Effect of 16-week dupilumab treatment on sinonasal respiratory symptoms and sense of smell in ethnically diverse patients with chronic rhinosinusitis and nasal polyposis (CRSwNP)

Study background and Rationale

Chronic rhinosinusitis (CRS) is one of the most common chronic diseases worldwide, with a prevalence of approximately 5% to 12% in the adult population.[1] Chronic rhinosinusitis with nasal polyps (CRSwNP) is an important phenotype of chronic rhinosinusitis.[2] Recent additions of biologic therapies to improve disease control have been shown to be effective in patients with CRSwNP.[3,4] These treatments are designed to improve objective measures of nasal polyps and patient-reported symptoms and their sense of smell among CRSwNP patients. They also prevent a repeated use of systemic corticosteroids and decrease the need in recurring sinus surgeries. Dupilumab has been shown to be effective in reducing nasal polyp size, radiologically assessed sinus opacification, and improving patient-reported symptom scores.[3,5] In patients with CRSwNP and comorbid asthma, dupilumab improves asthma control.[6,7]

While dupilumab efficacy in patients with CRSwNP has been confirmed in multiple studies, a notable weakness of these studies is that they have been conducted in a predominantly White populations or in populations of unknown ethnicity, making generalizability of the results difficult. If race or ethnicity were reported, the inclusion of non-White population was limited to one person in one study.[3] Black or African-American patients have not been specifically mentioned in the demographics tables of the SINUS 24 or SINUS 52 dupilumab trials. In the SINUS 24 trial, out of the 276 total patients, only 12 were listed as non-White ("Other"), which comprises only 4.3% of the patients.[5] Of note, in Figure S5A of this publication, the metric for Nasal Congestion and Nasal Polyp Score in Latino patients, indicated that dupilumab did not statistically significantly improved nasal congestion or nasal polyp score in six Latino study participants.

Therefore, our knowledge on how dupilumab works in CRSwNP patients from diverse ethnical background, is limited. To address this gap, we propose to recruit African American and Latino patients with CRSwNP from the Bronx borough of New York City.

There is no biologic evidence or clinical anecdotal cases to suggest that dupilumab would work differently in non-White patients, and our clinical experience in the Bronx thus far has shown us that dupilumab is therapeutically very efficacious in our minority patient populations. However, it is

important to acknowledge that CRS has several endotypes, which are separated by geography and ethnicities.[8] African-American patients with CRSwNP and comorbid asthma tend to have a more severe disease with increased asthma hospitalizations, compared to White patients.[9] The reasons for the differences in disease presentation are not well understood. Some biomarkers have different levels among White and other populations. For example, non-White patients with asthma tend to have higher immunoglobulin E (IgE) levels than White patients.[10-12] In addition, African-American patients with asthma exhibit greater eosinophilic airway inflammation than White patients with asthma.[12] It is not well understood what causes these differences, although socio-economic and environmental exposures could be at play.[13,14] African-American ancestry is associated with higher odds of asthma and asthma severity in Latinos.[15,16] In addition, African-American and Latino patients with AERD were more likely to have worsening of respiratory symptoms and a decline in FEV1 during aspirin treatment than White patients, indicating that their response to treatments may be different.[17]

Dupilumab treatment for patients with asthma affects both eosinophil counts and IgE levels. Its efficacy in asthma control has been shown to be higher in patients who had higher baseline peripheral blood eosinophil counts.[18] The median eosinophil count of the patients in LIBERTY ASTHMA trial was 255 cells/μL.[18] In two nasal polyposis studies on dupilumab efficacy (LIBERTY NP SINUS-24 and LIBERTY NP SINUS-52) the mean eosinophil count was 430 cells/μL (SD ±340).[5] In the dupilumab asthma study, the median IgE level was 167 IU/mL, the mean IgE level was 432 IU/mL (SD ±747), while in dupilumab nasal polyp study, mean IgE level was 229 IU/mL (SD ±318).[5,18]

To highlight the differences between the previously studied cohorts and the cohort of patients with CRSwNP from Bronx, NY, the mean is 644 cells/μL (SD ±550) and the median values of eosinophil counts in our cohort is 400 cells/μL (IQR 200-900). The mean IgE in our cohort is 821 IU/mL (SD ±1573), and the median is 312 IU/mL (IQR 121-756), nearly a double of what has been observed in previously studied populations. These characteristics indicate some of the specific differences in CRSwNP presentation between patients from the published cohorts and the patients from our cohort.

