Preventing Asthma in High Risk Kids (PARK) with Omalizumab: Design, Rationale, Methods, Lessons Learned and Adaptation

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Glossary of Abbreviations

Anti-IgE	Omalizumab
ATS	American Thoracic Society
CARE	Childhood Asthma Research Education
CASI	Composite Asthma Severity Index
ED	Emergency Department
ERS	European Respiratory Society
FDA	Food and Drug Administration
ICS	Inhaled Corticosteroids
IgE	Immunoglobulin E
IND	Investigational New Drug
INFANT	Individualized Therapy for Asthma in Toddlers
IRB	Institutional Review Board
LABA	Long-Acting Beta-Agonist
LTRA	Leukotriene Receptor Antagonist
MDI	Metered Dose Inhaler
MOP	Manual of Procedures
NAEPP	National Asthma Education and Prevention Program
NCICAS	National Cooperative Inner-City Asthma Study
NIAID	National Institute of Allergy and Infectious Diseases
PARK	Preventing Asthma in High Risk Kids
PEAK	Prevention of Early Asthma in Kids Study
PFS	Pre-Filled Syringe
PROSE	Preventative Omalizumab or Step-up Therapy for Severe Fall Exacerbations Trial

RTI	Respiratory Tract Illness (RTI)
SABA	Short-Acting Beta-Agonist
URECA	Urban Early Childhood Asthma study

Abstract: Asthma remains one of the most important challenges to pediatric public health in the US. A large majority of children with persistent and chronic asthma demonstrate aeroallergen sensitization, which remains a pivotal risk factor associated with the development of persistent, progressive asthma throughout life. In individuals with a tendency toward Type 2 inflammation, sensitization and exposure to high concentrations of offending allergens is associated with increased risk for development of, and impairment from, asthma. The cascade of biological responses to allergens is primarily mediated through IgE antibodies and their production is further stimulated by IgE responses to antigen exposure. In addition, circulating IgE impairs innate anti-viral immune responses. The latter effect could magnify the effects of another early life exposure associated with increased risk of the development of asthma – viral infections. Omalizumab binds to circulating IgE and thus ablates antigen signaling through IgE-related mechanisms. Further, it has been shown restore IFN-α response to rhinovirus and to reduce asthma exacerbations during the viral season.

We therefore hypothesized that early blockade of IgE and IgE mediated responses with omalizumab would prevent the development and reduce the severity of asthma in those at high risk for developing asthma. Herein, we describe a double-blind, placebo-controlled trial of omalizumab in 2-3 year old children at high risk for development of asthma to prevent the development and reduce the severity of asthma. We describe the rationale, methods, and lessons learned in implementing this potentially transformative trial aimed at prevention of asthma.

Introduction:

Asthma affects one out of 11 (7.1 million) U.S. children and is an incurable disease that degrades quality of life and may lead to significant, long term disability. A large majority of children with persistent and chronic asthma demonstrate aeroallergen sensitization (allergen-specific IgE antibody production), which remains a pivotal risk factor for developing persistent, progressive asthma throughout life[1, 2] . Furthermore, wheezing with viral infections during the first several years of life amplifies this effect[2-4]. In high risk children, aeroallergen sensitization precedes viral wheezing and is associated with the persistence and progression of the disease[5]. Most aeroallergen sensitization begins around age 1-3 years and escalates during school age[6]. Those who develop early sensitization are at greatest risk for persistent asthma and severe exacerbations of their disease[7-9]. The progression appears to be dependent on exposures to offending allergens-- the greatest incidence and impairment from asthma is seen in those who are sensitized and exposed to high concentrations of offending allergens with varying and complex relationships[10] [11-15]. Avoidance strategies aimed at environmental allergens are complex, time-consuming and often incomplete. However, several multi-faceted trials applying environmental exposure reduction in early life among at risk children have suggested long-term reduction in development and intensity of asthma [16-19] implicating that approaches to prevent allergic responses at a young age could potentially do more than control symptoms.

In addition to its role in mediating allergen-induced responses, IgE signals impair innate anti-viral immune responses[20, 21], which could lead to increased viral infections and thus potentially further enhance the cascade of progression to asthma. In experimental settings, IgE antibodies not only trigger mast cell-mediated hypersensitivity reactions but also act to promote Type 2 (allergic) immune responses and suppress the production protective Treg responses[22, 23]. This early Type 2 polarization

appears to prime children for asthma while augmenting susceptibility to viral induced lower respiratory infections, which can further induce the development of asthma.

Omalizumab (Xolair®) is a recombinant humanized monoclonal *anti-immunoglobulin E* (anti-IgE) antibody that works by blocking IgE-mediated processes and is FDA approved for children ages 6 years and above with allergic and severe asthma. It was shown in two NIAID funded asthma trials in inner-city children with asthma to markedly reduce asthma exacerbations in school aged children during the respiratory viral season[24, 25]. The Preventative Omalizumab or Step-up Therapy for Severe Fall Exacerbations (PROSE) trial showed that omalizumab treated children had restored anti-viral IFN-α response to rhinovirus (one of the most common causes of viral wheezing in school age children)[4, 25]. This suggests that omalizumab may prevent IgE driven responses to offending allergens and attenuate viral infections in those with Type 2 asthma. We describe the design, methods, and lessons learned of the PARK trial which tests the hypothesis that blockade of IgE in young children (age 2-3) at high risk for development of asthma will prevent asthma.

STUDY DESIGN AND METHODS:

Study Design

PARK is a 4 year multi-center, double-blinded, randomized, parallel design, placebo-controlled trial of omalizumab (anti-IgE) in children age 2-3 years at high-risk for developing asthma. Subjects are treated with omalizumab or placebo for 2 years and then followed for an additional 2 years to assess the development of asthma.

Study Population:

Rationale for Study Population

The proposed study population will be approximately 250 children aged 24 to 47 months of age at the time of screening who are at high risk for asthma. Potential participants will be assessed for criteria that

have been previously shown to correlate with subsequent persistent asthma in prospective studies of early childhood wheezing. The inclusion and exclusion criteria for this prevention study (see Table 1) were designed to strike a balance between indicating sufficient asthma risk (2 or more wheezing episodes) such that 2 years of injectable medication in small children was ethically acceptable and not such frequent symptoms (>4 wheezing episodes) that persistent asthma was already established.

Table 1. Inclusion/Exclusion Criteria

Inclusion Criteria	Individuals who meet all of the following criteria are eligible for enrollment as study participants: 1. Parent/guardian must be able to understand and provide signed and dated written informed consent; he/she must also be able to communicate with study staff 2. Age range: 24 through 47 months of age at the screening visit; participant must be under 4 years of age at the time of the randomization visit [exceptions for randomization of participants up to 48 months + 2 weeks may be allowed if an unforeseen situation occurs (e.g., lost samples, delays in IgE results, etc.)] 3. 2 to 4 wheezing episodes in the past year documented on physical examination by a health care provider. Wheezing events separated by at least 5 consecutive days without wheezing shall be counted as separate episodes. 4. Sensitization to one or more aeroallergens (by skin test wheal size at least 3 mm greater than negative control or allergen specific IgE ≥ 0.35 kU/L) 5. Diagnosis of asthma or allergy by a medical professional, or a positive test for allergy (skin test or serum test) in a first degree relative 6. Parent-reported history of participant having had either clinical varicella or administration of varicella vaccine 7. If participating in food immunotherapy treatment that is not part of a clinical trial, has been on an established maintenance regimen implemented continuously for a minimum of 2 months
Exclusion Criteria	Individuals who meet any of these criteria are not eligible for enrollment as study participants: 1. >4 episodes of wheezing in the past year 2. Use of Step 5 or Step 6 therapy (reference table 3) at the time of

	enrollment (Visit 0). (Previously use of inhaled steroids or inhaled steroids plus long acting beta agonists for respiratory symptoms for greater than 4 months in the past year was exclusionary - Adaptation 3/2020 and explained in Discussion) 3. Need for systemic corticosteroids or a hospitalization for respiratory symptoms within four weeks prior to screening 4. Three or more courses of systemic corticosteroids for wheezing illnesses in the last year 5. More than four days of symptoms of wheezing, or tightness in the chest or cough in the past two weeks causing at least minimal limitation of activity 6. More than four days of albuterol treatment (for symptoms) in the past two weeks 7. More than one night of symptoms of wheezing, chest tightness, or cough causing sleep disruption in the past two weeks 8. More than one night of albuterol treatment (for symptoms) in the past two weeks 9. Prematurity (<34 weeks gestation) 10. Need for oxygen for more than 5 days in the neonatal period 11. History of intubation or mechanical ventilation for respiratory illness 12. Other significant medical conditions, including but not limited to major congenital anomalies, cystic fibrosis, chronic pulmonary diseases, bronchopulmonary dysplasia, thoracic surgery, history of tuberculosis, immunodeficiency (primary or secondary), seizure disorders 13. Expecting to relocate within 4 years of study initiation to a place which would make in-person clinical visits impossible 14. Unable to adhere to study 15. activities 16. Prior aeroallergen immunotherapy or use of biologics including anti-IgE 17. Prior IVIG or systemic immunosuppressant other than corticosteroids 18. History of hypoxic seizures during a wheezing episode 19. Total IgE outside of the omalizumab dosing range 20. Enrolled in any other therapeutic interventional clinical trial within the past 30 days 21. Platelet count < 150 x 10^9/L at the screening visit 22. Past or current medical problems or findings from physical examination or laboratory testing that are not listed above, which, in the opinion of the investigator, may pose additional risks from participation in the study, may interfere with the participant's ability to comply with study requirements or may impact the quality or interpretation of the data obtained from the study. 23. History of severe anaphylactic/anaphylactoid reactions from any cause Exclusion criteria will be evaluated using 2-week recall If the participant meets any of the following criteria at any of the evaluations (screening, ICS step down run-in period or 4-week off ICS run-in period), then he/she is ineligible for randomization.

