CLINICAL RESEARCH IN INFECTIOUS DISEASES

STATISTICAL ANALYSIS PLAN
for
DMID Protocol: 18-0010

Study Title:
A Controlled Human Infection Study of Influenza A/Bethesda/MM2/H1N1 Virus (A/California/04/2009/H1N1-like) in Healthy Subjects to Assess the Effect of Pre-Existing Immunity on Symptomatic Influenza Virus Infection

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STUDY TITLE

Protocol Number Code:	**DMID Protocol: 18-0010**
Development Phase:	Phase 1 (Controlled human infection study)
Products:	Influenza A/Bethesda/MM2/H1N1virus
Form/Route:	Intranasal
Indication Studied:	Influenza
Sponsor:	Division of Microbiology and Infectious Diseases National Institute of Allergy and Infectious Diseases National Institutes of Health
Clinical Trial Initiation Date:	07OCT2019
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Version Number:	1.0

This study was performed in compliance with Good Clinical Practice.

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LIST OF ABBREVIATIONS

°C	Degree Celsius
°F	Degree Fahrenheit
AE	Adverse Event/Adverse Experience
ALT	Alanine Transaminase
ANC	Absolute Neutrophil Count
ASC	Antibody Secreting Cells
BMI	Body Mass Index
BP	Blood Pressure
CBC	Complete Blood Count
CFR	Code of Federal Regulations
CHI	Controlled Human Infection
cMRI	Cardiac MRI
CO_2	Carbon Dioxide
CoP	Correlation of Protection
COPD	Chronic Obstructive Pulmonary Disease
COVID-19	Coronavirus Disease 2019
Cr	Creatinine
CRF	Case Report Form
CXR	Chest X-Ray
DBP	Diastolic Blood Pressure
DCC	Data Coordinating Center
DMID	Division of Microbiology and Infectious Diseases, NIAID, NIH, DHHS
DSMB	Data and Safety Monitoring Board
ECG	Electrocardiogram
eCRF	Electronic Case Report Form
ELISA	Enzyme-linked Immunosorbent Assay
EPT	End Point Titers
ER	Emergency Room
FDA	Food and Drug Administration
GMFR	Geometric Mean Fold Rise

List of Abbreviations *(continued)*

GMP	Good Manufacturing Practice
GMT	Geometric Mean Titer
H1N1pdm09	A/California/04/2009 (H1N1)-like Influenza A Virus
HA	Hemagglutinin
HAI	Hemagglutination Inhibition
HR	Heart Rate
ICF	Informed Consent Form
ICH	International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use
IV	Intravenous
K	Potassium
Kg/m^2	Kilogram per Square Meter
mg	Milligram
mg/dL	Milligrams per Deciliter
mL	Milliliter
mm	Millimeter
mm^3	Cubic Millimeter
MMID	Mild-to-Moderate Influenza Disease
MN	Microneutralization
N	Number (typically refers to subjects)
NAI	Neuraminidase Inhibition
NIAID	National Institute of Allergy and Infectious Diseases, NIH, DHHS
NIH	National Institutes of Health
NLF	Nasal Lavage Fluid
NP	Nasopharyngeal
PBMC	Peripheral Blood Mononuclear Cells
PE	Physical Exam
PI	Principal Investigator
RR	Respiratory Rate
RT-PCR	Reverse Transcription – Polymerase Chain Reaction
SAE	Serious Adverse Event/Serious Adverse Experience

List of Abbreviations *(continued)*

SBP	Systolic Blood Pressure
SDCC	Statistical Data Coordinating Center
SE	Standard Error
sIgA	Secretory Immunoglobulin A
SMC	Safety Monitoring Committee
SpO_2	Saturation of Peripheral Oxygen
$TCID_{50}$	Median Tissue Culture Infective Dose
ULN	Upper Limit of Normal
US	United States
VTEU	Vaccine and Treatment Evaluation Units
WBC	White Blood Cell
wt	Wild-type

1. PREFACE

The Statistical Analysis Plan (SAP) for "A Controlled Human Infection Study of Influenza A/Bethesda/MM2/H1N1 Virus (A/California/04/2009/H1N1-like) in Healthy Subjects to Assess the Effect of Pre-Existing Immunity on Symptomatic Influenza Virus Infection" (DMID Protocol 18-0010) describes and expands upon the statistical information presented in the protocol.

This document describes all planned analyses and provides reasons and justifications for these analyses. It also includes sample tables, listings, and figures planned for the final analyses. Regarding the final analyses and Clinical Study Report (CSR), this SAP follows the International Conference on Harmonisation of Technical Requirements for Registration of Pharmaceuticals for Human Use (ICH) Guidelines, as indicated in Topic E3 (Structure and Content of Clinical Study Reports), and more generally is consistent with Topic E8 (General Considerations for Clinical Trials) and Topic E9 (Statistical Principles for Clinical Trials). The structure and content of the SAP provides sufficient detail to meet the requirements identified by the FDA and ICH, while all work planned and reported for this SAP will follow internationally accepted guidelines published by the American Statistical Association and the Royal Statistical Society for statistical practice.

This document contains four sections: (1) a review of the study design, (2) general statistical considerations, (3) comprehensive statistical analysis methods for efficacy and safety outcomes, and (4) a list of proposed tables and figures. Within the table, figure, and listing mock-ups (Appendices 1, 2, and 3), references to CSR sections are included. Any deviation from this SAP will be described and justified in protocol amendments and/or in the CSR, as appropriate. The reader of this SAP is encouraged to also review the study protocol for details on conduct of the study and the operational aspects of clinical assessments.

2. INTRODUCTION

The high morbidity and mortality associated with both pandemic and seasonal influenza, the continued threat of highly pathogenic avian influenza infections, the development and global spread of antiviral resistance, and the inadequacy of current influenza vaccines makes influenza a high research priority. Although prior influenza research has addressed numerous aspects of viral biology and pathogenesis, many questions remain unanswered. Advancing basic, translational and clinical research on influenza will inform the development of new and improved diagnostics, therapeutics, and vaccines [1].

Influenza human challenge models offer a unique opportunity to further study influenza pathogenesis. These models are conducted in a well-controlled challenge setting in which virus exposure, infection rate, virus shedding, and sampling are being monitored in real-time. Human challenge models provide a powerful tool to assess prevention and treatment options for influenza in a shorter time frame, with a smaller number of subjects and against a known virus strain. Due to the known time of infection, they allow for targeted collection of clinical samples, including prior to infection and in short time frames following inoculation. Human challenge models may be particularly useful to evaluate novel influenza vaccines that do not target the HA head region, and therefore cannot be judged based on current regulatory criteria for current inactivated influenza vaccines.

Since 2012, the Laboratory of Infectious Diseases (LID) at NIAID has been working to re-establish a controlled influenza A/H1N1 challenge model in healthy subjects in the US. The first dose-ranging study of 49 healthy subjects using a Good Manufacturing Practice (GMP) influenza A (H1N1pdm09) challenge strain demonstrated that mild to moderate influenza illness could be induced in 70% to 80% of healthy subjects and mimicked natural infection both clinically and immunologically [2]. Clinical symptoms of influenza occurred post-challenge with doses ranging from 10^3 to 10^7 $TCID_{50}$ but were most prevalent at the 10^6 and 10^7 $TCID_{50}$ doses. NIAID's second challenge study at LID using the H1N1 model evaluated anti-hemagglutinin (HA), anti-neuraminidase (NA) and anti-HA stalk antibodies as correlates of protection in H1N1 influenza illness in 74 healthy subjects, finding that while higher Hemagglutination inhibition (HAI) titers are indicative of some protection, Neuraminidase inhibition (NAI) titers may be more predictive of protection from influenza infection and reduced disease [3, 4]. These studies led to several ongoing laboratory studies using samples collected to address questions of intra-host evolution, gene expression, and immune response, many of which are expected to be published in the coming months.

In clinical studies the model is now being implemented to test new therapeutics and vaccines (NCT02371668 and NCT03845231). This virus has been used in 3 published trials and 1 unpublished trial to date in approximately 400 participants. No significant safety issues have been identified, no severe or complicated cases of influenza have occurred, and no transmission events outside of the study have been observed [2, 3, 5]. These studies also helped to define a clinical endpoint termed "mild-to-moderate influenza disease" (MMID) which included evidence of influenza A virus shedding by RT-PCR and any of a number of standardized signs, symptoms, and laboratory findings consistent with clinical influenza illness. MMID is assessed using a validated, patient-reported outcome (PRO) measure to standardize assessment of influenza (flu) symptoms in clinical research, i.e. FLU-PRO Survey Instrument [6].

2.1. Purpose of the Analyses

The scientific goal of this study is to conduct an influenza CHI study in healthy adults to evaluate the association of pre-existing HAI titers and the development of symptomatic RT-PCR-positive influenza virus infection in healthy adults after being challenged. The study will build upon the NIH Clinical Center's

influenza CHI model with influenza A/California/04/2009 (H1N1)-like virus [6]. Additional study objectives will assess clinical manifestations, viral shedding and immunological responses to the challenge virus.

3. STUDY OBJECTIVES AND OUTCOME MEASURES

Refer to Table 1 Study Objectives and Outcome Measures. Note that the outcome measures have been elaborated upon from what are given in the protocol.

3.1. Study Definitions and Derived Variables

3.1.1. Study Definitions

Baseline (pre-challenge) values for HAI, NAI, and MN antibody titer will be defined as measurements obtained within 48 hours before receiving the challenge virus (Study Day -2 or -1).

Seroprotection status will be defined as a binary variable ("Low < 1:40" *vs.* "High ≥ 1:40"), based on the 1:40 titer threshold for HAI, NAI, and MN assays.

Mild-to-moderate influenza disease (MMID) will be defined based on the presence of each of the following during the challenge period of the study (Day 1 through Day 8 or discharge from the challenge unit):

1. Viral shedding detected by any approved positive RT-PCR assay (qualitative or quantitative, as described below) from a NP swab.
2. Any one or more of the following symptoms or signs or laboratory findings, as related to the study agent: Arthralgia, Chest tightness, Chills, Conjunctivitis, Nasal Congestion, Sinus Congestion, Coryza, Decreased Appetite, Diarrhea, Dry Cough, Dyspnea/Shortness of Breath, Fatigue/Tiredness, Fever (>38.0°C), Headache, Lymphopenia (<1000 cells/mL), Myalgia, Nausea, Oxygen Saturation Decrease by ≥ 3% from baseline, Productive Cough, Rhinorrhea, Sore throat, and Sweats. The presence of arthralgia or myalgia will be determined by the FLU-PRO response for "body aches or pain".

Two thresholds for MMID will be applied:

- MMID-1: one or more positive RT-PCR assay and at least one of the symptoms, signs, and laboratory findings listed above, each occurring at any point during the challenge period of the study.
- MMID-2: two or more study days with a positive RT-PCR assay, and at least one of the symptoms signs, and laboratory findings listed above, each occurring at any point during the challenge period of the study.

MMID-1 will be considered the primary definition and used for the primary and secondary analyses. MMID-2 will be used in sensitivity analyses to assess changes in results from making the MMID definition more stringent.

Generally, viral shedding/detection will be defined as a binary ("RT-PCR Positive" *vs.* "RT-PCR Negative") variable, with positivity requiring at least one positive result (for quantitative RT-PCR, above a pre-determined threshold for positivity as determined by the lab(s) performing these) between quantitative PCR and qualitative multiplex respiratory virus assay on a NP swab specimen that includes one or more of the following results: Influenza A, Influenza A 2009 H1N1, and Influenza A H1. For certain analyses comparing results between assays, viral shedding status will be determined separately by assay, which will be indicated clearly in each relevant exhibit. Some analyses will include separate categories for those with one or more positive PCR tests and those with two or more tests (not mutually exclusive).

Symptom status will be defined as a binary variable, based on presence of any MMID-related symptom ("Asymptomatic" *vs.* "Ever Symptomatic") on any study day over the challenge period (Study Day 2 to 8).

For analyses relating to aggregate outcomes over the entire challenge period, infection status will be categorized into: RT-PCR Positive (at least once) Symptomatic, RT-PCR Positive Asymptomatic (for subjects with at least one PCR+ result but no symptoms reported), and RT-PCR Negative. For some analyses, results will also include a category for at least 2 RT-PCR positive tests and symptomatic, as part of the sensitivity analyses to determine if a more stringent threshold for classifying infection is needed.

3.1.2. Derived Variables

For individual subjects, fold rise will be calculated as $\frac{post-challenge\ titer}{pre-challenge\ titer}$, where the pre-challenge value is the result obtained at baseline.

Seroconversion for HAI, NAI, and MN assays will be defined as either a pre-challenge titer < 1:10 and a post-challenge titer ≥ 1:40, or a pre-challenge titer ≥ 1:10 and a minimum four-fold rise in post-challenge titer.

Viral copies/mL will be estimated as a mean across quantitative RT-PCR replicates. Some replicates may be dropped from the calculations if potential quality control issues are noted.

Duration of viral shedding will be defined as time from the initial positive NP swab until the day after the final positive NP swab, with viral shedding defined as above in Section 3.1.1.

Peak viral shedding will be calculated as the maximum viral concentration (RNA copies/mL) recorded the duration of the inpatient study period for each individual.

Total viral shedding over the inpatient period will be summarized by the area under the curve (AUC), approximated via the linear trapezoidal method, where the trajectory of an individual's viral concentration is partitioned into $n-1$ subareas until the last inpatient observation at time t_n. As the linear trapezoidal AUC method approximates the entire area by calculating the cumulative sum of the subareas, the formula that will be applied is:

$$\text{AUC}_i = \frac{1}{2} * \sum_{j=1}^{n} \left(t_{j+1} - t_j\right) * \left(C_{ij+1} + C_{ij}\right)$$

where $\left(t_{j+1} - t_j\right)$ is the difference between the *j*th and *j*+1th time points within the inpatient period, and C_{ij} and C_{ij+1} are the viral concentrations measured at the *j*th and *j*+1th time points for the *i*th individual. Intermittent missing values are expected to be minimal, so no imputation is planned.

Solicited symptoms of influenza are recorded daily via the 32-item FLU-PRO survey instrument across 6 domains: Nose (4 items), Throat (3 items), Eyes (3 items), Chest/Respiratory (7 items), Gastrointestinal (4 items), and Body Systemic (11 items) [7]. Respondents are asked to rate each symptom on a 5-point ordinal scale as follows:

Response[A]	**Score**
Not at all **or** Never **or** 0 times	0
A little bit **or** Rarely **or** 1 time	1
Somewhat **or** Sometimes **or** 2 times	2
Quite a bit **or** Often **or** 3 times	3
Very much **or** Always **or** 4 or more times	4
[A]See Appendix (Figure 2) for FLU-PRO questionnaire.	

For 27 of the items, symptoms are scaled in terms of extent (i.e., not at all, somewhat, quite a bit, etc.). For 5 items, severity is assessed in terms of numerical frequency, i.e., vomiting or diarrhea (0 times, 1 time, 2 times,

3 times, or 4 or more times); frequency of sneezing, coughing, and coughing up mucus or phlegm is evaluated on a scale from 0 ("Never") to 4 ("Always").

The FLU-PRO total severity score is defined as the mean score across all 32 items, with scores ranging from 0 to 4. Six domain scores are also computed as mean scores, representing symptom severity in each body system.

4. INVESTIGATIONAL PLAN

4.1. Overall Study Design and Plan

The goal of this CHI study is to characterize clinical manifestations, viral shedding and immunological response to Influenza A/Bethesda/MM2/H1N1. Influenza A/Bethesda/MM2/H1N1 challenge is performed using a 2 mL dose of approximately $5x10^6$ $TCID_{50}$/mL virus. A total of up to 80 participants who are in good health and meet all eligibility criteria were planned to be admitted to an inpatient challenge unit and receive CHI intranasally.

After CHI, subjects complete daily self-assessments using the Flu-Pro Survey Instrument and Validation Diary through 14 days post-challenge to generate symptom extent scores. Subjects clinical status is monitored closely. Subjects undergo scheduled blood draws, nasal lavages, and nasal swab collections for immunology and virologic laboratory testing, and safety labs are performed at scheduled times. All subjects are followed for approximately 90 days post-challenge.

Additional follow-up in-person visits occur on Visits 9 (Day 15), 10 (Day 29) and 11 (Day 61). Subjects undergo scheduled blood draws and nasal lavages for immunology and virologic laboratory testing during this follow-up period. A final follow-up by telephone occurs on Visit 12 (Day 91). Further details on the study design are presented in Figure 2.

4.2. Discussion of Study Design, Including the Choice of Control Groups

All participants receive the study influenza challenge virus. As such, there is no control group inherent in the study design.

4.3. Selection of Study Population

The study population is healthy subjects (male and non-pregnant, non-breastfeeding females) between the ages of 18 to 49 inclusive drawn from the general communities of each study site. Subjects must not have medical conditions at high risk for severe complications of influenza virus infection, as defined by the US Centers for Disease Control and Prevention (CDC). Eligibility criteria ensure that subjects have not received (and do not plan to receive) therapies that may be associated with impaired immune responsiveness, such as immunosuppressive therapies or cytotoxic drugs. Additional eligibility criteria are outlined in the study protocol Sections 6.1.1 and 6.1.2.

4.4. Interventions

4.4.1. Intervention Administered

Intranasal CHI is performed using a 2 mL dose of approximately $5x10^6$ $TCID_{50}$/mL virus. One mL of virus is administered in each nostril of a recumbent participant.

4.4.2. Identity of Investigational Product(s)

The challenge virus is nearly identical to circulating influenza A H1N1pdm09 viruses. There is no indication that the 2009 H1N1 Challenge Virus would behave any differently in humans than the wt influenza A/California/04/2009 (H1N1) virus, based on the in vitro and in vivo data. The A/Bethesda/MM2/H1N1 human challenge virus was manufactured in certified Vero cells by Charles River Laboratories in Malvern, PA from a reverse-genetics (RG) derived virus seed stock produced by Dr. Matthew Memoli and his

laboratory team in the Viral Pathogenesis and Evolution Section of the Laboratory of Infectious Diseases (NIAID).

