70\%$, sputum will be induced using 0.9% saline.

        - *Adults*: For participants with a post bronchodilator $FEV_1\% \geq 50\%$, sputum will be induced using 3% saline; for participants with a post bronchodilator $FEV_1\% < 50\%$, sputum will be induced using 0.9% saline.

    e. Exhaled breath condensate will be collected using RT-tubes, which condense air that is breathed out of the participants' lungs during a short period (~10 minutes). The participant is asked to breathe through a mouthpiece during this time. They can come off of the mouthpiece to cough and/or swallow. The sample will then be spun from the condensation tube and stored for shipment to the Biorepository Core Laboratory.
    f. Nasal swabs will allow for capture of samples from this region for biomarker studies (for example, measurement of eosinophil peroxidase, or EPX).

7. High Resolution CT scan of the chest will occur at the qualification visit following four puffs of albuterol before treatment start during the run-in period on all participants who meet entry criteria for severe asthma and compliance. In addition to providing a method to detect latent tuberculosis, noninvasive measures, such as imaging, can measure airway structure and remodeling, as well as air trapping, mucus plugs, and other parameters relevant to asthma pathophysiology. The PrecISE Network will work with an established Radiology Reading Center to provide standardization and harmonization of all CT imaging protocols. This Center will also provide initial quantitative CT analyses following the protocol outlined in the MOPs. The basic CT scan protocol will consist of obtaining multi-detector CT (MDCT) images of the entire lung at full inspiration (Total Lung Capacity = TLC) and at the end of expiration (Residual volume=RV). The images from the MDCT chest scans will be stored locally and sent electronically to the Radiology Core using software that assures protocol compliance and de-identification. Scans will be read by local radiologists to rule on the presence of clinically actionable conditions, such as pneumonia or nodules.
8. Study treatment administration will occur during the treatment periods as outlined in the visit structure. The study coordinators will provide the participants with the study treatment and instructions as to how it should be used. Both the coordinator and the participant will be blinded as to whether or not the participant will be receiving active treatment or placebo.
    a. Visits during the treatment period will be used to evaluate if the participant is compliant with the taking of their assigned active treatment/placebo.

8.3 ADVERSE EVENTS AND SERIOUS ADVERSE EVENTS

8.3.1 DEFINITION OF ADVERSE EVENTS (AE)

Adverse event means any untoward medical occurrence associated with the use of a drug in humans, whether or not considered drug related (21 CFR 312.32(a)). Adverse events occurring after the participant provides informed consent will be recorded.

8.3.2 DEFINITION OF SUSPECTED ADVERSE REACTION

Suspected adverse reaction means any adverse event for which there is a reasonable possibility that the drug caused the adverse event. For the purposes of IND safety reporting, ‘reasonable possibility’ means there is evidence to suggest a causal relationship between the drug and the adverse event. A suspected adverse reaction implies a lesser degree of certainty about causality than adverse reaction, which means any adverse event caused by a drug.

8.3.3 DEFINITION OF SERIOUS ADVERSE EVENTS (SAE)

An adverse event (AE) or suspected adverse reaction is considered "serious" if, in the view of either the investigator or sponsor, it results in any of the following outcomes:

- death
- a life-threatening adverse event
- inpatient hospitalization ≥ 24 hours or prolongation of existing hospitalization
- a persistent or significant incapacity or substantial disruption of the ability to conduct normal life functions, or
- a congenital anomaly/birth defect

- Is an important medical event that may not be immediately life-threatening or result in death or hospitalization but may jeopardize the patient and may require intervention to prevent one of the other outcomes listed in the definition above.

This definition permits either the Medical Monitor or the investigator to decide whether an event is serious. Serious adverse events are critically important for the identification of significant safety problems. Therefore, if either the Medical Monitor or investigator believes that the event is serious, the event must be considered serious and evaluated by the sponsor for expedited reporting (21 CFR 312.32(a) and 312.32(c)(1)).

8.3.4 RESPONSIBILITIES

Primary responsibility for AE identification, documentation, and assessment of severity and relationship to study agents can be determined by either the investigator or Medical Monitor. The medical personnel who perform these assessments must be directly involved in the clinical evaluation of the research participants.

8.3.5 CLASSIFICATION OF AN ADVERSE EVENT

8.3.5.1 SEVERITY OF EVENT

For adverse events (AEs), the following guidelines will be used to describe severity.

- **Mild** – Events require minimal or no treatment and do not interfere with the participant's daily activities.
- **Moderate** – Events result in a low level of inconvenience or concern with the therapeutic measures. Moderate events may cause some interference with functioning.
- **Severe** – Events interrupt a participant's usual daily activity and may require systemic drug therapy or other treatment. Severe events are usually potentially life-threatening or incapacitating. Of note, the term "severe" does not necessarily equate to "serious".

8.3.5.2 RELATIONSHIP TO STUDY INTERVENTION

The site investigator and/or Medical Monitor is responsible for assessing the relationship between the AE and the study agent(s). Site investigators must provide the initial assessment as to whether there is a reasonable possibility that the study agent(s) caused or contributed to a SAE. The relationship assessment, based on clinical judgment, often relies on the following:

- A temporal relationship between the event and administration of the study agent(s),
- A plausible biological mechanism for the agent to cause the AE,
- Another possible etiology for the AE,
- Previous reports of similar AEs associated with the study agent or other agents in the same class, and
- Recurrence of the AE after re-challenge or resolution after de-challenge, if applicable.

Further assessment of causality is provided by the DSMB based on accumulating safety reports between treatment groups.

The terms used to assess the relationship of an event to study agent are:

- Related – There is a reasonable possibility that the AE may be related to the study agent(s).
- Not Related – There is not a reasonable possibility that the AE is related to the study agent(s).

8.3.5.3 EXPECTEDNESS

Expected AEs are AEs that have been previously observed with use of the study agent(s) and are listed in the package insert or Investigator's Brochure. Expectedness is not based on what might be anticipated from the pharmacological properties of the study agent or for the disease or study population under study.

An adverse event or suspected adverse reaction is considered 'unexpected' if it is not listed in the investigator brochure or is not listed at the specificity or severity that has been observed. Further details are provided in the MOP Chapter 13, Safety and Adverse Events.

Blood draw: Participants may experience some lightheadedness or nausea while having blood drawn. There is a risk of bruising at the site where the needle enters the skin and a remote risk of infection.

Spirometry and maximum bronchodilation: Participants may experience shortness of breath while doing spirometry, or, very rarely, syncope. Temporary wheezing and chest tightness occur infrequently (5% of the time). Following the bronchodilator, a transient increase in heart rate and tremor is likely (>25% chance).

Methacholine Challenge: Participants may experience shortness of breath, coughing, lightheadedness and dizziness. In rare cases, some participants have experienced a severe asthma attack or a reaction to the methacholine. Participants with a pre-diluent FEV_1 of >50% predicted (>70% in children aged 12 to 17 years) and at least one liter will undergo the methacholine testing if they do not have a historical methacholine from testing performed under either the 2017 ERS technical standard[9] or the 1999 ATS Guidelines[10].

Sputum Induction: Participants may experience a salty after taste in the mouth, coughing, a feeling of needing to swallow, a sore throat, shortness of breath, wheezing, chest tightness, lightheadedness, nausea or headache. In rare cases, some participants have had a severe asthma attack or a reaction to the salty water that they breathe in. Bronchodilator treatment will be available if this occurs. The following safety procedures will be followed for the sputum induction procedure:

- Adolescent participants with a post bronchodilator $FEV_1\% \geq 70\%$, sputum will be induced using 3% saline; for adolescent participants with a post bronchodilator $FEV_1\% < 70\%$, sputum will be induced using 0.9% saline.
- Adult participants with a post bronchodilator $FEV_1\% \geq 50\%$, sputum will be induced using 3% saline; for adult participants with a post bronchodilator $FEV_1\% < 50\%$, sputum will be induced using 0.9% saline.
- a physician will be available during the challenge;
- study staff will calculate and record the peak flow and FEV_1 value that equals both a 10% and 20% fall in lung function based upon the recorded post-BD peak flow and FEV_1 values; and
- participants will not be discharged until their FEV_1 is within 10% of their post bronchodilator FEV_1.

