NON-INTERVENTIONAL (NI) STUDY PROTOCOL

Study information

Title	Viral SARS-CoV-2 rebounds in commercial pharmacy-based SARS-CoV-2 PCR testing
Protocol number	C4671048
Protocol version identifier	1.0
Date	08 February 2023
EU Post Authorization Study (PAS) register number	EUPAS103517
Active substance	Nirmatrelvir/Ritonavir (ATC code J05AE30)
Medicinal product	Paxlovid
Research question and objectives	The following are the research objectives that will be addressed in this study: •What is the risk of viral SARS-CoV-2 rebound, prior to the Omicron era and during the Omicron era, among high frequency (volume) SARS-CoV-2 diagnostic testers, and when stratified by variables such as age, comorbidities, vaccination status, co-infections, Paxlovid treatment status for SARS-CoV-2 and timing around viral SARS-CoV-2 rebounds? The following are the research objectives that will be addressed in this study: ***Primary Objectives:*** Objective 1: To estimate viral SARS-CoV-2 rebound rates, prior to the Omicron era and during the Omicron era (defined as 01 December 2021 to present), in high frequency (volume) SARS-CoV-2 diagnostic testers.

	Objective 2: To estimate viral SARS-CoV-2 rebound rates, prior to the Omicron era and during the Omicron era, in high frequency (volume) SARS-CoV-2 diagnostic testers, among those treated with Paxlovid/nirmatrelvir-ritonavir for SARS-CoV-2 versus those with no evidence of SARS-CoV-2 treatment (untreated for SARS-CoV-2), stratified by: a. Demographics of participants b. Time from first positive test to treatment (if treated). c. Participants at high-risk for progression to severe SARS-CoV-2 versus those not at high-risk, if sample size allows. If sample size is limited, age (<50 years of age versus 50+ years of age) will be used as a proxy for high-risk for progression to severe SARS-CoV-2 status) d. Co-infections e. Vaccinated against SARS-CoV-2 (has received at least 1 dose) and time since last vaccine dose > 6 months vs. ≤6 months f. Multiple SARS-CoV-2 treatment prescription status ***Secondary Objectives:*** Objective 1: As a sensitivity analysis, using an alternate less strict definition of high frequency (volume) SARS-CoV-2 diagnostic testing, to estimate viral SARS-CoV-2 rebound rates, prior to the Omicron era and during the Omicron era, in high frequency (volume) SARS-CoV-2 diagnostic testers (using the primary definition), among those treated with Paxlovid/nirmatrelvir-ritonavir for SARS-CoV-2 versus those with no evidence of SARS-CoV-2 treatment

	(untreated) for SARS-CoV-2, with the same stratifications as detailed in Primary Objective 2. Objective 2: As a sensitivity analysis, using an alternate stricter definition of high frequency (volume) SARS-CoV-2 diagnostic testing, to estimate viral SARS-CoV-2 rebound rates, prior to the Omicron era and during the Omicron era, in high frequency (volume) SARS-CoV-2 diagnostic testers, among those treated with Paxlovid/nirmatrelvir-ritonavir for SARS-CoV-2 versus those with no evidence of SARS-CoV-2 treatment (untreated) for SARS-CoV-2, with the same stratifications as detailed in Primary Objective 2. Objective 3: As a sensitivity analysis, under a less strict definition of viral SARS-CoV-2 rebound, to estimate viral SARS-CoV-2 rebound rates, prior to the Omicron era and during the Omicron era, in high frequency (volume) SARS-CoV-2 diagnostic testers, among those treated with Paxlovid/nirmatrelvir-ritonavir for SARS-CoV-2 versus those with no evidence of SARS-CoV-2 treatment (untreated) for SARS-CoV-2, with the same stratifications as detailed in Primary Objective 2.
Author	[redacted]

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2. LIST OF ABBREVIATIONS

Abbreviation	Definition
AE	Adverse event
CI	Confidence interval
COPD	Chronic obstructive pulmonary disease
COVID-19	Coronavirus disease 2019
CKD	Chronic kidney disease
EUA	Emergency use authorization
EPIC-HR	Evaluation of Protease Inhibition for Covid-19 in High-Risk Patients
ER	Emergency room
EU	European Union
FDA	Food and Drug Administration
GPP	Good Pharmacoepidemiology Practices
HIPAA	Health Insurance Portability and Accountability Act
HIV	Human immunodeficiency virus
ICD	International class disease
IEC	Independent Ethics Committee
IPTW	Inverse probability of treatment weighting
IRB	Institutional Review Board
NA	Not Applicable
NIH	National Institute of Health
NI	Non-interventional
PAS	Post-Authorization Study
PASS	Post-Authorization Safety Study

Abbreviation	Definition
PCR	Polymerase chain reaction
RWD	Real world data
SAP	Statistical Analysis Plan
SARS-CoV-2	Severe acute respiratory syndrome coronavirus 2
SMD	Standardized mean differences
US	United States
WHO	World Health Organization

3. RESPONSIBLE PARTIES

Principal Investigator(s) of the Protocol

Name, degree(s)	Job Title	Affiliation	Address
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4. ABSTRACT

In ANNEX 1. as standalone document.

5. AMENDMENTS AND UPDATES

Amendment number	Date	Protocol section(s) changed	Summary of amendment(s)	Reason

6. MILESTONES

Milestone	Planned date
Start of data collection	22 February 2023
Registration in the European Union (EU) PAS register	21 February 2023
End of data collection	15 March 2023
Final study report	15 February 2024

7. RATIONALE AND BACKGROUND

Coronavirus disease 2019 (COVID-19), caused by severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2), was declared a global pandemic by the World Health Organization (WHO) on 11 March 2020[6], and continues to be a serious global threat to public health and to health care systems. As of 07 March 2022, COVID-19 has infected at least 446 million people, and has led to at least 6 million deaths worldwide.[7]

In order to prevent SARS-CoV-2 infection, several COVID-19 vaccines have been approved or authorized for emergency use by Health Authorities and are being administered globally.[8]

In December 2021, the Food and Drug Adminitsration (FDA) granted emergency use authorization (EUA) for a 5 day course of treatment for 2 separate oral antiviral agents, nirmatrelvir/ritonavir[9] and molnupiravir[10], for the treatment of COVID-19 in adult and pediatric patients 12 years of age and older who weigh at least 40 kg, with mild to moderate COVID-19 who are at high risk for progressing to severe illness. In January 2022, remdesivir received approval for this same population. This treatment is administered by IV infusion over 3 days for nonhospitalized patients with mild to moderate COVID-19 who are at high risk for progressing to severe illness.