4.4.3. Method of Assigning Subjects to Intervention Groups (Randomization)

Not applicable. All subjects receive the challenge virus.

4.4.4. Selection of Doses in the Study

The first dose-ranging trial [NCT01646138] using a GMP influenza A (H1N1pdm09) challenge strain demonstrated that a total dose of $1x10^7$ $TCID_{50}$ induced MMID at a rate of 60% or higher in individuals who had pre-challenge HAI antibody titers of ≤ 1:40 [6].

4.4.5. Selection and Timing of Intervention Administration for Each Subject

On Day 1, eligibility and clinical status are reviewed. If criteria for proceeding with challenge are met, subjects receive A/Bethesda/MM2/H1N1 CHI.

4.4.6. Blinding

Not applicable. All subjects enrolled receive the challenge virus.

4.4.7. Prior and Concomitant Therapy

Concomitant medications are reviewed at every study visit through the end of the study. Women of childbearing potential in a heterosexual relationship must agree to use true abstinence or use at least one acceptable primary form of contraception through the end of the study.

Receipt of any influenza vaccine during the 2018/2019 influenza vaccine season regardless of the date of receipt are documented. Subject receipt of non-seasonal influenza vaccine, including those that are experimental, product type, vaccine virus strains and approximate date of vaccination (see Section 7.1.1 of study protocol) are also documented.

The following prescription or over-the-counter medications cannot be used within 7 days prior to admission to the inpatient unit and through the inpatient stay, unless approved by the investigator: oseltamivir, zanamivir, peramivir, baloxavir marboxil, amantadine (generic) and rimantadine (Flumadine and generic), aspirin, intranasal steroids, decongestants, antihistamines, and other non-steroidal anti-inflammatory drugs (NSAIDs).

Subjects who use asthma medications including inhaled, oral, or IV corticosteroids, leukotriene modifiers, long and short acting beta agonists, theophylline, ipratropium, biologics, will be excluded from the study.

Any medications that may be associated with impaired immune responsiveness including, but not limited to, corticosteroids exceeding 10 mg/day of prednisone equivalent, allergy injections, immunoglobulin, interferon, immunomodulators, cytotoxic drugs, or systemic corticosteroids or other similar or toxic drugs are prohibited during the preceding 12-month period prior to screening. Low dose topical and intranasal steroid preparations used for a discrete period of time are permitted.

Subjects should not have received any investigational drug/investigational vaccine/licensed vaccine within 30 days prior to the planned date of CHI. Subjects should not have received influenza vaccine six months prior to challenge and must not plan to receive an influenza vaccine 60 days post challenge. Subjects should not have received blood or blood products during the six months prior to the planned date of CHI.

4.4.8. Treatment Compliance

All subjects will be directly observed at the time of dosing by a member of the clinical research team who is licensed to administer the study product at the VTEU site.

4.5. Clinical, Immunogenicity and Safety Variables

See Table 2 for Schedule of Study Procedures.

4.5.1. Clinical Variables

Clinical responses after subjects are challenged, such as viral detection, viral shedding, infection status, as well as solicited symptoms and laboratory findings that are a part of MMID, will be assessed at each study day, and summarized from the time of CHI through study Day 8 or when the subject is discharged, whichever comes first.

Grading scales for FLU-PRO solicited symptoms are provided in Figure 3.

4.5.2. Immunogenicity Variables

Individual HAI, NAI and MN antibody results will be reported by the central immunology laboratory. Assay results are reported as a (endpoint) titer with values of $10*2^k$, where k=0, 1, 2, etc. The lower limit of detection for HAI, NAI and MN assays is 1:10; values below the limit of detection are reported as '<10', and for analysis are imputed as one-half the limit of detection (10/2 = 5). For analysis, the geometric mean of repeated results (i.e. valid replicates) for each sample will be computed and used as the response for all subsequent calculations. The variation arising from multiple measurements for an individual at a single time point is not used in subsequent calculations. See Section 3.1 for definitions of derived variables for the analysis of HAI, NAI and MN data.

All immunogenicity data will be uploaded into the SDCC's electronic data capture system. Data for other exploratory assays will be described in a separate analysis plan.

4.5.3. Safety Variables

Safety will be assessed by the frequency and severity of:

1. Study product-related serious adverse events occurring from the time of the challenge through the end of the study (approximately 3 months post-challenge).
2. Clinical safety laboratory adverse events occurring from the time of CHI through study Day 8. Parameters to be evaluated include: white blood cells (WBCs), absolute lymphocyte count, hemoglobin, platelets, and alanine transaminase (ALT), and creatinine (Cr).
3. Unsolicited Adverse Events – study product-related non-serious adverse events occurring from the time of CHI through approximately 30 days post-challenge.

Signs, symptoms, laboratory findings that are part of MMID from the time of CHI through 14 days post-challenge will not be considered adverse events. These signs, symptoms and laboratory findings (mild and moderate) which define MMID include the following: arthralgia, chest tightness, chills, conjunctivitis, nasal congestion, sinus congestion, coryza, decreased appetite, diarrhea, cough, dyspnea/shortness of breath, fatigue/tiredness, fever (>38.0°C), headache, lymphopenia (<1000 cells/mL), myalgia, nausea, oxygen saturation decrease by ≥3% from baseline, rhinorrhea, sore throat, and sweats.

Grading scales for vital signs and clinical laboratory parameters are provided in Table 6, Table 7, and Table 8.

5. SAMPLE SIZE CONSIDERATIONS

The study is planned to enroll up to 80 subjects, all of whom receive the study influenza challenge virus. While the study is not designed to test any specific null hypothesis or reach a pre-determined level of estimation precision, Table 3 and Table 4 present the precision and power for estimates and hypothesis tests of interest based on the proposed sample size.

Table 3 gives the power to detect varying decreases in the odds of a binary endpoint such as MMID with a one standard deviation (SD) increase in a continuous covariate (assumed to be normally distributed) among all subjects (n=80), via a Wald test from a logistic regression model at the α=0.05-level. This would roughly correspond to testing for an association between log-transformed baseline HAI titer and MMID.

Table 4 indicates the probability of observing one or more safety events, such as solicited symptom or an SAE of a particular classification in all subjects (n=80), given underlying event probabilities.

Binomial confidence intervals (CI) are widest (have the least precision) when the response rate is 50%. With this event rate among all subjects (40 events among n=80 subjects), the corresponding exact Clopper-Pearson CI would be (38.6%, 61.4%).

Adjustments to p-values in the presence of multiple comparisons may be made, depending on the objective, but these calculations do not account for this possibility. For further details on adjustment for multiple comparisons, refer to Section 6.8 of this document.

6. GENERAL STATISTICAL CONSIDERATIONS

6.1. General Principles

All continuous variables will be summarized using the following descriptive statistics: non-missing sample size (denoted by "n"), mean, standard deviation, median, maximum and minimum. Titers will be summarized with geometric means, first across replicates to compute one value for each individual sample and then across subjects within clinical outcome groups. The frequency and percentages (based on the non-missing sample size) of observed levels will be reported for all categorical measures. In general, all data will be listed, sorted by site or other grouping variable (e.g. infection status), subject, and by visit number within subject, where appropriate. All tables will be annotated with the total population size relevant to that table, including any missing observations.

If a substantial number of subjects meet eligibility criteria but had intercurrent infections that might affect the immunoassay results, sensitivity analyses will be performed excluding those cases and qualitative comparisons will be made between the Modified Intent-to-Treat (mITT) and Per Protocol analyses. See Section 6.3.3 below for additional details.

6.2. Timing of Analyses

No interim analysis is planned.

The CSR will be completed when all primary and secondary safety, clinical, and immunological endpoint data are available. Any available data from the exploratory endpoints may also be included. Additional exploratory endpoint data would be included in an addendum to the CSR.

6.3. Analysis Populations

6.3.1. Safety Population

The Safety Population will include all subjects who received the study influenza challenge.

6.3.2. Modified Intent-to-Treat (mITT) Population

The mITT population will include all subjects who received the study influenza challenge and have at least a baseline sample for immunogenicity. For analyses involving endpoints aggregated over the entire challenge period, only those followed for the entire duration of the study challenge period, or at least until the relevant endpoint can be calculated, will be included.

6.3.3. Per Protocol Population

The PP population may be used for sensitivity analyses if at least 10% of analysis data would be excluded per the reasons below. The PP population will include all subjects and visits in the mITT population with the following exclusions:

- Data from all available visits for a given subject found to be ineligible at baseline
- Data from any visit that occurs substantially out of window
    - For follow-up visits where immunogenicity data are collected, the following visit windows are defined in the protocol:
        - Visit 09: Day 15 ± 3 days

  - Visit 10: Day 29 ± 3 days
  - Visit 11: Day 60 ± 5 days

- Data from all visits subsequent to major protocol deviations:
  - Receipt of immunosuppression or any medications that may be associated with impaired immune responsiveness
  - Receipt of any investigational drug/investigational vaccine /licensed vaccine
  - Receipt of blood or blood products, or blood donation
  - Meeting other exclusion criteria that could potentially impact the integrity or interpretability of the results, per the study team or medical monitor

6.4. Covariates and Subgroups

Exploratory analysis on the association of MMID incidence and pre-challenge HAI antibody titer will include assessment of confounding by the following covariates: age (continuous), sex (male, female), prior receipt of seasonal influenza vaccine(s) (received 2018-2019, did not receive 2018-2019, unknown), and clinical site.

The protocol does not define any formal subgroup analyses, and the study was not powered to perform subgroup analyses.

6.5. Missing Data

All attempts are made to collect all data per protocol.

If FLU-PRO Survey Instrument data for one study day is missing, the missingness will be indicated in related summaries, but no imputation is planned for endpoints derived from these data (e.g. MMID). No imputation for missing quantitative RT-PCR, immunological or transcriptomic/genomic data is planned either.

Any data point that appears to be erroneous or inexplicable based on clinical judgment will be investigated as a possible outlier. If data points are identified as outliers, sensitivity analyses may be performed to examine the impact of including or excluding the outliers. Any substantive differences in these analyses will be reported.

6.6. Interim Analyses and Data Monitoring

Interim analyses are only used to terminate this trial if the study sponsor determines unanticipated safety events are of sufficient concern to require such action. These assessments are not made based on formal statistical hypothesis testing. As described in the study protocol (Section 9.7), DMID would convene an SMC to review participant, clinical, and safety data.

6.7. Multicenter Studies

Data will be pooled across all clinical sites. While clinical sites use standardized procedures and rely on central laboratories for the assessment of immunogenicity and clinical endpoints, assessments of symptoms and MMID will be described by site. See Section 8.2.1.1 and Section 8.3.3 for details.

6.8. Multiple Comparisons/Multiplicity

While the study is not designed to test any specific null hypothesis based on this proposed sample size, adjustments to p-values in the presence of multiple comparisons may be made for transcriptomics analysis, which will be further described in a separate SAP document. For other exploratory analyses across numerous endpoints (e.g. biomarker analyses), stricter thresholds for determining statistical significance may be used in place of p-value adjustment (e.g. $p<0.01$ or $p<0.001$), as appropriate for the number of individual endpoints tested. These will be determined ahead of conducting any hypothesis testing and will be clearly indicated in the final report(s).

7. STUDY SUBJECTS

7.1. Disposition of Subjects

Screening failures will be summarized in Table 12. A summary of subject disposition will be presented in Table 9, including the total number of subjects screened, enrolled, enrolled but not challenged, terminated early, or having completed the last follow-up visit.

The number and percentage of enrolled subjects excluded from each analysis population are presented in Table 10. A listing of subjects excluded from each analysis population will be presented in Listing 5.

A flowchart showing the disposition of study subjects, adapted from the CONSORT Statement, will be included and will present the number of subjects screened, challenged, lost to follow up, and analyzed (Figure 1).

A listing of subjects who discharged early or terminated from study follow-up and the reason will be included in Listing 2, where details about missed follow-up visits related to the Coronavirus Disease 2019 (COVID-19) pandemic will be included. Missed follow-up visits due to COVID-19 are likely to not substantially impact the planned analysis, however, as the most recent in-person clinic visits are expected to have been completed prior to the pandemic.

7.2. Demographic and Other Baseline Characteristics

Summaries of age, sex, ethnicity, race, prior receipt of seasonal vaccination, and baseline seroprotection status for HAI, NAI, and MN assays will be presented for all subjects challenged by site in Table 13 and Table 14. Age will be summarized as a continuous as well as categorical variable, categorized as 18-29, 30-39, and 40-49. Ethnicity is categorized as Hispanic or Latino, or not Hispanic and not Latino. In accordance with NIH reporting policy, subjects may self-designate as belonging to more than one race or may refuse to identify a race, the latter reflected in the CRF as "No" to each racial option. BMI will be summarized as a continuous as well as categorical variable, categorized as <30 and ≥ 30. Self-reported history of prior receipt of seasonal influenza vaccine will be categorized as follows: received 2018-2019 seasonal vaccine, did not receive 2018-2019 seasonal vaccine, or unknown. Baseline antibody titers will be summarized as a continuous as well as categorical variable, categorized according to baseline seroprotection status.

Individual subject listings will be presented for all demographics in Listing 6.

7.3. Measurements of Treatment Compliance

Subjects are directly observed at the time of dosing by a member of the clinical research team who is licensed to administer the study product. Table 11 summarizes the dates of challenge administration by study site. The number of subjects receiving the challenge inoculation will be presented as part of the subject disposition table (Table 9).

7.4. Protocol Deviations

A summary of subject-specific protocol deviations will be presented by the reason for the deviation and the deviation category for all subjects (Table 5). Major deviations that will be reviewed for possible exclusion of immunogenicity results from the PP population include deviations related to eligibility/enrollment, follow-up visit schedule, and receipt of exclusionary vaccines or medications.

All subject-specific protocol deviations and non-subject-specific protocol deviations will be included in Listing 3 and Listing 4, respectively.

8. CLINICAL AND IMMUNOLOGICAL EVALUATIONS

Analysis of clinical endpoints that do not involve immunoassays will be done in the Safety population. Analyses involving immune responses will be conducted using the mITT population, and if at least 10% of analysis data would be excluded by the reasons given in Section 6.3.3, they will be done in the PP population as well, as a sensitivity analysis.

8.1. Primary Analysis

The primary objective will be to evaluate the association of MMID post-challenge and pre-existing virus-specific HAI antibody titers in healthy subjects, which will be assessed in the mITT population via logistic regression:

$\text{logit}(p_i) = \beta_0 + \beta_1 * X_i$, where p_i = the probability of MMID through Day 8 for subject i, and X_i = baseline HAI antibody titer, log-2 transformed and assumed to be continuous.

Wald tests will be performed to evaluate whether MMID is associated with baseline HAI antibody titers. Odds ratio estimates, along with associated 95% confidence intervals and p-values, will be presented in Table 19. Model-based estimates for the probability of MMID based on baseline HAI antibody titer will also be presented in Figure 37. For the primary analysis, the MMID-1 definition will be used (see Section 3.1). Sensitivity analyses using the MMID-2 definition will also be conducted (Table 22).

8.2. Secondary Analyses

8.2.1. Mild-to-Moderate Influenza Disease

8.2.1.1. Baseline Microneutralization Titers

The associations between pre-existing virus-specific MN titers and the incidence of MMID post-challenge will be evaluated among subjects in the mITT population. Similar to that outlined for the primary analysis (Section 8.1), a univariable logistic regression model will be implemented with baseline MN antibody titer as the covariate and MMID-1 as the outcome variable. The estimated odds ratio, along with the associated 95% confidence interval and p-value, will be presented in Table 21. Model-based estimates for the probability of MMID based on baseline MN antibody titers will also be presented in Figure 39. A similar model will be fit using MMID-2 as the outcome, with results from that sensitivity analysis presented in Table 24.

8.2.2. Viral Shedding

Patterns in viral shedding will be described by assessing the frequency, timing, magnitude and duration daily from Days 2 to 8 of the challenge period. Viral shedding status will be summarized for each study day and over the entire challenge period based on detectable shedding results via qualitative and/or quantitative PCR at any point during the challenge period.

Data listings of quantitative RT-PCR and qualitative multiplex virus assay results will be provided in Listing 11, sorted by subject ID and study visit.

8.2.2.1. Frequency and Mean Viral Shedding Over Time

The number and percentage of subjects with detectable virus according to multiplex respiratory virus assay results (qualitative PCR), will be summarized by virus type, viral subtype, and study day (Day 2 to 8 and over the entire challenge period) in Table 33.

The number and percentage of subjects reporting viral shedding by quantitative PCR and/or qualitative PCR will be summarized for each study day (Day 2 to 8) and over the entire challenge period, by baseline HAI and MN seroprotection status in Table 34 and Table 36.

Bar charts for the frequencies of positive viral shedding results for each study day will be presented in Figure 26. Plots for the mean viral concentration by study day (Days 2 to 8) will be displayed among the mITT population stratified by symptom status and baseline seroprotection status for HAI and MN (Figure 27 and Figure 29).

8.2.2.2. Time to Viral Shedding from Challenge

Kaplan-Meier estimates of median time from challenge to viral shedding will be used to account for censored observations due to participants who terminated early or did not have virus detected during the data collection period. Table 37 will present mean and median times, in days, by baseline seroprotection status for each of HAI, NAI, and MN. Provided there is sufficient variability in event times, log-rank test results will be presented for differences in viral shedding times by baseline seroprotection status for HAI and MN (Figure 30 and Figure 32).

8.2.2.3. Duration and Total Days of Viral Shedding

The duration of viral shedding will be evaluated from the day of the initial positive result to the day of the last positive qualitative or quantitative PCR result, regardless of intermittent negative results. Table 41 will summarize the distribution of viral shedding duration, across all subjects in the mITT population who were followed for the entire challenge period and also stratified by baseline seroprotection status for HAI, NAI, and MN, separately.