Our established cohort is heavily represented by patients with aspirin-exacerbated respiratory disease (AERD), all of whom have CRSwNP and asthma, as well as aspirin-tolerant control patients with CRSwNP but without AERD. This cohort is one of the few organized cohorts of well-phenotyped AERD patients and the only organized cohort of AERD minority patients available for study in the U.S.[17,19,20] African-American and Latino patients make up 75% of our ethnically-diverse cohort.

With this study, we will evaluate some of the potential immune mechanisms that contribute to CRSwNP. A goal is to identify pathways by which dupilumab may "normalize" the immune response in patients with CRSwNP and prevent nasal polyp recurrence.

Several immunological features have been identified in CRSwNP that have been suggested as relevant drivers of pathology. CRSwNP patients have high systemic levels of pro-inflammatory eicosanoids prostaglandin D_2 (PGD_2) and leukotriene E_4 (LTE_4) that are especially elevated in patients with AERD.[21] AERD patients tend to have a greater recurrence rate of the nasal polyps and greater need for surgeries.[22-24] Published studies show that the change in PGD_2 metabolite levels during reactions to aspirin in AERD patients mirrors the pattern seen in anaphylactic reactions to an allergen.[25,26] This suggests that allergy to an environmental antigen or colonizing microbe may be involved in the mechanism of AERD. One candidate for this is *Staphylococcus (S.) aureus*, which is commonly present in nasal polyps, and is a known potential driver of Th2-responses.[27-34] We found a high frequency of *S. aureus*-specific serum immunoglobulin E (IgE) among CRSwNP patients. Results from studies by us and others show that after surgical removal of the nasal polyps, disease burden in most CRSwNP with AERD patients diminishes as reflected in better control of asthma and in reduced NSAID sensitivity. [19,35,36] These changes are accompanied by a decrease in urinary PGD_2-metabolite (PGDM) and $uLTE_4$ levels.[19] In patients with undetectable levels of PGD_2 in plasma following surgical polypectomy (more than one-third of the study population), clinical reactions to aspirin were completely eliminated.[19,35] These observations point towards an important contribution of PGD_2 to immune deregulation associated with nasal polyps.

To understand the causes of the immune deregulation in CRSwNP, we have been studying several specialized or unconventional subsets of lymphocytes that are known or suspected to be strong drivers of allergic inflammation. We have noticed a striking increase in a subset of the unconventional T lymphocytes known as iNKT cells, as well as in ILC2 cells and Th2A cells in the nasal polyp tissue, suggesting a direct role for them in inciting or amplifying the disease. An *in vitro* stimulation with PGD_2, *S. aureus* or the combination of both, led to an increased expression of CRTH2 receptors on peripheral blood iNKT cells, Th2A cells, and augmented IL-4- and IL-13 production by these lymphocyte subsets. *S. aureus* is a frequent colonizer of the nasopharynx,[27-30] and is known to stimulate Th2 and iNKT cell activation.[28,37,38]

Taken together, there are important differences between the previously studied and largely White populations, in which dupilumab efficacy has been established, and our diverse Bronx population of

patients with CRSwNP with and without comorbid asthma. The proposed study is designed to answer the important question of dupilumab efficacy in this ethnically diverse group of patients with CRSwNP that is characterized by higher baseline eosinophil counts and higher baseline serum IgE levels.

Hypothesis

The central hypothesis of this proposal is that the addition of dupilumab treatment onto standard-of-care intranasal corticosteroids will improve patient-reported measures of disease activity and sense of smell in a cohort of mostly ethnical and racial minority patients with CRSwNP.

The proposed study will address important research needs in the following ways. First, it will confirm the effectiveness of dupilumab the treatment of CRSwNP in ethnic and racial minority patients who have traditionally been underrepresented in existing clinical trials of biologics in CRSwNP (Primary Objective). Second, this research will establish a biomarker of therapeutic response to dupilumab (Secondary Objective) identifying biological effect of dupilumab in CRSwNP patients and its association with symptom improvement. We will determine the effect of dupilumab on PGD_2 receptor and cytokine expression by several lymphocyte subsets by analyzing changes to *in vitro* stimulation with *Staphylococcus (S.) aureus* and prostaglandin D_2 (PGD_2) and in patients treated with dupilumab. We will compare the effect of dupilumab on cellular function of lymphocyte subsets from CRSwNP patients to cellular function of lymphocytes from peripheral blood and sinonasal tissue of control patients who undergo endoscopic nasal surgeries for reasons other than CRSwNP (e.g., correction of deviated septum). Dupilumab has a strong effect against Th2-associated inflammation. Therefore, our hypothesis is that dupilumab will reduce the expression of Th2-proinflammatory markers on lymphocytes and lead to a “normalization” of the immune response in CRSwNP patients. Dupilumab-induced changes will be compared to cellular characteristics of the controls, who do not have CRSwNP.