Withholding medication: After consenting, participants will be asked to hold certain medications prior to each visit for the purpose of lung function testing. Participants may experience an increase in their asthma symptoms from holding their daily medications. If participants are unable to hold their medications, they are instructed to take the medication as needed and to call the study staff. This will also be documented and the participant will proceed with the scheduled procedure.

Conditions common in adolescents and adults who have severe asthma will be considered expected for IND reporting purposes. Examples include sinus infections, pneumonia, and sinus surgery.

For other events, a site investigator or sub-investigator will be responsible for determining whether an adverse event (AE) is expected or unexpected. An AE will be considered unexpected if the nature, severity, or frequency of the event is not consistent with the risk information previously described for the test treatment. Test intervention-specific expected adverse events are described in the appendices.

8.3.5.4 TIME PERIOD AND FREQUENCY FOR EVENT ASSESSMENT AND FOLLOW-UP

The occurrence of an adverse event (AE) or serious adverse event (SAE) may come to the attention of study personnel during study visits and interviews of a study participant presenting for medical care, or upon review by a study monitor.

All AEs including local and systemic reactions not meeting the criteria for SAEs will be captured on the appropriate case report form (CRF). Information to be collected includes event description, time of onset, clinician's assessment of severity, relationship to study intervention (assessed only by those with the training and authority to make a diagnosis), and time of resolution/stabilization of the event. All AEs must be documented appropriately regardless of relationship. All AEs will be followed to adequate resolution.

Any medical condition that is present at the time that the participant is screened will be considered as baseline and not reported as an AE. However, if the study participant's condition deteriorates at any time during the study, it will be recorded as an AE.

Changes in the severity of an AE will be documented to allow an assessment of the duration of the event at each level of severity to be performed. AEs characterized as intermittent require documentation of onset and duration of each episode.

A site physician investigator or sub-investigator will record all reportable events between the time the participant provides informed consent through 8 weeks after their last treatment or a period corresponding to five half-lives of the treatment last received, whichever is longer (Section 4.1). At each study visit, the investigator will inquire about the occurrence of AE/SAEs since the last visit. Events will be followed for outcome information until resolution or stabilization.

8.3.6 SERIOUS ADVERSE EVENT REPORTING

Study investigators will immediately notify the DMCC of any serious adverse event, whether or not considered test treatment-related, including those listed in the protocol or investigator brochure. The report must include an assessment of whether there is a reasonable possibility that the test treatment caused the event.

All serious adverse events (SAEs) will be followed until satisfactory resolution or until the site investigator deems the event to be chronic or the participant is stable. Other supporting documentation of the event may be requested by the DMCC and should be provided as soon as possible.

The DMCC will be responsible for notifying the FDA of any unexpected fatal or life-threatening suspected adverse reaction as soon as possible, but in no case later than 7 calendar days after initial receipt of the information. In addition, the DMCC must notify the FDA and all participating investigators in an IND safety report of potential serious risks, from clinical trials or any other source, as soon as possible, but in no case later than 15 calendar days after the DMCC determines that the information qualifies for reporting.

8.3.7 REPORTING EVENTS TO PARTICIPANTS

An unexpected finding may occur whenever safety testing or imaging (CT) of the lung are done. These unexpected findings can have clear or uncertain clinical significance. For example, clear clinical significance means that the testing shows a problem that may be treatable and that we generally know what the risks are of not treating the problem. Uncertain clinical significance means that the test result is unusual or outside the boundaries of "normal", but we do not know if it might affect the health of the participant, and treatment may not be appropriate or possible. In this study, the participant will be informed of any findings of clear clinical significance that may be discovered during the imaging procedure or safety laboratory test, but the participant will not be informed if there are findings of uncertain clinical significance. There may be benefits to learning such results (such as early detection and treatment of a medical condition), but there are risks as well (such as problems with getting insurance or employment, or becoming worried about a finding for which no treatment is required or appropriate). Participants may choose to have their physician informed of any findings of clear clinical significance that are reported. Participants will indicate their decision in the consent form regarding informing their physician and having the report placed in their medical record.

8.3.8 REPORTING OF PREGNANCY

Pregnancy, lactation or plans to get pregnant are exclusion criteria for this study. Females of child-bearing potential must use a medically accepted method of birth control from the time of enrollment until 16 weeks following the last dose of study drug. Urine pregnancy testing will be performed at regular intervals during the study on females of childbearing potential. If a participant becomes pregnant during the study, study treatment will be discontinued. If permission is granted, pregnant study participants or pregnant partners of study participants will be followed through pregnancy and delivery. Safety data will be collected during this interval.

8.4 UNANTICIPATED PROBLEMS

8.4.1 DEFINITION OF UNANTICIPATED PROBLEMS (UP)

Unanticipated Problem Involving Risk to Participants or Others (UPIRSO): Any incident, experience, or outcome that:

- Is unexpected (in terms of nature, severity, or frequency) given (a) the research procedures that are described in the protocol-related documents, such as the IRB-approved research protocol and informed consent document; and (b) the characteristics of the participant population being studied;
- Is related or possibly related to a participant's participation in the research; and

- Is serious or suggests that the research places participants or others at a greater risk of harm (including physical, psychological, economic, or social harm) than was previously known or recognized.

Unexpected AE: An AE, the nature or severity (intensity) of which is not consistent with the applicable agent information (Investigator's Brochure, package insert, or summary of agent characteristics).

8.4.2 UNANTICIPATED PROBLEM REPORTING

The investigator will report unanticipated problems (UPs) to the reviewing IRB and to the DMCC. The UP will be entered into the DMS and will include the following information:

- A detailed description of the event, incident, experience, or outcome;
- An explanation of the basis for determining that the event, incident, experience, or outcome represents an UP;
- A description of any changes to the protocol or other corrective actions that have been taken or are proposed in response to the UP.

To satisfy the requirement for prompt reporting, UPs will be reported to the IRB and to the DMCC as soon as possible after the investigator becomes aware of the event.

9 STATISTICAL CONSIDERATIONS

9.1 STATISTICAL HYPOTHESES

Two primary endpoints are specified to assess treatment efficacy (study objective #1):

1) **FEV_1 percent predicted,** assessed prior to bronchodilator administration
2) **Asthma symptom control,** assessed via the Juniper Asthma Control Questionnaire (ACQ-6)

FEV_1 percent predicted and ACQ-6 score will be measured at the start and the end of each treatment period. Efficacy analyses will compare the end-of-period outcome values between test treatment and placebo.

For the first primary study objective (overall assessment of efficacy), the following hypotheses will be tested:

1. Test treatment T does not improve FEV_1 percent predicted compared to placebo (the null hypothesis) versus T is different from placebo.
2. Test treatment T does not improve asthma symptom control assessed via ACQ-6 compared to placebo (the null hypothesis) versus T is different from placebo.

The second primary objective involves estimation of the optimal subgroup for targeting participants for each intervention. No formal statistical hypotheses will be tested for this objective.

9.2 SAMPLE SIZE DETERMINATION

An intervention is considered efficacious if significant benefit is shown for either of the two primary endpoints (FEV_1% predicted, and ACQ-6). Because there are multiple chances for a test treatment to

demonstrate efficacy, multiplicity adjustments are required. We will use the Hochberg method to adjust for multiplicity.