The total days of viral shedding will be calculated for each subject as the frequency of study days with positive results until discharge and categorized into the following time categories: 0, 1-2, 3-6, 7+, 1+, and 2+. Table 38 and Table 40 will present total days of viral shedding by HAI and MN baseline seroprotection status, respectively.

8.2.2.4. Peak and Total Viral Shedding

The distribution of peak and total viral shedding (log AUC), along with by-group mean differences, will be summarized by baseline seroprotection status for HAI, NAI, and MN and symptom status over the challenge period in Table 42 and Table 43, respectively. Differences in means will be calculated between high and low baseline titer groups along with associated 95% confidence intervals, and groups will be compared using two-sided t-tests, separately for each assay.

Figure 25 present scatterplots of total viral shedding vs. baseline HAI, NAI, and MN antibody titer, with separate panels for each assay, along with estimated Spearman rank-order correlations.

8.2.3. Baseline Microneutralization Titers and Infection Status

Provided there is a sufficient number of cases in both the RT-PCR negative and ever RT-PCR positive asymptomatic categories, the association between baseline MN antibody titer and infection status assessed through Day 8 will be assessed using a baseline-category logit model, where a set of three logit regressions will be simultaneously estimated. Letting p_1 = probability of never positive, p_2 = probability of RT-PCR positive asymptomatic, p_3 = probability RT-PCR positive symptomatic, and making RT-PCR negative the baseline category, the three logit regressions will be:

$\log(p_j / p_1) = \beta_0 + \beta_j * X_1$, for j = 2, 3 or 4, and X_1 = baseline MN antibody titer, log-2 transformed and assumed to be continuous.

Odds ratio estimates, empirical standard errors along with associated 95% confidence intervals, and p-values, will be presented in Table 28.

8.2.4. Immunological Evaluations

Data listings of HAIand MN assay results as reported by the central immunogenicity laboratory will be provided in Listing 8 and Listing 10. Listings will be sorted by subject ID and study visit.

HAI and MN antibody titers at baseline and Days 8, 29, and 61 will be summarized by viral shedding, symptom, and infection statuses, including the number and proportion of subjects achieving seroprotection (titer ≥ 1:40), the number and proportion achieving seroconversion, and GMFRs and GMTs, along with corresponding 95% confidence intervals in Table 16 and Table 18 among the mITT population.

The following boxplots will present longitudinal assessments of antibody GMT responses at baseline, Day 8, 29, and 61 stratified by:

- MMID status in Figure 4 and Figure 6
- Viral shedding status in Figure 7 and Figure 9
- Infection status in Figure 10 and Figure 12

Reverse cumulative distribution (RCD) plots will be presented for baseline and Days 8, 29, and 61, separately for HAI and MN antibody titer, with separate curves by MMID status (ever meeting MMID definition), in Figure 19 and Figure 21.

The correlation between baseline HAI and NAI antibody titers for each study day (baseline and Days 8, 29, and 61) will be presented in a scatterplot (Figure 23), along with estimated pairwise Spearman rank-order correlations and associated p-values.

8.3. Exploratory Analyses

8.3.1. Mild-to-Moderate Influenza Disease

The number and percentage of subjects meeting either MMID-1 or MMID-2 definitions, along with the associated 95% CIs, will be summarized by clinical site and baseline HAI, NAI and MN seroprotection status in Table 30, Table 31, and Table 32.

The percentage of subjects with MMID will be displayed by sex, receipt of prior seasonal vaccination, and age-category in Figure 33, Figure 34, and Figure 35, respectively. Figure 36 will display three-way stratifications of MMID proportions by sex, age-category, and receipt of prior seasonal vaccination.

The associations between MMID and baseline HAI antibody titers, along with other baseline covariates (Z) will be evaluated in the following multivariable logistic model:

$$\text{logit}(p_i) = \beta_0 + \beta_1 * X_{1i} + \ldots + \beta_p * Z_{pi},$$

where p_i = the probability of MMID through Day 8 for subject *I*, X_{1i} = baseline antibody titer, log-2 transformed and assumed to be continuous, and Z_{pi} includes potential confounders: age category (18-29 years, 30-39 years, 40-49 years), sex (male, female), BMI (< 30, ≥30), and prior seasonal vaccination (yes, no), and clinical site.

Subjects with missing baseline covariate values will be excluded from the analysis. Potential multicollinearity among covariates will be assessed and removed from the final model as appropriate. Modeling assumptions will be checked using standard diagnostic methods (e.g., INFLUENCE option in PROC LOGISTIC, or the LACKFIT option for Hosmer-Lemeshow goodness of fit results). Odds ratio estimates, along with associated 95% confidence intervals and p-values, will be presented in Table 25. A similar model will be fit using MMID-2 as the outcome, with results from that sensitivity analysis presented in Table 26.

8.3.2. Total Viral Shedding

To assess the association of total virus shed, log transformed, with baseline HAI seroprotection status, symptom status assessed through Day 8 and other baseline covariates (Z), the following multivariable linear regression model will be fit:

$$\log(AUC_j) = \beta_0 + \beta_1 * X_{1i} + \beta_2 * X_{2i} + \ldots + \beta_p * Z_{pi}, + \varepsilon,$$

where X_{1i} = baseline seroprotection status, with Low (<1:40) as the reference group, X_{2i} = Symptom status through Day 8 (ever symptomatic vs. asymptomatic), and Z_{pi} includes: age category (18-29 years, 30-39 years, 40-49 years), sex (male, female), BMI (< 30, ≥ 30), prior seasonal vaccination (yes, no), and clinical site.

Subjects with missing baseline covariate values will be excluded from the analysis. Modeling assumptions will be checked using standard diagnostic methods (e.g., INFLUENCE option in PROC GLM). Parameter estimates, along with associated 95% confidence intervals and p-values, will be presented in Table 29.

8.3.3. Comparison of Influenza Virus Detection Methods

To assess the agreement between qualitative and quantitative PCR results, summaries by qualitative PCR status across all visits during the inpatient period, with non-missing values for each pair of measurements in the mITT population, will be presented in Table 44.

8.3.4. FLU-PRO and Mild-to-Moderate Influenza Disease Symptoms

8.3.4.1. FLU-PRO Survey Instrument and Validation Diary

Solicited symptoms according to the FLU-PRO Survey Instrument will be summarized by the maximum extent for each study day, the maximum extent post-challenge, and the maximum extent over the entire study in Table 46 and Figure 40, for all subjects in the Safety population. The number and percentage of solicited symptoms by the maximum extent reported anytime post challenge will also be summarized by clinical site in Table 47, and by total days of shedding in Table 48. Listing 12 will include symptom data at any study day for subjects with at least one symptom extent score of 3 or 4.

Summaries of FLU-PRO total symptom severity scores, as well as severity scores by domain, will be presented by study day (Days 2 to 15) in Table 49. Corresponding plots for the change in mean severity scores over time will be presented among all subjects and by viral shedding status in Figure 41 and Figure 42, respectively. Table 50 will present summaries of FLU-PRO total symptom severity scores by viral shedding status, including medians, means, and 95% CIs for the mean, for each study day (Days 2 to 15).

A summary of the additional daily diary items will be presented in Table 58.

8.3.4.2. Mild-to-Moderate Influenza Disease Symptoms

Table 51 will summarize the number and percentage of MMID-related symptoms (see Section 3.1.1) among the mITT population stratified by clinical site. Each symptom will be derived from FLU-PRO responses or clinical and laboratory findings reported within the challenge period (Days 2 to 8) where applicable.

Symptoms will also be analyzed by taking the maximum response over the challenge period and dichotomizing into a binary variable ("Asymptomatic" vs. "Ever Symptomatic"). The number and percentage of subjects reporting each symptom will be presented by viral shedding status in Table 54, along with associated 95% CIs. Differences in proportions between groups will be compared using a Chi-square test, or Fisher's Exact test if the observed rates are low. Table 55, Table 56 and Table 57 will present the number and proportion of subjects, along with associated 95% CI, by viral shedding status for each symptom, stratified by baseline seroprotection status for HAI, NAI and MN assays, respectively.

To explore potential alternate case definitions for symptomatic influenza infection, a predictive model for RT-PCR-positivity will be developed based on maximum FLU-PRO symptom severity scores and the maximum severity of clinical labs used in the MMID definition, via 10-fold cross-validation. For symptoms that are not graded, presence/absence will be used instead. The data will be partitioned into 10 randomly selected subsets, and iteratively (10 times) 9 of them will be used as training data to develop a model, with 1 held out for testing. Within each training dataset, another 10-fold cross-validation step will be used to select the penalization hyperparameter (λ) for a logistic lasso model utilizing the maximum severity across the entire challenge period for each collected symptom. The value of λ minimizing the deviance will be used for shrinkage, and the model will be refit without penalization for unbiased estimates to apply to the testing data [8].

The area under the receiver operating characteristic (ROC) curve (AUC) will be estimated for each of the 10 iterations, and the optimal cutpoint for predicted probabilities within the test data – maximizing the distance to the sensitivity=specificity identity line – will be determined by Youden's J statistic, with sensitivity and specificity estimated at this optimal threshold [9, 10]. The 10 iterations will be summarized via means, minimums, medians, and maximums for estimated coefficients (log odds ratios), standard errors, AUCs, sensitivities, and specificities. The number of iterations with each symptom in the final model will be presented as well, to assess the stability of the final models across training partitions. The summary of the predictive modeling results will be presented in Table 52 and Table 53. The mean estimated coefficients will be used on the entire dataset, with AUC and the optimal cutpoint calculated as before, along with the sensitivity and specificity at the optimal cutpoint, for comparison with the results across 10-fold cross validation iterations. The ROC for the final mean model will be plotted along with the individual curves for each of the 10 iterations of cross-validation, in Figure 43. If computational or technical issues are encountered when utilizing lasso for model selection, other model selection approaches (e.g. forward selection) will be considered as part of this framework.

8.3.5. Additional Immunological Evaluations

Planned exploratory analyses for the neuraminidase inhibition assay (NAI) and antibody secretory cells (ASC; plasmablasts) are described here. If the data are available in time, these results will be included in the primary CSR; otherwise they will be included along with other exploratory assay analyses in future CSR addendum(a). Additional exploratory immunological analyses not described in this document will be described in an addendum to this SAP, and the results will be included in an addendum(a) to the CSR.

8.3.5.1. Neuraminidase Kinetics and Associations

Some NAI results will be included in previously described tables, figures, and listings, if those data are available in time for CSR compilation. Otherwise, those results will be presented along with the NAI analyses described below in an addendum to the CSR.

8.3.5.1.1. Baseline Neuraminidase Titers and Mild-to-Moderate Influenza Disease

The associations between pre-existing virus-specific NAI titers and the incidence of MMID post-challenge will be evaluated among subjects in the mITT population. A univariable logistic regression model will be fit using the MMID-1 definition, and odds ratio estimates, along with the associated 95% confidence interval and p-value, will be presented in Table 20. Model-based estimates for the probability of MMID based on baseline NAI titers will also be presented in Figure 38. A similar model with MMID-2 as the outcome instead will be presented in Table 23.

8.3.5.1.2. Immunological Evaluations

Data listings of NAI assay results as reported by the central immunogenicity laboratory will be provided in Listing 9. Listings will be sorted by subject ID and study visit.

Antibody titers at baseline and Days 8, 29, and 61 will be summarized by viral shedding, symptom, and infection statuses, including the number and proportion of subjects achieving seroprotection (titer ≥ 1:40), the number and proportion achieving seroconversion, and GMFRs and GMTs, along with corresponding 95% confidence intervals in Table 17 among the mITT population.

The following boxplots will present longitudinal assessments of antibody GMT responses at baseline, Day 8, 29, and 61 stratified by:

- MMID status in Figure 5
- Viral shedding status in Figure 8
- Infection status in Figure 11

Reverse cumulative distribution (RCD) plots will be presented for baseline and Days 8, 29, and 61 with separate curves by MMID status (ever meeting MMID definition) in Figure 20.

Scatterplots for pairwise correlations between baseline NAI antibody titers with HAI and MN for each study day (baseline and Days 8, 29, and 61) will be presented in Figure 22 and Figure 24, respectively, along with estimated pairwise Spearman rank-order correlations and associated p-values.

8.3.5.1.3. Viral Shedding

The number and percentage of subjects reporting viral shedding by quantitative PCR and/or qualitative PCR will be summarized for each study day (Day 2 to 8) and over the entire challenge period, by baseline NAI seroprotection status in Table 35. Plots for the mean viral concentration over time will be displayed among the mITT population by symptom status and baseline seroprotection status in Figure 28.

Table 39 will present total days of viral shedding by baseline NAI seroprotection status. Provided there is sufficient variability in event times, the log-rank test result will be presented for differences in time to viral shedding by baseline seroprotection status in Figure 31. Details for derivations and categories are referenced in Section 8.2.2.3.

8.3.5.1.4. Baseline Neuraminidase Titers and Infection Status

The association between baseline NAI antibody titer and infection status assessed through Day 8 will evaluated from a baseline-category logit model, with specifications similar to what is described in Section 8.2.3. Odds ratio estimates, empirical standard errors along with associated 95% confidence intervals, and p-values will be presented in Table 27.

8.3.5.2. Antibody Secreting Cells (aka Plasmablasts)

Antibody secreting cells (ASC) were measured from blood on Days 4, 6, and 8. The anti-HA, anti-NA, and total IgG and IgA ASC will be summarized by infection status in Table 45, including the number of subjects in group with available samples, the mean number of ASC per 10^6 cells and associated 95% CI, and the minimum, median, and maximum. The ratios of influenza-specific (anti-HA and anti-NA) IgA ASC to IgG ASC will be summarized similarly. Box plots of these data will be presented in Figure 13, Figure 14, Figure 15, Figure 16, Figure 17, and Figure 18.

8.3.6. Transcriptomics

To be described in an addendum to this SAP.

9. SAFETY EVALUATION

All summaries and analysis of safety data will be presented for the Safety population. The denominator for the percentages may be based on the number of non-missing observations for an assessment or based on the number of subjects in a population. This will be described for each table.

9.1. Prior and Concurrent Medical Conditions

All current illnesses and past or pre-existing medical conditions will be coded using Medical Dictionary for Regulatory Activities (MedDRA®) version 20.0 or higher.

Summaries of subjects' prior and concurrent medical conditions will be presented across all subjects by MedDRA system organ class in Table 15.

Individual subject listings will be presented for all reported medical history including prior and concurrent medical conditions in Listing 7.

9.2. Adverse Events

An overall summary of all adverse events will be presented in Table 59. A summary of adverse events occurring in at least 5% or more of subjects will be presented in Table 60.

9.2.1. Unsolicited Events

Unsolicited events are any other AEs not considered to be part of MMID that occur following administration of the challenge. These will be coded by Medical Dictionary for Regulatory Activities (MedDRA) for preferred term (PT) and system organ class (SOC).

The following summaries for unsolicited adverse events will be presented among all subjects who received the study challenge by MedDRA SOC and PT:

- Number of subjects reporting at least one event in each MedDRA SOC and PT, cross tabulated by severity and relationship to the influenza challenge in (Table 61);
- Relationship to the study challenge stratified by SOC, PT and by severity (Table 62);
- Bar chart of AE incidence by severity and MedDRA system organ class (Figure 44);
- Bar chart of total frequency of AEs by relationship to study product and MedDRA system organ class (Figure 45);
- Bar chart of AE incidence by relationship to study product and MedDRA system organ class (Figure 46)
- Number and percentage of unsolicited AEs stratified by serious and non-serious AEs for each study day, and at any time post challenge in Table 63.

A listing of unsolicited adverse events and their outcomes are presented in Listing 13.

9.3. Deaths, Serious Adverse Events and other Significant Adverse Events

Serious Adverse Events were documented from the time of challenge through the end of the study. A listing of deaths and other serious adverse events will be presented in Table 64, showing the event description, MedDRA PT and SOC, relevant dates (study challenge and AE), severity, relatedness, and outcome for each event.

9.4. Pregnancies

Pregnancies occurring in study subjects are reported via Advantage electronic data capture system (Advantage eClinical) on the Pregnancy Report form. Efforts are made to follow all pregnancies reported throughout the course this study to pregnancy outcome pending the subject's permission.

In addition, listings of pregnancies and their outcomes are presented in Listing 20, Listing 21, Listing 22, Listing 23, and Listing 24.

9.5. Clinical Laboratory Evaluations

Venous blood samples (approximately 10 mL) are collected from each subject approximately 8 days after the study challenge. These evaluations are performed by the central (clinical) laboratory. Clinical safety laboratory parameters that are evaluated after the study challenge include white blood cells (WBC), hemoglobin (HgB), platelets (PLT), absolute lymphocyte count (ALC), alanine aminotransferase (ALT), and creatinine (Cr). A by-subject listing of parameters for subjects who experience abnormal laboratory results at any time post-challenge administration will be presented in Table 65 and Table 66.

Table 67, Table 68, Table 69, Table 70, Table 71, and Table 72 will summarize the distribution of subjects who experienced a mild, moderate, or severe laboratory result for each day blood is drawn (baseline, Days 2, 4, and 8).

Descriptive statistics including the mean, standard deviation, median, minimum, and maximum by study day, will also be summarized for each parameter in Table 73, Table 74, Table 75, Table 76, Table 77, and Table 78. Graphical presentations will include box plots of the mean change from baseline for Days 2, 4, and 8 in Figure 47 and Figure 48.

Individual laboratory findings for chemistry and hematology will be presented in Listing 14 and Listing 15.