Finally, we will measure the effect of dupilumab on asthma symptoms and lung function in an ethnically diverse cohort of patients with CRSwNP and comorbid asthma. In accordance with NIH guidelines, ancestral background will be self-identified by the patients.[39]

Primary Objectives

1. Measure the effect of 16-week dupilumab treatment on nasal polyp symptoms in a cohort of mostly ethnical and racial minority patients.

1(a) To determine the effect of dupilumab treatment on Sinonasal Symptom Outcome Test (SNOT-22) scores in a diverse patient population with CRSwNP

2. Measure the effect of dupilumab on sense of smell in a cohort of mostly ethnical and racial minority patients with CRSwNP
   2(a) To determine dupilumab treatment effect on University of Pennsylvania Smell Identification Test (UPSIT™) and on nasal peak flow (NPF) values in patients with CRSwNP

Primary Endpoints

In patients with CRSwNP, we will
1(a) Measure SNOT-22 values at baseline, after 2 weeks, and after 16 weeks of dupilumab treatment
2(a) Measure UPSIT scores and NPF values at baseline, after 2 weeks, and after 16 weeks dupilumab treatment

Secondary Objectives

1. Measure the effect of 16-week dupilumab treatment on the biomarkers in a cohort of mostly ethnical and racial minority patients with CRSwNP
   1(a) To determine dupilumab treatment effect on urinary leukotriene E_4 ($uLTE_4$) levels in a cohort of mostly ethnical and racial minority patients with CRSwNP.
   1(b) To determine dupilumab treatment effect on peripheral blood eosinophil counts and serum immunoglobulin E (IgE) levels in a cohort of mostly ethnical and racial minority patients with CRSwNP.

2. Assess the effect of dupilumab on receptor expression and cellular function in several lymphocyte subsets from peripheral blood and nasal polyp tissue of patients with CRSwNP and controls to *in vitro* stimulation with *Staphylococcus (S.) aureus* and prostaglandin D_2 (PGD_2).

3. Determine the effect of dupilumab treatment for 16 weeks on the biomarkers in patients with CRSwNP

Secondary Endpoints

In patients with CRSwNP, we will

1(a) Compare changes in $uLTE_4$ levels from baseline, after 2 and after 16 weeks of dupilumab treatment

1(b) Compare peripheral blood eosinophil counts and IgE level change from baseline, after 2 and after 16 weeks of dupilumab treatment

2(a) Determine dupilumab effect *in vitro* on PGD_2 receptor expression (D-type prostanoid receptor 1 (DP1) and chemoattractant receptor-homologous molecule expressed Th2 cell (CRTH2) receptor) on lymphocyte subsets from peripheral blood and sinonasal tissue cells (type 2 innate lymphoid cells – ILC2, T-helper 2 allergic cells – Th2A, and invariant Natural Killer T (iNKT) cells) at baseline in patients with CRSwNP and compare to controls - patients without CRSwNP who undergo elective endoscopic nasal surgeries for reasons other than CRSwNP (e.g., correction of deviated septum).

2(b) Assess the effect of dupilumab on cellular function of blood and nasal polyp lymphocyte subsets from CRSwNP patients by measuring responses to *in vitro* stimulation with *S. aureus* and PGD_2, and assess changes in expression of GATA-3 (i.e., marker of Th2 differentiation), T-bet (i.e., marker of Th1 differentiation), FOXP3 (i.e., marker of T-reg differentiation), and cytokines, IL-4, -5, -9, -10, -13, and IFNγ.

3(a) Determine dupilumab effect on changes overtime in DP1 and CRTH2 receptor expression on rare lymphocyte subsets from peripheral blood in patients with CRSwNP treated with dupilumab for 4 weeks, 8 weeks, and 16 weeks

3(b) Determine dupilumab effect on changes overtime in expression of GATA-3, T-bet, FOXP3, IL-4, -5, -9, -10, -13, and IFNγ by several lymphocyte subsets from peripheral blood in patients with CRSwNP treated with dupilumab for 4 weeks, 8 weeks, and 16 weeks

Exploratory Objectives

1. To determine the effect of dupilumab on asthma symptoms in an ethnically diverse patient population with CRSwNP and comorbid asthma
2. To determine the effect of dupilumab on lung function, as measured by forced expiratory volume in 1 second (FEV1), in an ethnically diverse patient population with CRSwNP and comorbid asthma
3. To measure longitudinal changes from baseline in Nasal Fractioned Exhaled Nitric Oxide (N-FeNO) levels in a subset of CRSwNP patients with and without AERD
4. To determine the effect of dupilumab on tissue and peripheral blood lymphocyte subsets both, eicosanoid metabolism, T2-inflammatory markers, and sinonasal microbiome changes in patients treated with dupilumab and *in vitro.*