Case reports in the literature describe individuals who have experienced symptomatic relapses of SARS-CoV-2 infection following completion of a 5-day course of nirmatrelvir/ritonavir. In the available cases, patients are described as experiencing rapid improvement in systemic symptoms following initiation of treatment but then experience a rebound in COVID-19 symptoms after completing therapy. The time course of symptom rebound suggests that symptom recurrence is likely not related to re-exposure and that it does not reflect a potential re-infection event. In one case report, symptom relapse was accompanied by fluctuating reverse transcription polymerase chain reaction (RT-PCR) cycle thresholds and antigen testing suggesting the potential for a rebound in viral replication. In a separate case series of 7 patients with recurrent symptoms, median SARS-CoV-2 RNA at baseline was 6.1 $\log_{10}$ copies/mL (range 4.2-7.3) and enrollment viral cultures were positive in 3 of 7 individuals. Several of the published case reports on symptom recurrence included viral sequencing and these reports to date have not identified resistance mutations to nirmatrelvir.

A retrospective review of 483 high-risk patients treated with nirmatrelvir/ritonavir reported 4 (0.8%) of patients experienced rebound of symptoms. Median time to rebound was reported as 9 days.

Rebound symptoms were characterized as mild and resolving without additional COVID-19 therapy with no patient requiring hospitalization.[5]

Post-treatment increases in SARS-CoV-2 RNA levels (i.e., viral RNA rebound) in nasopharyngeal samples were observed on Day 10 or Day 14 in a subset of nirmatrelvir/ritonavir and placebo recipients in the EPIC-HR study. The frequency of detection of post-treatment viral RNA rebound varied according to analysis parameters but was generally similar among nirmatrelvir/ritonavir and placebo recipients, regardless of the rebound definition used. For participants who showed a half-$\log_{10}$ or greater increase relative to end of treatment and whose viral load was persistent through follow-up, the occurrence was 1.73% placebo and 2.32% nirmatrelvir/ritonavir. In addition to these, some participants had transient half-$\log_{10}$ or greater increases relative to end of treatment, the occurrence was 2.35% in placebo vs 4.65% nirmatrelvir/ritonavir. A similar or smaller percentage of placebo recipients compared to nirmatrelvir/ritonavir recipients had nasopharyngeal viral RNA results <LLOQ at all study timepoints in both the treatment and post-treatment periods. Post-treatment viral RNA rebound was not associated with the primary clinical outcome of COVID-19-related hospitalization or death from any cause through Day 28 following the single 5-day course of nirmatrelvir/ritonavir treatment. Post-treatment viral RNA rebound also was not associated with drug resistance as measured by mPro sequencing. The clinical relevance of post-treatment increases in viral RNA following nirmatrelvir/ritonavir or placebo treatment is unknown.

In response to the COVID-19 pandemic, Helix OpCo (Helix), an accredited laboratory (CAP #9382893, CLIA #05D2117342) based in San Diego, CA, with a suite of laboratory capabilities to support infectious disease programs and public health surveillance (including SARS-CoV-2 / Flu diagnostic testing, SARS-CoV-2 whole genome sequencing, and pan-respiratory virus whole genome sequencing), developed laboratory capabilities to support SARS-CoV-2 diagnostic testing. With funding from the NIH's Rapid Acceleration of Diagnostics (RADx) program, Helix became a national "mega-lab" for processing up to 150,000 SARS-CoV-2 real-time reverse transcription polymerase chain reaction (rRT-PCR) diagnostic tests per day. Helix also has partnered with the CDC to provide SARS-CoV-2 whole genome sequencing starting in December 2020 and continues to support public health sequencing efforts. Helix has processed over ten million SARS-CoV-2 RT-PCR diagnostic tests and hundreds of thousands of SARS-CoV-2 whole genome sequences. Helix has established registries, biorepositories, and Institutional Review Board (IRB) protocols for both human genomics and viral genomics programs, enabling performance of research initiatives and key analyses to inform public health decision-making.

In this study, we will leverage Helix's existing US-based Respiratory Registry (External Protocol #0003-0001) of in vitro diagnostic rRT-PCR testing for the qualitative detection of nucleic acid in anterior nasal swab specimens for the detection of the SARS-CoV-2 and Influenzas A/B with linkage to retrospective Komodo Healthcare claims data in order to further examine viral SARS-CoV-2 RNA rebound (SARS-CoV-2 rebound) patterns. Specifically, viral SARS-CoV-2 rebound will be examined after stratification of variables such as SARS-CoV-2 therapeutic treatment, viral load patterns, co-infections, and by timing around rebounds in relation to initial test positivity to treatment for SARS-CoV-2.

This non-interventional study (NIS) is designated as a Post-Authorization Safety Study (PASS) and is conducted voluntarily by Pfizer.

8. RESEARCH QUESTION AND OBJECTIVES

The following are the research objectives that will be addressed in this study:

Primary research questions:

- What is the risk of viral SARS-CoV-2 rebound, prior to the Omicron era and during the Omicron era, among high frequency (volume) SARS-CoV-2 diagnostic testers?
- What is the risk of viral SARS-CoV-2 rebound when stratified by variables such as age, comorbidities, vaccination status, co-infections, Paxlovid treatment status for SARS-CoV-2 and timing around viral SARS-CoV-2 rebounds, among high frequency (volume) SARS-CoV-2 diagnostic testers prior to the Omicron era and during the Omicron era?

Secondary research question:

- Under alternate definitions of viral SARS-CoV-2 rebound and high frequency (volume) testing, what is the risk of viral SARS-CoV-2 rebound, prior to the Omicron era and during the Omicron era, among high frequency (volume) SARS-CoV-2 diagnostic testers, including when stratified by variables such as age, comorbidities, vaccination status, co-infections, Paxlovid treatment status for SARS-CoV-2 and timing around viral SARS-CoV-2 rebounds?

Exploratory research questions:

- What are risk factors and clinical outcomes among high frequency (volume) SARS-CoV-2 diagnostic testers in relation to viral SARS-CoV-2 rebound?

The following are the research objectives that will be addressed in this study:

Primary Objectives:

Objective 1: To estimate viral SARS-CoV-2 rebound rates (primary definition: 1. a positive test (at the index date), followed by a single negative test, followed by a positive test - within 28 days, or 2. 1 or more positive tests at the index date, followed by 1 or more negative tests, followed by a positive test within same 28 day time span), prior to the Omicron era and during the Omicron era (defined as 01 December 2021 to present), in high frequency (volume) SARS-CoV-2 diagnostic testers (primary definition: 1 or more positive tests followed by 1 or more negative tests followed by 1 or more positives within same 28 day time span and median time between tests (positive or negative) is at most 10 days).