9.6. Vital Signs and Physical Evaluations

Vital sign metrics that are evaluated include pulse, systolic blood pressure, diastolic blood pressure, SpO_2, respiratory rate, and oral temperature. A by-subject listing of parameters for subjects who experience abnormal vital signs results at any time post-challenge administration will be presented in Table 79. The number and percentage of subjects who experienced a mild, moderate, or severe vital sign result for each study day (including baseline, and Days 1 to 8, 15, 29, and 61) will be presented in Table 80, Table 81, Table 82, Table 83, Table 84, Table 85, and Table 86. A listing of vital signs and results will be provided in Listing 16.

Targeted physical examinations are performed for each study day, including early termination or unscheduled visits, based on a subject's medical history. A listing of physical exam findings will be presented in Listing 17. Electrocardiogram (ECG) evaluations are performed on Day 6, or as clinically indicated otherwise. ECG findings will be provided in Listing 18.

9.7. Concomitant Medications

Prior and concomitant medications will be coded to the Anatomical Therapeutic Classification using the WHO Drug Dictionary. The use of concurrent medications during the study will be summarized by ATC1 and ATC2 code, study group, and stratum for the Safety population in Table 87.

A listing of concomitant medications will be presented in Listing 19.

10. REPORTING CONVENTIONS

P-values ≥0.001 and ≤0.999 will be reported to 3 decimal places; p-values less than 0.001 will be reported as "<0.001" The mean, standard deviation, and other statistics will be reported to 1 decimal place greater than the original data. The minimum and maximum will use the same number of decimal places as the original data. Proportions will be presented as 2 decimal places; values greater than zero but <0.01 will be presented as "<0.01". Percentages will be reported to the nearest whole number; values greater than zero but <1% will be presented as "<1"; values greater than 99% but less than 100% will be reported as >99%. Estimated parameters, not on the same scale as raw observations (e.g. regression coefficients) will be reported to 3 significant figures.

11. TECHNICAL DETAILS

SAS version 9.4 and R version 3.6 or above will be used to generate all tables, figures and listings.

12. SUMMARY OF CHANGES IN THE CONDUCT OF THE STUDY OR PLANNED ANALYSES

The analysis populations described in the protocol have been further clarified here. The mITT population will be used for analyses involving immunogenicity, and a baseline sample will be necessary for inclusion in those. While follow-up through the entire challenge period is needed for the calculation of some endpoints, other analyses include by-day presentations where valid data from subjects who withdraw later in the challenge period should not be excluded. Additionally, the Per Protocol population will only be used, as sensitivity analysis to compare to results from mITT, if at least 10% of the analysis data for mITT analyses would be excluded via the reasons given in Section 6.3.

13. REFERENCES

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14. LISTING OF TABLES, FIGURES, AND LISTINGS

Table, figure, and listing shells are presented in Appendices 1, 2, and 3.

APPENDICES

APPENDIX 1. TABLE MOCK-UPS

LIST OF TABLES

9.1 Overall Study Objectives, Design and Plan Description

Table 1: Study Objectives and Outcome Measures

Primary Objective	Primary Outcome Measure
To evaluate the association of symptomatic RT-PCR-positive influenza virus infection post-challenge and pre-existing HAI antibody titers	Percentage of healthy subjects reporting Mild-to-Moderate Influenza Disease (MMID) by baseline A/Bethesda/MM2/H1N1 hemagglutination inhibition (HAI) antibody seroprotection status (titer≥1:40 vs. titer<1:40). MMID is defined as the presence of both of the following reported post-challenge, evaluated through Day 8: Viral shedding detected by any approved positive RT-PCR test from NP swab, **and** Any one or more of the following symptoms or signs or laboratory findings, as related to the study agent; Arthralgia, Chest tightness, Chills, Conjunctivitis, Nasal congestion, Sinus Congestion, Coryza, Decreased appetite, Diarrhea, Dry Cough, Dyspnea/Shortness of Breath, Fatigue/Tiredness, Fever (>38.0°C), Headache, Lymphopenia (<1000 cells/mL), Myalgia, Nausea, Oxygen Saturation Decrease by ≥3% from baseline, Productive Cough, Rhinorrhea, Sore Throat, and Sweats. Association between baseline A/Bethesda/MM2/H1N1 hemagglutination inhibition (HAI) antibody titers and development of MMID post-challenge
Secondary Objectives	**Secondary Outcome Measures**
To describe viral recovery by qualitative and/or quantitative RT-PCR from study subjects at baseline and post-challenge	Frequency of viral shedding in Nasal Lavage Fluid (NLF) or Nasopharyngeal (NP) swab by baseline HAI seroprotection status Duration of viral shedding in Nasal Lavage Fluid (NLF) or Nasopharyngeal (NP) swab by baseline HAI seroprotection status Time until detectable viral shedding in Nasal Lavage Fluid (NLF) or Nasopharyngeal (NP) swab by baseline HAI seroprotection status Total viral shedding in Nasal Lavage Fluid (NLF) or Nasopharyngeal (NP) swab by baseline HAI seroprotection status
To describe serum HAI and MN antibody responses post-challenge in healthy subjects by infection status	HAI,and MN antibody GMTs from serum measured at baseline (Day -2) and at Days, 8, 29, and 61, by infection status post-challenge Percentage of subjects achieving HAI and MN seroconversion (defined as either a pre-challenge titer <1:10 and a post-challenge titer ≥1:40 or a pre-challenge titer ≥1:10 and a minimum four-fold rise in post-challenge antibody titer) from serum measured on Days 8, 29, and 61, by infection status post-challenge
To evaluate the association of asymptomatic RT-PCR-positive influenza virus infection (viral shedding) post-challenge and pre-existing HAI antibody titers	Percentage of subjects with HAI seroprotection from serum measured at baseline (Day -2), by infection status post-challenge
To evaluate the association of symptomatic RT-PCR-negative status post-challenge and pre-existing HAI antibody titers	Percentage of subjects reporting any FLU-PRO symptom during challenge period by viral shedding status and baseline HAI seroprotection status from serum measured at baseline (Day -2)
To determine the frequency of serious adverse events (SAE) post-challenge	Frequency of serious adverse events (SAE) post-challenge through the inpatient stay Frequency of serious adverse events (SAE) post-inpatient discharge through the duration of the study (approximately three months post-challenge)
Exploratory Objectives	**Exploratory Outcome Measures**
To describe serum NAI antibody responses post-challenge in healthy subjects by infection status	NAI antibody GMTs from serum measured at baseline (Day -2) and at Days, 8, 29, and 61, by infection status post-challenge Percentage of subjects with NAI seroprotection from serum measured at baseline (Day -2) and on Days 8, 29, and 61, by infection status post-challenge

Primary Objective	Primary Outcome Measure
	NAI antibody GMFRs from serum measured at Days, 8, 29, and 61, by infection status post-challenge Percentage of subjects achieving NAI seroconversion (defined as either a pre-challenge titer <1:10 and a post-challenge titer ≥1:40 or a pre-challenge titer ≥1:10 and a minimum four-fold rise in post-challenge antibody titer) from serum measured on Days 8, 29, and 61, by infection status post-challenge
To evaluate the association of symptomatic RT-PCR-positive influenza virus infection, asymptomatic RT-PCR-positive influenza virus infection (viral shedding only) and symptomatic RT-PCR-negative illness and pre-existing neuraminidase inhibition (NAI) antibody titers	Baseline A/Bethesda/MM2/H1N1 neuraminidase inhibition (NAI) antibody GMT measured at baseline (Day -2) association with infection status evaluated through Day 8
To evaluate mucosal sIgA immune response, at baseline and post-challenge	Positive, GMT and GMFR responses for A/Bethesda/MM2/H1N1-specific sIgA, nonnormalized and normalized for total IgA content, in NLF at baseline (Day -2), and Days 8, 29, and 61
To evaluate serum and mucosal cytokine and chemokine responses at baseline and post-challenge	Determine A/Bethesda/MM2/H1N1 induced cytokines and chemokines in serum at baseline (Day -2), and Days 2, 4, 6, 8, and 15 Determine A/Bethesda/MM2/H1N1 induced cytokines and chemokines in NLF by study arm at baseline (Day -2), and approximately on Days 2, 4, 6, 8, and 15
To assess the effects of age, sex, baseline antibody titers and prior receipt of seasonal influenza vaccine on symptomatic RT-PCR-positive influenza infection post-challenge	Evaluate the association between baseline A/Bethesda/MM2/H1N1 hemagglutination inhibition (HAI) GMT, age, sex, and receipt of seasonal influenza vaccine in the previous year, and the development of MMID post-challenge through Day 8 by multivariate logistic regression
To compare the clinical features of symptomatic RT-PCR-positive and RT-PCR-negative illness post-challenge	Self-report of clinical symptoms and their severity as measured by component question in FLU-PRO Survey Instrument by post-challenge day and infection status
To explore alternative case definitions for symptomatic RT-PCR-positive influenza virus infection	The combination of symptoms and symptom severity as determined by self-report using the Flu- PRO instrument will be evaluated to develop clinical case definitions which optimize sensitivity, specificity, and both for influenza virus infection among subjects with evidence of RT-PCR-positive influenza virus infection post-challenge
To evaluate the plasmablast response post-challenge in a subset of study subjects	Determine the kinetics of A/Bethesda/MM2/H1N1-specific plasmablasts, their specific-antibody (*e.g.*, anti-H1, -N1 and - H1N1 virus) production and isotype (*e.g.*, IgG, IgA) using an enzyme-linked immunospot (ELISpot) assay in PBMCs on Days 4, 6, and 8 in a subset of subjects
To describe systemic transcriptional responses post-challenge in a subset of study subjects	Determine changes in peripheral blood gene expression profiling based on sequencing on Days 2, 4, 8, and 15 relative to baseline (Days -2 and 1 pre-challenge) in a subset of subjects
To describe mucosal and systemic human immune cells in blood and NLF at baseline and post-challenge in a subset of study subjects	Determine the frequency, duration, timing and phenotypic characteristics of immune cells in the blood of subjects at baseline (Day -2) and at approximately on Days 4, 6, 8, 15, 29, and 61 in a subset of subjects Determine the frequency, duration, timing and phenotypic characteristics of immune cells in the NLF in subjects at baseline (Day -2) and Days 2, 4, 6 and 8 in a subset of subjects
To describe anti-HA-stalk antibody titer at baseline and post-challenge	For HA-stalk specific response, the GMTs and GMFR to group 1 stem domains will be measured at baseline (Day -2), and at approximately on Days 29 and 61
To assess the association of human leukocyte antigen (HLA) class I and II alleles with clinical, immune and viral responses	Determine the frequency of HLA class I and II alleles as measured by genetic testing at Day 61. To assess the association of subject HLA class I and II alleles with development of MMID post-challenge through Day 8, pre-existing immune

Primary Objective	Primary Outcome Measure
	status, and magnitude and breadth of the elicited immune responses post-challenge
To evaluate cellular immune response at baseline and post-challenge	Summarize the data on systemic and mucosal cytokines and chemokines induced by challenge and define their possible role in the process by which influenza exposure leads to infection, disease outcomes, and seroconversion at baseline and daily through Day 8 Frequency of influenza-specific B cell subsets in circulation (PBMC) at baseline (Day -2), and at approximately on Days 6, 8, 15, 29 and 61 Magnitude of influenza-specific T cell responses in circulation (PBMC) to conserved T cell epitopes and inactivated virus at baseline (Day -2), and at approximately on Days 4, 6, 8, 15, 29, and 61

9.5.1 Efficacy/Immunogenicity and Safety Measurements Assessed and Flow Chart

Table 2: Schedule of Study Procedures

Evaluation	Enrollment/Screening Study Visit 00A Day -30 to -3	Inpatient Study Visit 00B Day -2	Inpatient Study Visit 00C Day -1	Inpatient Study Visit 1 Day 1	Inpatient Study Visit 2 Day 2	Inpatient Study Visit 3 Day 3	Inpatient Study Visit 4 Day 4	Inpatient Study Visit 5 Day 5	Inpatient Study Visit 6 Day 6	Inpatient Study Visit 7 Day 7	Inpatient Study Visit 8 Day 8	Follow-up Study Visit 9 Day 15±3	Follow-up Study Visit 10 Day 29±3	Follow-up Study Visit 11 Day 61±5	Final Study Visit 12 Day 91 ± 7	Early Termination Visit	Unscheduled Visit
Visit type[1]	c	i	i	i	i	i	i	i	i	i	i	c	c	c	p	c	c
Screening for	x																
Signed informed	x																
Review/confirm	x[2]	x	x	x													
Review/confirm		x[3]		x													
Inpatient period		x[4]	x	x	x	x	x	x	x	x	x						
Discharge from											x[5]						
Treatment with											x[6]						
Demographics	x																
Medical History	x	x		x													
Height	x[7]																
Weight	x[7]	x[7]															
Concomitant	x	x	x	x	x	x	x	x	x	x	x	x	x	x		x	X
Physical Exam	x																
Targeted physical		x	x	x	x	x	x	x	x	x	x	x	x	x		x[22]	x[22]
Vital signs[9]	x	x	x	x	x	x	x	x	x	x	x	x	x	x		x[22]	x[22]
SpO_2[10]	x	x	x	x	x	x	x	x	x	x	x						
Influenza				x													
FLU-PRO Survey			x[11]	x	x	x	x	x	x	x	x					x[11]	x[11]
Collect FLU-PRO												x					
Adverse event				x	x	x	x	x	x	x	x	x	x			x[22]	x[22]
SAE review				x	x	x	x	x	x	x	x	x	x	x	x	x	X
ECG (12 lead)	x[13]								x[13]								
CXR (PA and	x																
Blood for HIV,	x																
Serum HCG	x																
Urine HCG[14]	x	x															
Urine toxicology[15]	x	x															
Blood for Safety	x																
Blood for Safety					x		x				x					x[22]	
Sera for cytokine		x			x		x		x		x	x	x	x			
Blood for ASC							x		x		x						
Blood for innate and/or T and B		x					x		x		x	x	x	x			
Blood for T and B immunology		x					x		x		x	x	x	x		x[22]	x[22]

Evaluation	Enrollment/Screening Study Visit 00A Day -30 to -3	Inpatient Study Visit 00B Day -2	Inpatient Study Visit 00C Day -1	Inpatient Study Visit 1 Day 1	Inpatient Study Visit 2 Day 2	Inpatient Study Visit 3 Day 3	Inpatient Study Visit 4 Day 4	Inpatient Study Visit 5 Day 5	Inpatient Study Visit 6 Day 6	Inpatient Study Visit 7 Day 7	Inpatient Study Visit 8 Day 8	Follow-up Study Visit 9 Day 15±3	Follow-up Study Visit 10 Day 29±3	Follow-up Study Visit 11 Day 61±5	Final Study Visit 12 Day 91 ± 7	Early Termination Visit	Unscheduled Visit
Blood for		x		x[20]	x		x				x	x				x[22]	x[22]
NP swab for		x	x		x	x	x	x	x	x	x						
NLF for cytokine		x			x		x		x		x	x	x	x		x[22]	x[22]
NLF for other		x			x		x		x		x					x[22]	x[22]
HLA Typing														x		x[22]	x[22]

Evaluation	Enrollment/Screening Study Visit 00A Day -30 to -3	Inpatient Study Visit 00B Day -2	Inpatient Study Visit 00C Day -1	Inpatient Study Visit 1 Day 1	Inpatient Study Visit 2 Day 2	Inpatient Study Visit 3 Day 3	Inpatient Study Visit 4 Day 4	Inpatient Study Visit 5 Day 5	Inpatient Study Visit 6 Day 6	Inpatient Study Visit 7 Day 7	Inpatient Study Visit 8 Day 8	Follow-up Study Visit 9 Day 15±3	Follow-up Study Visit 10 Day 29±3	Follow-up Study Visit 11 Day 61±5	Final Study Visit 12 Day 91 ± 7	Early Termination Visit	Unscheduled Visit

1. Visit type: c=clinic, i=inpatient unit, p=phone call.
2. Eligibility criteria may change from the day of enrollment to Day -3. To avoid admitting subjects with new exclusion criteria into the inpatient unit, the eligibility criteria will be reviewed and confirmed before the inpatient stay.
3. Subjects will have approximately a 48-hour window to decide if they would like to drop out of the study and leave the inpatient unit before challenge virus is administered.
4. Eligible subjects will be admitted to the inpatient unit on Day -2 (which is two days prior to the planned challenge).
5. Subjects will remain in the inpatient unit for a minimum of seven days after the date of challenge. Subjects will leave the unit after they meet the following discharge criteria: two consecutive negative NP swabs (that are performed on Day 7 or thereafter) for influenza A by qualitative RT-PCR performed by the local clinical laboratory, are afebrile, have $SpO_2 \geq 95\%$ on room air, show no moderate or severe influenza signs or symptoms by clinical evaluation, and are clinically and hemodynamically stable for 48 hours. Subjects who do not meet discharge criteria on Day 8 will remain in the inpatient unit until the criteria are met. Study procedures for the inpatient unit will be the same as on Day 8, except for no additional blood draws unless clinically indicated.
6. A single dose of baloxavir marboxil will be offered to all subjects who do not have two consecutive negative NP swabs (on Day 7 or thereafter) for influenza A by qualitative RT-PCR.
7. Height and weight will be measured at initial Screening. Weight will be documented a second time on admission to the inpatient unit. BMI calculation for the purpose of Eligibility will be based on the first measured weight. BMI for the purpose of analyses will be calculated with the second measured weight.
8. Clinical evaluation (including lung, heart, oral/pharyngeal, and neck exams), and symptom evaluation daily while in the inpatient unit.
9. Oral temperature, blood pressure, pulse, and respiratory rate will be assessed three times per day while the subjects are awake, and as clinically indicated.
10. Peripheral mixed venous oxygen saturation (SpO_2) on room air at screening, on admission, and when oral temperature, blood pressure, pulse, and respiratory rate are assessed through the inpatient stay.
11. Training on FLU-PRO Survey Instrument and Validation Diary for evaluation of severity score. Self-assessment using FLU-PRO and diary to be performed in the morning pre-challenge on Day 1, and at approximately 5 PM through day 14 during the inpatient stay and following discharge to home.
12. Unsolicited AEs will be monitored for approximately 30 days post challenge. Review of FLU-PRO for indicators of possible severe influenza associated illness (signs, symptoms, or lab findings) not consistent with MMID.
13. ECG to be performed at screening, on Day 6, and as clinically indicated. VTEU sites will consult a cardiologist if ECG is abnormal to determine clinically necessary work up.
14. Among female subjects of childbearing potential, a repeat urine pregnancy test will be performed locally before CXR performed if > 7 days have passed since the negative serum pregnancy test was drawn.
15. Negative drug urine toxicology result (amphetamines, cocaine, and opiates,) required on screening performed at the Central Lab, and (amphetamines, cocaine, opiates, and cannabinoids) on admission to the challenge unit performed locally.
16. Screening Safety Labs: white blood cells (WBCs), absolute lymphocyte count, hemoglobin, platelets, alanine transaminase (ALT), and creatinine (Cr).
17. Follow-Up Safety Labs: Days 2, 4, and 8, and as clinically indicated: white blood cells (WBCs), absolute lymphocyte count, hemoglobin, platelets, and alanine transaminase (ALT), and creatinine (Cr).
18. Including HAI, MN and NAI antibody titers at admission to the inpatient unit (Day -2) and Visits 8,10 and 11.
19. Will be drawn from at least a subset of participants for local testing
20. Blood draw to occur prior to challenge
21. Multiplex respiratory virus assay to be performed at each site. Positive results prior to influenza virus challenge will result in subject exclusion from the study. No respiratory virus testing will be done on the day of challenge.
22. If indicated.