Exploratory Endpoints

In patients with CRSwNP and comorbid asthma we will:

1. Compare change from baseline in asthma control questionnaire (ACQ) score after 2 and after 16 weeks of dupilumab treatment
4. Compare change from baseline in FEV1 after 2 and after 16 weeks of dupilumab treatment
5. In a subset of CRSwNP patients with and without AERD (10 to 15 patients per group) we will determine longitudinal changes in N-FeNO during dupilumab treatment
6. In patients with CRSwNP we will collect sinonasal tissue, nasal fluid, and peripheral blood samples during dupilumab treatment and assess dupilumab effects on changes in lymphocyte activation, eicosanoid metabolism, T2-inflammatory markers, and sinonasal microbiome changes, both in vivo and in vitro in a future study.

Study Design

At Montefiore AERD Center and the Combined Chronic Rhino-Sinusitis Clinic, we have a large population of patients with CRSwNP. We will offer study participations to the patients on an ongoing basis during their scheduled visits at either clinic. The first 60 patients interested in the proposed study will be included in it. We anticipate enrolling 20 Latino, 20 African American, non-Latino, and 20 White, non-Latino patients with CRSwNP.

We evaluate 5-10 patients with CRSwNP per week. Despite COVID-19 restrictions we continue seeing approximately the same number of patients in the last 3 months. Therefore, we conservatively anticipate that we would be able to meet recruitment goals within the 18 months. Since this is a “real-world” observational study, approval-related delays could potentially impact the enrollment. Therefore, we suggest a total study duration of 24 months.

- The patient sample size of 60 was chosen in part due to feasibility (we estimate 100 patients will meet inclusion criteria and expect about half of them to be interested in participation in this study), and in part due to sufficiency in sample size as these patients have been chosen to be on the relatively severe spectrum of disease (have sought out medical care at a tertiary care CRSwNP Center, and on at least one controller medication for asthma if patients have a co-morbid asthma) – this will allow us to gather the most robust data regarding therapeutic efficacy and side effects, and will allow us to see any window of change in the high level of healthcare utilization for patients who initiate biologics.

- We will also recruit 10 individuals in the control group who have no history of nasal polyps or asthma and undergo sinonasal surgery for other reasons, for example deviated septum.
- Based on our previous recruitment experience, the estimated drop-out rate should be <10%. Since study visits are aligned with the clinical visits, patients are likely to come for follow-ups.
- The study is a prospective, longitudinal 16-weeks study.

This is a single-site study. All participants are patients of Montefiore Medical Center Allergy and Immunology and/or Otolaryngology clinics.

Inclusion Criteria

1. Patients with physician-diagnosed CRSwNP, with or without comorbid asthma that meet indication criteria for FDA-approved use of Dupixent.
2. Patients aged 18 years and older.
3. Patient willing to provide consent to be a participant in the study.
4. Patients with insurance that allows Dupixent coverage or Dupixent coverage obtained through Dupixent MyWay Program

Exclusion Criteria

1. Age under 18
2. Suspected or diagnosed allergic fungal rhinosinusitis.
3. Suspected or diagnosed cystic fibrosis.
4. Dupixent coverage denied through insurance or Dupixent MyWay Program
5. Patients who required a steroid taper in the preceding 30 days. However, patients on chronic steroids equal to ≤20 mg of prednisone daily, are eligible.
6. Patients who were on a different biologic medication in the preceding 3 months.
7. Patients with a diagnosis of EGPA/Churg-Strauss Sydnrome
8. Pregnant patients
9. Patients with inverted papilloma growth

Visit description for patients with nasal polyps with or without asthma:

1 - screening visit

2 - baseline visit, at which medication will be administered for the first time and patient will be trained to self-administer Dupixent

3 - follow-up visit in 2 weeks from baseline

4 - follow-up visit in 16 weeks from baseline

5 – long term follow-up visits in 36 weeks

6 – long term follow-up visits in 52 weeks

Additional Study Details

- The estimated per-participant study duration is 16 weeks, with secondary follow-up visits at 36 and 52 weeks
- Anticipated Study Start/Study Activation: July 2021
- Study Completion: 36 months from start date, July 2024
- Questionnaires and sample collections will be completed throughout the duration of the study (5 time points = week 0, 2, and 16) and in a long-term follow up for 36 and 52 weeks

Assessments to be performed

- SNOT-22, ACQ, UPSIT
- FEV_1, NPF, N-FENO, absolute eosinophil count, IgE levels, urine collection for $uLTE_4$ analysis, nasal tissue, fluid, and microbiome collection. The nasal inferior turbinate tissue, fluid, and microbiome collection will be done at baseline, at 16, 36, and 52 weeks. If sinus surgery is clinically indicated and the patient with nasal polyps undergoes an endoscopic sinus surgery, we will collect less than a quarter of the sinonasal tissues such as inferior turbinate, uncinate process, nasal polyp, and microbiome swabs during surgery after the remaining has been used for diagnostic purposes.