	1. More than four days of symptoms of wheezing, or tightness in the chest or cough in the past two weeks causing at least minimal limitation of activity 2. More than four days of albuterol treatment (for symptoms) in the past two weeks 3. More than one night of symptoms of wheezing, or tightness in the chest or cough causing sleep disruption in the past two weeks 4. More than one night of albuterol treatment (for symptoms) in the past two weeks 5. Symptomatic to the point of requiring controller medication [prednisone/prednisolone per the detailed rescue algorithm in the protocol, or inhaled corticosteroids with or without long-acting beta-agonists, other systemic corticosteroids, formoterol, theophylline, cromolyn, leukotriene antagonists (for wheezing), or salmeterol prescribed outside of the protocol] during the run-in/washout period. For participants in the ICS step down run-in, an increase in ICS step above the current step or the addition of other controller medication will be exclusionary. 6. Hospitalized for respiratory symptoms Patients may be re-enrolled into the run-in period if the subject fulfills all other inclusion/exclusion criteria and has not required hospitalization or systemic corticosteroids for four weeks prior to re-enrollment. Participants who come off controller medications or have their ICS decreased in order to enroll in the study may be enrolled in the run-in a maximum of two times.

Exclusion Criteria During Run-In	Enrolled participants enter a run-in period to characterize stability off asthma controllers and level of control at baseline prior to randomization. Those who exacerbate during the run-in period used to assess stability of respiratory symptoms to further evaluate eligibility for study participation. The length of the run-in and the schedule for respiratory symptom assessment depends on the medications a participant is taking and how long they have been taken at the time of the enrollment: • For participants not taking any controller medications at enrollment, and who have been stable for 2 weeks off controller medication, a single evaluation will occur after a 2-week run-in period (+1 week window). • Participants who are on ICS medication at a dose equivalent to Step 1 (as in Table 3) and/or are on a non-ICS controller medication (e.g., LTRA) at enrollment will enter a 4-week run-in period off all controllers. • Participants who have taken ICS medication at a dose equivalent to Steps 2, 3 or 4 for more than 4 consecutive months at enrollment will decrease by one step at 2-week intervals until they reach Step 0 (no controllers). A

	telephone contact will take place to assess stability before each dose reduction, and an in-clinic visit will occur before beginning the 4-week run-in period without ICS. The participant will be assessed at enrollment for stable respiratory symptoms and, if stable, will start the run-in by decreasing their ICS one step (e.g., a child who was taking Step 2 ICS will begin the run-in on Step 1 ICS). (Adapted March 2020 when protocol amended to include children on > 4 months ICS) • Participants who have taken ICS medications at a dose equivalent to Steps 2, 3, or 4 for less than or equal to 4 consecutive months at enrollment can be either stepped down as described above or have the medication discontinued at enrollment based on the judgment of the study clinician. • Once on Step 0 (no controllers), there will be two consecutive 2-week evaluations to confirm that the participant remained stable off ICS for a full 4-week period prior to randomization.

Study Procedures

Allergen Skin Testing: Skin testing will be performed to cat, dog, mouse, oak, aspergillus, alternaria, German/American cockroach mix, *D. farinae/D. pteronyssinus* mix, timothy grass and a clinic-specific battery of locally relevant allergens (not to exceed 16 tests in total including the positive and negative controls) at the PARK clinical centers by certified personnel**.** Participants will be asked to stop taking antihistamines for 5-7 days (dependent on the medication) prior to the visits at which the skin testing is performed to limit interference with the results of the skin test.

Pulmonary Function Test (Full-volume spirometry): Beginning at the week 80 visit, sites will begin teaching spirometry to participants. All spirometry will be conducted using the Global Lungs Initiative equations for predicted normal values. Pre- and Post- bronchodilator (4 puffs albuterol) spirometry will be conducted at Study Visits at weeks 92, 144 and 192.

At sites with airway oscillometry testing systems available, oscillometry will be performed consistent with ATS/ERS guidelines for preschool testing.

Venipuncture: A venous blood sample will be obtained at the initial screen visit, two months after initiation of treatment and then annually and analyzed for safety labs and other mechanistic labs as shown in Table 2 Schedule of Events.

Questionnaires: Respiratory questionnaire/history will be conducted in conjunction with monthly study visits during the treatment period and either via phone or clinic visits during the observation period using well validated surveys [24, 26, 27]. The same questionnaire scripts will be used by research clinic personnel during both visits and telephone interviews. Self-report accuracy will be enhanced by asking the parent/legal guardian to estimate medication use during the previous 1-week period. Two-week recall has been adopted for symptom assessment given the greater reliability of this interval compared to 1-month recall, based on the success of its use in the PEAK Trial[26] , the National Cooperative Inner-City Asthma Study (NCICAS)[27], and the Inner-City Asthma Study[24, 28]. Questionnaires evaluating the child's home environment, food allergies, eczema and rhinitis will be administered at baseline and at the end of each 48-week period. Standardized and validated tools for assessing outcomes will be utilized, such as the Composite Asthma Severity Index CASI)[29, 30].

At the monthly injection visit, surveys are administered to ascertain symptoms, all wheezing episodes, and clinically significant wheezing episodes. During the 96 week observation period, monthly phone calls will be conducted to ascertain symptoms and wheezing episodes and research clinic visits will be performed every 4 months.

House Dust Sampling Environment Assessment: In addition to reported allergen and tobacco smoke exposure through frequent surveys, which provide reasonable markers for exposure, we will improve our precision[31-33] with objective measures. Environmental home dust allergens will be collected at baseline, during treatment and observation periods, and measured using standard methods as

published by the Boston group[34] and others[35]. All vacuum dust samples will be collected in a standard protocol[27] that avoids contamination through use of new, clean, disposable collector filters by the participant's family or site staff and brought into clinic using a standardized and validated protocol[27] [34]. If the participant moves within the area, then one additional home sample will be collected in the same fashion. Additional dust samples from baseline, end of treatment and end of observation will be banked appropriately, providing opportunities for future sequencing of the environmental microbiome, should funding become available.

Study Visits and Assessments

Screening Visit (Visit 0): Children 2-3 years of age with parental report or documentation of episodes of wheezing in the past year will be invited for a screening visit. During this visit, we will obtain informed consent and further review inclusion and exclusion criteria.

Children who fulfill screening criteria by survey and whose parents agree to participation will receive allergy skin testing to relevant aeroallergens, provide serum for total and specific IgE, and have height and weight measured. Open-label albuterol metered dose inhaler (MDI) with AeroChamber® (Allergan, Dublin, Ireland) spacer and two courses of prednisolone/prednisone will be dispensed along with instructions on their use. Parents will also receive a specific respiratory symptoms action plan/rescue algorithm and instructions on how to recognize onset of respiratory signs and symptoms, and the protocol for contacting study staff/physician.

Run-In (2-10 Weeks between Screening Visit and Visit 1 Randomization):

Participants who are on controller medications at enrollment must be stable off all controller medications for at least 4 weeks prior to randomization. Participants on ICS medications at a dose equivalent to PARK Step 2, 3 or 4 for more than 4 consecutive months at the time of enrollment will

have their treatment decreased by one step every two weeks as described in **Table 1**. Participants who enroll on ICS will demonstrate at least a 4-week final run-in period to confirm stability off ICS prior to randomization. Those who are not on any controllers at enrollment will have a 2-week run-in. The length of the run-in will be two-to-ten weeks depending on controller medication usage, duration of use and step level of ICS treatment reported at the enrollment visit.

When total and specific IgE results become available, subjects who demonstrated either a positive skin test or positive allergen specific IgE ≥ 0.35 kU/L to at least one aeroallergen AND who are still eligible based on concentration of total IgE and other inclusion/exclusion criteria as detailed in **Table 1** will be invited to return to the clinic for the randomization visit (Visit 1).

Randomization and Treatment Phase Visits (Years 1 and 2; Visits 1-25):

Children between the ages of 24 and 48 months who satisfy the eligibility criteria during the run-in period for being at risk for the development of asthma will be randomized to omalizumab (anti-IgE) or its placebo, with clinical center, age (2-<3 years vs ≥3 years at randomization), total IgE (≤100 IU/mL and >100 IU/mL), and gender as stratifying variables, and using the adaptive randomization approach of Hu et al[36] to minimize treatment imbalance across strata. Roughly speaking, covariate adaptive randomization seeks to reduce treatment arm imbalance, with respect to important covariates, by altering the randomization probability when imbalance is detected. Unlike commonly employed block randomization, which reduces imbalance in a deterministic fashion by forcing at least one subject per block into the treatment arm that has fewer subjects, the covariate adaptive method preserves randomization for every subject, although not always 50:50. Also unlike block randomization, which focuses on imbalance at the stratum level alone, covariate adaptive randomization also reacts to imbalances at the factor margin level and at the overall level. The stratum level corresponds to the full cross classification of the stratification factors, in this case age, gender, IgE and clinical center. The factor

margin level corresponds to each factor individually, for example, balance across males and females irrespective of the other factors. Overall level corresponds to the overall number of subjects assigned to each treatment arm irrespective of all factors. Covariate adaptive randomization is achieved by calculating imbalance, the difference in the number of subjects currently assigned to each treatment arm, at each level (stratum, factor margin, and overall) and then summing those differences in a weighted fashion. When imbalance is present, treatment arm assignment for the next subject uses unequal randomization probabilities, also called biasing probabilities, in order to increase the chances that imbalance will be reduced. When imbalance is not present, treatment arm assignment for the next subject uses equal randomization probabilities. For the PARK study, we wished to avoid treatment arm imbalance on four different factors simultaneously (age, gender, IgE, and center) so that blocked randomization would have been impractical because there are 96 total strata (2 x 2 x 2 x 12). However, this is an ideal setting for covariate adaptive randomization. We chose to employ biasing probabilities of 0.15 and 0.85, along with a weighting scheme that gave equal weight (1/3 each) to imbalance at the strata level, at the factor margin level, and at the overall level. Our choice of biasing probabilities and weights was driven by suggestions in the adaptive randomization literature. We performed a simulation study to demonstrate that the scheme would prevent gross imbalances with high probability, but we did not investigate the performance characteristics of different biasing probabilities or weights.