9.7.1 Sample Size

Table 3: Sample Size/Probability Estimates

	Power (%)			
Prevalence*	OR=0.4	OR=0.5	OR=0.6	OR=0.7
0.5	96.1	82.8	59.1	34.2
0.6	95.5	81.3	57.3	32.9
0.7	94.9	79.6	55.1	31.3
*The probability of a binary outcome such as MMID within the study population				

Table 4: Probability (%) of Observing at Least One Safety Event, Given Varying Underlying Event Probabilities and Group Sizes

True Event Probability	All Subjects (N=80)
10% (very common)	>99.9
1% (common)	55.2
0.1% (uncommon)	7.7
0.01% (rare)	0.8

10.2 Protocol Deviations

Table 5: Summary of Protocol Deviations by Category and Type

[Implementation Note: Include all deviation types collected in the study, even if not listed here; do not include categories with zero deviations reported for All Subjects.]

Category	Deviation Type	All Subjects (N=X)	
		No. of Subjects	No. of Deviations
Eligibility/enrollment	Any type		
	Did not meet inclusion criterion	x	x
	Met exclusion criterion		
	ICF not signed prior to study procedures		
	Other		
Challenge administration schedule	Any type		
	Missed visit/visit not conducted		
	Other		
Follow-up visit schedule	Any type		
	Out of window visit		
	Missed visit/visit not conducted		
	Other		
Protocol procedure/assessment	Any type		
	Incorrect version of ICF signed		
	Blood not collected		
	Urine not collected		
	Other specimen not collected		
	Too few aliquots obtained		
	Specimen result not obtained		
	Required procedure not conducted		
	Required procedure done incorrectly		
	Study product temperature excursion		
	Specimen temperature excursion		
	Other		
Challenge administration	Any type		
	Required procedure done incorrectly		
	Study product temperature excursion		
	Other		
Note: N = Number of subjects enrolled.			

12.2.2 Displays of Adverse Events

Table 6: Severity Grading for Oral Temperature

Systemic (Quantitative)	Mild (Grade 1)	Moderate (Grade 2)	Severe (Grade 3)
Fever* - oral†	38.1°C – 38.4°C 100.6°F – 101.1°F	38.5°C – 38.9°C 101.2°F – 102.0°F	>38.9°C >102.0°F
*A fever can be considered not related to the study product if an alternative etiology can be documented. †Subjects must not eat or drink anything hot or cold, or smoke within 10 minutes prior to taking oral temperature			

Table 7: Pulse, Blood Pressure, Respiratory Rate and Saturation of Peripheral Oxygen (SpO_2) Adverse Event Grading Scale

Physiologic Parameter	Mild (Grade 1)	Moderate (Grade 2)	Severe (Grade 3)
Bradycardia – beats per minute	45 – 46	40 – 44	<40
Tachycardia – beats per minute	100 – 130	131 – 155	>155
Hypotension (systolic) mmHg	80 – 84	75 – 79	<75
Hypotension (diastolic) mmHg	50 – 54	45 – 49	<45
Hypertension (systolic) mmHg	140 – 155	156 – 160	>160
Hypertension (diastolic) mmHg	90 – 100	101 – 110	>110
SpO_2 (%)	92-94	89-91	<89
RR (increase) (bpm)	21-24	25-29	>30
RR (decrease) (bpm)	9-11	6-8	<6
Note: RR = Respiratory Rate; SpO_2 = Saturation of Peripheral Oxygen			

12.4.1 Individual Laboratory Measurements and Abnormal Laboratory Values

Table 8: Clinical Safety and Laboratory Adverse Event Grading Scale

Hematology	**Mild (Grade 1)**	**Moderate (Grade 2)**	**Severe (Grade 3)**
WBC x10^3/µL (Decrease)	2.50 – 3.69	1.50 – 2.49	<1.50
WBC x10^3/µL (Increase)	11.01 – 15.09	15.10 – 20.00	>20.00
Absolute Lymphocyte count x 10^3/µL (Decrease)	0.75 - 0.99	0.50 - 0.74	< 0.50
Hgb g/dL (Decrease) (Female)	10.1 – 10.9	8.5 – 10.0	<8.5
Hgb g/dL (Decrease) (Male)	11.0 – 12.4	9.5 – 10.9	<9.5
Platelets cell x 10^3/µL (Decrease) EDTA	140 – 162	100 – 139	<100
Platelets x 10^3/µL (Increase) EDTA	376 – 550	551– 750	>750
Platelets K/cu mm (Decrease) Citrate	115-124	100-114	<100
Platelets K/cu mm (Increase) Citrate	376 - 550	551-750	>750
Chemistry	**Mild (Grade 1)**	**Moderate (Grade 2)**	**Severe (Grade 3)**
ALT IU/L (Increase) (Female)	44 – 100	101 – 200	>200
ALT IU/L (Increase) (Male)	61- 138	139 – 275	>275
Creatinine mg/dL (Increase)	1.5 – 1.8	1.9 – 2.2	> 2.2

14.1 Description of Study Subjects

14.1.1 Disposition of Subjects

Table 9: Subject Disposition

Subject Disposition	All Subjects (N=X)	
	n	%
Screened	x	N/A
Enrolled/Received Challenge	x	100
Left Isolation Unit Prior to Fulfilling Discharge Criteria		
Completed Challenge Period[a]		
Early Termination[b]		
Completed Study		

Note: N = Number of subjects enrolled; N/A = not applicable.

[a] Challenge period duration defined from Days 2 to 8.

[b] For additional details about subjects who terminated early, refer to "Subjects Discharged Early or Terminated Early" listing.

Table 10: Analysis Populations

Analysis Populations	Reason Subjects Excluded[a]	All Subjects (N=X)	
		n	%
Safety Population	Any Reason	x	xx
	No Study Challenge Received		
Modified Intent-to-Treat, Challenge Period	Any Reason		
	No Study Challenge Received		
	Baseline Result Not Reported by Lab		
	Early Termination		
Per Protocol, Challenge Period	Any Reason		
	No Study Challenge Received		
	Baseline Result Not Reported by Lab		
	Ineligible Baseline Results		
	Early Termination		
Per Protocol, Day 15	Any Reason		
	No Study Challenge Received		
	Baseline Result Not Reported by Lab		
	Ineligible at Baseline		
	Early Termination		
	Receipt of Non-Study Vaccination		
	Receipt of Immunosuppressive Medication		
	Study Day 15 Result Not Reported by Lab		
	Study Day 15 Visit Out of Window		
Per Protocol, Day 29	Any Reason		
	No Study Challenge Received		
	Baseline Result Not Reported by Lab		
	Ineligible at Baseline		
	Early Termination		
	Receipt of Non-Study Vaccination		
	Receipt of Immunosuppressive Medication		
	Day 29 Result Not Reported by Lab		
	Day 29 Visit Out of Window		
Per Protocol, Day 61	Any Reason		
	No Study Challenge Received		
	Baseline Result Not Reported by Lab		
	Ineligible at Baseline		
	Early Termination		

Analysis Populations	Reason Subjects Excluded[a]	All Subjects (N=X)	
		n	%
	Receipt of Non-Study Vaccination		
	Receipt of Immunosuppressive Medication		
	Day 61 Result Not Reported by Lab		
	Day 61 Visit Out of Window		

Note: N = Number of subjects enrolled.
[a] More than one reason for exclusion may apply for each subject and visit.

Table 11: Dates of Challenge Administration by Site

Dates of Dosing	Saint Louis University (N=X)	Cincinnati Hospital (N=X)	University of Maryland (N=X)	Duke University (N=X)	All Subjects (N=X)
Total (Entire period of enrollment)					
DDMMMYYYY-DDMMMYYYY [categorize based on length of enrollment period]	x	x	x	x	
Note: N = Number of subjects challenged.					

Table 12: Ineligibility Summary of Screen Failures

Inclusion/Exclusion Category	Inclusion/Exclusion Criterion	n[a]	%[b]
Inclusion and Exclusion	Number of subjects failing any eligibility criterion	x	xx
Inclusion	Any inclusion criterion	x	xx
	[inclusion criterion 1]	x	xx
	[inclusion criterion 2]	x	xx
	[inclusion criterion 3]	x	xx
Exclusion	Any exclusion criterion	x	xx
	[exclusion criterion 1]	x	xx
	[exclusion criterion 2]	x	xx
	[exclusion criterion 3]	x	xx
Eligible but Not Enrolled		x	xx

[a] More than one criterion may be marked per subject.

[b] Denominator for percentages is the total number of screen failures.

14.1.2 Demographic Data

Table 13: Summary of Categorical Demographic and Baseline Characteristics by Site

Variable	Characteristic	Saint Louis University (N=X)		Cincinnati Hospital (N=X)		University of Maryland (N=X)		Duke University (N=X)		All Subjects (N=X)	
		n	%	n	%	n	%	n	%	n	%
Sex	Male	x	xx	x	xx	x	xx	x	xx	x	xx
	Female										
Age	18-29										
	30-39										
	40-49										
BMI	<30										
	≥30										
Ethnicity	Not Hispanic or Latino										
	Hispanic or Latino										
	Not Reported										
	Unknown										
Race	American Indian or Alaska Native										
	Asian										
	Native Hawaiian or Other Pacific Islander										
	Black or African American										
	White										
	Multi-Racial										
	Unknown										
Receipt of Prior Seasonal Influenza Vaccination (2018-2019)	Yes										
	No										
Receipt of Prior Non-Seasonal Influenza Vaccination	Yes										
	No										
Baseline Seroprotection (HAI)	Low < 1:40										
	High ≥ 1:40										

Variable	Characteristic	Saint Louis University (N=X)		Cincinnati Hospital (N=X)		University of Maryland (N=X)		Duke University (N=X)		All Subjects (N=X)	
		n	%	n	%	n	%	n	%	n	%
Baseline Seroprotection (NAI)	Low < 1:40										
	High ≥ 1:40										
Baseline Seroprotection (MN)	Low < 1:40										
	High ≥ 1:40										

Note: N = Number of subjects enrolled.

HAI = Hemagglutination Inhibition; NAI = Neuraminidase Inhibition; MN = Microneutralization.

Table 14: Summary of Continuous Baseline Demographic and Clinical Characteristics by Site

Variable	Statistic	Saint Louis University (N=X)	Cincinnati Hospital (N=X)	University of Maryland (N=X)	Duke University (N=X)	All Subjects (N=X)
Age (years)	Mean	x.x	x.x	x.x	x.x	x.x
	Standard Deviation	x.x	x.x	x.x	x.x	x.x
	Median	x.x	x.x	x.x	x.x	x.x
	Minimum	x	x	x	x	x
	Maximum	x	x	x	x	x
BMI	Mean	x.x	x.x	x.x	x.x	x.x
	Standard Deviation	x.x	x.x	x.x	x.x	x.x
	Median	x.x	x.x	x.x	x.x	x.x
	Minimum	x	x	x	x	x
	Maximum	x	x	x	x	x
HAI titer	GMT					
	Minimum					
	Median					
	Maximum					
NAI titer	GMT					
	Minimum					
	Median					
	Maximum					
MN titer	GMT					
	Minimum					
	Median					
	Maximum					

Note: N = Number of subjects enrolled.
HAI = Hemagglutination Inhibition; NAI = Neuraminidase Inhibition; MN = Microneutralization.

14.1.3 Prior and Concurrent Medical Conditions

Table 15: Summary of Subjects with Pre-Existing Medical Conditions by MedDRA System Organ Class

MedDRA System Organ Class	All Subjects (N=X)	
	n	%
Any SOC	x	xx
[SOC 1]		
[SOC 2]		

Note: N = Number of subjects in the Safety population; n = Number of subjects reporting medical history within the specified SOC. A subject is only counted once per SOC.

14.1.4 Clinical and Immunogenicity Data

Table 16: Hemagglutination Inhibition Antibody Against A/Bethesda/MM2/H1N1 Geometric Mean Titer, Geometric Mean Fold Rise, Seroprotection, and Seroconversion Results by Study Day, by Infection Status, Symptom Status, Viral Shedding, and Mild-to-Moderate Influenza Disease Status, Modified Intent-to-Treat Population

		Infection Status[a]			Viral Shedding[a]			Symptom Status[a]	
Study Day	Statistic	RT-PCR Positive Symptomatic (MMID-1)	At Least Twice RT-PCR Positive Symptomatic (MMID-2)	RT-PCR Positive Asymptomatic	RT-PCR Negative	RT-PCR Positive (One or More)	RT-PCR Positive (Two or More)	Symptomatic	Asymptomatic
Baseline	n	x	x	x	x	x	x	x	x
	GMT (95% CI)[b]	x.x (x.x, x.x)	x.x (x.x, x.x)	x.x (x.x, x.x)	x.x (x.x, x.x)	x.x (x.x, x.x)	x.x (x.x, x.x)	x.x (x.x, x.x)	x.x (x.x, x.x)
	Seroprotection - % (95% CI)[c]	x.x (x.x, x.x)	x.x (x.x, x.x)	x.x (x.x, x.x)	x.x (x.x, x.x)	x.x (x.x, x.x)	x.x (x.x, x.x)	x.x (x.x, x.x)	x.x (x.x, x.x)
Day 8	n	x	x	x	x	x	x	x	x
	GMT (95% CI)	x.x (x.x, x.x)	x.x (x.x, x.x)	x.x (x.x, x.x)	x.x (x.x, x.x)	x.x (x.x, x.x)	x.x (x.x, x.x)	x.x (x.x, x.x)	x.x (x.x, x.x)
	GMFR (95% CI)[b]	x.x (x.x, x.x)	x.x (x.x, x.x)	x.x (x.x, x.x)	x.x (x.x, x.x)	x.x (x.x, x.x)	x.x (x.x, x.x)	x.x (x.x, x.x)	x.x (x.x, x.x)
	Seroprotection - % (95% CI)	x.x (x.x, x.x)	x.x (x.x, x.x)	x.x (x.x, x.x)	x.x (x.x, x.x)	x.x (x.x, x.x)	x.x (x.x, x.x)	x.x (x.x, x.x)	x.x (x.x, x.x)
	Seroconversion - % (95% CI)[c]	x.x (x.x, x.x)	x.x (x.x, x.x)	x.x (x.x, x.x)	x.x (x.x, x.x)	x.x (x.x, x.x)	x.x (x.x, x.x)	x.x (x.x, x.x)	x.x (x.x, x.x)
Day 29	n								
	GMT (95% CI)								
	GMFR (95% CI)								
	Seroprotection - % (95% CI)								
	Seroconversion - % (95% CI)								
Day 61	n								
	GMT (95% CI)								
	GMFR (95% CI)								
	Seroprotection - % (95% CI)								
	Seroconversion % (95% CI)								

Note: n = Number of subjects in the Modified Intent-to-Treat population with available results; GMT = Geometric Mean Titer; GMFR = Geometric Mean Fold Rise.
[a] If ever reported during the challenge period (Days 2 to 8).
[b] Confidence Interval calculated based on the student's t-distribution.
[c] Exact binomial confidence interval calculated using the Clopper-Pearson methodology.

Tables with similar format:

Table 17: Neuraminidase Inhibition Antibody Against A/Bethesda/MM2/H1N1 Geometric Mean Titer, Geometric Mean Fold Rise, Seroprotection, and Seroconversion Results by Study Day and by Infection Status, Symptom Status and Viral Shedding, Modified Intent-to-Treat Population

Table 18: Microneutralization Antibody Against A/Bethesda/MM2/H1N1 Geometric Mean Titer, Geometric Mean Fold Rise, and Seroconversion Results by Study Day and by Infection Status, Symptom Status and Viral Shedding, Modified Intent-to-Treat Population

Table 19: Univariable Logistic Regression Model to Evaluate the Relationship of Baseline Hemagglutination Inhibition Antibody Against A/Bethesda/MM2/H1N1 with Mild-to-Moderate Influenza Disease, Modified Intent-to-Treat Population

Model Parameter	Parameter Estimate	SE	p-value	Odds Ratio	95% CI
Intercept	x.xx	x.xx	x.xxx	-	-
Baseline log-2 Titer	x.xx	x.xx	x.xxx	x.xx	(x.xx, x.xx)
Note: XX subjects in the Modified Intent-to-Treat population included in the model. MMID-1 definition used: one or more PCR+ tests and meeting the symptom threshold at any point during the challenge period.					