A sample size of 111 participants is required to achieve 80% power to detect a treatment effect with respect to at least one of the two primary endpoints equal to 0.3 times the standard deviation (SD) of that endpoint, taking into account the possibility to stop for futility. The Type I error probability will be controlled under the global null hypothesis (no effect on either endpoint) at $\alpha \leq 0.10$. Power was estimated under the assumption of a within-participant, between-period correlation of responses on consecutive placebo and test treatment, τ, of 0.70 and a treatment period discontinuation rate of 0.10. The required sample size for interventions with treatment period drop-out rates significantly different from 0.10, assessed based on pooled placebo and active treatment data, may be adjusted during the trial, prior to any unblinding. If futility analysis is not feasible for an intervention due to difficulty in participant accrual, 93 participants (2:1 ratio of biomarker positive to negative participants) are required to provide the same power described above.

Examples of treatment effects yielding a 0.3 standardized effect size include: a difference of 4.3 percent predicted FEV_1 between intervention and placebo with a SD of 14.5 percent predicted; and a difference of 0.18 in average ACQ-6 symptom scores between the intervention and placebo with a SD of 0.6.

Given the experimental nature of the trial, the Type I error rate will be controlled at the two-sided 0.1 level. Our analysis method ensures a strong control of the Type I error rate at this level. Taking into account our stopping rule for futility, the Type I error rate is equal to about 0.05, two-sided, for $\tau = 0.70$.

Based on participant accrual to date (at the time of protocol version 3.1) and assuming continued accrual of 12 participants per month, the target number of 395 participants and a study duration of at least 50 months should result in enough participant-periods to support the evaluation of 5 interventions. These calculations take into account a study discontinuation rate of 0.02 every 4 weeks (or approximately 24% per year). That is, the probability for a participant to permanently drop-out of the study in any 4-week time period is 0.02.

9.3 DESIGN ADAPTATIONS

For each intervention, an a priori best subgroup is specified based on the literature and data available at the time the intervention enters the trial. Participants belonging to this a priori best subgroup are referred to as biomarker positive for that intervention. Treatment assignment probabilities will be initially set to achieve an approximate 2:1 ratio of biomarker positive and negative participants for each intervention. This treatment allocation will allow us to gather information on the association of biomarkers and interventions early in the study. A single interim analysis for futility may be performed for each intervention (independently of the others) after test treatment and placebo (matched or unmatched) data are available for approximately 34 biomarker positive participants. Those interventions that have not been eliminated due to either safety concerns or futility will continue to be assigned to participants for the remainder of the study until the required number of participants is assigned to each intervention. A futility analysis will be also performed in biomarker negative patents. Further enrollment to test treatments demonstrating futility in biomarker negative subgroup will be limited to biomarker positive participants.

An interim analysis to estimate the within-participant (between-period) correlation, pooling data across test and placebo treatment periods while maintaining masking of treatment assignments, was planned and has now been conducted (at the time of protocol version 3.1). The within-participant (between-period) correlation for FEV1 and ACQ was much higher than 0.38, originally hypothesized; therefore, the required sample size for the final analysis has been reduced.

The DSMB will review interim analysis data for purposes of stopping an intervention due to futility, in addition to performing their other safety oversight responsibilities (see Section 10.1.6). A second independent advisory group, the Protocol and Adaptations Review Committee or PARC, will advise on study adaptations and on new interventions that may be considered for entry into the study. The PARC will not review any unblinded data or comparative analyses to avoid the potential for bias in making their recommendations.

9.4 POPULATIONS FOR ANALYSES

Screening Population

Participants who are screened but not randomized will be reported, but not included in any efficacy or safety analyses of study data. SAEs occurring during the screening period will also be reported.

Safety Population

The Safety Population for each intervention will consist of all participants randomized to that intervention who received at least one dose of study medication. This population will form the basis for all safety analyses and selected secondary efficacy analyses of the intervention.

Intention-to-Treat (ITT) Population

The ITT population for each intervention will consist of all participants randomized to that intervention.

The primary and secondary analyses will be conducted using the modified intention-to-treat (mITT) population, where a participant receives study drug (test treatment or placebo) and contributes at least one post-baseline measurement for each of the primary clinical endpoints during the treatment period in which the participant receives the intervention.

Per-Protocol Population

The per protocol population for each intervention consists of all participants in the mITT population who complete study treatment and are in reasonable compliance with the protocol during the treatment period in which the participant received the intervention and in at least one placebo period. Additional details are contained in the Statistical Analysis Plan.
Membership in the per-protocol population will be determined prior to unmasking of treatment assignments to avoid the potential for bias. The population will form the basis for selected secondary efficacy analyses.

9.5 STATISTICAL ANALYSES

9.5.1 GENERAL APPROACH

An intervention will be considered effective if it is effective on at least one of the two primary outcomes. The analysis model for the final efficacy analysis will take into account the correlations on outcomes assessed for the same patient across the different treatment periods. The final analysis will include pairs of the treatment and matching placebo from the first two periods of the trial, as well as unmatched observations on the same patient. Mixed model repeated measures methods will be used for the primary analysis.

A single futility analysis may be conducted for each intervention during the trial. The objective of the futility analysis is to stop interventions showing no signal of efficacy on either of the two primary endpoints. The final precision medicine analyses will use machine learning methods to optimize the target subgroup definition for each intervention. These methods rely on outcomes assessed for patients both within and outside the target subgroups and may result in changes to the cut-points for a particular biomarker used to delineate the positive and negative patients, or to changes in the biomarker themselves.

9.5.2 ANALYSIS OF THE PRIMARY EFFICACY ENDPOINT(S)

The final analysis to assess the efficacy of an intervention will be performed when accrual and follow-up for that intervention is complete. Treatment effects with respect to each of the two primary endpoints will be estimated using a mixed-model for repeated measures (MMRM) that appropriately takes into account the within-participant correlations between periods. A detailed description of the analysis model is provided in the Statistical Analysis Plan.

No adjustment for multiplicity with respect to the multiple interventions in the study is planned. Success is defined independently for each intervention, and interventions will not be compared to each other in the efficacy analyses. Adjustment for multiplicity across the two primary outcomes will be performed using the Hochberg method within each intervention.

9.5.2.1 MISSING DATA

We will use multiple imputation to handle missing data for the primary efficacy outcomes in a treatment period. Details of the planned imputation procedures and associated sensitivity analyses are provided in the Statistical Analysis Plan. Analysis of the Secondary Endpoint(s)

Secondary endpoints such as CompEx, FVC pre-BD, FEV_1 post-BD, symptom free days (defined using diary/CompEx data) and asthma free days will be analyzed similar to the primary analysis model. Details of secondary analyses are provided in the Statistical Analysis Plan.

9.5.3 SAFETY ANALYSES

Safety data will be evaluated on an ongoing basis. Monthly reports of serious adverse events will be provided to the chair of the DSMB. The DSMB will be unblinded to assigned treatment. More comprehensive reports will be provided to all DSMB members on a regular schedule as determined by the DSMB. These reports will include incidence of treatment-emergent adverse events by assigned treatment group. Adverse events will be coded using the Medical Dictionary for Regulatory Activities (MedDRA). Adverse event rates (and severity non-serious adverse events) will be compared between active and placebo periods for each intervention. Note that for a particular intervention, all placebo periods for participants receiving the intervention will be included, and not just those where the matching placebo was received.

Based on the results of these reviews, the DSMB will recommend to the NHLBI whether to continue or discontinue each test treatment. In addition, the DSMB will be monitoring all of the safety data throughout the course of the trial and will be notified as soon as possible of any SAE that is deemed both unexpected and related to treatment. All SAEs will be reviewed at each DSMB meeting.

Final safety analyses will be performed for each intervention, once the intervention completes and all follow-up data are obtained. Descriptive summaries (tables and graphs) of safety endpoints, including adverse events, laboratory parameters, and other safety biomarkers, will be generated by test

treatment versus placebo, and hypothesis tests for differences between groups will be generated for selected parameters.