Epinephrine auto injectors (Auvi-Q®, Kaleo inc., Richmond, VA, USA) 0.1 mg <15 kg, 0.15 mg 15-30 kg and 0.3 mg >30 kg) and education on their use will be provided to the families as a precautionary measure in the rare event of delayed anaphylaxis. The day after each investigational product administration, the parents will be contacted by telephone or e-mail/text (parent preference) to inquire about adverse events. If any problems are noted via e-mail or text, a phone call will take place to gather the details of adverse events and to ensure that the child has received any necessary treatment.

Dosing: Dosing was carefully considered for this age range which has not been extensively studies previously. Randomization of eligible participants was to subcutaneous omalizumab or placebo (1:1 randomization allocation ratio) every 4 weeks ensuring at least 0.016 mg/ kg/IU total IgE (measured at screening) in 75 mg increments utilizing pre-filled 75 mg and 150 mg syringes (PFS) with a maximum dose of 25 mg/kg. The dosing utilized was established in studies for the treatment of asthma in older children and adults and is described in the FDA approved package insert. The maximum dose is 10% of the dose at which thrombocytopenia was seen in non-human primates. The dosing interval was limited to every 4 weeks to improve acceptability for caregivers and participants. The use of a dosing algorithm will allow the widest range of children to be covered. Using this plan, we anticipated being able to provide sufficient omalizumab coverage to adequately dose 86% of children who would have met eligibility criteria based on data from the Childhood Asthma Research and Education (CARE) Network and The Urban Environment and Childhood Asthma (URECA)[35] cohort. We plotted weight and IgE levels in our cohort studies to determine the percentage of children eligible for dosing based on our criteria. Using data from the CARE network and URECA on similar aged children with similar risk factors, we projected that most children would start and remain on one injection every 4 weeks during the treatment phase. Depending on the patient's body weight and serum total IgE at the screening visit, the dose will vary between75 mg and 600 mg every 4 weeks.

The dosing of each subject is determined by computer algorithm using information on current weight and enrollment total IgE gathered from electronic case report forms. The computer algorithm will allow the investigators to estimate the number of shots the child will receive at the visits over the course of treatment, based on the child's weight percentile from CDC growth charts, and provide this estimate to parents prior to randomization. In the package insert, adjustments for weight are recommended in the treatment of asthma, although no specific interval for such adjustment is given. During the 24-month treatment phase, children in this study would be expected to reach 127% to 138%

of their enrollment weight. Doses will be adjusted every 24 weeks at Study Weeks 24, 48 and 72. Dose adjustments will be based on the most recent body weight measured in the clinic at one of the two preceding monthly visits.

Observation Phase (Years 3 and 4; Visits 25-32):

Participants will return to the research site every four months for clinical assessments and management of respiratory symptoms. Beginning at Week 100, participants will be contacted by phone each month, except when the calls would coincide with a clinic visit. The occurrence of respiratory symptoms and medication use will be obtained. The management of respiratory symptoms will also be conducted as described in a subsequent section. If indicated, a participant will be brought into clinic for an unscheduled visit.

Table 2. Schedule of Events

		Treatment Phase (96 weeks)														
Week		0	4 T4	**8&12** T8/12	16 T16	**20,24&28** **T20, T24, T28**	32 T32	**36, 40 &44** **T36 T40 T44**	48 T48	**52,56&60** **T52 T56 T60**	64 T64	**68, 72 & 76** **T68 T72 T76**	80 T80	**84,88** T84 T88	92 T92	96 T96 O 0
Visit #	0	1	V2	V3-4	V5	V6-8	V9	V10-12	V13[3]	V14-16	V17	V18-20	V21	V22-23	V24[3]	V25
Run In	+[1]															
Informed Consent	+															
Medical History	+															
Physical Exam Heart/Lungs/Skin/ENT	+					T24			+			T 72				+

		Treatment Phase (96 weeks)														
Week		0	4 T4	**8&12** T8/12	16 T16	**20,24&28** **T20, T24, T28**	32 T32	**36, 40 &44** **T36 T40 T44**	48 T48	**52,56&60** **T52 T56 T60**	64 T64	**68, 72 & 76** **T68 T72 T76**	80 T80	**84,88** T84 T88	92 T92	96 T96 O 0
Visit #																
Stadiometry and Weight[5]	+				+	T20		T40, 44			+	T68			+	+
Inhaler Technique Assessment[4]	+	+							+						+	
Blood Collection:																
Specific/total IgE	+															
Free IgE									+						+	
Safety Labs - CBC with differential and platelets, eosinophil count	+			T8					+						+	
Blood for Mechanistic Studies	+								+						+	
Allergy Skin Prick Test	+															
SQ Study Drug Administered[2]		+	+	+	+	+	+	+	+	+	+	+	+	+	+	
Study Drug Dose Determination		+				T24			+			T72				
Medical Management of Respiratory Symptoms		+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Spirometry/ Oscillometry Train													+	+	+	+
Spirometry															+	+
Post-Bronchodilator Spirometry (post 4 puffs albuterol)															+	
Oscillometry (if available)															+	+
CASI	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Questionnaires: ICAC/ISAAC/Home/Food		+							+						+	
Respiratory Action Plan Reviewed	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+

	Observation Phase (96 weeks)

Week	104 O 8	112 O 16	128 O32	144 O48	160 O64	176 O80	192 O96
Visit #	V26	V27	V28	V29[1]	V30	V31	V32
Physical Exam Heart/Lungs/Skin/ENT				+			+
Stadiometry and Weight	+	+	+	+	+	+	+
Inhaler Technique Assessment[2]				+			+
Blood: Specific, total IgE safety, mechanistics				+			+
Allergy Skin Prick Test							+
Medical Management of Respiratory Symptoms [4]	+	+	+	+	+	+	+
Spirometry/Oscillometry Train	+	+	+	+	+	+	+
Spirometry	+	+	+	+	+	+	+
Post-Bronchodilator Spirometry (post 4 puffs albuterol)				+			+
Oscillometry (if available)	+	+	+	+	+	+	+
CASI	+	+	+	+	+	+	+
Questionnaires: ICAC/ISAAC/Home/Food				+			+
Respiratory Action Plan Reviewed	+	+	+	+	+	+	

Study Management of Respiratory Symptoms

During the entire study, respiratory medications will be managed according to an algorithm based on the National Asthma Education and Prevention Program expert panel report 3(NAEPP EPR-3) guidelines for the diagnosis and management of asthma.

Because PARK will enroll children 2-3 years of age, we expect that nearly all subjects will experience at least one respiratory tract illness (RTI) during the course of this trial given the high prevalence of RTIs in this age group. While many of these RTIs will be limited to the upper airways, some RTIs may involve the lower airways and may therefore trigger or worsen respiratory symptoms in affected subjects[37]. A

rescue algorithm will be implemented which is similar to the approach used in the Prevention of Early Asthma in Kids PEAK trial[26], AsthmaNet preschool studies [38] and in the Inner City Asthma Consortium.

Each subject enrolled in this study will receive a respiratory symptoms action plan (PARK ACTION PLAN) outlining important triggers for starting treatment, including wheeze, increased work of breathing, or persistent disruptive cough.

Parents will be asked to contact the PARK Center as follows:

1. To inform them of worsening or persistent respiratory symptoms, or
2. If respiratory symptoms do not improve after prednisolone/prednisone treatment, or
3. After any unscheduled visit for respiratory symptoms, to either a primary care physician, sub-specialty physician, urgent care facility, or emergency department, or
4. After hospitalization for any reason, or
5. Whenever they have specific questions or concerns.

Parents will also be instructed by study staff, and directed by a respiratory symptoms action plan, to seek care immediately (e.g., urgent care or emergency department) for any symptoms requiring immediate medical attention, such as severe respiratory distress or rapidly progressive symptoms.

Parents will be instructed to call the investigative site to inform the study personnel that emergency care was sought, after the child's status has improved.

We will also assess criteria that indicate the need for immediate medical attention at all study visits and direct the family to seek emergency care if not already obtained.

Protocol for prednisolone/prednisone initiation for acute symptoms

Parents will be instructed to call the investigative site or the on-call study clinician if, according to the PARK respiratory symptoms action plan, they have followed instructions and believe that further treatment is indicated for the treatment of their child's respiratory symptoms.

Information will be obtained from the phone call to ascertain whether any one of the following situations exist:

A) Albuterol has been needed by inhaler/spacer or by nebulization for six or more individual treatments in the past 24 hours

B) Symptoms of wheezing, shortness of breath, or tightness in the chest or cough or severe pain that do not significantly improve after 3 or more doses of albuterol administered every 20 minutes over a period of 1 hour

C) wheezing, or tightness in the chest, or cough, pain, or shortness of breath, plus associated activity limitations with or without albuterol occurs for at least 5 of the preceding 7 days.

Parents will be instructed to initiate administration of the study-provided prednisolone/prednisone course only after the information obtained from the parent is reviewed by a study clinician. The prednisolone/prednisone course will consist of a 4-day course of oral prednisolone/prednisone: 2 mg/kg/day for 2 days (maximum 60 mg/day), followed by 1 mg/kg/day for 2 days (maximum 30 mg/day).

Follow-up of prescribed prednisolone/prednisone burst

If prednisolone/prednisone is recommended by PARK Clinical Center medical personnel, these personnel will telephone the parents 3-5 days after the initiation of the prednisolone/prednisone to reassess the

child's condition and determine whether an extension of prednisolone/prednisone courses may be warranted.

If the child is still symptomatic during the 3-5 days phone call and the PARK Clinical Center medical personnel are comfortable with telephone management of the child (based on their medical judgment), the prednisolone/prednisone course will be repeated (i.e., 2 mg/kg/day for 2 days [maximum 60 mg/day], followed by 1 mg/kg/day for 2 days [maximum 30 mg/day]). The study physician will make decisions on an extended course of therapy based on the child's clinical response to rescue therapy or if symptoms worsen, determine whether the child should be assessed in the clinical center or referred to urgent care or the emergency department for additional evaluation.