Tables with similar format:

Table 20: Univariable Logistic Regression Model to Evaluate the Relationship of Baseline Neuraminidase Inhibition Antibody Against A/Bethesda/MM2/H1N1 with Mild-to-Moderate Influenza Disease, Modified Intent-to-Treat Population

Table 21: Univariable Logistic Regression Model to Evaluate the Relationship of Baseline Microneutralization Antibody Against A/Bethesda/MM2/H1N1 with Mild-to-Moderate Influenza Disease, Modified Intent-to-Treat Population

Table 22: Univariable Logistic Regression Model to Evaluate the Relationship of Baseline Hemagglutination Inhibition Antibody Against A/Bethesda/MM2/H1N1 with Alternatively Defined Mild-to-Moderate Influenza Disease, Modified Intent-to-Treat Population

Table 23: Univariable Logistic Regression Model to Evaluate the Relationship of Baseline Neuraminidase Inhibition Antibody Against A/Bethesda/MM2/H1N1 with Alternatively Defined Mild-to-Moderate Influenza Disease, Modified Intent-to-Treat Population

Table 24: Univariable Logistic Regression Model to Evaluate the Relationship of Baseline Microneutralization Antibody Against A/Bethesda/MM2/H1N1 with Alternatively Defined Mild-to-Moderate Influenza Disease, Modified Intent-to-Treat Population

[Implementation note: Tables 22-24 will be the same as Tables 20-22, above, but with MMID-2 definition used. Footnotes should be updated to reflect this.]

Table 25: Multivariable Logistic Regression Model to Evaluate the Relationship of Baseline Hemagglutination Inhibition Antibody Against A/Bethesda/MM2/H1N1 with Mild-to-Moderate Influenza Disease, Controlling for Age, Sex, Body Mass Index, Prior Vaccinations, and Clinical Site, Modified Intent-to-Treat Population

Model Parameter	Parameter Level	Parameter Estimate	SE	p-value	Odds Ratio	95% CI
Intercept	N/A	x.xx	x.xx	x.xxx	-	-
Baseline log-2 Titer	N/A	x.xx	x.xx	x.xxx	x.xx	(x.xx, x.xx)
Age category	18-29 (reference)	-	-	-	-	-
	30-39	x.xx	x.xx	x.xxx	x.xx	(x.xx, x.xx)
	40-49	x.xx	x.xx	x.xxx	x.xx	(x.xx, x.xx)
Sex	Male (reference)	-	-	-	-	-
	Female	x.xx	x.xx	x.xxx	x.xx	(x.xx, x.xx)
BMI	< 30 (reference)	-	-	-	-	-
	≥ 30	x.xx	x.xx	x.xxx	x.xx	(x.xx, x.xx)
Receipt of Prior Seasonal Influenza Vaccination (2018-2019)	No (reference)	-	-	-	-	-
	Yes	x.xx	x.xx	x.xxx	x.xx	(x.xx, x.xx)
Clinical Site	University of Maryland (reference)	-	-	-	-	-
	Saint Louis University	x.xx	x.xx	x.xxx	x.xx	(x.xx, x.xx)
	Cincinnati Hospital	x.xx	x.xx	x.xxx	x.xx	(x.xx, x.xx)
	Duke University	x.xx	x.xx	x.xxx	x.xx	(x.xx, x.xx)

Note: XX subjects in the Modified Intent-to-Treat population included in the model. MMID-1 definition used: one or more PCR+ tests and meeting the symptom threshold at any point during the challenge period.

Tables with similar format:

Table 26: Multivariable Logisitc Regression Model to Evaluate the Relationship of Baseline Hemagglutination Inhibition Antibody Against A/Bethesda/MM2/H1N1 with Alternatively Defined Mild-to-Moderate Influenza Disease, Controlling for Age, Sex, Body Mass Index, Prior Vaccinations, and Clinical Site, Modified Intent-to-Treat Population

[Implementation note: Table 26 will be the same as Table 25, above, but with MMID-2 definition used. Footnote should be updated to reflect this.]

Table 27: Baseline Category Logit Model to Evaluate the Relationship of Baseline Microneutralization Antibody Against A/Bethesda/MM2/H1N1 with Infection Status, Modified Intent-to-Treat Population

	RT-PCR+ Symptomatic vs. RT-PCR Negative					RT-PCR+ Asymptomatic vs. RT-PCR Negative				
Model Parameter	**Parameter Estimate**	**SE**	**p-value**	**Odds Ratio**	**95% CI**	**Parameter Estimate**	**SE**	**p-value**	**Odds Ratio**	**95% CI**
Intercept	x.xx	x.xx	x.xxx	-	-	x.xx	x.xx	x.xxx	-	-
Baseline log-2 Titer	x.xx	x.xx	x.xxx	x.xx	(x.xx, x.xx)	x.xx	x.xx	x.xxx	x.xx	(x.xx, x.xx)
Note: XX subjects in the Modified Intent-to-Treat population were included in the model.										

Tables with similar format:

Table 28: Baseline Category Logit Model to Evaluate the Relationship of Baseline Neuraminidase Inhibition Antibody Against A/Bethesda/MM2/H1N1 with Infection Status, Modified Intent-to-Treat Population

Table 29: Multivariable Linear Regression Model to Evaluate the Relationship of Total Viral Shedding with Baseline Hemagglutination Inhibition Seroprotection Status Against A/Bethesda/MM2/H1N1 and Symptom Status Controlling for Potential Confounders, Modified Intent-to-Treat Population

Model Parameter	Parameter Level	Parameter Estimate	SE	p-value	95% CI
Intercept	N/A	xxx.x	xxx.x	x.xxx	-
Baseline Seroprotection Status	Low < 1:40 (reference)	-	-	-	-
	High ≥ 1:40	xxx.x	xxx.x	xxx.x	(xx, xx)
Symptom Status	Asymptomatic (reference)	-	-	-	-
	Symptomatic	xxx.x	xxx.x	xxx.x	(xx, xx)
Age Category	18-29 (reference)	-	-	-	-
	30-39	xxx.x	xxx.x	xxx.x	(xx, xx)
	40-49	xxx.x	xxx.x	xxx.x	(xx, xx)
Sex	Male (reference)	-	-	-	-
	Female	xxx.x	xxx.x	xxx.x	(xx, xx)
BMI	< 30 (reference)	-	-	-	-
	≥ 30	xxx.x	xxx.x	xxx.x	(xx, xx)
Receipt of Prior Seasonal Influenza Vaccination (2018-2019)	No (reference)	-	-	-	-
	Yes	xxx.x	xxx.x	xxx.x	(xx, xx)
Clinical Site	University of Maryland (reference)	-	-	-	-
	Saint Louis University	xxx.x	xxx.x	xxx.x	(xx, xx)
	Cincinnati Hospital	xxx.x	xxx.x	xxx.x	(xx, xx)
	Duke University	xxx.x	xxx.x	xxx.x	(xx, xx)
Note: XX subjects in the Modified Intent-to-Treat population with missing covariate data were excluded from this analysis.					

Table 30: Mild-to-Moderate Influenza Disease by Clinical Site and Baseline Hemagglutination Inhibition Seroprotection Status Against A/Bethesda/MM2/H1N1, Safety Population

Baseline HAI Titer	Mild-to-Moderate Disease	Saint Louis University (N=X)			Cincinnati Hospital (N=X)			University of Maryland (N=X)			Duke University (N=X)			All Subjects (N=X)		
		n	%	95% CI	n	%	95% CI	n	%	95% CI	n	%	95% CI	n	%	95% CI
High ≥ 1:40	MMID-1	x	x.x	x.x, x.x	x	x.x	x.x, x.x	x	x.x	x.x, x.x	x	x.x	x.x, x.x	x	x.x	x.x, x.x
	No MMID-1	x	x.x	x.x, x.x	x	x.x	x.x, x.x	x	x.x	x.x, x.x	x	x.x	x.x, x.x	x	x.x	x.x, x.x
	MMID-2	x	x.x	x.x, x.x	x	x.x	x.x, x.x	x	x.x	x.x, x.x	x	x.x	x.x, x.x	x	x.x	x.x, x.x
	No MMID-2	x	x.x	x.x, x.x	x	x.x	x.x, x.x	x	x.x	x.x, x.x	x	x.x	x.x, x.x	x	x.x	x.x, x.x
Low < 1:40	MMID-1															
	No MMID-1															
	MMID-2															
	No MMID-2															

Note: N = Number of subjects in the Safety population.

[a] Results reported for each subject at any timepoint within the challenge period (Days 2 to 8).

Tables with similar format:

Table 31: Mild-to-Moderate Influenza Disease by Clinical Site and Baseline Neuraminidase Inhibition Seroprotection Status Against A/Bethesda/MM2/H1N1, Safety Population

Table 32: Mild-to-Moderate Influenza Disease by Clinical Site and Baseline Microneutralization Inhibition Seroprotection Status Against A/Bethesda/MM2/H1N1, Safety Population

Table 33: Multiplex Respiratory Virus Assay Results by Virus, Subtype, and Study Day, Safety Population

Virus	Subtype[a]	Day 2 (N=X)		Day 3 (N=X)		Day 4 (N=X)		Day 5 (N=X)		Day 6 (N=X)		Day 7 (N=X)		Day 8 (N=X)		Days 9+ (N=X)		Anytime Post-Challenge (N=X)	
		n	%	n	%	n	%	n	%	n	%	n	%	n	%	n	%	n	%
Influenza	Any Subtype	x	x.x	x	x.x	x	x.x	x	x.x	x	x.x	x	x.x	x	x.x	x	x.x	x	x.x
	Influenza A, Influenza A 2009 H1N1 or Influenza H1[b]																		
	Influenza A																		
	Influenza A 2009 H1N1																		
	Influenza A H1																		
	Influenza A H3																		
	Influenza B																		
Other	Any Other Virus																		
	Coronavirus NL68																		
	Coronavirus OC43																		
	Human Rhinovirus/Enterovirus																		
	Legionella Pneumophila																		
	Mycoplasma Pneumoniae																		
	Parainfluenza Virus 1																		
	Parainfluenza Virus 2																		
	Parainfluenza Virus 3																		
	Parainfluenza Virus 4																		
	Respiratory Syncytial Virus (unspecified)																		
	Respiratory Syncytial Virus A																		
	Respiratory Syncytial Virus B																		
	Rhinovirus/Enterovirus																		

Note: N = Number of subjects in the Safety population with available data; n = Number of subjects in the Safety population reporting positive results.

[a] More than one viral result may apply for each subject and study day.

[b] Results used as part of the viral shedding definition.

Table 34: Viral Shedding by Baseline Hemagglutination Inhibition Seroprotection Status Against A/Bethesda/MM2/H1N1 and Study Day, Modified Intent-to-Treat Population

Study Day	Baseline Seroprotection Status	N	RT-PCR Positive		RT-PCR Negative	
			n	%	n	%
Day 2	Low < 1:40	X	x	x.x	x	x.x
	High ≥ 1:40	X	x	x.x	x	x.x
Day 3	Low < 1:40					
	High ≥ 1:40					
Day 4	Low < 1:40					
	High ≥ 1:40					
Day 5	Low < 1:40					
	High ≥ 1:40					
Day 6	Low < 1:40					
	High ≥ 1:40					
Day 7	Low < 1:40					
	High ≥ 1:40					
Day 8	Low < 1:40					
	High ≥ 1:40					
Anytime Post-Challenge[a]	Low < 1:40					
	High ≥ 1:40					

Note: N = Number of subjects in the Modified Intent-to-Treat population with available results.

[a] The number of subjects with viral shedding reported at any time point within the challenge period (Days 2 to 8).

Tables with similar format:

Table 35: Viral Shedding by Baseline Neuraminidase Inhibition Seroprotection Status Against A/Bethesda/MM2/H1N1 and Study Day, Modified Intent-to-Treat Population

Table 36: Viral Shedding by Baseline Microneutralization Seroprotection Status Against A/Bethesda/MM2/H1N1 and Study Day, Modified Intent-to-Treat Population

Table 37: Time to Viral Shedding from Challenge by Assay Type and Baseline Seroprotection Status Against A/Bethesda/MM2/H1N1, Modified Intent-to-Treat Population

Assay Type	Baseline Seroprotection Status	N	Median[a] (95% CI)	n	Mean[b]
Hemagglutination Inhibition	Low < 1:40	X	x.x (x.x, x.x)	X	x.x
	High ≥ 1:40				
Neuraminidase Inhibition	Low < 1:40				
	High ≥ 1:40				
Microneutralization	Low < 1:40				
	High ≥ 1:40				
All Subjects	-				

Note: N = Number of subjects in the Modified Intent-to-Treat population; n = Number of subjects reporting viral shedding at any point during the challenge period; NC = Not Calculable.

[a] Kaplan-Meier estimate among all challenged subjects, adjusted for censoring.

[b] Mean time to viral shedding for subjects with viral shedding reported at any point during the challenge period.

Table 38: Total Days of Viral Shedding by Baseline Hemagglutination Inhibition Seroprotection Status Against A/Bethesda/MM2/H1N1, Modified Intent-to-Treat Population

Number of Days	Low < 1:40 (N=X)		High ≥ 1:40 (N=X)		All Subjects (N=X)	
	n	%	n	%	n	%
0	xx	xx.x	xx	xx.x	xx	xx.x
1						
2						
3 - 6						
7+						
1+						
2+						
Note: N = Number of subjects in the Modified Intent-to-Treat population followed for the entire challenge period						

Tables with similar format:

Table 39: Total Days of Viral Shedding by Baseline Neuraminidase Inhibition Seroprotection Status Against A/Bethesda/MM2/H1N1, Modified Intent-to-Treat Population

Table 40: Total Days of Viral Shedding by Baseline Microneutralization Seroprotection Status Against A/Bethesda/MM2/H1N1, Modified Intent-to-Treat Population

Table 41: Summary of the Duration of Viral Shedding by Baseline Seroprotection Status Against A/Bethesda/MM2/H1N1, Modified Intent-to-Treat Population

Assay Type	Baseline Seroprotection Status	N	n	Mean (95% CI)	Median	Min, Max
Hemagglutination Inhibition	Low < 1:40	x	x	x.x (x.x, x.x)	x.x	x.x, x.x
	High ≥ 1:40					
Neuraminidase Inhibition	Low < 1:40					
	High ≥ 1:40					
Microneutralization	Low < 1:40					
	High ≥ 1:40					
All Subjects	-					

Note: N = Number of subjects in the Modified Intent-to-Treat population followed for the entire challenge period; n = Number of subjects with detected virus at any time during challenge period (Days 2 to 8).

Table 42: Summaries of Peak and Total Viral Shedding by Assay Type and Baseline Seroprotection Status Against A/Bethesda/MM2/H1N1, Modified Intent-to-Treat Population

Parameter	Baseline Seroprotection Status	N	Mean	Median	Min, Max	Difference in mean (95% CI)
Hemagglutination Inhibition						
Peak	Low < 1:40	X	x.x	x.x	x.x, x.x	-
	High ≥ 1:40	X	x.x	x.x	x.x, x.x	x.x (x.x, x.x)
Log AUC	Low < 1:40					
	High ≥ 1:40					
Neuraminidase Inhibition						
Peak	Low < 1:40					
	High ≥ 1:40					
Log AUC	Low < 1:40					
	High ≥ 1:40					
Microneutralization						
Peak	Low < 1:40					
	High ≥ 1:40					
Log AUC	Low < 1:40					
	High ≥ 1:40					

Note: N = Number of subjects in the Modified Intent-to-Treat population who were followed for the entire challenge period.

Table 43: Summaries of Peak and Total Viral Shedding by Symptom Status through Day 8, Modified Intent-to-Treat Population

Parameter	Symptom Status	N	Mean	Median	Min, Max	Difference in mean (95% CI)
Peak	Asymptomatic	X	x.x	x.x	x.x, x.x	-
	Symptomatic	X	x.x	x.x	x.x, x.x	x.x (x.x, x.x)
	All Subjects					
Log AUC	Asymptomatic					
	Symptomatic					
	All Subjects					

Note: N = Number of subjects in the Modified Intent-to-Treat population who were followed for the entire challenge period.

Table 44: Comparison Between Qualitative Multiplex Respiratory Virus Assay and Quantitative PCR for Influenza, Modified Intent-to-Treat Population

	Qualitative PCR Positive	Qualitative PCR Negative
Quantitative PCR (copies/mL)		
n	X	X
Minimum	xx	xx
Median	xx.x	xx.x
Mean	xx.x	xx.x
Maximum	xx	xx
# Below LOD (%)	xx	xx
Note: n = Number of samples across all subjects in the Modified Intent-to-Treat population and all study days during the inpatient period; column percentages presented.		