Objective 2: To estimate viral SARS-CoV-2 rebound rates (using the primary definition of viral SARS-CoV-2 rebound), prior to the Omicron era and during the Omicron era, in high frequency (volume) SARS-CoV-2 diagnostic testers (using the primary definition of high frequency testing), among those treated with Paxlovid/nirmatrelvir-ritonavir for SARS-CoV-2 versus those with no evidence of SARS-CoV-2 treatment (untreated for SARS-CoV-2), stratified by:

a. Demographics of participants

b. Time from first positive test to treatment (if treated).

c. Participants at high-risk for progression to severe SARS-CoV-2 versus those not at high-risk, if sample size allows. If sample size is limited, age (<50 years of age versus 50+ years of age) will be used as a proxy for high-risk for progression to severe SARS-CoV-2 status)

d. Co-infections

e. Vaccinated against SARS-CoV-2 (has received at least 1 dose) and time since last vaccine dose > 6 months vs. ≤6 months

f. Multiple SARS-CoV-2 treatment prescription status

Secondary Objectives:

Objective 1: As a sensitivity analysis, using an alternate less strict definition of high frequency (volume) SARS-CoV-2 diagnostic tester (defined as tested for SARS-CoV-2 at least 3 times and median time between tests (positive or negative) is at most 14 days), to estimate viral SARS-CoV-2 rebound rates (using the primary definition), prior to the Omicron era and during the Omicron era, in high frequency (volume) SARS-CoV-2 diagnostic testers, among those treated with Paxlovid/nirmatrelvir-ritonavir for SARS-CoV-2 versus those with no evidence of SARS-CoV-2 treatment (untreated) for SARS-CoV-2, stratified by:

a. Demographics of participants

b. Time from first positive test to treatment (if treated).

c. Participants at high-risk for progression to severe SARS-CoV-2 versus those not at high-risk, if sample size allows. If sample size is limited, age (<50 years of age versus 50+ years of age) will be used as a proxy for high-risk for progression to severe SARS-CoV-2 status)

d. Co-infections

e. Vaccinated against SARS-CoV-2 (has received at least 1 dose) and time since last vaccine dose >6 months vs. ≤6 months

f. Multiple SARS-CoV-2 treatment prescription status

Objective 2: As a sensitivity analysis, using an alternate stricter definition of high frequency (volume) SARS-CoV-2 diagnostic tester (defined as tested for SARS-CoV-2 at least 3 times and median time between tests (positive or negative) is at most 7 days), to estimate viral SARS-CoV-2 rebound rates (using the primary definition), prior to the Omicron era and during the Omicron era, in high frequency (volume) SARS-CoV-2 diagnostic testers, among those treated with Paxlovid/nirmatrelvir-ritonavir for SARS-CoV-2 versus those with no evidence of SARS-CoV-2 treatment (untreated) for SARS-CoV-2, stratified by:

a. Demographics of participants

b. Time from first positive test to treatment (if treated).

c. Participants at high-risk for progression to severe SARS-CoV-2 versus those not at high-risk, if sample size allows. If sample size is limited, age (<50 years of age versus 50+ years of age) will be used as a proxy for high-risk for progression to severe SARS-CoV-2 status)

d. Co-infections

e. Vaccinated against SARS-CoV-2 (has received at least 1 dose) and time since last vaccine dose >6 months vs. ≤6 months

f. Multiple SARS-CoV-2 treatment prescription status

Objective 3: As a sensitivity analysis, under a less strict definition of viral SARS-CoV-2 rebound (defined as two positives more than 14 days apart), to estimate viral SARS-CoV-2 rebound rates, prior to the Omicron era and during the Omicron era, in high frequency (volume) SARS-CoV-2 diagnostic testers (using the primary definition), among those treated with Paxlovid/nirmatrelvir-ritonavir for SARS-CoV-2 versus those with no evidence of SARS-CoV-2 treatment (untreated) for SARS-CoV-2, stratified by:

a. Demographics of participants

b. Time from first positive test to treatment (if treated).

c. Participants at high-risk for progression to severe SARS-CoV-2 versus those not at high-risk, if sample size allows. If sample size is limited, age (<50 years of age versus 50+ years of age) will be used as a proxy for high-risk for progression to severe SARS-CoV-2 status)

d. Co-infections

e. Vaccinated against SARS-CoV-2 (has received at least 1 dose) and time since last vaccine dose >6 months vs. ≤6 months

f. Multiple SARS-CoV-2 treatment prescription status

Exploratory Objectives:

Objective 1: To estimate viral SARS-CoV-2 rebound rates (using the primary definition of viral SARS-CoV-2 rebound), prior to the Omicron era and during the Omicron era, in high frequency (volume) SARS-CoV-2 diagnostic testers (using the primary definition of high frequency testing), among those treated with Paxlovid/nirmatrelvir-ritonavir for SARS-CoV-2 versus those with no evidence of SARS-CoV-2 treatment (untreated) for SARS-CoV-2, stratified by:

a. Demographics of participants

b. Time from first positive test to treatment (if treated).

c. Participants at high-risk for progression to severe SARS-CoV-2 versus those not at high-risk, if sample size allows. If sample size is limited, age (<50 years of age versus 50+ years of age) will be used as a proxy for high-risk for progression to severe SARS-CoV-2 status)

d. Co-infections

e. Vaccinated against SARS-CoV-2 (has received at least 1 dose) and time since last vaccine dose > 6 months vs. ≤ 6 months

f. Multiple SARS-CoV-2 treatment prescription status

Objective 2: To examine risk factors and clinical outcomes of viral SARS-CoV-2 rebounders (using the primary definition of viral SARS-CoV-2 rebound), prior to the Omicron era and during the Omicron era in high frequency (volume) SARS-CoV-2 diagnostic testers (using the primary definition of high frequency testing), among those treated with Paxlovid/nirmatrelvir-ritonavir for SARS-CoV-2, those untreated for SARS-CoV-2 versus those with no evidence of SARS-CoV-2 treatment (untreated) for SARS-CoV-2, as defined below:

a. Risk factors
    i. High-risk factors
    ii. Demographics
    iii. Other risk factors to be determined
b. Clinical outcomes, based on healthcare utilization during follow-up post-index date
    i. Hospitalization (including length of stay)
    ii. Hospitalizations with invasive mechanical ventilation
    iii. Emergency room visits
    iv. Urgent care visits
    v. All-cause mortality

9. RESEARCH METHODS

9.1. Study design

This is a non-interventional, retrospective cohort study using secondary data sources containing structured data from the US. This study will include adults (≥18 years of age) and pediatric participants (<18 years of age) with a positive SARS-CoV-2 test (e.g., positive polymerase chain reaction (PCR) of direct SARS-CoV-2 viral testing).