Randomization Method and Blinding: Not applicable

Study design for the control group:

The control group will include patients without polyps, but having sinonasal surgery for conditions in which a small amount of mucosa may be removed incidentally, such as [correction of a deviated septum, repair of the facial trauma with sinus fracture, CSF rhinorrhea, or a tumor of the nasal cavity or paranasal sinuses], a portion of which is not submitted for pathology diagnosis and could be used for research. Sinonasal tissues such as inferior turbinate, uncinate process, nasal polyp, and microbiome swabs will be collected during the surgery. We will collect the remnant tissue for research after the remaining has been used for diagnostic purposes. The control group will not have any subsequent visits associated with the study.

Materials collected:

The tissue samples and nasal secretions will be collected and stored for future research purposes. Nasal polyp tissue and turbinate scrapping collections will be done by the ENT surgeons. The rest will be done by the study team. The sinonasal tissues once collected will be analyzed in the Porcelli/Jerschow lab,

Nasal scraping:

In order to obtain nasal tissues using the scraping method, plastic disposable nasal curettes (Rhino-probe, VWR, Radnor, PA, USA) will be used which has been developed specifically to obtain cytological samples for research and diagnostic purposes. Nasal scraping can be used for any site in the sinonasal cavity. The protocol described below is considered to be reproducible, fast and in minimally traumatic[40]:

1. Once the patients clear their nose of any excess secretions, the tip of the probe is passed gently along the medial surface of the inferior turbinate. This will be performed by direct visualization.
2. The samples will be obtained by two or three scrapes and will then be spread on microscope slides fixed with 95% ethyl alcohol which will be stored for later use.
3. These samples may be sent for diagnosis if clinically indicated, but since they are not part of the usual diagnostic workup, may be used exclusively for research.

Nasal mucosal biopsy:

A nasal biopsy will be performed as described below.[41] Only nasal polyps will be biopsied. Since nasal polyps have no innervation, their biopsy is usually painless. Topical administration of cophenylcaine (5% lidocaine with 0.5% phenylephrine) will be done to achieve local decongestion and anesthesia by placing cotton wool pledgets in the nasal cavities for 10 minutes.

1. Using an up-cutting 45-degree Blakesley forceps, biopsies will be taken under direct visualization from a mucosal fold on the lateral nasal wall anterior to the inferior turbinate.
2. Gauze will then be placed for 10 minutes in the nasal cavity for hemostasis.

If practical, the biopsies may be divided and a small portion of one or more sent to pathology for diagnosis and correlation with research findings.

<u>Patient recruitment and retention plan</u>

All patients will be recruited by PI, Dr. Elina Jerschow in the Montefiore Allergy and Asthma clinic.

The patients will be provided with informed consent forms and will be assured that they will not lose any services, benefits or rights or access to care that they would normally have if you choose not to volunteer.

All study patients will be recruited from this center and while they complete all study visits described in this proposal, the PI maintains contact with them during their regular follow-up visits in clinics scheduled as part of their medical care. The PI will make it to her priority to personally meet with every prospective study participant to answer any questions and ensure that every patient is comfortable with the research plans.

Although patients could voluntarily withdraw from the study at any time, to minimize dropouts during the initial recruitment, the following retention plan has been developed: 1) during the initial visit's informed consent discussion, we will discuss study expectations will all participants; 2) we will explain in detail the nature of the standard-of-care and aspirin desensitization and treatments that are applied during this study, 3) between study visits, the PI will promptly respond to patient questions and concerns, 4) the PI and/or the study coordinator remind patients of upcoming study visits; and 5) in scheduling study visits, the PI will try to accommodate patient schedules.

The patient's information and research records will be kept confidential. The study data file will be de-identified and will contain the responses to the questionnaires, laboratory, spirometry, and FeNO results. The clinical database including consent forms, questionnaires, and study report forms will be stored in a locked cabinet in a locked office in a security-protected office space of Montefiore Medical Center. The study data will be stored on a password-protected computer. Every effort will be made to protect confidential information such as subject identity. The study information will be kept as long as they are useful for the research.