Management of Controller Medications for Persistent Symptomology

Controller therapy will be provided by the study and will be initiated when one of three conditions is met:

A. Persistent symptoms*, as described below, for at least 2 weeks after a 4-8 day corticosteroid burst

B. Two or more symptomatic wheezing episodes that require a corticosteroid burst in a six-month period

C. Need for a hospitalization for a symptomatic wheezing episode since the last assessment.

* Symptoms include –

a) daytime wheezing, chest tightness, or cough which occurs nine or more days in the past 2 weeks and causes at least minor activity limitation, or

b) nighttime symptoms of wheezing, chest tightness, or cough that disrupt sleep occurring at least three nights in the past 2 weeks, or

c) SABA use for symptom control (not prevention of exercise induced bronchospasm) nine or more days in the past two weeks.

Once the child meets criteria for the initiation of controller therapy, the child will be started on Step 1 therapy and adjusted as indicated (see Table 3. Medication Regimens)**.**

Table 3. Medication Regimens

Step	**Medication Equivalents**
0	SABA only
1	Fluticasone (Flovent®) MDI HFA 44 mcg/inh (1p BID)
2	Fluticasone (Flovent®) MDI HFA 44 mcg/inh (2p BID)
3	Fluticasone (Flovent®) MDI HFA 110 mcg/inh (2 puffs bid)
4	Fluticasone (Flovent®) MDI HFA 110 mcg/inh (2 puffs bid) plus Montelukast (Singulair®) 4mg po qd for 2- 5 year olds Montelukast (Singulair®) 5mg po qd for 6 years old and above
5	Fluticasone propionate and salmeterol 115/21 (Advair HFA) at 2 inhalations bid
6	Fluticasone propionate and salmeterol 230/21 (Advair HFA) at 2 inhalations bid

Adjustment of therapy after controller medication initiated

To accommodate for variation in the course of recurrent wheeze and preschooler respiratory symptoms, the protocol has established criteria for increasing and tapering asthma controller medications based on clinical symptoms and NAEPP EPR-3 guidelines [39].

Each month, the clinician will use information obtained via study questionnaires to determine the highest "symptom level" of the participant's morbidity described in **Table 4- Determination of Symptom Level**. During the treatment phase, the assessment will be made at each scheduled clinic visit. During the observation phase, the questionnaire will be completed by telephone every month, except when a clinic visit is scheduled which will allow for completion at the study site. Additionally, evaluation of a subject's respiratory status can be performed at an unscheduled clinic visit.

At the time of each assessment, the study clinician's decision to increase, decrease or keep same the controller medication is determined by applying the participant's "symptom level" to the treatment algorithm (**Table 5– Treatment Algorithm**). Adherence to the prescribed controller regimen further directs the treatment algorithm and will be obtained preferably from the medication counters, but per verbal report, if necessary.

Based on the Treatment Algorithm, the clinician then selects the appropriate treatment step (**Table 3 Medication Regimens**). The medication step level may be increased on a monthly basis as indicated by symptoms. Medication step levels cannot be decreased after a step level increase until a participant has been stable for at least 6 weeks; generally this will be two consecutive monthly assessments using 2-week recall, unless there is a 6 week duration between when the step level was increased and the following clinic visit/phone contact.

Table 4 - Determination of Symptom Level (2-week recall for symptoms and activity limitation)

Symptom Level	**# days with wheezing, or tightness in the chest or cough / two weeks and at least minimal limitation of activity**	**# days with rescue albuterol use/ two weeks**	**# nights of sleep disruption due to wheezing, or tightness in the chest or cough / two weeks**	**# nights use of albuterol for awakening / two weeks**	**Courses of systemic steroids for respiratory needs since last assessment**
Well Controlled (Symptom level 1)	0-4 days	0-4 days	0-1 night	0-1 night	0
Not Well Controlled (Symptom level 2)	≥5	≥5	≥2 nights	≥2 nights	≥1

Table 5 Controller Medication Treatment Algorithm (Applied only to subjects currently taking controller medications)

Symptom Level (from Table 4)	**Treatment Algorithm for Participants with Acceptable Adherence (≥50% of prescribed controller doses taken)**	**Treatment Algorithm for Participants with Unacceptable Adherence (< 50% of prescribed controller doses taken)**
Well Controlled (Symptom level 1)	If participant has been on their current Step (1-6) for <6 weeks – **no change** in treatment If participant has been on their current Step (1-6) for ≥6 weeks **– decrease** treatment by one step **unless** the participant has taken prednisolone for respiratory symptoms within the previous 28 days. Assess acute symptoms for initiation of prednisone	If participant has been on their current Step (1-6) for <6 weeks – **no change** in treatment If participant has been on their current Step (1-6) for ≥6 weeks **– decrease** treatment by one step **unless** the participant has taken prednisolone for respiratory symptoms within the previous 28 days. Assess acute symptoms for initiation of prednisone

Symptom Level (from Table 4)	Treatment Algorithm for Participants with Acceptable Adherence (≥50% of prescribed controller doses taken)	Treatment Algorithm for Participants with Unacceptable Adherence (< 50% of prescribed controller doses taken)
Not Well Controlled (Symptom level 2)	If currently on Steps 1-5, **increase** controller regimen by 1 Step or, If currently on Step 6, **continue** Step 6 Assess acute symptoms for initiation of prednisone	Continue same controller regimen or place on Step 2 therapy, whichever is higher Assess acute symptoms for initiation of prednisone

TREATMENT FAILURE

A participant will be considered a treatment failure and will be treated per physician discretion if any of the following criteria are met: 1) a participant requires 2 hospitalizations extending across more than one night per hospitalization for wheezing or asthma in a 12 month period, 2) requires intubation for acute wheezing or asthma exacerbation at any time, 3) has a hypoxic seizure during a wheezing or asthma exacerbation at any time, or 4) has more than 4 systemic steroid treatments for wheezing or asthma exacerbations within a 12 month period. If a treatment failure occurs then study drug will be stopped, but the child will continue to be followed in the study to evaluate long term safety and outcomes and to facilitate clinical care in concert with the participant's primary care or specialist provider.

Primary Outcomes: There are two primary outcomes: a predefined asthma outcome based on the NIAID funded URECA cohort[35] and asthma severity in the children who develop asthma assessed by the CASI[40]. The analysis plan for our proposed hypothesis of the two primary outcomes of asthma prevention and reduction of asthma severity in those who do develop asthma is described in the statistics and analysis section.

1. The diagnosis of current asthma at the end of the observation period.

A. 1 or more hospitalizations for wheezing/ asthma, or

B. 6 or more months of asthma controller use, or

C. 2 or more wheezing episodes, or

D. 2 or more Dr. or ED visits for asthma, or

E. FEV_1 reversibility $\geq$ 10% after 4 puffs albuterol PLUS

- 1 or more wheezing episodes
- 1 or more physician or ED visits for wheezing/ asthma

* A wheezing episode is defined as parental or documented report of an episode of wheezing or whistling in the chest that lasts at least 24 hours. Wheezing events separated by at least 5 consecutive days without wheezing shall be counted as separate episodes.

2. Average CASI score over the final three clinic study visits with the score set to zero for children without diagnosis of current asthma.

Secondary outcomes:

1. Number of wheezing episodes analyzed in each consecutive 48 week period (treatment and observation phases)
2. Number of clinically significant wheezing episodes analyzed in each consecutive 48 week period (treatment and observation phases)
3. Development of sensitization to new allergens as measured by in vitro (ImmunoCAP) and in-vivo (skin testing) assessment of allergen-specific IgE at the end of the observation period

Safety outcomes:

1. Number of adverse events will be analyzed by the frequency of adverse events

Exploratory Outcomes:

Exploratory outcomes to be analyzed separately in both the treatment (96 weeks) and observation (96 weeks) periods (unless otherwise limited) will include:

1. Time to first wheezing episode or clinically significant wheezing episode during the intervention and observation period
2. Lung function (FEV_1, FEV_1/FVC, FEF_{25-75}, bronchodilator responsiveness (% change in FEV_1 post albuterol))[41] at 92 weeks and during the observation period
3. Asthma-like symptom days in the past 2 weeks as modeled in the Inner-City Asthma Consortium defined as the largest value among three variables: number of days with wheezing, tightness in the chest or cough, number of nights with disturbed sleep as a result of asthma, and number of days on which the child had to slow down or discontinue play activities because of asthma[28]
4. Number of systemic corticosteroid courses
5. Time to first systemic corticosteroid course
6. Medical/health care utilization (i.e. number of hospitalizations for wheezing during the intervention and observation periods and Number of ED and unscheduled medical visits for wheezing during the intervention and observation periods)
7. Growth and BMI
8. Proportion of participants with physician diagnosed atopic dermatitis, allergic rhinitis, asthma, or food allergy throughout the trial
9. Proportion of participants with asthma primary outcome at the end of the first 48 weeks of the observation phase of the trial

10. Proportion of participants with atopic dermatitis defined as doctor's diagnosis of eczema and current symptoms of itchy rash in areas typical of eczema during the observation period
11. Proportion of participants with allergic rhinitis (seasonal or perennial rhinitis by questionnaire, and corresponding allergen specific IgE by skin tests and/or allergen specific IgE).
12. Number of food-induced allergic reactions in those participants who have physician diagnosed food allergy during the intervention and observation periodsAnalysis Plan

Analysis Population

For the 96-week observation extension, the enrollment target sample size is 250 participants, among whom we account for 20% dropout, giving an expected analysis sample of at least 200. The projected attrition is based on other similar studies such as Prevention of Early Asthma in Kids Study (PEAK), which had 10% dropout.

We will follow the intention-to-treat principle for the primary analysis, attributing the randomly assigned treatment to each subject regardless of adherence. Thus, the available data from all randomized children will be included in the statistical analyses, regardless of treatment failure status. As secondary analyses we will compare outcomes according to level of adherence, and also conduct an alternative analysis in which data collected after the assignment of treatment failure status are excluded.