9.2. Setting

This study will be conducted using SARS-CoV-2 diagnostic testing data from the Helix Respiratory Registry with linkage to the Komodo Healthcare claims data during the period of 01 June 2020, to 28 February 2023. Details about the database are found in Section 9.7. Further details will be outlined in the Statistical Analysis Plan (SAP).

9.2.1. Study population and cohorts

The study population will be generated from the two US databases described in Section 9.2.

The main cohort will include all participants meeting inclusion and exclusion criteria outlined in Section 9.3.

The high-risk subcohort (if sample size is limited, age (<50 years of age versus 50+ years of age) will be used as a proxy for high-risk for progression to severe SARS-CoV-2 status) will include all participants in the main cohort with high-risk clinical characteristics (comorbidities) for progression to severe SARS-CoV-2, as outlined in Section 9.4.

9.2.2. Time periods

9.2.2.1. Study period

The study will include an overall study period of June 01, 2020, to February 28, 2023.

9.2.2.2. Baseline period

The 365 days before index date will be the baseline period.

9.2.2.3. Index date

Index date is defined as first confirmed positive SARS-CoV-2 rRT-PCR test within a positive - negative – positive test sequence that occurs within 28 days during the study period (by omicron era status). Only the first infection for each patient during the study period (by omicron era status) will be examined.

9.2.2.4. Follow-up

The follow-up period starts at index date and participants will be followed until the date of the first occurring of the following events:

- Death
- At the end of their database disenrollment from the database
- At the end of study period
- Occurrence of the outcome/endpoint of interest

Further details will be defined in SAP.

9.3. Inclusion and Exclusion Criteria

9.3.1. Inclusion Criteria

Participants in the main cohort must meet all of the following inclusion criteria to be eligible for inclusion in the study:

1. Age ≥ 12 years on the index date (i.e., first confirmed positive SARS-CoV-2 rRT-PCR test within a positive - negative – positive sequence that occurs within 28 days)
2. High frequency (volume) tester, as defined as tested for SARS-CoV-2 at least 3 times (including the test on the index date) within 28 days of the index date and median time between tests (positive or negative) is at most 10 days. Alternative definitions using maximum and minimum boundaries for time between tests may be explored.
3. Sufficient enrollment and clinical encounter data in Komodo Healthcare claims, through 6-months of continuous medical and/or pharmacy claims enrollment (≤ 30 day gaps allowed) will be required, to allow at least 1 year of a baseline period prior to the index date to assess medical history.

In analyses involving dichotomous stratification by high-risk status for progression to severe SARS-CoV-2, inclusion/exclusion criteria similar to EPIC-HR (Evaluation of Protease Inhibition for Covid-19 in High-Risk Patients) trial criteria (e.g., diabetes, smoking status, obesity, chronic lung disease (including asthma), chronic kidney disease, chronic liver disease, immunosuppressive disease or immunosuppressive treatment, cardiovascular diseases, hypertension, sickle cell disease, neurodevelopmental disorders, active cancer, medically-related technological dependence, or were 60 years of age and older regardless of comorbidities), will be implemented if sample size allows. These variables are detailed in Section 9.4. If sample size is limited, age (<50 years of age versus 50+ years of age) will be used as a proxy for high-risk for progression to severe SARS-CoV-2 status).

Inclusion criteria will vary in sensitivity analyses based on alternate definitions of viral SARS-CoV-2 rebound and high frequency (volume) testing. Further details will be defined in SAP.

9.3.2. Exclusion Criteria

Participants in all cohorts are excluded from the study if the following criterion applies:

1. None / NA

9.4. Variables

All study variables of interest, including demographics, diagnostic (testing and disease), therapeutic and outcomes/endpoints are listed below. Index date is defined as first confirmed positive SARS-CoV-2 rRT-PCR test within a positive - negative – positive test sequence that occurs within 28 days. Baseline period will vary by patient, based on how much history is available and index date.

Endpoints

The following are the primary endpoint:

- Viral SARS-CoV-2 rebound*, where the main definition defines rebound as 1.) a positive test (at the index date), followed by a single negative test, followed by a positive test - within 28 days, or 2.) 1 or more positive tests starting at the index date, followed by 1 or more negative tests, followed by a positive test within same 28-day time span. Index date will be defined as first positive test date.

The following are the exploratory endpoints:

- Clinical outcomes (within 30 days of index date), based on healthcare utilization following viral SARS-CoV-2 rebound
    - Hospitalization (including length of stay)
    - Hospitalizations with invasive mechanical ventilation
    - Emergency room visits
    - Urgent care visits
    - All-cause mortality

*Alternate definitions of viral SARS-CoV-2 rebound will be included in sensitivity analyses and detailed in the SAP.

Covariates

Covariates will include baseline demographic, high-risk clinical characteristics (comorbidities) for progression to severe SARS-CoV-2, medications, and health utilization, as well as those needed for the study cohort inclusion and exclusion criteria. These are not intended to be the exclusive lists. Adjustments of the covariates may be made, and additional variables may be added in SAP as needed over time. The code lists of conditions and medications (if applicable) will be provided in SAP.

Demographics (prior to index date):

- Age/Age group (age groups to be defined in the SAP)
- Gender
- Race/ethnicity (if available)
- Location/region (if available)

High-risk clinical characteristics (comorbidities) for progression to severe SARS-CoV-2 (prior to index date):

- Immunosuppressive disease or immunosuppressive treatment.
- Cancer (active) other than localized skin cancer
- Cardiovascular conditions (Cerebrovascular disease or stroke, coronary heart disease, cardiomyopathies, heart failure, etc.)
- Chronic kidney disease (CKD)
- Deyo-Charlson Comorbidity Index Score
- Diabetes mellitus
- HIV
- Hypertension

- Chronic liver disease (cirrhosis; non-alcoholic fatty liver disease; alcoholic liver disease; autoimmune hepatitis, etc.)
- Neurodevelopmental disorders
- Obesity
- Respiratory conditions (Asthma, Chronic obstructive pulmonary disease (COPD), Interstitial lung disease (ILD), Chronic lung disease, etc.)
- Sickle cell disease
- Smoking status (current or former)

Prior healthcare characteristics (prior to index date):

- Prior-year healthcare utilization
- Prior SARS-CoV-2 infection/s
- Prior SARS-CoV-2 vaccination/s (never vaccinated, ≥1 dose with most recent dose given within 6 months of index date, ≥1 dose with most recent dose >6 months from index date)
- Prior influenza vaccinations

Risk factors to be examined (prior to index date):

- All demographics, high-risk clinical characteristics (comorbidities) for progression to severe SARS-CoV-2 and prior healthcare characteristics mentioned above

Diagnostic Testing Characteristics (on or after index date):

- High frequency (volume) SARS-CoV-2 diagnostic testing status**

Medications for treatment of COVID-19 (on or after index date):

- Anti-SARS-CoV-2 Monoclonal Antibodies
  - Bamlanivimab/Etesevimab
  - Casirivimab/Imdevimab
  - Sotrovimab
  - Tixagevimab/Cilgavimab
  - Bebtelovimab
- Antiviral therapy
  - Paxlovid/nirmatrelvir-ritonavir
  - Lagevrio/molnupiravir
  - Veklury/remdesivir [IV]

**Alternate definitions of high frequency (volume) SARS-CoV-2 diagnostic testing will be included in sensitivity analyses and detailed in the SAP.