The only people who can see the research records are:

- Researchers and other individuals who work with the researchers
- Organizations and institutions involved in this research, including those that fund the research, if applicable
- Groups that review research such as central reviewers, Institutional Review Boards, the Office for Human Research Protections, the US Food and Drug Administration, data coordinating centers, and domestic and foreign agencies that regulate research.

Informed Consent

The informed consent will be obtained by Dr. Elina Jerschow, the principal investigator. The consent will be obtained at allergy and combined chronic rhinosinusitis clinics where patients are seen for their

condition. No minors will be included in the study as stated by the inclusion criteria. Once the patients consent to their participation in the study and are recruited, they will receive $50 for each visit. There are no additional costs associated with the study. The routine blood tests (CBC, Chemistry) that are usually collected during follow-up visit in patients with CRSwNP on or off dupilumab will be billed to their insurance.

Key Personnel: Dr. Elina Jerschow, Dr. Esha Sehanobish, Dr. Mohammad Asad

Statistical analysis

We will evaluate the SNOT-22 and UPSIT at 16 weeks, compute the changes from the baseline for each participant, and report the means of changes, and the standard errors. From what is reported by SINUS-24 and SINUS-52 at 24 week endpoints, we estimate that the standard deviation of changes is about 20 for SNOT-22 and about 9 for UPSIT, which correspond to standard errors at 2.6 and 1.2 respectively, given the proposed sample size of 60. The standard errors at such level are adequate compared to the mean changes from the baseline at 16 weeks as reported by SINUS-24 and SINUS-52, which is about -25 for SNOT-22 and 10 for UPSIT.
We will report the mean changes from the baseline at 24 and 52 weeks with standard errors.

Primary Objectives:
Non-inferiority tests will be performed to compare the results from this study to the results reported by the Phase 3 clinical trials, with the margin of non-inferiority is set at 75% of reported effect size. We will also compare the outcomes between Caucasian and non-Caucasian patients in this study. We don't expect them to be different but will report if significant differences are found.

Secondary Objectives: Paired T-tests will be used to assess the effect of dupilumab on receptor expression and cellular function before and after the in vitro stimulation. Paired T-tests will also be used to assess the changes of biomarkers in patients between the baseline and 16 weeks after taking dupilumab. Log transformation will be applied to the outcome variables when appropriate.

Power Estimation
Primary Objective 1(a):In the Phase 3 trials of nasal polyposis, 6 months of dupilumab induced a decline in SNOT-22 score by a LS mean of 30.5 (S.E. =1.54) from the baseline in the treatment group, compared to a drop in LS mean score of 9.31 in the placebo group.[5] We will compare the mean drop

in SNOT-22 score from the baseline of our 60 patients to the reported drop of 30 in the previous Phase 3 trials using a t-test. At significance level 0.05, given our sample size, and assume a standard deviation at 18.5 as reported by the above mentioned clinical trials, we have 95% power to detect non-inferiority with a margin of non-inferiority of 7.5, that is, 75% of reported effect size corresponding to a drop in SNOT-22 score by 22.5 from the baseline.

Primary Objective 1(b):In the Phase 3 trials of nasal polyposis, 6 months of dupilumab induced an increase in UPSIT score by a LS mean of 11 (S.E.=0.67) from the baseline in the treatment group.[5] At significance level 0.05, given our sample size, and assumed standard deviation at 8 as reported by the above mentioned clinical trials, we have 88% power to detect non-inferiority with a margin of non-inferiority of 2.75, that is 75% of reported effect size, corresponding to a mean increase of 8.25 from the baseline.

Secondary Objective: Given a sample size of 20 patients with CRSwNP and significance level 0.05, we have 80% power to detect a mean change of 0.5σ, where σ is the standard deviation of within patient variation.

Statistical analysis of the exploratory objectives will be similar to those of primary and secondary objectives.

Data Management

Upon expressing an interest to participate in the study, the patients will be provided with an informed consent explaining the details of the study. Each subject will be assigned a study identification number at the time of consent and recruitment. RedCap database will be used for capturing of the study data.

Data management and analysis will be carried out by Esha Sehanobish, PhD (Postdoctoral Research Fellow, Albert Einstein College of Medicine), Elina Jerschow, MD (Attending Allergy/Immunology, Montefiore Medical Center).