9.5.4 BASELINE DESCRIPTIVE STATISTICS

The run-in period is considered to be the baseline evaluation period. The initial statistical analysis will focus on summarizing the baseline characteristics of the study participants. Details of these analyses are provided in the Statistical Analysis Plan.

9.5.5 PLANNED INTERIM ANALYSES

Details of the planned interim analyses for futility are provided in the Statistical Analysis Plan.

9.5.6 OPTIMIZATION OF TARGET SUBGROUPS

We designed the precision medicine analysis to highlight the best treatments for key patient subgroups. The therapies selected for PrecISE might only work in their predictive biomarker subgroups; their effect on participants outside of the best subgroup may be rather small. Primary and secondary predictive biomarkers are specified for each intervention. The a priori best subgroup is specified as a function of the primary predictive biomarker for each intervention before the study begins (or the intervention enters the study) based on the best available information at that time. More details are provided in the Statistical Analysis Plan.

9.5.7 TABULATION OF INDIVIDUAL PARTICIPANT DATA

Data listings of individual participant data that include treatment assignment will not be shared with investigators outside the DMCC until database lock. Data listings of individual SAEs will be prepared by unblinded DMCC statisticians and programmers, and shared with the DSMB in confidential reports. The data will be scrubbed of identifiable information prior to release to a public data repository.

9.5.8 EXPLORATORY ANALYSIS

Exploratory analyses will be performed at the end of the study to identify the best target subgroup for each intervention using all available data and to explore individualized treatment rules for interventions with overlapping target subgroups. Details of the planned exploratory analyses are provided in the Statistical Analysis Plan.

10 SUPPORTING DOCUMENTATION AND OPERATIONAL CONSIDERATIONS

10.1 REGULATORY, ETHICAL, AND STUDY OVERSIGHT CONSIDERATIONS

10.1.1 INFORMED CONSENT PROCESS

Potential participants will be asked to provide consent to the master protocol at the initial screening visit at week 0 through a master consent document. Our general approach to the consent process is to explain the design of the master protocol to each potential participant, why this design is ideal for testing novel therapies and determining which therapies work for which patients, how participation in

the master protocol will allow participants to "try out" multiple therapies, why their participation is important, and how their data will contribute to the research. The goal is to obtain buy-in from potential participants on the study design as a whole and explain clearly what they can expect from participation in the study (e.g., expected study duration given their enrollment date, opportunity for multiple sequential treatments, possibilities of placebo, etc.). The master consent form will provide details about the possible interventions each potential participant may receive.

Separate master consent forms will be used for adults and adolescents. The master consent document for adults will describe the interventions that are being studied in PrecISE at the time the participant is enrolled, and explain that the participant may receive any of these interventions during their participation in the study through a series of re-randomizations and treatment assignments (provided no safety exclusions specific to any of the interventions apply). Their consent applies to the set of available interventions, and if they are not willing to provide consent, they are not eligible for the study. Consent will not be obtained for each intervention individually, to avoid the potential for bias. In a similar manner, a parent or guardian will provide consent for each adolescent's participation in the study; adolescent participants will provide assent.

New participants will be consented to the new set of possible interventions, and active participants will be re-consented to the new set of possible interventions. Again, this consent will be to the master protocol (the set of possible interventions) and not to any individual intervention, because at any point of re-randomization, a participant may receive any one of the interventions. If an active participant refuses to be re-consented, they may continue their participation in the study, but with a reduced set of possible interventions reflecting those remaining after the treatment determined to be futile was discontinued from the study and before introducing the new agent. Participants who refused to consent to some interventions but then changed their mind and are willing to take any of the five interventions will be allowed to do so.

Parents or guardians of adolescents will receive a different consent form that will specify the set of interventions approved for study in that population. A corresponding assent form will be provided to adolescents. Each adolescent recruited to PrecISE will be asked to provide assent to receive either of the two interventions or placebo at any of the three treatment periods for which they will remain in the study (because no participant will be assigned to the same treatment twice, adolescents would only remain in the study for three periods, if two treatments are approved for them).

Note that participants will also be asked to consent to storage of and future use of their biospecimens separately from their consent to study participation. Participants will be able to consent to the study (including use of biospecimens in PrecISE) but opt out of having their biospecimens stored for future use.

This study will be reviewed under a single IRB at Vanderbilt University. All other US sites will rely on Vanderbilt University's IRB, and an IRB Authorization Agreement (IAA), also known as a reliance agreement, will be established for each site institution. Once fully executed, a copy of the IAA will be submitted to the DMCC, with the original agreement filed on site. The DMCC will ensure that every consent form meets federal requirements and presents accurate information, as determined by the protocol team and NIH regulatory officials, and the current consent form will be kept on file and available for review.

In obtaining and documenting informed consent, PrecISE study investigators and study teams will comply with applicable regulatory requirements, namely, ICH, GCP and other regulatory requirements including Title 45 Part 46 of the Code of Federal Regulations (45 CFR 46), 21 CFR 11 (Electronic Records),

21 CFR 50 (Protection of Human Subjects), 21 CFR 54 (Financial Disclosure of Clinical Investigators), 21 CFR 56 (Institutional Review Boards), and 21 CFR 312 (Investigational New Drug Application).

10.1.1.1 CONSENT/ASSENT AND OTHER INFORMATIONAL DOCUMENTS PROVIDED TO PARTICIPANTS

Consent forms describing in detail the study intervention, study procedures, and risks are given to the participant and written documentation of informed consent is required prior to starting administration of the study intervention. The following consent materials are submitted with this protocol:

- Adult Consent Form_English
- Adult Consent Form_Spanish
- Minor Subjects Assent Form_English
- Minor Subjects Assent Form_Spanish

Basic consent elements and appropriate additional elements as outlined in 45 CFR §46.116 and 21 CFR §50.25 include:

- Statement that the study involves research;
- Explanation of the purposes of the research, the expected duration of the subject's participation, and a description of the procedures to be followed;
- A statement of the conditions and period of time under which the participant's data will be stored and will be accessible;
- A description of any foreseeable risks or discomforts to the participant;
- Identification of any procedures which are experimental;
- Reasonably expected benefits to participant or others;
- Disclosure of alternative procedures;
- Confidentiality measures;
- Explanation of compensation and medical treatment, if any, for injury;
- A statement that participation is voluntary, refusal to participate will involve no penalty or loss of benefits and that the participant may discontinue participation in any time without penalty or loss of benefits; and
- An explanation of whom to contact for answers to pertinent questions about the research and research participant's rights, and whom to contact in the event of a research-related injury to the participant, for research questions, or for questions about rights.

Additional elements include:

- Unforeseeable risks to the participant and/or to the fetus or embryo in case of pregnancy;
- Circumstances under which participation may be terminated without regard to participant's consent;
- Additional costs to participant resulting from study participation;
- Consequences of the decision to withdraw and procedures for orderly termination of participation by the participant;
- New study findings which may affect the participant's decision to continue; and
- Approximate number of study participants.

Informed consent form requirement for clinical trials:

- The FDA and NIH have mandated that all informed consent documents inform patient volunteers that trial data will be added to the national clinical trial registry databank at ClinicalTrials.Gov. The exact wording to be included in the documents reads: "A description of

this clinical trial will be available on http://www.ClinicalTrials.gov, as required by U.S. Law. This Web site will not include information that can identify you. At most, the Web site will include a summary of the results. You can search this Web site at any time."