9.5. Data sources

This study will be conducted using SARS-CoV-2 diagnostic testing data from the Helix Respiratory Registry with linkage to the retrospective Komodo Healthcare claims data during the period of

01 June 2020, to 28 February 2023, after appropriate de-identification certification, data privacy, and security compliance in accordance with Pfizer and Helix policies and industry best practices.

9.6. Study size

This is a descriptive study with no minimum sample size. All eligible participants during the relevant study periods will be included. This is a NI study with no a priori hypotheses specified; therefore, sample size calculations are not applicable.

Preliminary feasibility counts provided to Pfizer, Inc. by Helix indicate that 1,244,791 SARS-CoV-2 diagnostic tests performed from 10 October 2020, to 10 August 2022, among individuals who have tested with a Helix SARS-CoV-2 diagnostic test through a retail pharmacy in the US, 558,340 of these tests were positive. These positive tests represent 449,759 unique infections, defined as positive tests more than 28 days apart. Among high frequency (volume) SARS-CoV-2 diagnostic testers (under the primary definition), there were 21,991 unique infections during the same time period.

9.7. Data management

All data exists as structural data originating from two sources; part one from Helix's Respiratory Registry database (AWS Redshift), and the second part from Komodo's insurance claim database (Snowflake). Both datasets are routinely deidentified via Datavant's tokenization solution. Helix's dataset is version controlled on both the extraction date and the extract transform load (ETL) code side. Extracted flat files used for encrypted transit into Komodo's environment Sentinel are furthermore integrity tested by checksums (SHA256 and AWS ETag) and scanned for sensitive data before ingested into a study-specific Snowflake SQL (v6.41.2) environment. A concatenated ID string based on Datavant's tokens 1 and 2 is used for joining the two datasets throughout the study. Further data will be added by either vendor's data analytics team as needed or in case of relevance. As the database will be regularly updated, date and version of the database will be specified in the report of the study.

Originating data from Helix are under Helix's data management policies and procedures, developed by Helix's information and security teams. Data at Helix is stored following industry best practices. Following implemented comprehensive physical, organizational, technical, and administrative controls to ensure that data collection, storage, and use of directly identifiable information adheres to stringent security requirements and complies with applicable law, in accordance with Health Insurance Portability and Accountability Act (HIPAA) and National Institute of Standards and Technology (NIST) standards.

The study will be conducted by the analytic team at Helix. Personalized credentials will be used by each member of the Helix team for security and audibility when accessing the Sentinel environment. Retention of study-related data, documents, and other materials will be governed by Pfizer Policy on Records and Information Management, and per this policy, will remain effective for a period of five years from the date of project initiation. Amendments must be made only with the prior approval of Pfizer.

9.8. Data analysis

Descriptive statistics among individuals with SARS-CoV-2 in the Helix Respiratory Registry that overlap with the retrospective Komodo Healthcare claims data will be summarized. Means with standard deviations, medians with interquartile ranges will be provided for continuous variables. Numbers and percentages will be provided for dichotomous variables or categorical variables.

For exploratory analyses, logistic regression or Cox proportional hazards regression models will be used to identify risk factors and outcomes for viral SARS-CoV-2 rebound in adjusted analyses. Model selection will balance biological and contextual knowledge for potential confounders with statistical considerations for model fit and the bias versus variance tradeoff.

An analysis comparing viral SARS-CoV-2 rebound, between participants treated with Paxlovid, untreated participants (and pending sufficient sample size, comparator SARS-CoV-2 treatment group(s) described above) will be conducted. Viral SARS-CoV-2 rebound and other outcomes in participants who received Paxlovid compared with untreated participants (and/or comparator SARS-CoV-2 treatment group(s), pending sufficient sample size) will be evaluated using multivariable Cox regression models with propensity score inverse probability of treatment weighting (IPTW) with a robust variance estimator having age, sex, week and year of initial positive SARS-CoV-2 test, and potentially the aforementioned baseline clinical characteristics and comorbidities as independent variables to account for time-varying confounding. Standardized mean differences (SMDs) will be calculated to check whether IPTW yielded comparable cohorts, considering an SMD below 0.1 as indicating satisfactory covariate balance.

Detailed methodology for summary and statistical analyses of data collected in this study will be documented in a statistical analysis plan (SAP), which will be dated, filed, and maintained by the sponsor. The SAP may modify the plans outlined in the protocol; any major modifications of primary endpoint definitions or their analyses would be reflected in a protocol amendment.

9.9. Quality control

Analyses are programmed according to the specifications in the protocol, and the SAP, and documented in a programming plan as appropriate. Data imported into Komodo's environment Sentinel from Helix is processed by an ETL codebase developed following an industry best practices approach. Including, test-driven development with unit/integration tests and a software development lifecycle, which includes source control and reviewed pull requests. Data integrity controls are performed by checksums (SHA256 and AWS ETag) during and after an encrypted transfer between Helix and Komodo's platforms. Final deliverables will be reviewed and verified by a second, independent programmer who may also perform double programming. All quality checks will be documented in the programming plan.

9.10. Limitations of the research methods

COVID-19 is a new disease with evolving diagnostic testing, disease mechanisms and treatments. Many infected persons are thought to have mild to unrecognizable symptoms and may therefore not present for testing. Thus, participants with these infections will not be identifiable in the datasets, especially if they did not undergo repeated diagnostic testing for SARS-CoV-2. However, as high-

volume repeat testers are more likely to undergo a positive-negative-positive test pattern, which will result in an overestimate of the true proportion of participants who experience viral SARS-CoV-2 rebound (along with SARS-CoV-2 treatment rebound (including Paxlovid)) as the worst-case scenario, yet these results should form a conservative upper-limit estimate.