Data safety and monitoring

All subjects will be from within Montefiore Allergy/Immunology or Otolaryngology clinics. Subjects will be provided with the informed consent form when the investigators determined they may be an acceptable candidate for the study and that they met the eligibility criteria. The forms, aims of the study, data collection, and all potential risks and benefits of the study will be discussed with each

subject by qualified study site personnel. No subjects will be enrolled without documentation of informed consent, and no waivers of this process will be sought or granted. All data collected will be observational and does not alter standard-of-care treatment in these patients. Risk of data confidentiality: data capture system. All users of the REDCap database system will be provided access in a secure fashion and can be tracked with user specific audit trails. To provide the best protection to research participants, subjects will be assured of complete confidentiality or test results. As with all research data, information gathered by the study will be used only for aggregate analysis; it will not be released with any information that identifies research participants. The PI for the study will monitor the study. The study is not interventional. It is observational and follows standard of care. The medication is FDA approved for the indication for which it is used in this study.

Independent study monitor: Dr. Golda Hudes, MD, PhD, is an Associate Professor in the Departments of Medicine (Allergy and Immunology) and Otorhinolaryngology – Head and Neck Surgery at Albert Einstein College of Medicine/Montefiore Medical Center. Dr. Hudes has a breadth of experience in clinical trials in the field of asthma, nasal polyposis, and eczema. She will serve as an independent study monitor.

Responsibilities: The independent study monitor will review the research protocol and evaluate the progress of the clinical study every six months. Dr. Jerschow will provide a study progress summary to the independent study monitor every six months. The study monitor will assess data quality, timeliness, recruitment, and safety of the study participants. The independent study monitor will provide recommendations regarding continuation, modification, and/or termination. All adverse events will be compiled every six months and at the conclusion of the study.

Stopping rules for individual subjects

Any subject enrolled in the study may choose to voluntarily end their participation at any point, without affecting their medical care or treatment. For safety reasons, participants may discontinue their participation in the study if they develop any study-related serious adverse events (SAEs). At the conclusion of the study, subjects will continue on standard therapy based on clinical response and in consultation with their physician.

Study termination criteria

The study PI may terminate this study at any time. Reasons for termination may include, but are not limited to, the incidence or severity of AEs in this study indicating a potential health hazard. The PI will

promptly inform all other participating investigators if the study is suspended or terminated for safety reasons and will also inform the IRB and regulatory authorities of the suspension or termination of the study and the reason(s) for the action.

In addition, the study may be terminated for reasons including but not limited to:

1. Investigator request to withdraw from study participation.
2. Serious and/or persistent noncompliance by the investigator with the protocol, the clinical research agreement, or other local applicable regulatory guidelines in conducting the study.
3. An IRB/ISM decision to terminate or suspend approval for the investigation or the investigator.

Reporting adverse events (AE)

An AE is defined as any undesirable event, whether or not related to the investigational protocol, which occurs to a subject during the clinical study any time after the subject signs the informed consent form, until the last visit, or a premature discontinuation visit. The following precautions are will be in place during the recruitment:

Serious adverse events (SAE)

An adverse event is '**serious**' if it:

- Results in death
- Is life-threatening: when the individual is, in the opinion of the investigator, at immediate risk of death from the event as it occurs (this definition does not include an event that hypothetically might have caused death if it were more severe)
- Requires hospitalization or prolongation of existing hospitalization
- Results in persistent or significant disability or incapacity (i.e., substantial disruption of a person's ability to conduct normal life functions)
- Consists of a congenital abnormality or birth defect in the offspring of study participant's or participant's partner
- Is a medically important event, which may jeopardize the person and may require medical or surgical intervention to prevent one of the outcomes listed above.

Eliciting adverse event Information: The period of observation for AEs/SAEs extends from the time the subject signs the informed consent form until their exit from the study. At each study visit and telephone contact, subjects will be asked standard questions to elicit medically related changes in their well-being. They will also be asked if they have been hospitalized, had any accidents, used any new medications, or changed concomitant medication regimens (both prescription and over the-

counter). In addition to observations by subjects, AEs will be documented from data collected on the AE log (e.g., laboratory values, physical examination findings, medical history).

AE logs/SAE Case Report recording - AEs and SAEs are to be recorded from the time the subject signs the informed consent form until their last visit or a premature discontinuation visit. Signs and symptoms of each AE were to be described in detail, including onset time/date, offset time/date, severity, relationship to dupilumab treatment, and outcome. Action taken are defined as none, medication administration, hospitalization, other.

Potential Benefits and knowledge to be gained from the Study

The study will help us identify the effects of 16-week dupilumab treatment on nasal polyp symptoms in an ethnically diverse population. It will help us understand the effects of dupilumab on sense of smell in the same cohort with CRSwNP. It will also help us understand the effects of the 16-week dupilumab treatment on biomarkers such as peripheral blood eosinophil counts, serum IgE and also urinary levels of LTE_4. We also hope to determine the effects of dupilumab treatment on lung function and on the tissue and peripheral blood lymphocytes.