A per protocol analysis will be performed in a similar manner to the intention-to-treat except the population analyzed will be those participants who receive at least 75% of injections, have missed 3 or fewer consecutive doses and have either completed visit 32 or have a phone interview in its place.

The safety population will include all participants who are randomized and received at least one dose of the investigational product.

Descriptive Analyses

Descriptive statistics will be calculated for all baseline measurements and characteristics, continuous variables (means and standard deviations, or medians and inter-quartile ranges) and categorical variables (frequencies). The descriptive statistics will be calculated based on the treatment arm allocation, in order to assess whether the two groups differ prior to the treatment period. Given the target sample size for this trial of 250 randomized children, it is expected that the two groups will be balanced with respect to demographics and prognostic variables.

Primary Analysis of Primary Outcomes

The first primary clinical trial outcome is dichotomous: diagnosis of current asthma during the final 48 weeks of the follow-up period off therapy. This will be assessed at the end of the trial since our definition includes diagnosis and symptoms/medication requirements in the past year. We will compare the proportion of subjects with current asthma diagnosis between treatment arms using the chi-square test with significance level 0.04.

The second primary clinical trial outcome, asthma burden, is semi-continuous: the CASI score averaged over the final 3 clinic study visits. The CASI score is a measure of asthma severity that takes integer values between 0 and 20 inclusive so the average CASI score over the final 3 visits is also constrained by the limits 0 and 20. Because the elements that comprise the CASI score are the same as those that determine asthma diagnosis: use of controller medication, severe wheezing episodes, hospitalization and lung function, the CASI score will necessarily be greater than zero for children diagnosed with asthma. It is possible for a child who does not have asthma to have an observed non-zero CASI score due to the occurrence of “asthma-like symptoms”. However, since we are interested in true asthma

burden, not the occurrence of asthma-like symptoms, the asthma burden score will be zero for all children without asthma diagnosis, regardless of what their CASI score would have been had they been diagnosed with asthma. We will compare the asthma burden score between treatment arms using the Wilcoxon test with significance level 0.01.

Analytic approaches for Secondary, Mechanistic, and Exploratory Outcomes

Secondary outcomes, listed above, include dichotomies, counts, time to event, and continuous measures. Dichotomous outcomes will be analyzed by logistic regression. Counts, typically small integers tabulated over a fixed period, will be analyzed by log-linear regression based on negative binomial likelihood, using covariates and effect modifiers as with the dichotomies and an offset to account for variable assessment periods. Continuous outcomes determined at baseline and follow-up, including the primary mechanistic endpoint (T-cell proliferation assay), will be analyzed by factorial repeated measures analysis of variance (ANOVA) with time × treatment interaction as the featured hypothesis, assessing the between-arm difference in change between baseline and follow-up. Skewed variables will be appropriately transformed for analysis. Covariate adjustments and tests of heterogeneity of effect will be applied as with the dichotomies and count data. Time-to-event outcomes will be compared between arms by Cox (proportional-hazards) regression, similarly to the above regression methods. For outcomes determined at multiple intervals during the trial, we will also employ repeated-measures ANOVA to compare the entire time course of the endpoint between treatment arms, forming and testing pertinent contrasts at time points of interest from parameters of the fitted longitudinal model. We will account for serial correlation in individuals' measurements by use of a compound-symmetric or autoregressive covariance structure, as appropriate for the endpoint. For the more frequently measured variables (e.g., growth) we will construct spline models, comparing the magnitude and placement of nonlinear features of the time course as well as the overall trend.

Supportive Analyses of the Primary and Secondary Outcomes

Supportive analyses of the primary outcomes will include further modeling approaches incorporating covariate adjustments and tests for interaction (effect modification, heterogeneity of effect). The first primary outcome can be modeled using logistic regression. Since the second primary outcome is a combination of binary and semi-continuous components, there is no natural location parameter to model. We will follow the approach of Vermeulen et al and model the marginal probabilistic index, which is the probability that a randomly chosen subject from the omalizumab group has a lower outcome than a randomly chosen subject from the placebo group.[42] In order to understand the treatment effect gradient, we will employ a pre-specified approach to assessing *heterogeneous treatment effect*, using baseline risk factors.[43] These will be assessed by interaction tests (covariate × treatment group). Of particular interest is the question of risk × treatment interaction, which addresses the possibility that the treatment could be more efficacious for more severely affected participants. A finding of significant heterogeneity in this regard would have important clinical implications. Further analysis, if we make such a finding, could include testing individual domains of the primary outcome and/or secondary outcomes that have multiple domains, such as the individual components of the CASI, to determine whether particular components may be driving the heterogeneity of response. A strength of the proposed study is that we have access to external tools (CASI) comprising several domains, which can be applied straightforwardly for a test of interaction with treatment[44]. If the interaction is significant we can, as suggested by Wang and Ware[45], divide the risk scale into categories and provide category-specific estimates of treatment effect. The same sort of analysis can be conducted with individual domain scores.

Also worth pursuing is development of an internal risk score, which has the advantage of *prima facie* pertinence to the study sample. To carry out this alternative assessment of risk we will use the

proportional-interaction model of Kovalchik[46] and Follman and Proschan[47]. Burke et al[48] obtained the best results using data from both arms to develop the internal multivariable score. To avoid overfitting, they recommend having at least 10 control-arm events per predictor variable. We project 50% will have diagnosis of current asthma in 100 placebo subjects, or 50 events; thus an internal tool would be limited to 5 predictors, not a severe constraint as CASI comprises only 5 domains, and other similar studies have even successfully employed a subset[49].

Exploratory analyses will also include examination of seasonal effects, allergen exposure concentrations, sensitization, and the combination of exposure + sensitization. These analyses will again include interaction terms in models to determine of any of these factors predict responsiveness to the intervention.

Missing Data

We will follow recently published guidelines for the handling of missing data[50]. The potential influence of missing covariates will be evaluated by first comparing dropouts to completers with respect and baseline or, if available, more recent characteristics. If no measured characteristics distinguish the groups, that constitutes evidence supporting the assumption that the missing data can be considered missing completely at random (MCAR), and the analysis methods described above can be relied on to produced valid estimates of treatment effect. If the groups differ with respect to recorded covariates only (missing at random, MAR), then the missing covariates can be filled in by multiple imputation methods. If the occurrence of missing values is not adequately explained by recorded variables, or the assumption of MAR is not adequately supportable (missing not at random, MNAR), we will assess the sensitivity of the results to hypothetical scenarios for biased dropout. The potential influence of missing endpoints will be assessed by conducting an alternative inverse-probability-weighted analysis. Weights are derived from a model of the full data in which the occurrence of a missing endpoint is the

dependent variable (rather than the value itself), and all available covariates are predictors. In addition, as noted above, lack of relationship between missingness and measured characteristics does not guarantee MCAR. Therefore, additional sensitivity analyses will be conducted by imputing the limiting cases for missing values under the null hypotheses (i.e., all missing values imputed as positive for asthma diagnosis or all missing values imputed as negative for asthma diagnosis).

Statistical Hypotheses

Our primary null hypotheses are:

1. That a current diagnosis of asthma at the end of the final observation year (Year 4), following 96 weeks' treatment of randomly assigned omalizumab or placebo and an intervening off-treatment year, will be equally frequent among trial subjects in the two groups. We will test this hypothesis with a two-sided test Type I error probability of 0.04 as described below. The alternative hypothesis is thus that the current diagnosis of asthma is not equal in the two groups, allowing for the outcome to be more frequent either in the omalizumab group or in the placebo group.
2. That the total asthma burden, combination of diagnosis and severity, at the end of the final observation year (Year 4), following 96 weeks' treatment of randomly assigned omalizumab or placebo and an intervening off-treatment year, will be equal among trial subjects in the two groups. We will test this hypothesis with a two-sided Type I error probability of 0.01 as described below. The alternative hypothesis is thus that the asthma burden is not equal in the two groups, allowing for the outcome to be higher in the omalizumab group or in the placebo group.

The significance levels 0.04 for the prevention outcome and 0.01 for the burden outcome were chosen to ensure that the total familywise type I error rate for the two primary null hypothesis tests will not exceed 0.05. In order to maximize the power for each test, decision rules for rejections will be based on the closure principle as shown below. If either the prevention null hypothesis or the burden null hypothesis is rejected, then the trial will be considered successful in terms of demonstrating a treatment effect. As a sensitivity analysis, we will repeat the primary analyses with p-values calculated using the permutation testing approach instead of the population sample likelihood approach.

Table 6. Power

Prevention outcome p-value	Burden outcome p-value	Reject Prevention Null Hypothesis	Reject Burden Null Hypothesis	Trial Successful
< 0.04	< 0.01	YES	YES	YES
0.04 – 0.05	< 0.01	YES	YES	YES
> 0.05	< 0.01	NO	YES	YES
< 0.04	0.01 – 0.05	YES	YES	YES
0.04 – 0.05	0.01 – 0.05	NO	NO	NO
> 0.05	0.01 – 0.05	NO	NO	NO
< 0.04	> 0.05	YES	NO	YES
0.04 – 0.05	> 0.05	NO	NO	NO
> 0.05	> 0.05	NO	NO	NO

For secondary and exploratory endpoints, our hypothesis is that the likelihood (for dichotomous endpoints), mean count (for countable endpoints), hazard (for time-to-event endpoints), mean value (for continuous endpoints), or time course (for repeated measures) is equal in the two treatment groups, with a two-sided alternative. We will use Type I error 0.05 as the criterion for statistical

significance without adjustment for multiple testing, following the rationale of Glantz and Slinker[51] by which each of the above planned comparisons represents a separate hypothesis of scientific interest.

We will use a more stringent criterion for exploratory hypotheses and apply formal methods such as the Holm step-down procedure[52] only in cases of pre-planned multiple testing, such as comparing levels of a multi-category predictor or assessing multiple simultaneously assayed markers. In those cases we will apply a familywise Type I error rate 0.05 and calculate the False Discovery Rate at each step by the method of Benjamini and Yekutieli[53].

