Study Protocol Synopsis of the RESPIRE Study

Title of clinical trial	Evaluation of Efficacy and Safety of RespiraFloxInhale™ in Patients with RespiraFlox (RESPIRE Study)
Site Locations	20 centres that enrolled and randomised patients in country X, Y, and Z
Study period	First patient first visit was conduted on 01Jan2020 and last patient last visit on 31Dec2024
Objectives	The primary objective of this study is to assess the efficacy of the RespiraFloxInhale™ in improving respiratory symptoms in patients with RespiraFlox, as measured by changes in cough frequency and severity from baseline to Week 12. The secondary objectives of the study are: • To assess the impact of the RespiraFloxInhale™ on additional respiratory symptoms, quality of life measures, and pulmonary function tests • To explore the pharmacokinetics of the RespiraFloxInhale™ in patients with RespiraFlox
Study design	This is a phase 2, multi-center, international, randomized, double-blind, placebo-controlled study aimed at demonstrating the efficacy of RespiraFloxInhale™ in improving respiratory symptoms in patients with RespiraFlox. **Study Structure:** The study comprises three periods: 1. Screening Period**:** This initial phase will determine the eligibility of participants, including diagnostic testing to assess RespiraFlox levels in the blood. 2. Double-blind Treatment Period**:** Eligible participants will be randomly assigned to receive either RespiraFloxInhale™ or a placebo. The first dose will be administered on day 1, followed by twice-daily doses throughout the study duration. 3. Safety Follow-up Period**:** Following the treatment period, there will be a 40-week-off-drug follow-up period to monitor safety outcomes. Patients who fulfill all the eligibility criteria will be randomised and receive their first dose of assigned study dryg on day 1, with subsequent doeses adminsitred twice daily of the duration of the study. Following that will be a 40-week-off-drug follow-up period. **Assessment:** • Efficacy: The effectiveness of the treatment will be evaluated by measuring the number of patients with positive test results, along with laboratory findings, responses to self-reported questionnaires, and pulmonary function tests. • Safety: Safety assessments will be conducted throughout the study, comparing the frequency and severity of adverse events (AEs) between the

	RespiraFloxInhale™ and placebo groups. This will involve physical examinations, laboratory tests, and vital sign measurements.
Study duration	Participants will receive treatment with the RespiraFloxInhale™ or placebo for a maximum duration of 12 weeks.
Population	**Sample size:** Approximately 200 patients **Target population:** Inclusion Criteria: • Adults aged 18-65 diagnosed with RespiraFlox, experiencing moderate to severe respiratory symptoms • Medically stable • Able to understand and comply with study requirements/procedures based on the assessment of the investigator Exclusion Criteria: History of severe respiratory disease, significant comorbidities, use of certain medications, pregnancy, breastfeeding.
Treatment	**Study drug (Dose/route/Schedule)**: 100 mg of RespiraFloxInhale™ administered via inhalation twice daily. Inhalation (±3 days) **Placebo (Route/Schedule)**: Matching Placebo via inhalation twice daily (±3 days)
Endpoints	**Primary Endpoint**: The primary endpoint of the study is the change in cough frequency and severity from baseline to Week 12. Data on cough frequency and severity will be collected using electronic diaries (e-diaries), with participants recording their daily experiences of cough symptoms throughout the study period. Follow-up assessments will be conducted at Week 6 and Week 12 to evaluate changes in cough frequency and severity over time. **Secondary Endpoints**: • Safety Assessments: Adverse events and serious adverse events will be monitored and recorded throughout the study. • Additional Respiratory Symptoms: Data on additional respiratory symptoms will be collected through participant self-reporting and clinical assessments. • Quality of Life Measures: Quality of life measures will be assessed using validated questionnaires administered at baseline, Week 6, and Week 12. • Pulmonary Function Tests: Pulmonary function tests, including forced expiratory volume (FEV), will be conducted at baseline and Week 12 to evaluate changes in lung function over time.
Statistical Plan	Primary Analysis: • Objective: Evaluate the change in cough frequency and severity from baseline to Week 12.

	• Statistical Test: Paired t-test or Wilcoxon signed-rank test (depending on the distribution of the data) to compare mean or median cough frequency and severity scores between baseline and Week 12. • Handling Missing Data: Missing data for the primary endpoint will be handled using imputation techniques. **Secondary Analyses**: 1. Safety Assessments: o Descriptive statistics (frequencies, percentages) for adverse events and serious adverse events. o Chi-square test or Fisher's exact test to compare the incidence of adverse events between treatment groups. 2. Additional Respiratory Symptoms: o Descriptive statistics for individual respiratory symptoms. o Analysis of covariance (ANCOVA) or logistic regression to assess the effect of treatment on individual symptoms, adjusting for baseline severity. 3. Quality of Life Measures: o Analysis of variance (ANOVA) or linear regression to compare changes in quality of life scores between treatment groups. 4. Pulmonary Function Tests: o Analysis of covariance (ANCOVA) to compare changes in forced expiratory volume (FEV) between treatment groups at Week 12, adjusting for baseline FEV. o Subgroup analyses by disease severity (e.g., mild, moderate, severe) to explore treatment effects across different patient populations. **Handling Missing Data**: • Missing data for secondary endpoints will be handled using appropriate imputation methods, such as last observation carried forward (LOCF) or multiple imputation, depending on the nature and extent of missingness. **Subgroup Analyses**: • Subgroup analyses will be conducted to evaluate treatment effects in predefined subgroups based on demographic characteristics (e.g., age, gender), disease severity, and baseline cough frequency and severity.