Participants with chronic diseases or in inpatient settings, the elderly, or other risk groups may be more likely to be under ongoing care or thought to be at higher risk and therefore may be more likely to seek care or be referred for testing. Differences among at-risk subpopulations may arise from access to care/health care seeking behaviors rather than true differences in risk. Generalizability may be limited as SARS-CoV-2 testing patterns and treatment are evolving over time, and health care seeking behaviors among participants at high-risk of SARS-CoV-2 progression may differ from those who are not at high-risk .

Additional limitations are similar to those from observational studies, including the inability to obtain detailed clinical information on disease severity and activity due to the nature of secondary databases. However, the Komodo Healthcare claims data provides a more accurate record of all prescriptions given compared to electronic health records (EHRs), which it is able to indicate whether or not a prescription is filled, including refills. Being able to access this additional component provides the opportunity to calculate additional measures (e.g., if refills were filled on time). Finally, there is a lack of complete similarity between the criteria used to construct the high-risk subcohort in this study and the high-risk criteria implemented in the EPIC-HR clinical trial.

9.11. Other aspects

Not applicable.

10. PROTECTION OF HUMAN SUBJECTS

10.1. Patient information

This study involves data that exist in anonymized structured format and contain no patient personal information.

10.2. Patient consent

As this study involves anonymized structured data, which according to applicable legal requirements do not contain data subject to privacy laws, obtaining informed consent from patients by Pfizer is not required.

10.3. Institutional review board (IRB)/Independent ethics committee (IEC)

Independent Ethics Committee (IEC)/ Institutional Review Board (IRB) review is not required.

10.4. Ethical conduct of the study

The study will be conducted in accordance with legal and regulatory requirements, as well as with scientific purpose, value and rigor and follow generally accepted research practices described in Guidelines for Good Pharmacoepidemiology Practices (GPP) issued by the International Society for Pharmacoepidemiology (ISPE).

11. MANAGEMENT AND REPORTING OF ADVERSE EVENTS/ADVERSE REACTIONS

This study involves the use of diagnostic testing data and claims data that exist as structured data by the time of study start or a combination of existing structured data and unstructured data, which will be converted to structured form during the implementation of the protocol solely by a computer using automated/algorithmic methods, such as natural language processing.

In these data sources, individual patient data are not retrieved or validated, and it is not possible to link (i.e., identify a potential association between) a particular product and medical event for any individual. Thus, the minimum criteria for reporting an adverse event (AE) (i.e., identifiable patient, identifiable reporter, a suspect product, and event) cannot be met.

12. PLANS FOR DISSEMINATING AND COMMUNICATING STUDY RESULTS

A final study report will be generated at the end of the study. Publications, and potentially pre-print manuscripts, may be written throughout the course of the study.

In the event of any prohibition or restriction imposed (e.g., clinical hold) by an applicable competent authority in any area of the world, or if the party responsible for collecting data from the participant is aware of any new information which might influence the evaluation of the benefits and risks of a Pfizer product, Pfizer should be informed immediately.

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14. LIST OF FIGURES

No table of figures entries found.

15. ANNEX 1. LIST OF STAND ALONE DOCUMENTS

Number	Document reference number	Date	Title
1	NA	08-Feb-2022	Abstract: Viral SARS-CoV-2 rebounds in commercial pharmacy-based SARS-CoV-2 PCR testing

16. ANNEX 2. ENCEPP CHECKLIST FOR STUDY PROTOCOLS

Not applicable.

15. ANNEX 1. LIST OF STAND ALONE DOCUMENTS

NON-INTERVENTIONAL STUDY PROTOCOL ABSTRACT

Title: Viral SARS-CoV-2 rebounds in commercial pharmacy-based SARS-CoV-2 PCR testing

Date: 08 February 2023,
Protocol 08 February 2023, version 1

Name and affiliation of the main author:

[redacted]

Rationale and background:

Coronavirus disease 2019 (COVID-19), caused by severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2), was declared a global pandemic by the WHO on 11 March 2020[6], and continues to be a serious global threat to public health and to health care systems. As of 07 March 2022, COVID-19 has infected at least 446 million people, and has led to at least 6 million deaths worldwide.[7]

In order to prevent SARS-CoV-2 infection, several COVID-19 vaccines have been approved or authorized for emergency use by Health Authorities and are being administered globally.[8]

In December 2021, the FDA granted emergency use authorization (EUA) for a 5 day course of treatment for 2 separate oral antiviral agents, nirmatrelvir/ritonavir[9] and molnupiravir[10], for the treatment of COVID-19 in adult and pediatric patients 12 years of age and older who weigh at least 40 kg, with mild to moderate COVID-19 who are at high risk for progressing to severe illness. In January 2022, remdesivir received approval for this same population. This treatment is administered by IV infusion over 3 days for nonhospitalized patients with mild to moderate COVID-19 who are at high risk for progressing to severe illness.

Case reports in the literature describe individuals who have experienced symptomatic relapses of SARS-CoV-2 infection following completion of a 5-day course of nirmatrelvir/ritonavir. In the available cases, patients are described as experiencing rapid improvement in systemic symptoms following initiation of treatment but then experience a rebound in COVID-19 symptoms after completing therapy. The time course of symptom rebound suggests that symptom recurrence is likely not related to re-exposure and that it does not reflect a potential re-infection event. In one case report, symptom relapse was accompanied by fluctuating RT-PCR cycle thresholds and antigen testing suggesting the potential for a rebound in viral replication. In a separate case series

of 7 patients with recurrent symptoms, median SARS-CoV-2 RNA at baseline was 6.1 log_{10} copies/mL (range 4.2-7.3) and enrollment viral cultures were positive in 3 of 7 individuals. Several of the published case reports on symptom recurrence included viral sequencing and these reports to date have not identified resistance mutations to nirmatrelvir.

A retrospective review of 483 high-risk patients treated with nirmatrelvir/ritonavir reported 4 (0.8%) of patients experienced rebound of symptoms. Median time to rebound was reported as 9 days. Rebound symptoms were characterized as mild and resolving without additional COVID-19 therapy with no patient requiring hospitalization.[5]

Post-treatment increases in SARS-CoV-2 RNA levels (i.e., viral RNA rebound) in nasopharyngeal samples were observed on Day 10 or Day 14 in a subset of nirmatrelvir/ritonavir and placebo recipients in the EPIC-HR study. The frequency of detection of post-treatment viral RNA rebound varied according to analysis parameters but was generally similar among nirmatrelvir/ritonavir and placebo recipients, regardless of the rebound definition used. For participants who showed a half-log_{10} or greater increase relative to end of treatment and whose viral load was persistent through follow-up, the occurrence was 1.73% placebo and 2.32% nirmatrelvir/ritonavir. In addition to these, some participants had transient half-log_{10} or greater increases relative to end of treatment, the occurrence was 2.35% in placebo vs 4.65% nirmatrelvir/ritonavir. A similar or smaller percentage of placebo recipients compared to nirmatrelvir/ritonavir recipients had nasopharyngeal viral RNA results <LLOQ at all study timepoints in both the treatment and post-treatment periods. Post-treatment viral RNA rebound was not associated with the primary clinical outcome of COVID-19-related hospitalization or death from any cause through Day 28 following the single 5-day course of nirmatrelvir/ritonavir treatment. Post-treatment viral RNA rebound also was not associated with drug resistance as measured by mPro sequencing. The clinical relevance of post-treatment increases in viral RNA following nirmatrelvir/ritonavir or placebo treatment is unknown.