Table 45: Summary of Antibody Secreting Cells (Plasmablasts) by Study Day and Infection Status

Study Day	Measure	Statistic	RT-PCR Positive Symptomatic[a]	RT-PCR Positive Asymptomatic[a]	RT-PCR Negative[a]	All Subjects
Day 4	Anti-HA IgG ASC (per 10^6 cells)	N	xx	xx	xx	xx
		Mean (95% CI)	xx.x (xx.x, xx.x)	xx.x (xx.x, xx.x)	xx.x (xx.x, xx.x)	xx.x (xx.x, xx.x)
		Minimum, Median, Maximum	xx.x, xx.x, xx.x	xx.x, xx.x, xx.x	xx.x, xx.x, xx.x	xx.x, xx.x, xx.x
	Anti-NA IgG ASC (per 10^6 cells)	N	xx	xx	xx	xx
		Mean (95% CI)	xx.x (xx.x, xx.x)	xx.x (xx.x, xx.x)	xx.x (xx.x, xx.x)	xx.x (xx.x, xx.x)
		Minimum, Median, Maximum	xx.x, xx.x, xx.x	xx.x, xx.x, xx.x	xx.x, xx.x, xx.x	xx.x, xx.x, xx.x
	Total IgG ASC (per 10^6 cells)	N	xx	xx	xx	xx
		Mean (95% CI)	xx.x (xx.x, xx.x)	xx.x (xx.x, xx.x)	xx.x (xx.x, xx.x)	xx.x (xx.x, xx.x)
		Minimum, Median, Maximum	xx.x, xx.x, xx.x	xx.x, xx.x, xx.x	xx.x, xx.x, xx.x	xx.x, xx.x, xx.x
	Anti-HA IgA ASC (per 10^6 PBMC)	N	xx	xx	xx	xx
		Mean (95% CI)	xx.x (xx.x, xx.x)	xx.x (xx.x, xx.x)	xx.x (xx.x, xx.x)	xx.x (xx.x, xx.x)
		Minimum, Median, Maximum	xx.x, xx.x, xx.x	xx.x, xx.x, xx.x	xx.x, xx.x, xx.x	xx.x, xx.x, xx.x
	Anti-NA IgA ASC (per 10^6 PBMC)	N	xx	xx	xx	xx
		Mean (95% CI)	xx.x (xx.x, xx.x)	xx.x (xx.x, xx.x)	xx.x (xx.x, xx.x)	xx.x (xx.x, xx.x)
		Minimum, Median, Maximum	xx.x, xx.x, xx.x	xx.x, xx.x, xx.x	xx.x, xx.x, xx.x	xx.x, xx.x, xx.x
	Total IgA ASC (per 10^6 PBMC)	N	xx	xx	xx	xx
		Mean (95% CI)	xx.x (xx.x, xx.x)	xx.x (xx.x, xx.x)	xx.x (xx.x, xx.x)	xx.x (xx.x, xx.x)
		Minimum, Median, Maximum	xx.x, xx.x, xx.x	xx.x, xx.x, xx.x	xx.x, xx.x, xx.x	xx.x, xx.x, xx.x
	Anti-HA IgA ASC/Anti-HA IgG ASC	N	xx	xx	xx	xx
		Mean (95% CI)	xx.x (xx.x, xx.x)	xx.x (xx.x, xx.x)	xx.x (xx.x, xx.x)	xx.x (xx.x, xx.x)
		Minimum, Median, Maximum	xx.x, xx.x, xx.x	xx.x, xx.x, xx.x	xx.x, xx.x, xx.x	xx.x, xx.x, xx.x
	Anti-NA IgA ASC/Anti-NA IgG ASC	N	xx	xx	xx	xx
		Mean (95% CI)	xx.x (xx.x, xx.x)	xx.x (xx.x, xx.x)	xx.x (xx.x, xx.x)	xx.x (xx.x, xx.x)
		Minimum, Median, Maximum	xx.x, xx.x, xx.x	xx.x, xx.x, xx.x	xx.x, xx.x, xx.x	xx.x, xx.x, xx.x
[Repeat for Days 6 and 8]						

Note: N = Number of subjects in the Modified Intent-to-Treat population with available results
[a] Viral shedding and symptoms reported as of the given study day.

14.3.1.1 Solicited Symptoms

Table 46: Number and Percentage of FLU-PRO Solicited Symptoms by Study Day, Symptom, and Extent, Safety Population

Symptom	Extent[a]	Baseline (N=X)		Day 1 (Post-Challenge) (N=X)		Day 2 (N=X)		Day 3 (N=X)		Day 4 (N=X)		Day 5 (N=X)		Day 6 (N=X)		Day 7 (N=X)		Day 8 (N=X)		Days 9+ (N=X)		Anytime Post-Challenge (N=X)	
		n	%	n	%	n	%	n	%	n	%	n	%	n	%	n	%	n	%	n	%	n	%
Any Symptom	0	x	x.x	x	x.x	x	x.x	x	x.x	x	x.x	x	x.x	x	x.x	x	x.x	x	x.x	x	x.x	x	x.x
	1																						
	2																						
	3																						
	4																						
Nose																							
Any Symptom	0																						
	1																						
	2																						
	3																						
	4																						
[Symptom 1]	0																						
	1																						
	2																						
	3																						
	4																						
Throat																							
Any Symptom	0																						
	1																						
	2																						
	3																						
	4																						

Symptom	Extent[a]	Baseline (N=X)		Day 1 (Post-Challenge) (N=X)		Day 2 (N=X)		Day 3 (N=X)		Day 4 (N=X)		Day 5 (N=X)		Day 6 (N=X)		Day 7 (N=X)		Day 8 (N=X)		Days 9+ (N=X)		Anytime Post-Challenge (N=X)	
		n	%	n	%	n	%	n	%	n	%	n	%	n	%	n	%	n	%	n	%	n	%
[Symptom 1]	0																						
	1																						
	2																						
	3																						
	4																						
Eyes																							
Any Symptom	0																						
	1																						
	2																						
	3																						
	4																						
[Symptom 1]	0																						
	1																						
	2																						
	3																						
	4																						
[Repeat for all solicited FLU-PRO symptoms and separate by Body System/Domain]																							

Note: N = Number of subjects in the Safety population with available data.

[a]Extent is the maximum symptom score reported post-challenge for each subject.

Table 47: Number and Percentage of FLU-PRO Solicited Symptoms Anytime Post-Challenge by Study Site, Symptom, and Maximum Extent, Safety Population, Safety Population

Symptom	Extent[a]	Saint Louis University (N=X)		Cincinnati Hospital (N=X)		University of Maryland (N=X)		Duke University (N=X)		All Subjects (N=X)	
		n	%	n	%	n	%	n	%	n	%
Any Symptom	0	x	x.x	x	x.x	x	x.x	x	x.x	x	x.x
	1										
	2										
	3										
	4										
Nose											
Any Symptom	0										
	1										
	2										
	3										
	4										
[Symptom 1]	0										
	1										
	2										
	3										
	4										
Throat											
Any Symptom	0										
	1										
	2										
	3										
	4										
[Symptom 1]	0										

Symptom	Extent[a]	Saint Louis University (N=X)		Cincinnati Hospital (N=X)		University of Maryland (N=X)		Duke University (N=X)		All Subjects (N=X)	
		n	%	n	%	n	%	n	%	n	%
	1										
	2										
	3										
	4										
Eyes											
Any Symptom	0										
	1										
	2										
	3										
	4										
[Symptom 1]	0										
	1										
	2										
	3										
	4										
[Repeat for all solicited FLU-PRO symptoms and separate by Body System/Domain]											

Note: N = Number of subjects in the Safety population with available data.

[a] Extent is the maximum symptom score reported post-challenge for each subject.

Table 48: Number and Percentage of FLU-PRO Solicited Symptoms Anytime Post Challenge by Total Days of Viral Shedding, Symptom, and Maximum Extent, Safety Population

Symptom	Extent[a]	0 Days (N=X)		1 Day (N=X)		2 Days (N=X)		3-6 Days (N=X)		7+ Days (N=X)		1+ Days (N=X)		2+ Days (N=X)	
		n	%	n	%	n	%	n	%	n	%	n	%	n	%
Any Symptom	0	x	x.x	x	x.x	x	x.x	x	x.x	x	x.x	x	x.x	x	x.x
	1														
	2														
	3														
	4														
Nose															
Any Symptom	0														
	1														
	2														
	3														
	4														
[Symptom 1]	0														
	1														
	2														
	3														
	4														
Throat															
Any Symptom	0														
	1														
	2														
	3														
	4														
[Symptom 1]	0														

Symptom	Extent[a]	0 Days (N=X)		1 Day (N=X)		2 Days (N=X)		3-6 Days (N=X)		7+ Days (N=X)		1+ Days (N=X)		2+ Days (N=X)	
		n	%	n	%	n	%	n	%	n	%	n	%	n	%
	1														
	2														
	3														
	4														
Eyes															
Any Symptom	0														
	1														
	2														
	3														
	4														
[Symptom 1]	0														
	1														
	2														
	3														
	4														
[Repeat for all solicited FLU-PRO symptoms and separate by Body System/Domain]															

Note: N = Number of subjects in the Safety population follow for the entire challenge period.
[a]Extent is the maximum symptom score reported post-challenge for each subject.

Table 49: Summary of FLU-PRO Symptom Severity Scores by Body System/Domain and Study Day, Safety Population

Body System/Domain	Statistic	Baseline	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	Day 8	Day 9	Day 10	Day 11	Day 12	Day 13	Day 14	Day 15
Nose	N	x	x	x	x	x	x	x	x	x	x	x	x	x	x	x	x
	Mean	x.x	x.x	x.x	x.x	x.x	x.x	x.x	x.x	x.x	x.x	x.x	x.x	x.x	x.x	x.x	x.x
	SD	(x.x)	(x.x)	(x.x)	(x.x)	(x.x)	(x.x)	(x.x)	(x.x)	(x.x)	(x.x)	(x.x)	(x.x)	(x.x)	(x.x)	(x.x)	(x.x)
	Median (IQR)	x.x (x.x, x.x)	x.x (x.x, x.x)	x.x (x.x, x.x)	x.x (x.x, x.x)	x.x (x.x, x.x)	x.x (x.x, x.x)	x.x (x.x, x.x)	x.x (x.x, x.x)	x.x (x.x, x.x)	x.x (x.x, x.x)	x.x (x.x, x.x)	x.x (x.x, x.x)	x.x (x.x, x.x)	x.x (x.x, x.x)	x.x (x.x, x.x)	x.x (x.x, x.x)
Throat	N																
	Mean																
	SD																
	Median (IQR)																
Eyes	N																
	Mean																
	SD																
	Median (IQR)																
[Repeat for remaining Body Systems/Domains]																	
Total Severity Score	N																
	Mean																
	SD																
	Median (IQR)																
Note: N = Number of subjects in the Safety population with available data.																	

Table 50: FLU-PRO Total Symptom Severity Scores by Viral Shedding Status and Study Day, Safety Population

Study Day	Viral Shedding Status	N	Median	Mean (95% CI)
Day 2	RT-PCR Negative	X	x.x	x.x (x.x, x.x)
	RT-PCR Positive	X	x.x	x.x (x.x, x.x)
Day 3	RT-PCR Negative			
	RT-PCR Positive			
Day 4	RT-PCR Negative			
	RT-PCR Positive			
Day 5	RT-PCR Negative			
	RT-PCR Positive			
Day 6	RT-PCR Negative			
	RT-PCR Positive			
Day 7	RT-PCR Negative			
	RT-PCR Positive			
Day 8	RT-PCR Negative			
	RT-PCR Positive			
Day 9	RT-PCR Negative			
	RT-PCR Positive			
Day 10	RT-PCR Negative			
	RT-PCR Positive			
Day 11	RT-PCR Negative			
	RT-PCR Positive			
Day 12	RT-PCR Negative			
	RT-PCR Positive			
Day 13	RT-PCR Negative			
	RT-PCR Positive			
Day 14	RT-PCR Negative			
	RT-PCR Positive			
Day 15	RT-PCR Negative			
	RT-PCR Positive			
Note: N = Number of subjects in the Safety population with available data.				

Table 51: Number and Percentage of Subjects Reporting Mild-to-Moderate Influenza Disease Solicited Symptoms Anytime Post-Challenge (Days 2 to 8) by Clinical Site and Symptom, Modified Intent-to Treat Population

Influenza Symptom	Saint Louis University (N=X)		Cincinnati Hospital (N=X)		University of Maryland (N=X)		Duke University (N=X)		All Subjects (N=X)	
	n	%	n	%	n	%	n	%	n	%
Body Aches or Pain[a]										
Chest Tightness										
Chills										
Conjunctivitis[b]										
Nasal Congestion										
Sinus Congestion										
Coryza[b]										
Decreased Appetite										
Diarrhea										
Dry Cough										
Dyspnea										
Fatigue										
Headache										
Fever (>38.0°C)										
Lymphopenia[d]										
Nausea										
Oxygen Saturation Decrease by ≥ 3%[c]										
Productive Cough										
Rhinorrhea										
Sore Throat										
Sweats										

Note: N = Number of subjects in the Modified Intent-to-Treat population.

[a] Includes any report of arthralgia or myalgia as determined by clinician during targeted physical examinations, or patient-reported "Body Aches or Pains" responses via FLU-PRO questionnaire.

[b] Clinical findings reported during targeted physical examinations. For subject-specific details refer to Listing 17.

[c] Saturation of Peripheral Oxygen percent decrease of 3 or more relative to most recent assessment prior to challenge administration.

[d] Absolute Lymphocyte Count laboratory result of grade 1 or higher reported on Day 2, 4 or 8. For subject-specific details refer to Listing 15.

Table 52: Influenza Symptom Scoring Algorithm – Symptoms Predicting RT-PCR Positive Influenza Infection Based on 10-Fold Cross Validation, Safety Population

Symptom	Number of Iterations with Nonzero Coefficient[a]	Mean Estimated Coefficient (min, median, max)	Corresponding Odds Ratio (min, median, max)	Mean Standard Error (min, median, max)	Standard Deviation of Estimated Coefficients
[Symptom 1]	x	x.xx (x.xx, x.xx, x.xx)	x.xx (x.xx, x.xx, x.xx)	x.xx (x.xx, x.xx, x.xx)	x.xx
[Symptom 2]					
[Include all solicited symptoms and labs used in the MMID definition]					
Note: N = X subjects in the Safety population with available symptom data included in the analysis. [a] Out of 10 cross validation iterations.					

Table 53: Influenza Symptom Scoring Algorithm – Operating Characteristics

Statistic	Value
Mean AUC (min, median, max)[a]	0.xx (0.xx, 0.xx, 0.xx)
Mean Specificity (min, median, max)[a]	0.xx (0.xx, 0.xx, 0.xx)
Mean Sensitivity (min, median, max)[a]	0.xx (0.xx, 0.xx, 0.xx)
AUC of final model on entire data set[b]	0.xx
Specificity of final model on entire data set[b]	0.xx
Sensitivity of final model on entire data set[b]	0.xx

Note: N = X subjects in the Safety population with available symptom data included in the analysis.
[a] Across 10-fold cross-validation steps
[b] Internal performance of mean coefficients for predicting RT-PCR positivity, based on the optimal cutpoint as determined by Youden's J statistic

Table 54: Comparison of the Proportion of Subjects Experiencing Symptoms by Viral Detection Status, Safety Population

		Comparison 1		Comparison 2	
Symptom	**Statistic**	**RT-PCR Positive (One or More) (N=X)**	**RT-PCR Negative (None Positive) (N=X)**	**RT-PCR Positive (Two or More) (N=X)**	**RT-PCR Negative (One or Less Positive) (N=X)**
Any Symptom	Proportion (95% CI)	x.x (x.x, x.x)	x.x (x.x, x.x)	x.x (x.x, x.x)	x.x (x.x, x.x)
	Difference[a] (95% CI)	x.x (x.x, x.x)	N/A	x.x (x.x, x.x)	N/A
	p-value	0.xx	N/A	N/A	N/A
[Symptom 1]	Proportion (95% CI)	x.x (x.x, x.x)	x.x (x.x, x.x)	x.x (x.x, x.x)	x.x (x.x, x.x)
	Difference[a] (95% CI)	x.x (x.x, x.x)	N/A	x.x (x.x, x.x)	N/A
	p-value	0.xx	N/A	N/A	N/A
[Repeat for all solicited symptoms and clinical labs included in the MMID definition]					

Note: N = Number of subjects in the Safety population.
[a]Difference in proportions between RT-PCR Positive and RT-PCR Negative groups.

Table 55: Number and Percentage of Subjects Reporting Symptoms, by Viral Detection Status, Symptom, and Baseline Hemagglutination Inhibition Seroprotection Status Against A/Bethesda/MM2/H1N1, Modified Intent-to-Treat Population

Symptom	Baseline Seroprotection Status	RT-PCR Positive (One or More) (N=X)			RT-PCR Positive (Two or More) (N=X)			RT-PCR Negative (N=X)		
		n	%	95% CI	n	%	95% CI	n	%	95% CI
No Symptoms	Low < 1:40	x	x.x	x.x, x.x	x	x.x	x.x, x.x	x	x.x	x.x, x.x
	High ≥ 1:40	x	x.x	x.x, x.x	x	x.x	x.x, x.x	x	x.x	x.x, x.x
Any Symptom	Low < 1:40									
	High ≥ 1:40									
[Symptom 1]	Low < 1:40									
	High ≥ 1:40									
[Symptom 2]	Low < 1:40									
	High ≥ 1:40									
[Repeat for all symptoms]										
Note: N = Number of subjects in the Modified Intent-to-Treat population.										

Tables with similar format:

Table 56: Number and Percentage of Viral Shedding Status by Symptom and Baseline Neuraminidase Inhibition Seroprotection Status Against A/Bethesda/MM2/H1N1, Modified Intent-to-Treat Population

Table 57: Number and Percentage of Viral Shedding Status by Symptom and Baseline Microneutralization Seroprotection Status Against A/Bethesda/MM2/H1N1, Modified Intent-to-Treat Population

Table 58: Overall Summary of Additional Daily Diary Items for Flu-Pro Validation, Safety Population

[Implementation Note: A "Not Reported" row should be added for any survey item that has no data reported for at least one subject post the relevant challenge day.]