Our calculations of detectable effect (below) uniformly assume power 0.9 (Type II error 0.1), reflecting a conviction that a study of this scale should provide low chance of failing to demonstrate clinically or biologically significant treatment effects whenever such effects are present.

SAS software (version 9.4 et seq.) and R will be used for statistical computations.

Sample Size Considerations

For the following calculations we set the Type I error rate (0.04, 0.01, or 0.05 as described above) and determined the magnitude of effect detectable with power 0.9 (Type II error 0.1), given our anticipated sample size (allowing conservatively for 20% attrition). We can thus argue that the study is adequately powered to demonstrate effects as small as those described here, covering the clinically or biologically significant range and justifying the large-scale effort that the study entails.

Clinical Trial.

Our sample of 250, allocated 1:1 (omalizumab: placebo) and allowing conservatively for 20% dropout, gives us 100 treated patients and 100 controls to compare.

- With respect to the prevention null hypothesis: If the risk of current asthma diagnosis is 0.50 in the placebo group, we will have power of at least 0.9 to detect an absolute risk decrease of 0.23 for omalizumab compared to placebo. This corresponds to a relative risk of 0.54 for omalizumab compared to placebo. The assumption for placebo is based on unpublished data from the NIAID URECA birth cohort, in which children who fulfilled the PARK inclusion/exclusion criteria at age 2-3 years of age demonstrated a 50% risk of developing asthma as defined by URECA at age 6-7. Assuming the known treatment effects of anti-IgE in school aged children (50-80% reductions in exacerbation rates)[24, 54] in comparison with the observed risk of current asthma diagnosis at age 6-7 in URECA, we are being appropriately conservative in our powered effect size. Furthermore, we conducted a sensitivity analysis of a range of effects based on treatment effects of other therapies in the PARK age range and their observed outcomes in the placebo arms (see section below).
- With respect to the burden null hypothesis: If the risk of current asthma diagnosis is 0.50 in the placebo group and the absolute risk decrease for omalizumab compared to placebo is 0.23, and if the average CASI score for those with asthma diagnosis in the placebo group is 3.5 and the average CASI score for those with asthma diagnosis in the omalizumab group is 2.7, then we will have will have power of at least 0.9 to detect a decrease in asthma burden for omalizumab compared to placebo. This corresponds to an absolute decrease of 0.8 in the average CASI score for those with asthma diagnosis, which is slightly smaller than the published Minimally Important Difference of 0.9. As noted above, the prevention null hypothesis is independent of any changes in severity among those with asthma diagnosis, whereas the burden null hypothesis depends on changes in both asthma risk and severity. Thus, it is possible to achieve high power for the burden outcome even if there is very little prevention effect as illustrated in the figures below. Although the maximum CASI score is 20, that value represents very severe asthma (daily

symptoms, more than 3 nighttime awakenings per week, lung function less than 70% of predicted, high-dose ICS controller therapy, and recent hospitalization). We anticipate relatively low CASI scores in the PARK study. We assumed a mean CASI score of 3.4 in the placebo group, which corresponds, for example, to low-dose ICS controller therapy with lung function greater than 85% of predicted, symptoms 3 days per week and one nighttime awakening per week. For the purposes of power calculations, we assumed that the CASI scores followed a negative binomial distribution with mean 3.4 and variance 3.6, which has a 95^{th} percentile of 7. So although the negative binomial would theoretically have to be truncated at 20, our assumptions entail a very low likelihood of CASI scores greater than 10. The Inner City Asthma Consortium, which developed the CASI score, suggested an MID of 0.9 with Hedges g statistic of 0.36 (JACI 2017). Our target effect size is 0.8 for power 0.9.

- With respect to secondary and exploratory outcomes: These can be grouped in the following four classes: dichotomous conditions assessed at the end of follow-up; count data measured in a fixed period; time-to-event endpoints; and measured variables, assessed in both groups at baseline and the end of the trial (including T-cell proliferation, the main mechanistic endpoint).
    - Dichotomous outcomes: The expected proportions drive the power calculation. For outcomes with relatively low expected proportions, such as 0.3, our sample size is adequate to detect a risk decrease of 65% (relative risk 0.35), but for outcomes with higher expected proportions, such as 0.7, our sample size is adequate to detect a risk decrease of only 35% (relative risk 0.65).
    - Count data outcomes: The expected rates drive the power calculation. For outcomes with a relatively low rate, such as 1.0, our sample size is adequate to detect a rate decrease of 50%, but for outcomes with a higher rate, such as 4.0, our sample size is adequate to detect a rate decrease of only 25%.

- Time to event outcomes: The expected numbers of events drive the power calculation. For relatively uncommon events, our sample size is adequate only to detect several-fold relative hazard; but for a more common event, likely to occur in about half the sample, a more modest effect size, around two-fold, will be demonstrable.
- Continuous outcomes: The standard deviation of individuals' pre-post change is $SD\Delta=SD\times(2(1-\rho))1/2$, where ρ is pre-post correlation. For (approximately) Gaussian distributed outcomes, the detectable difference in mean change between treatment groups lies between 0.38 and 0.58 SD, depending on pre-post correlation. These differences correspond to a shift in distribution, such that the mean in one group aligns roughly with the upper tertile boundary in the other group (percentile 64-72). This is a relatively small but potentially clinically significant shift, as one-sixth of the shifted group are newly located above the median of the other group. For outcomes that require a logarithmic transformation, detectable effect sizes are shown relative to the coefficient of variation (given as percentage).

Sensitivity analysis

The power of this study design to detect smaller or larger treatment effects for the first primary outcome was investigated across a range of risks in the placebo group as shown in the figure below. The target sample size of 250, with 20% attrition, will provide power of at least 0.79 for the targeted effect size even if the risk in the placebo group is as low as 0.40 (20% smaller than expected), and power at least 0.70 of the targeted placebo risk even if the relative risk is as high as 0.64 (20% smaller treatment effect than expected).

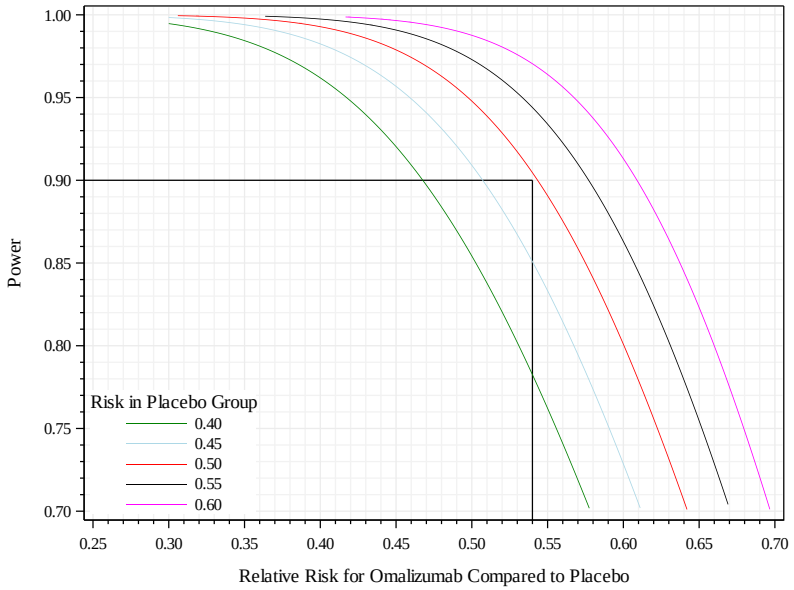

Figure 1.

Our assumptions are appropriately conservative by targeting a population at most risk for persistent, progressive disease that will also be directly targeted by our intervention. This allows us to focus on a population most likely to respond to the therapy but not so far along with established asthma that modification of progression or prevention will be less precise to detect.

The figure below illustrates the effect of asthma burden effect under different asthma prevention scenarios. If the true omalizumab relative risk is 0.54, the value on which the prevention outcome power is based, then the burden outcome power is high regardless of whether omalizumab affects the CASI score. If the true omalizumab prevention effect is weaker, for example relative risk is 0.65 (in which case the power for the prevention outcome is only 0.7), the power for the burden effect can still be adequate if omalizumab has an important effect on the CASI score. The overall probability of a successful trial (either or both null hypothesis rejected) is at least as high as the higher of the pertinent curves in the two panels below. For example, if the omalizumab prevention effect relative risk is 0.65 and the CASI effect is 1.0, then the overall probability of a successful trial is at least 0.80.