Potential risks

Risk of dupilumab treatment: Dupilumab is used as an add-on maintenance treatment in adult patients with inadequately controlled CRSwNP. However, the risks during the dupilumab treatment includes hypersensitive reactions such as urticaria, rash, erythema, anaphylaxis, nodosum (skin inflammation), and serum sickness. There may be an increase in eosinophilic conditions as evident from rashes, worsening pulmonary symptoms and/or neuropathy in cases of reduced oral corticosteroid use. There may be cases of new onset or worsening conjunctivitis and keratitis.

Risks of ACT and sinus and nasal symptom score (ACQ, SNOT) questionnaires: Each subject will complete these questionnaires at their study visits. Investigators do not interpret the questionnaires in any manner. There are no risks associated with administration of these questionnaires.

Risks of blood draws: Risks associated with drawing blood include pain and discomfort that usually lasts for several seconds when the needle is inserted. There is a small risk of bruising and/or infection at the place where the needle enters the arm. Some people may experience

lightheadedness, nausea or fainting. It is possible to develop an infection, but this is rare and can be treated.

Risks of spirometry: Occasionally individuals may develop a slight dizziness and/or temporary cough and/or chest discomfort when performing breathing tests. These tests are part of routine care for asthma patients and used in clinical settings where asthma care is provided.

Risks of tissue donation during endoscopic sinus surgery: The collection of nasal or sinus tissue for research purposes will not affect the surgery or diagnostic pathology, since the latter requires only 2 cassettes of tissue (4 grams), which almost always results in left over tissue adequate in quantity for research. If the surgeon performing the polypectomy feels that the removed tissue may contain a lesion requiring more pathology workup, such as cancer or inverted papilloma, it will all be submitted for pathology diagnosis and not collected for this study.

Risks of sinonasal epithelium collection with Rhino-Pro Currette: We chose the use of Rhino-Pro® Currettes as sinonasal epithelium collection tool for several reasons: (i) it is safe; (ii) it causes no to minimal discomfort to the patient; (iii) it has been used as a collection tool in previous studies.2 The sinonasal epithelium collections will be performed during routine post-surgical care The use of the curette may potentially lead to irritation of the nasal mucosa, bleeding, and, rarely, an infection that may require antibiotic treatment. Participants may have some soreness around the nose or sinuses for 1 or 2 days after the endoscopy. To minimize patient discomfort, we will use a local anesthetic (topical lidocaine, 4%), as it is a routine during rhinoscopy. A minimal bleeding may occur and will be treated with vasoconstrictor oxymethazoline.

Adequacy of Protection Against Risks

Recruitment and Informed Consent

Upon expressing interest to participate, the subject will participate in a full, unpressured explanation of the study. Informed consent will be obtained from all study subjects who meet eligibility criteria and participate in the study. The investigational nature of the study, its objectives, the procedures involved, and their potential risks and benefits will be explained to the participants using simple, direct language with full explanation of any medical terms involved (either in Spanish or English). The procedures will also be described in the consent form in the same direct language with complete description of the procedures entailed. Participants who will consent to the study will sign the informed consent form. A notification of study participation will be placed in the medical records. A copy of the

signed document will be given to the study participant and another copy will be kept in the locked cabinet in the PI's office to ensure confidentiality. Informed consent will be obtained before any study data are captured and before any specimens are collected.

Protection Against Risks

This study will be conducted in compliance with the protocol, Good Clinical Practice and all applicable regulatory requirements. All research personnel have received the required education and training needed for conducting clinical research related to the protection of human subjects and personal health information according to the Institutional Review Board and Health Insurance Portability and Accountability Act of 1996 Public Law 104-191. As per clinical routine, the study visits will take place within the confines of the Montefiore Medical Center, with all levels of medical aide and emergency care immediately available, for any serious adverse consequences.

Adverse events due to dupilumab treatment: Any adverse reactions during the dupilumab treatment such as hypersensitive reactions and conjunctivitis can be easily treated by the use of eye drops, and prednisone for eosinophilia with systemic symptoms. If deemed necessary for a patient, usage of dupilumab will be discontinued.

Risk to Data Confidentiality: To mitigate potential risk to data confidentiality, two separate data files will be maintained. The clinical data file will contain the link between the subject and the study identification number. The study data file will be de-identified and will contain the responses to the questionnaires, laboratory, and spirometry results. The clinical database including consent forms, questionnaires, and study report forms will be stored in a locked cabinet in a locked office in a security-protected office space of Montefiore Medical Center. The study data will be stored on a password-protected computer. Every effort will be made to protect confidential information such as subject identity.

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