10.1.1.2 CONSENT PROCEDURES AND DOCUMENTATION

Informed consent is a process that is initiated prior to the individual's agreeing to participate in the study and continues throughout the individual's study participation. Consent forms will be IRB-approved and the participant will be asked to read and review the document. The investigator or study coordinator will explain the research study to the participant and answer any questions that may arise. A verbal explanation will be provided in terms suited to the participant's comprehension of the purposes, procedures, and potential risks of the study and of their rights as research participants, this includes explaining the study in Spanish, for Spanish speakers. In addition, study staff will request assent from minors after receiving parental permission. Participants will have the opportunity to carefully review the written consent form and ask questions prior to signing. The participants should have the opportunity to discuss the study with their family or surrogates or think about it prior to agreeing to participate. The participant will sign the informed consent document prior to any procedures being done specifically for the study. Participants must be informed that participation is voluntary and that they may withdraw from the study at any time, without prejudice. A copy of the informed consent document will be given to the participants for their records. The informed consent process will be conducted and documented in the source document (including the date), and the form signed, before the participant undergoes any study-specific procedures. Consent may also be obtained remotely. In this case, the consent form will be provided to the participant. It will be reviewed with the participant over the phone or during a video call. The participant may sign the consent form and return it to the site, or the form may be signed electronically, provided a software tool that is compliant with the Code of Federal Regulations (CFR) 21 Part 11 concerning electronic signatures is available at the site. The rights and welfare of the participants will be protected by emphasizing to them that the quality of their medical care will not be adversely affected if they decline to participate in this study.

10.1.2 STUDY DISCONTINUATION AND CLOSURE

The study or any of the interventions included in PrecISE may be temporarily suspended or prematurely terminated if there is sufficient reasonable cause (see examples below). Written notification, documenting the reason for study suspension or termination, will be provided by the suspending or terminating party to study participants, investigators, the NHLBI, the Food and Drug Administration and other relevant regulatory authorities. If the study is prematurely terminated or suspended, the Principal Investigator at each study site will promptly inform study participants, the IRB, and the sponsor and will provide the reason(s) for the termination or suspension. Study participants will be contacted, as applicable, and be informed of changes to study visit schedule. Please refer to **Section 7, Study Intervention Discontinuation and Participant Discontinuation/Withdrawal**, for handling of enrolled study participants.

Circumstances that may warrant termination or suspension of the study include, but are not limited to:

- Determination of unexpected, significant, or unacceptable risk to participants
- Insufficient compliance to protocol requirements
- Data that are not sufficiently complete and/or evaluable

The study may resume once concerns about safety, protocol compliance, and data quality are addressed, and satisfy NHLBI, the IRB and/or other regulatory agencies.

There are multiple interventions in this study, and it may be that a specific intervention will be terminated due to safety or futility, based on recommendations from the DSMB (Section 10.1.6). In this case, the parent study will continue with the other active and planned interventions.

10.1.3 CONFIDENTIALITY AND PRIVACY

Participant confidentiality and privacy is strictly held in trust by the participating investigators, their staff, and the sponsor(s) and their test treatments. This confidentiality is extended to cover testing of biological samples and genetic tests in addition to the clinical information relating to participants. Therefore, the study protocol, documentation, data, and all other information generated will be held in strict confidence. No information concerning the study or the data will be released to any unauthorized third party without prior written approval of the sponsor.

All research activities will be conducted in as private a setting as possible.

The study monitor, other authorized representatives of the sponsor, representatives of the IRB, or regulatory agencies may inspect all documents and records required to be maintained by the investigator, including but not limited to, medical records (office, clinic, or hospital) and pharmacy records for the participants in this study. The clinical study site will permit access to such records.

The study participant's contact information will be securely stored at each clinical site for internal use during the study. At the end of the study, all records will continue to be kept in a secure location for as long a period as dictated by the reviewing IRB, Institutional policies, or sponsor requirements.

Study participant research data, which is for purposes of statistical analysis and scientific reporting, will be transmitted to and stored at the DMCC. The study data entry and study management systems used by clinical sites and by the DMCC will be secured and password protected (see Section 2.3 for additional details). At the end of the study, all study databases will be de-identified and archived at the NHLBI data repository.

Certificate of Confidentiality

To further protect the privacy of study participants, a Certificate of Confidentiality will be issued by the National Institutes of Health (NIH). This certificate protects identifiable research information from forced disclosure. It allows the investigator and others who have access to research records to refuse to disclose identifying information on research participation in any civil, criminal, administrative, legislative, or other proceeding, whether at the federal, state, or local level. By protecting researchers and institutions from being compelled to disclose information that would identify research participants, Certificates of Confidentiality help achieve the research objectives and promote participation in studies by helping assure confidentiality and privacy to participants.

10.1.4 FUTURE USE OF STORED SPECIMENS AND DATA

PrecISE will be collecting specimens on all participants. Strict confidentially standards are in place and will be maintained to protect the privacy of PrecISE participants. Biospecimen samples will be labeled with a unique identifier that does not contain any protected health information or otherwise identify a participant. Biospecimens collected will not be stored long-term at the clinical sites but rather will be used immediately for analyses (sent to a Clinical Laboratory Vendor) or deposited in batches at PrecISE

study-approved Central Core laboratories. These specimens will not be individually identifiable by the laboratories, clinical centers, or DMCC personnel. The DMCC will develop and maintain a tracking system whereby study participants can modify their level of consent for their use of any stored samples for future studies. Participants can ask that any specimens still in storage be destroyed and not included in future analyses. The request to remove stored specimens will be made at the clinical centers to preserve participant confidentiality.

Genetic studies utilizing the participant's DNA will be conducted in PrecISE. The participant will be fully informed of the intended extent of these studies, which range from single nucleotide polymorphism testing for specific variants to inform randomization to potentially full genome sequencing to inform baseline characterization and treatment responses. In the event a new genetic marker is found during the course of PrecISE, participants with this hypothetical marker may be notified if they consent to this notification. Some of the analyses conducted as part of PrecISE are not intended to be diagnostic, and may not take place in a Clinical Laboratory Improvement Amendments (CLIA) certified lab. Some analyses, however, will be conducted in CLIA labs and may potentially include participant notification as part of the study protocol.

Beyond DNA, other "omics" studies will also be conducted in PrecISE. These may include studies to evaluate the RNA transcriptome and the microbiome, and they could potentially include studies involving the metabolome, the epigenome, and the proteome. The participant will be fully informed of the intended and/or potential extent of these studies.

Data collected for this study will be analyzed and stored at the study's DMCC (Collaborative Studies Coordinating Center, Department of Biostatistics, Gillings School of Global Public Health, The University of North Carolina at Chapel Hill). After the study is completed, the de-identified, archived data will be transmitted to and stored at NHLBI's Biologic Specimen and Data Repository Information Coordinating Center (BioLINCC) for use by other researchers including those outside of the study. Permission to transmit data to BioLINCC will be included in the informed consent.

With the participant's approval and as approved by the study's single IRB, de-identified biological samples will also be stored at BioLINCC after the study is completed. These samples could be used for future research.

During the conduct of the study, an individual participant can choose to withdraw consent to have biological specimens stored for future research. However, withdrawal of consent with regard to biosample storage may not be possible after the study is completed. When the study is completed, access to study data and/or samples will be provided through BioLINCC.