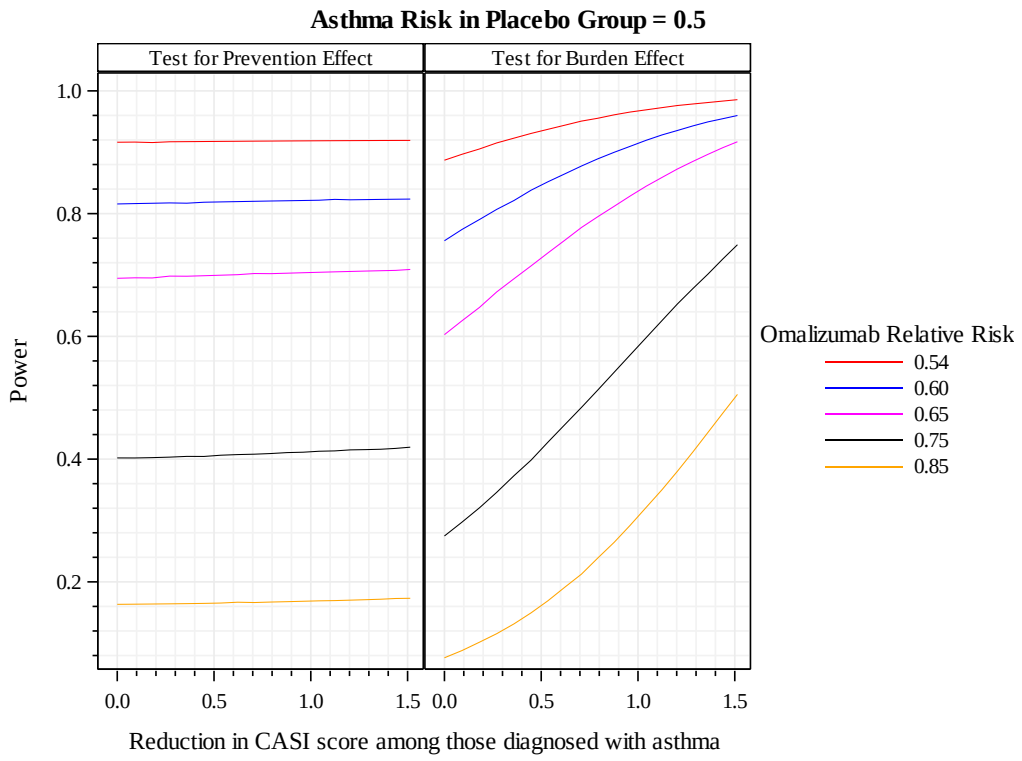

Figure 2

DISCUSSION

The PARK study is a unique intervention trial targeting IgE, the central antibody related to allergic sensitization and recently implicated in innate viral response, to determine if asthma can be prevented or the severity modified in young children at high risk of the developing the disease. PARK brings together a team of established childhood asthma investigators to design and implement the intervention trial that is challenged by maintaining scientific rigor while achieving feasibility of recruiting young children to receive serial injections in a placebo controlled trial. Even prior to starting the clinical trial, It took over a decade of working with the Food and Drug Administration and amassing real world omalizumab safety data for nearly two decades before an Investigational New Drug (IND) was granted down to age 2 years.

Our study is focused on prevention. The ability to assess this endpoint involved establishing eligibility criteria that met FDA requirements on treating a group of children at high enough risk to justify such a

treatment who did not already have established persistent asthma. We acknowledge the population treated within these constraints may limit the generalizability of our results. However, this will provide important insight into the role of early immunomodulation by treatment with omalizumab in such a population and leverage our understanding for further study.

Since starting this trial, there have been several lessons learned. Prevention studies in young children face unique challenges to enrollment compared to many other clinical trials. Prevention studies require the prospective participant's parents and caregivers to do a more complex and less concrete weighing of potential risk versus potential benefit over the long term as opposed to trials addressing an established need, potential for immediate symptom relief, and often shorter period of involvement for a pre-existing disease. Add to that the young age of the children and parenteral treatment, the decision faced by a parent or guardian is difficult. To date, the majority of participants have been recruited from the allergy/asthma specialty clinics and frequently have other allergic conditions. Family history of severe disease in the parents or older siblings is also a motivating factor. The most successful recruiters have been strong physician advocates. A physician champion in clinic who could clearly and confidently explain the rationale and goals behind the study is by far the most effective recruiting tool and this approach works well for most clinical trials since it is important for families to have an explanation for the study from someone who understands the treatment. Such enthusiasm might increase a placebo effect in shorter trials but we think it unlikely to have a substantial effect in a trial of this duration. Additionally, the protocolized asthma treatment algorithm should limit the placebo effect on outcome.

Online recruitment tools have also been helpful. We have added a multi-center website (https://parkstudy.org/) explaining in simple terms the study with tabs by location for interested participants to enter to obtain more information. Facebook and other awareness strategies are being

used and a simple blog https://vector.childrenshospital.org/2016/08/asthma-prevention-xolair/ made by the Boston Children's Hospital has been effective in increasing interest.

Once interested in participating, talented staff foster trusting relationships with the families and maximize sustained involvement. In our centers with a Spanish speaking populations, we adapted study materials, including the electronic CRFs, to Spanish and our committed, multi-lingual, cross-cultural study staff have been creative in ways to implement strategies for translation when bilingual staff and physicians were not available[35, 55].

Performing serial injections in a young child for clinical research can be traumatizing. To decrease discomfort and fear of the monthly injections, we've offered multiple strategies including numbing cream and Buzzy® (Pain care Labs inc., Atlanta GA, USA) distractors.

The protocol has needed modification as practice pattersn have changed. The focus is prevention and yet appreciable risk is required to justify the intervention, so, initially a "window of opportunity" was set requiring children to have 2 or 3 wheezing episodes but excluding those on > 4months of inhaled corticosteroids in the past year. Given the early use of daily inhaled corticosteroid recommended under current guidelines[56] , the restriction to 4 months of ICS impaired our ability to recruit and excluded children who might benefit the most from this approach. The current widespread ICS use does not necessarily identify patients who have asthma and the increase in the use of ICS in young children during a time of decreased incidence of asthma suggests that some of these children are overprescribed this type of therapy[57, 58]. Finally, it has been noted that in children aged 1- 4 years, wheezing phenotypes are often unstable. As an example, multi-trigger wheeze, often considered a surrogate for an asthma diagnosis in pre-schoolers, and episodic viral wheeze are not stable phenotypes, with 32% of children changing phenotype over time[59]. In support of this, the Individualized Therapy for Asthma in Toddlers (INFANT) trial enrolled children on EPR-3 Step 2 guideline

therapy for one year of treatment. The children were symptomatic at baseline (required an average of 2 oral corticosteroid courses in the 6 months prior to enrollment, and had an average of 5 wheezing episodes prior to study entry, with 15% hospitalized for wheezing) yet during the trial 1/3 of these children had very low rates of symptoms throughout the study (mean asthma control days above 95% during all treatment periods) [60]. This suggests that if we exclude such patients symptomatic to the point of needing step 2 therapy at baseline, we likely will exclude children without stable asthma who might benefit from the trial. For these reasons, we approached the FDA to modify the ICS exclusion criterion and were allowed to change the protocol as presented with the addition of protocolized monitoring to safely wean children currently using ICS >4 months (excluding children on step 5 or 6 at enrollment) and give them the opportunity to demonstrate stability off medications prior to enrollment.

In early 2020 the COVID-19 pandemic caused new safety challenges and suspension of enrollment. The focus shifted to providing therapy to children already randomized and obtaining safety labs. The pandemic and its attendant changes in standards for patient contact limited our ability to do some procedures and sampling. We worked to maximize remote participation. The FDA cleared and the IRB subsequently allowed us to offer home drug administration during the pandemic as long as 1) the child had no prior history of anaphylaxis; 2) at least 3 doses of omalizumab had been given in clinic; 3) training was provided to the parent/caregiver to recognize and manage signs and symptoms of a severe hypersensitivity reaction, including anaphylaxis, and 4) parent/caregiver was confirmed to be able to perform injections with omalizumab PFS with proper technique, based on the guidelines of Genentech and the FDA advisory due to the pandemic[61, 62]. We also added provisions for remote monitoring during injection. Delivering omalizumab or placebo to a family was a challenge due to temperature control requirements, and we provided an option for drive through pick up or courier service. As of this writing, we were able to continue to provide injections for most of the participants during the pandemic with in-person visits as first choice. Given the extra challenges and logistics in doing home injection, this

is reserved as a final resort for families who simply will not come to the research clinic during the pandemic.

PARK is a potentially paradigm-shifting study. The trial's innovation focuses upon early life intervention to modify the development and progression to disease during a critical time in lung growth and immune development, and relies on evidence that omalizumab completely blocks the effects of IgE-allergen interactions that 1) critically regulate progression to persistent asthma in susceptible individuals, 2) contribute to susceptibility to virally-induced exacerbations—both of which are fundamental in progression to established disease. If successful, omalizumab (anti-IgE) therapy will be the first intervention to significantly alter the progression from wheeze to established pediatric asthma. It will have tremendous impact on pharmacotherapeutic development and basic/translational approaches to asthma. If positive, the results could strengthen rationale for IgE targeted interventions by not only antibody-based mechanisms but further development of small molecule therapeutics, which are currently in the early stages of development. No matter what our results, the outcomes of this trial will provide significant insights into the pathobiology of asthma and potentially other IgE-mediated allergic diseases (i.e. food allergy and allergic rhinitis), and prepare us and others to identify the next steps in tackling asthma prevention, one of the most important unmet needs in asthma.

References:

1. Spahn, J.D. and R. Covar, *Clinical assessment of asthma progression in children and adults.* J Allergy Clin Immunol, 2008. **121**(3): p. 548-57; quiz 558-9.
2. Sly, P.D., et al., *Early identification of atopy in the prediction of persistent asthma in children.* Lancet, 2008. **372**(9643): p. 1100-6.
3. Kusel, M.M., et al., *Early-life respiratory viral infections, atopic sensitization, and risk of subsequent development of persistent asthma.* J Allergy Clin Immunol, 2007. **119**(5): p. 1105-1110.
4. Jackson, D.J., et al., *Wheezing rhinovirus illnesses in early life predict asthma development in high-risk children.* Am J Respir Crit Care Med, 2008. **178**(7): p. 667-672.
5. Jackson, D.J., et al., *Evidence for a causal relationship between allergic sensitization and rhinovirus wheezing in early life.* Am J Respir Crit Care Med, 2012. **185**(3): p. 281-5.