In response to the COVID-19 pandemic, Helix OpCo (Helix), an accredited laboratory (CAP #9382893, CLIA #05D2117342) based in San Diego, CA, with a suite of laboratory capabilities to support infectious disease programs and public health surveillance (including SARS-CoV-2 / Flu diagnostic testing, SARS-CoV-2 whole genome sequencing, and pan-respiratory virus whole genome sequencing), developed laboratory capabilities to support SARS-CoV-2 diagnostic testing. With funding from the NIH's Rapid Acceleration of Diagnostics (RADx) program, Helix became a national "mega-lab" for processing up to 150,000 SARS-CoV-2 rRT-PCR diagnostic tests per day. Helix also has partnered with the CDC to provide SARS-CoV-2 whole genome sequencing starting in December 2020 and continues to support public health sequencing efforts. Helix has processed over ten million SARS-CoV-2 RT-PCR diagnostic tests and hundreds of thousands of SARS-CoV-2 whole genome sequences. Helix has established registries, biorepositories, and IRB protocols for both human genomics and viral genomics programs, enabling performance of research initiatives and key analyses to inform public health decision-making.

In this study, we will leverage Helix's existing US-based Respiratory Registry (External Protocol #0003-0001) of in vitro diagnostic real-time reverse transcription polymerase chain reaction (rRT-PCR) testing for the qualitative detection of nucleic acid in anterior nasal swab

specimens for the detection of the SARS-CoV-2 and Influenzas A/B with linkage to retrospective Komodo Healthcare claims data in order to further examine viral SARS-CoV-2 RNA rebound (SARS-CoV-2 rebound) patterns. Specifically, viral SARS-CoV-2 rebound will be examined after stratification of variables such as SARS-CoV-2 therapeutic treatment, viral load patterns, co-infections, and by timing around rebounds in relation to initial test positivity to treatment for SARS-CoV-2.

This non-interventional study (NIS) is designated as a Post-Authorization Safety Study (PASS) and is conducted voluntarily by Pfizer.

Research question and objectives

Research Questions

Primary research questions:

- What is the risk of viral SARS-CoV-2 rebound, prior to the Omicron era and during the Omicron era, among high frequency (volume) SARS-CoV-2 diagnostic testers?
- What is the risk of viral SARS-CoV-2 rebound when stratified by variables such as age, comorbidities, vaccination status, co-infections, Paxlovid treatment status for SARS-CoV-2 and timing around viral SARS-CoV-2 rebounds, among high frequency (volume) SARS-CoV-2 diagnostic testers prior to the Omicron era and during the Omicron era?

Secondary research question:

- Under alternate definitions of viral SARS-CoV-2 rebound and high frequency (volume) testing, what is the risk of viral SARS-CoV-2 rebound, prior to the Omicron era and during the Omicron era, among high frequency (volume) SARS-CoV-2 diagnostic testers, including when stratified by variables such as age, comorbidities, vaccination status, co-infections, Paxlovid treatment status for SARS-CoV-2 and timing around viral SARS-CoV-2 rebounds?

Research Objectives

Primary Objectives:

Objective 1: To estimate viral SARS-CoV-2 rebound rates (primary definition: 1. a positive test (at the index date), followed by a single negative test, followed by a positive test - within 28 days, or 2. 1 or more positive tests at the index date, followed by 1 or more negative tests, followed by a positive test within same 28 day time span), prior to the Omicron era and during the Omicron era (defined as 01 December 2021 to present), in high frequency (volume) SARS-CoV-2 diagnostic testers (primary definition: 1 or more positive tests followed by 1 or more negative tests followed by 1 or more positives within same 28 day time span and median time between tests (positive or negative) is at most 10 days).

Objective 2: To estimate viral SARS-CoV-2 rebound rates (using the primary definition of viral SARS-CoV-2 rebound), prior to the Omicron era and during the Omicron era, in high frequency (volume) SARS-CoV-2 diagnostic testers (using the primary definition of high frequency

testing), among those treated with Paxlovid/nirmatrelvir-ritonavir for SARS-CoV-2 versus those with no evidence of SARS-CoV-2 treatment (untreated for SARS-CoV-2), stratified by:

a. Demographics of participants

b. Time from first positive test to treatment (if treated).

c. Participants at high-risk for progression to severe SARS-CoV-2 versus those not at high-risk, if sample size allows. If sample size is limited, age (<50 years of age versus 50+ years of age) will be used as a proxy for high-risk for progression to severe SARS-CoV-2 status)

d. Co-infections

e. Vaccinated against SARS-CoV-2 (has received at least 1 dose) and time since last vaccine dose > 6 months vs. ≤ 6 months

f. Multiple SARS-CoV-2 treatment prescription status

Secondary Objectives:

Objective 1: As a sensitivity analysis, using an alternate less strict definition of high frequency (volume) SARS-CoV-2 diagnostic tester (defined as tested for SARS-CoV-2 at least 3 times and median time between tests (positive or negative) is at most 14 days), to estimate viral SARS-CoV-2 rebound rates (using the primary definition), prior to the Omicron era and during the Omicron era, in high frequency (volume) SARS-CoV-2 diagnostic testers, among those treated with Paxlovid/nirmatrelvir-ritonavir for SARS-CoV-2 versus those with no evidence of SARS-CoV-2 treatment (untreated) for SARS-CoV-2, stratified by:

a. Demographics of participants

b. Time from first positive test to treatment (if treated).

c. Participants at high-risk for progression to severe SARS-CoV-2 versus those not at high-risk, if sample size allows. If sample size is limited, age (<50 years of age versus 50+ years of age) will be used as a proxy for high-risk for progression to severe SARS-CoV-2 status)

d. Co-infections

e. Vaccinated against SARS-CoV-2 (has received at least 1 dose) and time since last vaccine dose > 6 months vs. ≤ 6 months

f. Multiple SARS-CoV-2 treatment prescription status

Objective 2: As a sensitivity analysis, using an alternate stricter definition of high frequency (volume) SARS-CoV-2 diagnostic tester (defined as tested for SARS-CoV-2 at least 3 times and median time between tests (positive or negative) is at most 7 days), to estimate viral SARS-CoV-