*See **Section 10.1.3, Confidentiality and Privacy** and **Section 10.1.9, Data Handling and Record Keeping**, for further information on future use of study records.*

10.1.5 KEY ROLES AND STUDY GOVERNANCE

Study Leadership include:

NHLBI	
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Steering Committee Chairs	
Steve Georas, MD *University of Rochester Medical Center* *601 Elmwood Avenue* *Rochester, NY 14620* *585-472-0362* *Steve_georas@urmc.rochester.edu*	*Rosalind Wright, MD* *Mount Sinai School of Medicine* *145 Beach Avenue* *Larchmont, NY 10538* *781-771-9034* *Rosalind.wright@mssm.edu*
Data, Modeling and Coordinating Center	
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Stanley Szefler, MD *Children's Hospital Colorado* *13123 E. 16th* *Aurora, CO 80045* *720-777-0985* *Stanley.szefler@childrenscolorado.org*	*Michael Wechsler, MD* *National Jewish Health* *1400 Jackson Street, J215* *Denver, CO 80206* *617-285-4987* *wechslerm@njhealth.org*
Arizona/Wake Forest	
Eugene Bleecker, MD *University of Arizona* *1230 N. Cherry, 2nd Floor, Room 2964* *Tucson, AZ 85721* *520-626-8624* *erbleecker@email.arizona.edu*	*Monica Kraft, MD* *Mount Sinai School of Medicine* *1468 Madison Ave* *New York, NY 10029* *Monica.kraft@mssm.edu*
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Praveen Akuthota, MD *UC at San Diego* *9500 Gilman Drive, MC 0643* *La Jolla, CA 92093* *858-822-4106* *pakuthota@ucsd.edu*	*Sonia Jain, PhD* *UC at San Diego* *9500 Gilman Drive, MC 0725* *La Jolla, CA 92093* *858-822-2388* *sojain@ucsd.edu*

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University of Kansas/Vanderbilt	
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Study leadership includes an Executive Committee and a Steering Committee. The Executive Committee is composed of members from NHLBI, the DMCC, the chairs of the Steering Committee and two representatives from the clinical centers. The Steering Committee consists of all members of the Executive Committee and all PIs from the clinical centers.

10.1.6 SAFETY OVERSIGHT

An independent data and safety monitoring board (DSMB) will be established and charged with oversight of data quality, study integrity, and safety of participants. In addition to monthly reports of serious adverse events (SAEs), the DSMB will review unblinded data reports semi-annually and make recommendations to the NHLBI to ensure that participants are not exposed to undue risks. Summary reports of recommendations will be sent to site investigators and the central IRB.

For each intervention, the PrecISE DSMB will make recommendations as to whether the intervention should be stopped for safety reasons based on periodic monitoring of adverse events and other safety parameters. Regularly scheduled meetings will be held to review safety data reports prepared by the DMCC that include summary statistics by treatment group for adverse events, laboratory parameters, and other safety outcomes. Ad hoc meetings to review safety data can also occur if triggered by unusually high or unexpected SAE reporting.

The DSMB will also determine if an intervention should be recommended for discontinuation due to futility based on the results of a single interim analysis conducted for this purpose, approximately half-way through recruitment for that intervention. DSMB meetings to discuss futility analyses are triggered by enrollment targets and data availability. The DMCC will prepare a report summarizing the futility analysis results and corresponding stopping boundaries for each intervention in advance of the meeting (see Section 9.5.6). If the results of the futility interim analysis for an intervention are on or near the stopping boundary, the DSMB will weigh the benefit and risks of the intervention based on data accumulated to date, and make their recommendation accordingly.

Once a determination is made to recommend discontinuation of an intervention for either reason (futility or safety), the DSMB will summarize their deliberations to arrive at the recommendation,

and the DMCC will prepare a report for the NHLBI Project Office and the Steering Committee that summarizes the results of analyses supporting the recommendations, the stopping boundaries or rules used, and the DSMB's summary of their deliberations to arrive at the decision. The DMCC will then work with the Steering Committee to develop an appropriate communication plan for the various stakeholders.

A second independent advisory group, the Protocol and Adaptations Review Committee (PARC), will be established to advise on study adaptations and other protocol amendments that may be needed throughout the study. The adaptive design of PrecISE allows for adaptation to the subgroups targeted for each intervention following pre-specified adaptation rules (see Section 9.3). Consistent with good clinical practice and FDA guidance on data monitoring committees, DSMBs privy to unblinded comparative results by treatment group should not be involved in other decisions about changes to the study design or protocol, to minimize the possibility of bias.[3,16-18] The PARC will be formed to oversee study adaptations and recommend other changes to the study protocol, including the addition of new interventions to the study. This group will not review any unblinded data or comparative analyses to avoid the potential for bias in making their recommendations.

DSMB Members

Members of the PrecISE DSMB are independent experts chosen on the basis of their expertise and scientific rigor. They are not associated with the trial or with the pharmaceutical companies that supply the study agents. Committee members' areas of expertise span the disciplines relevant to the conduct of this clinical trial, including clinical trials, pharmacology, biostatistics, and clinical care of participants with asthma.

DSMB Mandates

The DSMB has the responsibility to review the research protocol and to evaluate the progress of the trial overall and at each participating clinical center. This includes accrual, data quality and completeness, episodes of exacerbations, hospitalization, mortality, other toxicities, and protocol violations. Serious unexpected and related events will be disclosed to the committee between meetings. The DSMB will also review interim endpoint data (both primary and secondary endpoints) to assess futility of each intervention.

Concurrently, the DSMB will evaluate the safety of each intervention studied under the Master Protocol as the trial progresses, considering evolving scientific discoveries or treatment options that may affect the desirability of continued treatment with any one of the interventions. At the conclusion of each meeting, the DSMB will recommend whether each intervention be continued. If an intervention proves to be more harmful than expected in terms of mortality or severe morbidity, we will stop treatment of all participants currently assigned to the intervention, and that intervention will be removed from the possible re-randomization assignments for ongoing participants. New enrollees will receive a master consent form reflecting the removal of any intervention discontinued for safety or futility reasons. This decision will be made by NHLBI on recommendation of our DSMB.

Frequency of DSMB Meetings

The DSMB will meet via webinar every 3-6 months to review study progress and safety for each intervention. Additional meetings to review the futility of each intervention will be scheduled based on

accrual rates and data availability for the interventions. These meetings will likely take place via webinars, due to the need to schedule them at different times for different interventions. Ad hoc meetings may also be scheduled via webinar, if particular safety issues for any of the interventions arises in between the regularly scheduled meetings.

DSMB Meeting Structure

The PrecISE principal investigators, the DMCC statisticians (blinded and unblinded), NHLBI representatives and on occasion other key personnel, will participate in the meetings' open sessions. Open sessions include consideration of recruitment, retention, and general scientific issues. The DSMB's voting members will discuss the unblinded by-treatment data for each intervention in a closed session. These data include adverse events, and material that should be kept confidential from the investigators.

Frequency and content of reports to the DSMB

In advance of each meeting, the unblinded statistician at the DMCC will prepare a report for DSMB review. The report will contain the following categories of information aggregated by treatment group, where applicable:

- Current enrollment status and timeline for completion of follow-up
- Adverse events reported, both serious and non-serious, including hospitalizations and any mortality
- Major and minor protocol violations and deviations
- Numbers of participants whose study medication is discontinued

In the safety reports, the DMCC will label the treatment groups as "A", "B", etc. to lessen the chance of accidental unmasking. The unblinded DMCC statistician will provide the treatment for each code to DSMB members as needed, and the same code mapping will be used for all meetings.

Serious events are reported to the DSMB monthly, and serious adverse events that are unexpected and related to treatment are reported within 15 days from the time the DMCC is notified of the event.

Frequency, content, and distribution of meeting reports

Following each meeting of the DSMB, its chair will prepare a summary of the questions raised by committee members, monitoring recommendations, and recommendations for the continuation of treatments. This report will be distributed confidentially to meeting participants. The committee's secretary also prepares a redacted summary of this report, focusing on safety issues, for distribution to PrecISE investigators and their IRBs.

10.1.7 CLINICAL MONITORING

Clinical site monitoring is conducted to ensure that the rights and well-being of study participants are protected, that the reported study data are accurate, complete, and verifiable, and that the conduct of the study is in compliance with the currently approved protocol/amendment(s), with International Conference on Harmonisation Good Clinical Practice (ICH GCP), and with applicable regulatory requirement(s). Monitoring for this study will be performed by the DMCC.

Areas of Focus

- Staff training
- Human subjects protection
- Protocol compliance
- Regulatory compliance
- Laboratory SOPs and compliance.