6. Arbes, S.J., Jr., et al., *Prevalences of positive skin test responses to 10 common allergens in the US population: results from the third National Health and Nutrition Examination Survey.* J Allergy Clin Immunol, 2005. **116**(2): p. 377-83.
7. Simpson, A., et al., *Beyond atopy: multiple patterns of sensitization in relation to asthma in a birth cohort study.* Am J Respir Crit Care Med, 2010. **181**(11): p. 1200-1206.
8. Stoltz, D.J., et al., *Specific patterns of allergic sensitization in early childhood and asthma & rhinitis risk.* Clin Exp Allergy, 2013. **43**(2): p. 233-41.
9. Havstad, S., et al., *Atopic phenotypes identified with latent class analyses at age 2 years.* J Allergy Clin Immunol, 2014. **134**(3): p. 722-727 e2.
10. Bacharier, L.B., et al., *Longitudinal Phenotypes of Respiratory Health in a High-Risk Urban Birth Cohort.* Am J Respir Crit Care Med, 2019. **199**(1): p. 71-82.
11. Sporik, R., M.D. Chapman, and T.A. Platts-Mills, *House dust mite exposure as a cause of asthma.* Clinical and Experimental Allergy, 1992. **22**(10): p. 897-906.
12. Sporik, R., et al., *Exposure to house-dust mite allergen (Der p I) and the development of asthma in childhood. A prospective study.* New England Journal of Medicine, 1990. **323**(8): p. 502-7.
13. Sporik, R. and T.A. Platts-Mills, *Allergen exposure and the development of asthma.* Thorax, 2001. **56**(Suppl 2): p. ii58-63.
14. Matricardi, P.M., et al., *Wheezing in childhood: incidence, longitudinal patterns and factors predicting persistence.* Eur Respir J, 2008. **32**(3): p. 585-92.
15. Illi, S., et al., *Perennial allergen sensitisation early in life and chronic asthma in children: a birth cohort study.* Lancet, 2006. **368**(9537): p. 763-70.
16. Arshad, S.H., et al., *Prevention of allergic disease during childhood by allergen avoidance: the Isle of Wight prevention study.* J Allergy Clin Immunol, 2007. **119**(2): p. 307-13.
17. Scott, M., et al., *Multifaceted allergen avoidance during infancy reduces asthma during childhood with the effect persisting until age 18 years.* Thorax, 2012. **67**(12): p. 1046-51.
18. Chan-Yeung, M., et al., *A randomized controlled study on the effectiveness of a multifaceted intervention program in the primary prevention of asthma in high-risk infants.[see comment].* Archives of Pediatrics and Adolescent Medicine, 2000. **154**(7): p. 657-63.
19. Chan-Yeung, M., et al., *The Canadian Childhood Asthma Primary Prevention Study: outcomes at 7 years of age.* J Allergy Clin Immunol, 2005. **116**(1): p. 49-55.
20. Durrani, S.R., et al., *Innate immune responses to rhinovirus are reduced by the high-affinity IgE receptor in allergic asthmatic children.* J Allergy Clin Immunol, 2012. **130**(2): p. 489-95.
21. Gill, M.A., et al., *Counterregulation between the FcepsilonRI pathway and antiviral responses in human plasmacytoid dendritic cells.* J.Immunol., 2010. **184**(11): p. 5999-6006.
22. Burton, O.T. and H.C. Oettgen, *Beyond immediate hypersensitivity: evolving roles for IgE antibodies in immune homeostasis and allergic diseases.* Immunological reviews, 2011. **242**(1): p. 128-43.
23. Logsdon, S.L. and H.C. Oettgen, *Anti-IgE therapy: clinical utility and mechanistic insights.* Curr Top Microbiol Immunol, 2015. **388**: p. 39-61.
24. Busse, W.W., et al., *Randomized trial of omalizumab (anti-IgE) for asthma in inner-city children.* N Engl J Med, 2011. **364**(11): p. 1005-15.
25. Teach, S.J., et al., *Preseasonal treatment with either omalizumab or an inhaled corticosteroid boost to prevent fall asthma exacerbations.* J Allergy Clin Immunol, 2015.
26. Guilbert, T.W., et al., *The Prevention of Early Asthma in Kids study: design, rationale and methods for the Childhood Asthma Research and Education network.* Control Clin Trials, 2004. **25**(3): p. 286-310.
27. Mitchell, H., et al., *Design and methods of the National Cooperative Inner-City Asthma Study.* Pediatric Pulmonology, 1997. **24**(4): p. 237-52.

28. Morgan, W.J., et al., *Results of a home-based environmental intervention among urban children with asthma.* N Engl J Med, 2004. **351**(11): p. 1068-80.
29. Wildfire, J.J., et al., *Development and validation of the Composite Asthma Severity Index--an outcome measure for use in children and adolescents.* J Allergy Clin Immunol, 2012. **129**(3): p. 694-701.
30. Krouse, R.Z., et al., *Minimally important differences and risk levels for the Composite Asthma Severity Index.* J Allergy Clin Immunol, 2017. **139**(3): p. 1052-1055.
31. Chew, G.L., et al., *Limitations of a home characteristics questionnaire as a predictor of indoor allergen levels.* American Journal of Respiratory and Critical Care Medicine, 1998. **157**(5 Pt 1): p. 1536-41.
32. Phipatanakul, W., et al., *Predictors of indoor exposure to mouse allergen in urban and suburban homes in Boston.* Allergy, 2005. **60**(5): p. 697-701.
33. Mascola, M.A., et al., *Exposure of young infants to environmental tobacco smoke: breast-feeding among smoking mothers.* Am J Public Health, 1998. **88**(6): p. 893-6.
34. Phipatanakul, W., et al., *The school inner-city asthma study: design, methods, and lessons learned.* J Asthma, 2011. **48**(10): p. 1007-14.
35. Gern, J.E., et al., *The Urban Environment and Childhood Asthma (URECA) birth cohort study: design, methods, and study population.* BMC Pulm Med, 2009. **9**: p. 17.
36. Hu, Y. and F. Hu, *Assymptomatic properties of covariate adaptive randomization.* Ann Stat 2012. **40**: p. 1794-1815.
37. van Wonderen, K.E., et al., *Stability and predictiveness of multiple trigger and episodic viral wheeze in preschoolers.* Clin Exp Allergy, 2016. **46**(6): p. 837-47.
38. Bacharier, L.B., et al., *Early Administration of Azithromycin and Prevention of Severe Lower Respiratory Tract Illnesses in Preschool Children With a History of Such Illnesses: A Randomized Clinical Trial.* JAMA, 2015. **314**(19): p. 2034-44.
39. *National Asthma Education and Prevention Program Expert Panel. Guidelines for the diagnosis and management of asthma - update on selected topics 2007. Bethesda, MD: US Department of Health and Human Services National Heart, Lung and Blood Institute.* 2007.
40. Wildfire, J.J., et al., *Development and validation of the Composite Asthma Severity Index-an outcome measure for use in children and adolescents.* J Allergy Clin Immunol, 2012. **129**(3): p. 694-701.
41. Tepper, R.S., et al., *Asthma outcomes: pulmonary physiology.* J Allergy Clin Immunol, 2012. **129**(3 Suppl): p. S65-87.
42. Vermeulen, K., O. Thas, and S. Vansteelandt, *Increasing the power of the Mann-Whitney test in randomized experiments through flexible covariate adjustment.* Stat Med, 2015. **34**(6): p. 1012-30.
43. Tsiatis, A.A., et al., *Covariate adjustment for two-sample treatment comparisons in randomized clinical trials: a principled yet flexible approach.* Stat Med, 2008. **27**(23): p. 4658-77.
44. Mathias, T.L., et al., *Cardiac Function and Short-Term Outcome in Patients with Acute Ischemic Stroke: A Cross-Sectional Study.* J Cardiovasc Dis, 2013. **1**(2): p. 26-29.
45. Wang, R. and J.H. Ware, *Detecting moderator effects using subgroup analyses.* Prev Sci, 2013. **14**(2): p. 111-20.
46. Kovalchik, S.A., R. Varadhan, and C.O. Weiss, *Assessing heterogeneity of treatment effect in a clinical trial with the proportional interactions model.* Stat Med, 2013. **32**(28): p. 4906-23.
47. Follmann, D.A. and M.A. Proschan, *A multivariate test of interaction for use in clinical trials.* Biometrics, 1999. **55**(4): p. 1151-5.
48. Burke, J.F., et al., *Using internally developed risk models to assess heterogeneity in treatment effects in clinical trials.* Circ Cardiovasc Qual Outcomes, 2014. **7**(1): p. 163-9.

49. Lemanske, R.F., Jr., et al., *Step-up therapy for children with uncontrolled asthma receiving inhaled corticosteroids.* N Engl J Med, 2010. **362**(11): p. 975-85.
50. Little, R.J., et al., *The prevention and treatment of missing data in clinical trials.* N Engl J Med, 2012. **367**(14): p. 1355-60.
51. Glantz, S.A. and B.K. Slinker, *Primer of Applied Regression and Analysis of Variance*. 1990: New York.
52. Holm, S., *A simple sequentially rejective multiple test procedure.* Scand J Statist, 1979. **6**: p. 65-70.
53. Benjamini, Y. and D. Yekutieli, *The control of the false discovery rate in multiple testing under dependency.* Ann Stat, 2001. **29**(4): p. 1165-1188.
54. Milgrom, H., et al., *Treatment of childhood asthma with anti-immunoglobulin E antibody (omalizumab).* Pediatrics, 2001. **108**(2): p. E36.
55. Zook, P.M., et al., *Retention strategies and predictors of attrition in an urban pediatric asthma study.* Clin Trials, 2010. **7**(4): p. 400-10.
56. *Expert Panel Report 3 (EPR-3): Guidelines for the Diagnosis and Management of Asthma-Summary Report 2007.* J Allergy Clin Immunol, 2007. **120**(5 Suppl): p. S94-138.
57. Bloom, C.I., et al., *Changing prevalence of current asthma and inhaled corticosteroid treatment in the UK: population-based cohort 2006-2016.* Eur Respir J, 2019. **53**(4).
58. Yang, C.L., et al., *Misdiagnosis of asthma in schoolchildren.* Pediatr Pulmonol, 2017. **52**(3): p. 293-302.
59. Raaymakers, M.J.A., et al., *Episodic viral wheeze and multiple-trigger wheeze in preschool children are neither distinct nor constant patterns. A prospective multicenter cohort study in secondary care.* Pediatr Pulmonol, 2019. **54**(9): p. 1439-1446.
60. Fitzpatrick, A.M., et al., *Individualized therapy for persistent asthma in young children.* J Allergy Clin Immunol, 2016. **138**(6): p. 1608-1618 e12.
61. *Genentech.*
62. Shaker, M., et al., *Estimation of Health and Economic Benefits of Clinic Versus Home Administration of Omalizumab and Mepolizumab.* J Allergy Clin Immunol Pract, 2020. **8**(2): p. 565-572.

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