2 rebound rates (using the primary definition), prior to the Omicron era and during the Omicron era, in high frequency (volume) SARS-CoV-2 diagnostic testers, among those treated with Paxlovid/nirmatrelvir-ritonavir for SARS-CoV-2 versus those with no evidence of SARS-CoV-2 treatment (untreated) for SARS-CoV-2, stratified by:

a. Demographics of participants

b. Time from first positive test to treatment (if treated).

c. Participants at high-risk for progression to severe SARS-CoV-2 versus those not at high-risk, if sample size allows. If sample size is limited, age (<50 years of age versus 50+ years of age) will be used as a proxy for high-risk for progression to severe SARS-CoV-2 status)

d. Co-infections

e. Vaccinated against SARS-CoV-2 (has received at least 1 dose) and time since last vaccine dose > 6 months vs. ≤ 6 months

f. Multiple SARS-CoV-2 treatment prescription status

Objective 3: As a sensitivity analysis, under a less strict definition of viral SARS-CoV-2 rebound (defined as two positives more than 14 days apart),, to estimate viral SARS-CoV-2 rebound rates, prior to the Omicron era and during the Omicron era, in high frequency (volume) SARS-CoV-2 diagnostic testers (using the primary definition), among those treated with Paxlovid/nirmatrelvir-ritonavir for SARS-CoV-2 versus those with no evidence of SARS-CoV-2 treatment (untreated) for SARS-CoV-2, stratified by:

a. Demographics of participants

b. Time from first positive test to treatment (if treated).

c. Participants at high-risk for progression to severe SARS-CoV-2 versus those not at high-risk, if sample size allows. If sample size is limited, age (<50 years of age versus 50+ years of age) will be used as a proxy for high-risk for progression to severe SARS-CoV-2 status)

d. Co-infections

e. Vaccinated against SARS-CoV-2 (has received at least 1 dose) and time since last vaccine dose > 6 months vs. ≤ 6 months

f. Multiple SARS-CoV-2 treatment prescription status

Study design

This is a non-interventional, retrospective cohort study using secondary data sources containing structured data from the US. This study will include adults (≥18 years of age) and pediatric

participants (<18 years of age) with a positive SARS-CoV-2 test (e.g., positive polymerase chain reaction (PCR) of direct SARS-CoV-2 viral testing).

Study population and cohorts
This study will be conducted using SARS-CoV-2 diagnostic testing data from the Helix Respiratory Registry with linkage to the retrospective Komodo Healthcare claims data during the period of June 01, 2020, to February 28, 2023.

The main cohort will include all participants meeting inclusion and exclusion criteria. The high-risk subcohort (if sample size is limited, age (<50 years of age versus 50+ years of age) will be used as a proxy for high-risk for progression to severe SARS-CoV-2 status) will include all participants in the main cohort with high-risk clinical characteristics (comorbidities) for progression to severe SARS-CoV-2.

Index date is defined as first confirmed positive SARS-CoV-2 rRT-PCR test within a positive - negative – positive test sequence that occurs within 28 days during the study period (by omicron era status). Only the first infection for each patient during the study period (by omicron era status) will be examined. Baseline period will vary by patient, based on how much history is available and index date. The follow-up period starts at index date and participants will be followed until the date of the first occurring of the following events: 1) Death, 2) At the end of their database disenrollment from the database, 3) At the end of study period, 4) Occurrence of the outcome/endpoint of interest.

Variables
Study variables include demographics, diagnostic (testing and disease), high-risk clinical characteristics (comorbidities) for progression to severe SARS-CoV-2, SARS-CoV-2 medications, and health utilization.

The primary endpoint/safety events of interest is viral SARS-CoV-2 rebound, where the main definition defines rebound as: 1.) a positive test (at the index date), followed by a single negative test, followed by a positive test - within 28 days, or 2.) 1 or more positive tests starting at the index date, followed by 1 or more negative tests, followed by a positive test within same 28-day time span. Index date will be defined as first positive test date. Alternate definitions of SARS-CoV-2 rebound will be included in sensitivity analyses and detailed in the SAP.

Data Sources

This study will be conducted using SARS-CoV-2 diagnostic testing data from the Helix Respiratory Registry with linkage to the retrospective Komodo Healthcare claims data during the period of June 01, 2020, to February 28, 2023, after appropriate de-identification certification, data privacy, and security compliance in accordance with Pfizer and Helix policies and industry best practices.

Study Size

This is a descriptive study with no minimum sample size. All eligible participants during the relevant study periods will be included. This is a NI study with no a priori hypotheses specified; therefore, sample size calculations are not applicable.

Data Analysis

Descriptive statistics among individuals with SARS-CoV-2 in the Helix Respiratory Registry that overlap with the retrospective Komodo Healthcare claims data will be summarized. Means with standard deviations, medians with interquartile ranges will be provided for continuous variables. Numbers and percentages will be provided for dichotomous variables or categorical variables.

For exploratory analyses, logistic regression or Cox proportional hazards regression models will be used to identify risk factors and outcomes for viral SARS-CoV-2 rebound in adjusted analyses. Model selection will balance biological and contextual knowledge for potential confounders with statistical considerations for model fit and the bias versus variance tradeoff.

An analysis comparing viral SARS-CoV-2 rebound, between participants treated with Paxlovid, untreated participants (and pending sufficient sample size, comparator SARS-CoV-2 treatment group(s) described above) will be conducted. Viral SARS-CoV-2 rebound and other outcomes in participants who received Paxlovid compared with untreated participants (and/or comparator SARS-CoV-2 treatment group(s), pending sufficient sample size) will be evaluated using multivariable Cox regression models with propensity score inverse probability of treatment weighting (IPTW) with a robust variance estimator having age, sex, week and year of initial positive SARS-CoV-2 test, and potentially the aforementioned baseline clinical characteristics and comorbidities as independent variables to account for time-varying confounding. Standardized mean differences (SMDs) will be calculated to check whether IPTW yielded comparable cohorts, considering an SMD below 0.1 as indicating satisfactory covariate balance.

Detailed methodology for summary and statistical analyses of data collected in this study will be documented in a statistical analysis plan (SAP), which will be dated, filed, and maintained by the sponsor. The SAP may modify the plans outlined in the protocol; any major modifications of primary endpoint definitions or their analyses would be reflected in a protocol amendment.

Milestones

Milestone	Planned date
Start of data collection	22 February 2023
Registration in the European Union (EU) PAS register	21 February 2023
End of data collection	15 March 2023
Final study report	15 February 2024