- Quality assurance
- Safety
- Adverse event reporting
- Integrity of research data and samples

Two types of data monitoring will be conducted during the study: on-site monitoring and central monitoring. On-site monitoring refers to a review of the data that takes place at the clinical site whereas central monitoring refers to activities that can be conducted at the data coordinating center.

Monitoring Activities will include:

- Review of credentials, training records, and delegation of duties logs.
- Review of 100% of Consent Forms.
- Review of reports on missed events, missing data, protocol deviations, and unanticipated problems.
- Comparison of CRFs to source documentation to ensure data are accurate and complete.
- Review of documentation for AEs, SAEs, and UPs.
- Review of critical fields such as eligibility, study endpoints, and SAEs.
- Regulatory Files: Limited reviews at interim visits, e.g., IRB annual reviews, safety reporting, IRB submissions of protocol deviations, and updated essential documents.
- Laboratory Review: Full laboratory review of processing and storage of specimens at first and close-out visits and at least biannually. Assessment of laboratory specimens stored at the clinical site.

The DMCC will provide copies of on-site monitoring reports within 10 business days of the visit.

Details of clinical site monitoring are documented in the PrecISE Clinical Monitoring Plan (CMP). The CMP describes in detail who will conduct the monitoring, at what frequency monitoring will be done, at what level of detail monitoring will be performed, and the distribution of monitoring reports.

10.1.8 QUALITY ASSURANCE AND QUALITY CONTROL

Each clinical site will perform internal quality management of study conduct, data and biological specimen collection, documentation and completion. An individualized quality management plan will be developed to describe a site's quality management.

Quality control (QC) procedures will be implemented beginning with the data entry system. The database will include real-time data QC checks. Any missing data or data anomalies will be communicated to the site(s) for clarification/resolution.

Following written Standard Operating Procedures (SOPs), the monitors will verify that the clinical trial is conducted and data are generated and biological specimens are collected, documented (recorded), and reported in compliance with the protocol, International Conference on Harmonization Good Clinical Practice (ICH GCP), and applicable regulatory requirements (e.g., Good Laboratory Practices (GLP), Good Manufacturing Practices (GMP)).

The investigational site will provide direct access to all trial related sites, source data/documents, and reports for the purpose of monitoring and auditing by the DMCC, and inspection by local and regulatory authorities.

10.1.9 DATA HANDLING AND RECORD KEEPING

10.1.9.1 DATA COLLECTION AND MANAGEMENT RESPONSIBILITIES

Data collection is the responsibility of the clinical trial staff at the site under the supervision of the site investigator. The investigator is responsible for ensuring the accuracy, completeness, legibility, and timeliness of the data reported.

All source documents should be completed in a neat, legible manner to ensure accurate interpretation of data.

Data recorded in the electronic case report form (eCRF) derived from source documents should be consistent with the data recorded on the source documents.

Adverse events will be coded using the Medical Dictionary for Regulatory Activities (MEDDRA) and concomitant medications will be coded using the National Drug Code (NDC) Directory.

Clinical data (including adverse events (AEs), concomitant medications, and expected adverse reactions data) and clinical laboratory data will be entered into the PrecISE Data Management System, a 21 CFR Part 11-compliant data capture system provided by the PrecISE DMCC. The data system includes password protection and internal quality checks, such as automatic range checks, to identify data that appear inconsistent, incomplete, or inaccurate. Clinical data will be entered directly from the source documents.

10.1.9.2 STUDY RECORDS RETENTION

Study documents should be retained for a minimum of 3 years after the completion of research in accordance with HHS regulations. These documents will be retained for a longer period, however, if required by local regulations. No records will be destroyed without the written consent of NHLBI. It is the responsibility of NHLBI to inform the investigators when these documents no longer need to be retained.

10.1.10 PROTOCOL DEVIATIONS

A protocol deviation is any noncompliance with the clinical trial protocol, International Conference on Harmonization Good Clinical Practice (ICH GCP), or Manual of Procedures (MOP) requirements. The noncompliance may be either on the part of the participant, the investigator, or the study site staff. As a result of deviations, corrective actions are to be developed by the site and implemented promptly.

These practices are consistent with ICH GCP:

- 4.5 Compliance with Protocol, Sections 4.5.1, 4.5.2, and 4.5.3
- 5.1 Quality Assurance and Quality Control, Section 5.1.1
- 5.20 Noncompliance, Sections 5.20.1, and 5.20.2.

Protocol deviations require reporting to the DMCC, and to the oversight IRB, and additional source/supporting documents may be requested and should be kept in the participant's record. Sites should notify the DMCC in accordance with the Manual of Procedures.

It is the responsibility of the site investigator to use continuous vigilance to identify and report deviations within 15 working days of identification of the protocol deviation, or within 15 working days of the scheduled protocol-required activity. All deviations must be addressed in study source documents and reported to the DMCC. Protocol deviations must be sent to the reviewing IRB per their policies. The site investigator is responsible for knowing and adhering to the reviewing IRB requirements. Further details about the handling of protocol deviations will be included in the MOP.

10.1.11 PUBLICATION AND DATA SHARING POLICY

This study will be conducted in accordance with the following publication and data sharing policies and regulations:

National Institutes of Health (NIH) Public Access Policy, which ensures that the public has access to the published results of NIH funded research. It requires scientists to submit final peer-reviewed journal manuscripts that arise from NIH funds to the digital archive PubMed Central upon acceptance for publication.

This study will comply with the NIH Data Sharing Policy and Policy on the Dissemination of NIH-Funded Clinical Trial Information and the Clinical Trials Registration and Results Information Submission rule. As such, this study will be registered at ClinicalTrials.gov, and results information from this trial will be submitted to ClinicalTrials.gov. In addition, every attempt will be made to publish results in peer-reviewed journals. Data from this study may be requested from other researchers after the completion of the primary endpoint manuscript by contacting NHLBI's BioLINCC. Industry partners who are providing study agents will be provided with serious adverse events reports that occur when participants are receiving their product. Additional details about the data sharing policy are provided in MOP Appendix 9.

In addition, this study will comply with the NIH Genomic Data Sharing Policy, which applies to all NIH-funded research that generates large-scale human or non-human genomic data, as well as the use of these data for subsequent research. Large-scale data include genome-wide association studies (GWAS), SNP arrays, and genome sequence, transcriptomic, epigenomic, and gene expression data.

The Publications Committee will be responsible for developing publication procedures and resolving authorship issues.

10.1.12 CONFLICT OF INTEREST POLICY

The independence of the PrecISE study from any actual or perceived influence, such as by the pharmaceutical industry, is critical. Therefore, any actual conflict of interest of investigators who have a role in the design, conduct, analysis, publication, or any aspect of this trial will be disclosed and managed. Furthermore, investigators who have a perceived conflict of interest will be required to have such conflicts managed in a way that is appropriate to their participation in the design and conduct of this trial. The study leadership in conjunction with the NHLBI has established policies and procedures for investigators to disclose all conflicts of interest and has established a mechanism for the management of all reported dualities of interest. These policies are outlined in the PrecISE Manual of Procedures.

10.2 ADDITIONAL CONSIDERATIONS

None

10.3 ABBREVIATIONS

The list below includes abbreviations utilized in this template. However, this list should be customized for each protocol (i.e., abbreviations not used should be removed and new abbreviations used should be added to this list).

ACE	Angiotensin-converting enzyme (ACE) inhibitors
ACQ-6	Asthma Control Questionnaire
ACRN TALC	Asthma Clinical Research Network Tiotropium Bromide as an Alternative to Increased Inhaled Corticosteroid in Patients Inadequately
AE	Adverse Event
ANCOVA	Analysis of Covariance
AQLQ	Asthma Quality of Life Questionnaire
ATS	American Thoracic Society
BD	Bronchodilator
BDR	Bronchodilator Response
BioLINCC	Biologic Specimen and Data Repository Information Coordinating Center
CARE	Childhood Asthma Research and Education Network Trial
CC	Coordinating Center
CF	Cystic Fibrosis
CFR	Code of Federal Regulations
CLIA	Clinical Laboratory Improvement Amendments
CMP	Clinical Monitoring Plan
CNS	Central Nervous System
COC	Certificate of Confidentiality
CONSORT	Consolidated Standards of Reporting Trials
COPD	Chronic Obstructive Pulmonary Disease
CRF	Case Report Form
DCC	Data Coordinating Center
DHHS	Department of Health and Human Services
DLCO	Diffusing Capacity for Carbon Monoxide
DMCC	Data, Modeling and Coordinating Center
DMS	Data Management System
DPI	Dry Powder Inhaler
DSMB	Data Safety Monitoring Board
DRE	Disease-Related Event
EC	Ethics Committee
eCRF	Electronic Case Report Forms
ED	Emergency Department
FDA	US Food and Drug Administration
FDAAA	US Food and Drug Administration Amendments Act of 2007
FFR	Federal Financial Report
FeNO	Fractional exhaled Nitric Oxide
GCP	Good Clinical Practice
GERD	Gastroesophageal Reflux Disease
GLP	Good Laboratory Practices

GMP	Good Manufacturing Practices
GWAS	Genome-Wide Association Studies
HADS	Hospital Anxiety and Depression Scale
HFA	Hydrofluoroalkane
HHS	Health and Human Services
HIPAA	Health Insurance Portability and Accountability Act
IAA	IRB Authorization Agreement
IB	Investigator's Brochure
ICH	International Conference on Harmonisation
ICMJE	International Committee of Medical Journal Editors
ICS	Inhaled Corticosteroids
ICU	Intensive Care Unit
IDE	Investigational Device Exemption
IND	Investigational New Drug Application
IRB	Institutional Review Board
ISM	Independent Safety Monitor
ISO	International Organization for Standardization
ITT	Intention-To-Treat
LABA	Long Acting Beta Agonists
LAMA	Long Acting Muscarinic
LSMEANS	Least-squares Means
MDCT	Multi-detector CT
MDI	Metered Dose Inhaler
MedDRA	Medical Dictionary for Regulatory Activities
MMRM	Mixed-Model for Repeated Measures
MOP	Manual of Procedures
MSDS	Material Safety Data Sheet
NCT	National Clinical Trial
NDC	National Drug Code
NHLBI	National Heart Lung and Blood Institute
NIH	National Institutes of Health
NIH IC	NIH Institute or Center
NSAIDS	Nonsteroidal Anti-Inflammatory Drugs
OHRP	Office for Human Research Protections
PAC	Participant Advisory Council
PEF	Peak Expiratory Flows
PFT	Pulmonary Function Test
PI	Principal Investigator
PrecISE	Precision Interventions for Severe and/or Exacerbation-Prone Asthma Network
QA	Quality Assurance
QC	Quality Control
SAE	Serious Adverse Event
SAP	Statistical Analysis Plan
SARP	Severe Asthma Research Program
SD	Standard Deviation
SFAR	Score for Allergic Rhinitis

sIRB	Single Institutional Review Board
SMC	Safety Monitoring Committee
SNP	Single Nucleotide Polymorphism
SNQ5	Sinunasal Questionnaire
SOA	Schedule of Activities
SOC	System Organ Class
SOP	Standard Operating Procedure
UNC	University of North Carolina
UNC-CH	University of North Carolina at Chapel Hill
UP	Unanticipated Problem
UPIRSO	Unanticipated Problem Involving Risk to Subjects or Other
US	United States
VCD	Pittsburgh Vocal Cord Dysfunction Index
WPAI	Work Productivity and Activity Impairment Questionnaire

10.4 PROTOCOL AMENDMENT HISTORY

The table below is intended to capture changes of IRB-approved versions of the protocol, including a description of the change and rationale. A Summary of Changes table for the current amendment is located in the Protocol Title Page.

Version	Date	Description of Change	Brief Rationale
2.5	9/10/2021	1.3 Schedule of Activities A triglycerides measurement was added at Visit 0 so that participants with high baseline triglyceride levels could be excluded from the clazakizumab treatment arm.	Safety
2.5	9/10/2021	5.1 Inclusion Criteria The inclusion criteria for stable asthma medications was clarified.	Clarification
2.5	9/10/2021	5.3 Randomization Criteria The randomization criteria for stable asthma meds in the 4 weeks prior to randomization was clarified.	Clarification
2.5	9/10/2021	6.3 Measures to Minimize Bias: Randomization and Blinding The text was clarified to specify that participants will not receive more than three placebos during their participation in the network.	Clarification
2.5	9/10/2021	8.2 Safety and Other Assessments The text was revised to clarify that spirometry may be conducted at home or in the clinic.	Clarification
3.0	11/19/2021	The sample size has been reduced from 800 to 600, the number of interventions has been reduced from six to five and the duration of the study has been extended from 42 months to 54 months. The text has been revised to clarify that participants will receive no more than three placebos during the study.	Clarification
3.0	11/19/2021	A training session was added at V0 to accommodate remote spirometry. Safety labs (pregnancy test, CBC and chemistry panel), FeNO and biorepository specimens are no longer	Clarification

		required for adolescents at visits X.2 and X.4. PBMCs will not be required at visit X.1 except visit 1.1. Some procedures (sputum, CT scans, PBMCs) will not be collected at all sites.	
3.0	11/19/2021	For participants whose asthma diagnosis has been verified at VSA/VSB but whose measure of poor asthma control at V0 is reliant on low lung function, the definition of the potential for improvement on study has been modified.	Recruitment
3.0	11/19/2021	The definition of participants at risk for bronchopulmonary dysplasia has been revised.	Recruitment
3.0	11/19/2021	The upper limits of tobacco consumption and vaping session frequency have been modified.	Recruitment
3.0	11/19/2021	The kidney function exclusion criteria (eGFR) have been revised.	Recruitment
3.0	11/19/2021	The kidney function criteria for discontinuing a participant's intervention was revised.	Retention
3.0	11/19/2021	The criteria for discontinuing a participant who meets the study's safety exclusion criteria was revised.	Retention
3.0	11/19/2021	The study now has a statistical analysis plan; much of the text has been moved from Section 9 of the protocol to the analysis plan.	Simplification
3.1	2/23/2023	1.1 Synopsis The sample size has been reduced from 600 to 395 and the duration of the study has been extended from 54 months to 62 months. The CompEx events endpoint has been moved from a primary endpoint to a secondary endpoint.	Clarification
3.1	2/23/2023	1.3 Schedule of Activities Physical exams are no longer required for adolescents at visits X.2 and X.4.	Clarification

3.1	2/23/2023	3 Objectives and Endpoints The CompEx events endpoint has been moved from a primary endpoint to a secondary endpoint.	Clarification
3.1	2/23/2023	6.5.4 Major Changes in Therapy The addition, dose adjustment or discontinuation of a beta blocker during the study is now considered a major change in therapy. Participants with a major change in therapy will discontinue treatment immediately and then withdraw the study.	Clarification
3.1	2/23/2023	8.1 Efficacy Assessments The CompEx events endpoint has been moved from a primary endpoint to a secondary endpoint.	Clarification
3.1	2/23/2023.	9.2 Sample Size Determination The sample size has been reduced from 600 to 395. The size required to enroll to each intervention has been reduced from 200 to 111.	Clarification
3.1	2/23/2023	5.2 Exclusion Criteria The smoking history exclusion criteria has been revised	Clarification
3.1	2/23/2023	6.5.4 Major Changes in Therapy It was clarified that the addition, dose adjustment, or discontinuation of an additional asthma controller is a major change in therapy	Clarification
3.1	2/23/2023	9.5.1 Statistical Analysis – General Approach	Clarification

		The secondary endpoint CompEx was removed from the interim futility analyses.	

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