	Day 2		Day 3		Day 4		Day 5		Day 6		Day 7		Day 8	
Subjects reporting	n	%	n	%	n	%	n	%	n	%	n	%	n	%
[a] RT-PCR Positive (N=X)														
Medication use for flu symptoms	x	x	x	x	x	x	x	x	x	x	x	x	x	x
Severity of flu symptoms today														
No flu symptoms today	x	x	x	x	x	x	x	x	x	x	x	x	x	x
Mild														
Moderate														
Severe														
Very Severe														
Severity of flu symptoms compared to previous day														
Much better														
Somewhat better														
A little better														
About the same														
A little worse														
Somewhat worse														
Much worse														
Extent flu symptoms interfered with usual activities today														
Not at all														
A little bit														
Somewhat														
Quite a bit														
Very much														
Rate of physical health today														
Excellent														
Very Good														
Good														
Fair														

	Day 2		Day 3		Day 4		Day 5		Day 6		Day 7		Day 8	
Subjects reporting	n	%	n	%	n	%	n	%	n	%	n	%	n	%
Poor														
Return to usual activities today														
Return to usual health today														
[a] RT-PCR Negative (N=X)														
[Repeat items for RT-PCR negative]														

N = Number of subjects in Safety population; n = Number of subjects reporting each validation diary answer
[a] Viral shedding status on given day

14.3 Safety Data

14.3.1 Displays of Adverse Events

Table 59: Overall Summary of Adverse Events, Safety Population

	All Subjects (N = X)	
Subjects[a]	**n**	**%**
At least one unsolicited adverse event	x	x
At least one related unsolicited adverse event	x	x
Mild (Grade 1)	x	x
Moderate (Grade 2)	x	x
Severe (Grade 3)	x	x
At least one severe (Grade 3) unsolicited adverse event		
Related		
Unrelated		
At least one serious adverse event		
At least one related, serious adverse event		
At least one adverse event leading to early termination[b]		
At least one hematology laboratory adverse event[c]		
Mild (Grade 1)		
Moderate (Grade 2)		
Severe (Grade 3)		
At least one chemistry laboratory adverse event[c]		
Mild (Grade 1)		
Moderate (Grade 2)		
Severe (Grade 3)		
At least one abnormal vital sign[c]		

Note: N = Number of subjects in the Safety population.
[a] Subjects are counted once for each category regardless of the number of events.
[b] As reported on the Adverse Event eCRF.
[c] Only findings that are not part of Mild-to-Moderate Influenza Disease and occurred post-challenge are considered.

Table 60: Adverse Events Occurring in At Least 5% of Subjects by MedDRA System Organ Class and Preferred Term, Safety Population

[Implementation Note: This table should include a row for All PT/SOC and for any PT/SOC reported by ≥[threshold]% subjects in any group. This includes all adverse events collected (e.g., solicited, unsolicited, AESI, laboratory adverse events, etc.), regardless of relationship to study product, so this summary should combine data collected across multiple eCRFs/domains.]

Preferred Term	MedDRA System Organ Class	All Subjects (N=X)		
		n	%	Events
All	All	x	x	x
PT1	SOC1	x	x	x
Etc.	Etc.			
Notes: N = Number of subjects in the Safety population; n = Number of subjects reporting event; Events = Total frequency of events reported.				

14.3.1.2 Unsolicited Adverse Events

Table 61: Summary of All Unsolicited Adverse Events by MedDRA System Organ Class and Preferred Term, Maximum Severity, and Relationship to Study Challenge, Safety Population

MedDRA Classification		Severity (N = X)			Relationship to Study Challenge (N=X)	
		Mild	Moderate	Severe	Not Related	Related
		n	n	n	n	n
System Organ Class	Preferred Term					
Any SOC	Any PT					
[SOC 1]	Any PT					
	[PT 1]					
	[PT 2]					
[SOC 2]	Any PT					
	[PT 1]					
	[PT 2]					
Note: N = Number of subjects in the Safety population.						

Table 62: Unsolicited Adverse Events by MedDRA System Organ Class and Preferred Term, Maximum Severity, and Relationship to Study Challenge, Safety Population

MedDRA System Organ Class	Preferred Term	Severity	All Subjects					
			Related (N = X)		Not Related (N = X)		Total (N = X)	
			n	%	n	%	n	%
Any SOC	Any PT	Any Severity	x	xx	x	xx	x	xx
		Mild	x	xx	x	xx	x	xx
		Moderate	x	xx	x	xx	x	xx
		Severe	x	xx	x	xx	x	xx
[SOC 1]	[PT 1]	Any Severity	x	xx	x	xx	x	xx
		Mild	x	xx	x	xx	x	xx
		Moderate	x	xx	x	xx	x	xx
		Severe						
	[PT 2]	Any Severity						
		Mild						
		Moderate						
		Severe						

Note: N = Number of subjects in the Safety population.

Table 63: Unsolicited Adverse Events by MedDRA System Organ Class and Preferred Term, and Study Day Post-Challenge, Safety Population

		Day 1 (N=X)		Day 2 (N=X)		Day 3 (N=X)		Day 4 (N=X)		Day 5 (N=X)		Day 6 (N=X)		Day 7 (N=X)		Day 8 (N=X)		Anytime Post-Challenge (N=X)	
MedDRA System Organ Class	**MedDRA Preferred Term**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**
Serious Adverse Events																			
Any SOC	Any PT	x	xx	x	xx	x	xx	x	xx	x	xx	x	xx	x	xx	x	xx	x	xx
[SOC 1]	Any PT																		
	[PT 1]																		
	[PT 2]																		
[SOC 2]	Any PT																		
	[PT 1]																		
	[PT 2]																		
Non-serious Adverse Events																			
Any SOC	Any PT																		
[SOC 1]	Any PT																		
	[PT 1]																		
	[PT 2]																		

Note: N = Number of subjects in the Safety population. This table presents number and percentage of subjects. For each time point, a subject is only counted once per PT, separately for SAEs and non-serious AEs.

14.3.2 Listing of Deaths, Other Serious and Significant Adverse Events

Table 64: Listing of Serious Adverse Events

[Implementation Note: This listing is included in the table shells document, as it is included in the body of the CSR. If the event is ongoing (no stop date), indicate "ongoing" for the "Duration". If more than one reason is selected for the reason reported as an SAE, list all reasons in the column, separated by a comma. In the "If Not Related, Alternate Etiology" column, merge the 2 data fields for collecting alternate etiology, separate by a colon. If there are no comments for an event, populate 'Comments' row with 'None'. Add columns for MedDRA HLT or LLT depending on halting criteria or other study needs. In the CSR, Subject ID should be USUBJID (not PATID) for purposes of de-identification. Listing should be sorted by Subject ID, Associated with Dose No., and No. of Days Post Associated Dose.]

Adverse Event	No. of Days Post Associated Dose (Duration)	No. of Days Post-Challenge the Event Became Serious	Reason Reported as an SAE	Severity	Relationship to Study Treatment	If Not Related, Alternative Etiology	Subject Discontinued Due to AE	Outcome	MedDRA System Organ Class	MedDRA Preferred Term
Subject ID: , AE Number:										
Comments: [Verbatim comments from the clinical site are displayed here]										
Subject ID: , AE Number:										
Comments: [Verbatim comments from the clinical site are displayed here]										

14.3.3 Narratives of Deaths, Other Serious and Significant Adverse Events

(not included in SAP, but this is a placeholder for the CSR)

14.3.4 Abnormal Laboratory Value Listings (by Subject)

Table 65: Listing of Abnormal Laboratory Results - Hematology

[Implementation Note: This listing is included in the table shells document, as it is included in the body of the CSR. This listing should include all hematology results for any subject that had at least one abnormal chemistry laboratory result. A complete listing of all laboratory results is included in the listings document. In the Laboratory Parameter column, indicate the units after the parameter, e.g., Hemoglobin (g/dL). This listing is not color-coded, but the severity should be included in parentheses after the result, e.g., 16.2 (mild).In the "If Not Related, Alternate Etiology" column, merge the 2 data fields for collecting alternate etiology, separate by a colon. In the CSR, Subject ID should be USUBJID (not PATID) for purposes of de-identification.]

Subject ID	Sex	Age (years)	Planned Study Day	Actual Study Day	Laboratory Parameter (Units)	Result (Severity)	Relationship to Challenge	If Not Related, Alternate Etiology	Action Taken with Study Challenge	Subject Discontinued Due to Result?

Table 66: Listing of Abnormal Laboratory Results - Chemistry

Subject ID	Sex	Age (years)	Planned Study Day	Actual Study Day	Laboratory Parameter (Units)	Result (Severity)	Relationship to Challenge	If Not Related, Alternate Etiology	Action Taken with Study Challenge	Subject Discontinued Due to Result?

14.3.5 Displays of Laboratory Results

14.3.5.1 Hematology and Biochemistry Results

Table 67: White Blood Cell Results (10^9/L) by Severity and Study Day, Safety Population

Study Day	N	None		Mild/ Grade 1 (Low)		Mild/ Grade 1 (High)		Moderate/ Grade 2 (Low)		Moderate/ Grade 2 (High)		Severe/ Grade 3 (Low)		Severe/ Grade 3 (High)		Missing	
		n	%	n	%	n	%	n	%	n	%	n	%	n	%	n	%
Baseline	x	x	x.x	x	x.x	x	x.x	x	x.x	x	x.x	x	x.x	x	x.x	x	x.x
Day 2	x	x	x.x	x	x.x	x	x.x	x	x.x	x	x.x	x	x.x	x	x.x	x	x.x
Day 4																	
Day 8																	
Maximum Severity Post-Challenge[a]																	

Note: Note: N = Number of subjects in the Safety population.

[a] Maximum severity experienced by each subject at any time point post-challenge, including unscheduled assessments.

Tables with similar format:

Table 68: Hemoglobin (g/dL) Results by Severity and Study Day, Safety Population

Table 69: Platelet (10^9/L) Results by Severity and Study Day, Safety Population

Table 70: Absolute Lymphocyte Count (10^9/L) Results by Severity and Study Day, Safety Population

Table 71: Alanine Aminotransferase (U/L) Results by Severity and Study Day, Safety Population

Table 72: Creatinine (mg/dL) Results by Severity and Study Day, Safety Population

Table 73: Laboratory Summary Statistics by Parameter and Study Day – White Blood Cells (10^9/L), Safety Population

Study Day	N	Mean	Standard Deviation	Median	Min, Max
Baseline	x	xx.x	xx.x	xx.x	xx.x, xx.x
Day 2	x	xx.x	xx.x	xx.x	xx.x, xx.x
Day 4					
Day 8					
Max Severity Post-Challenge[a]					

Note: N = Number of subjects in the Safety population.
[a] Maximum severity experienced by each subject at any time point post-challenge, including unscheduled assessments.

Tables with similar format:

Table 74: Laboratory Summary Statistics by Study Day – Hemoglobin (g/dL), Safety Population

Table 75: Laboratory Summary Statistics by Study Day – Platelets (10^9/L), Safety Population

Table 76: Laboratory Summary Statistics by Study Day – Absolute Lymphocyte (10^9/L), Safety Population

Table 77: Laboratory Summary Statistics by Study Day – Alanine Aminotransferase (U/L), Safety Population

Table 78: Laboratory Summary Statistics by Study Day – Creatinine (mg/dL), Safety Population

Table 79: Listing of Abnormal Vital Signs

[Implementation Note: This listing is included in the table shells document, as it is included in the body of the CSR. This listing should include all hematology results for any subject that had at least one abnormal hematology laboratory result. A complete listing of all laboratory results is included in the listings document. In the Laboratory Parameter column, indicate the units after the parameter, e.g., Hemoglobin (g/dL). This listing is not color-coded, but the severity should be included in parentheses after the result, e.g., 16.2 (mild). In the "If Not Related, Alternate Etiology" column, merge the 2 data fields for collecting alternate etiology, separate by a colon. In the CSR, Subject ID should be USUBJID (not PATID) for purposes of de-identification.]

Subject ID	Planned Study Day	Actual Study Day	Assessment Time	Temperature (°C)	Systolic Blood Pressure (mmHg)	Diastolic Blood Pressure (mmHg)	Heart Rate (beats/min)	Respiratory Rate (breaths/min)	Saturation of Peripheral Oxygen (%)

14.3.6 Displays of Vital Signs

Table 80: Vital Signs by Severity and Study Day – Any Assessment, Safety Population

Study Day	N	None		Mild		Moderate		Severe	
		n	%	n	%	n	%	n	%
Baseline[a]	x	x	xx	x	xx	x	xx	x	xx
Day 1									
Day 2									
Day 3									
Day 4									
Day 5									
Day 6									
Day 7									
Day 8									
Day 15									
Day 29									
Day 61									
Max Severity Post-Challenge[b]									

Note: Note: N = Number of subjects in Safety population with available data. Maximum severity reported each day is presented.

[a] Day 1 assessment prior to challenge.

[b] Maximum severity experienced by each subject at any time point post-challenge, including unscheduled assessments.

Tables with similar format:

Table 81: Vital Signs by Severity and Study Day – Pulse, Safety Population

Table 82: Vital Signs by Severity and Study Day – Systolic Blood Pressure, Safety Population

Table 83: Vital Signs by Severity and Study Day – Diastolic Blood Pressure, Safety Population

Table 84: Vital Signs by Severity and Study Day – Oral Temperature, Safety Population

Table 85: Vital Signs by Severity and Study Day – Respiratory Rate, Safety Population

Table 86: Vital Signs by Severity and Study Day – Saturation of Peripheral Oxygen (SpO_2), Safety Population

[Implementation note: SpO_2 to include baseline and Day 1 to 8 results.]

14.4 Summary of Concomitant Medications

Table 87: Number and Percentage of Subjects with Prior and Concomitant Medications by WHO Drug Classification, Safety Population

WHO Drug Code Level 1, Anatomic Group	WHO Drug Code Level 2, Therapeutic Subgroup	All Subjects (N=X)	
		n	%
Any Level 1 Codes	Any Level 2 Codes	x	xx
[ATC Level 1 - 1]	Any [ATC 1 – 1]		
	[ATC 2 - 1]		
	[ATC 2 - 2]		
	[ATC 2 - 3]		
[ATC Level 1 – 2]	[ATC 2 - 1]		
	[ATC 2 - 2]		
	[ATC 2 - 3]		

Note: N = Number of subjects in the Safety Population; n = Number of subjects reporting taking at least one medication in the specific WHO Drug Class.

APPENDIX 2. FIGURE MOCK-UPS

LIST OF FIGURES

10.1 Disposition of Subjects

Figure 1: CONSORT Flow Diagram

[Implementation Note: This is just an example; a similar figure will be generated for this study. No allocation row will be included since no randomization occurred in this study.

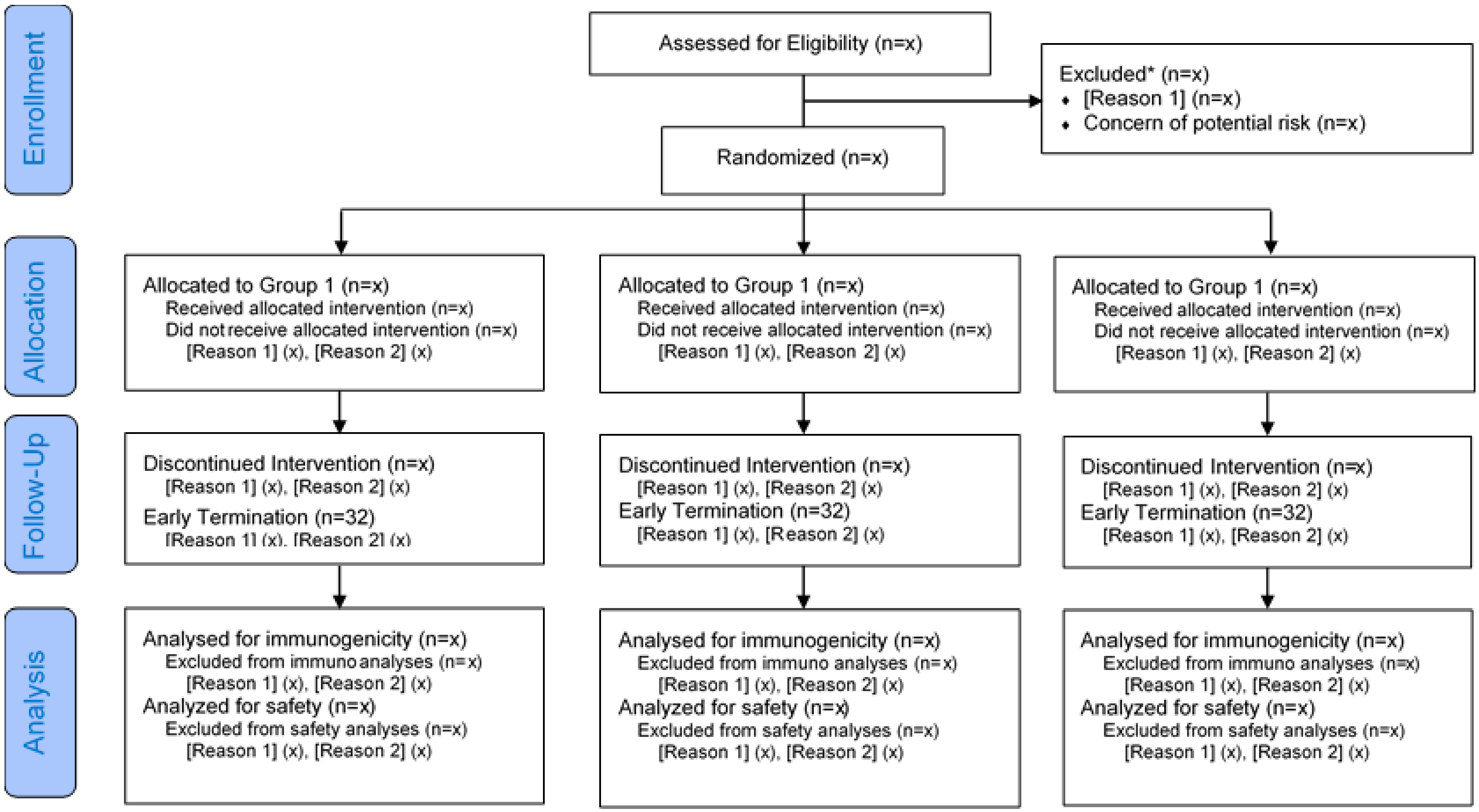

Figure 2: Study Design

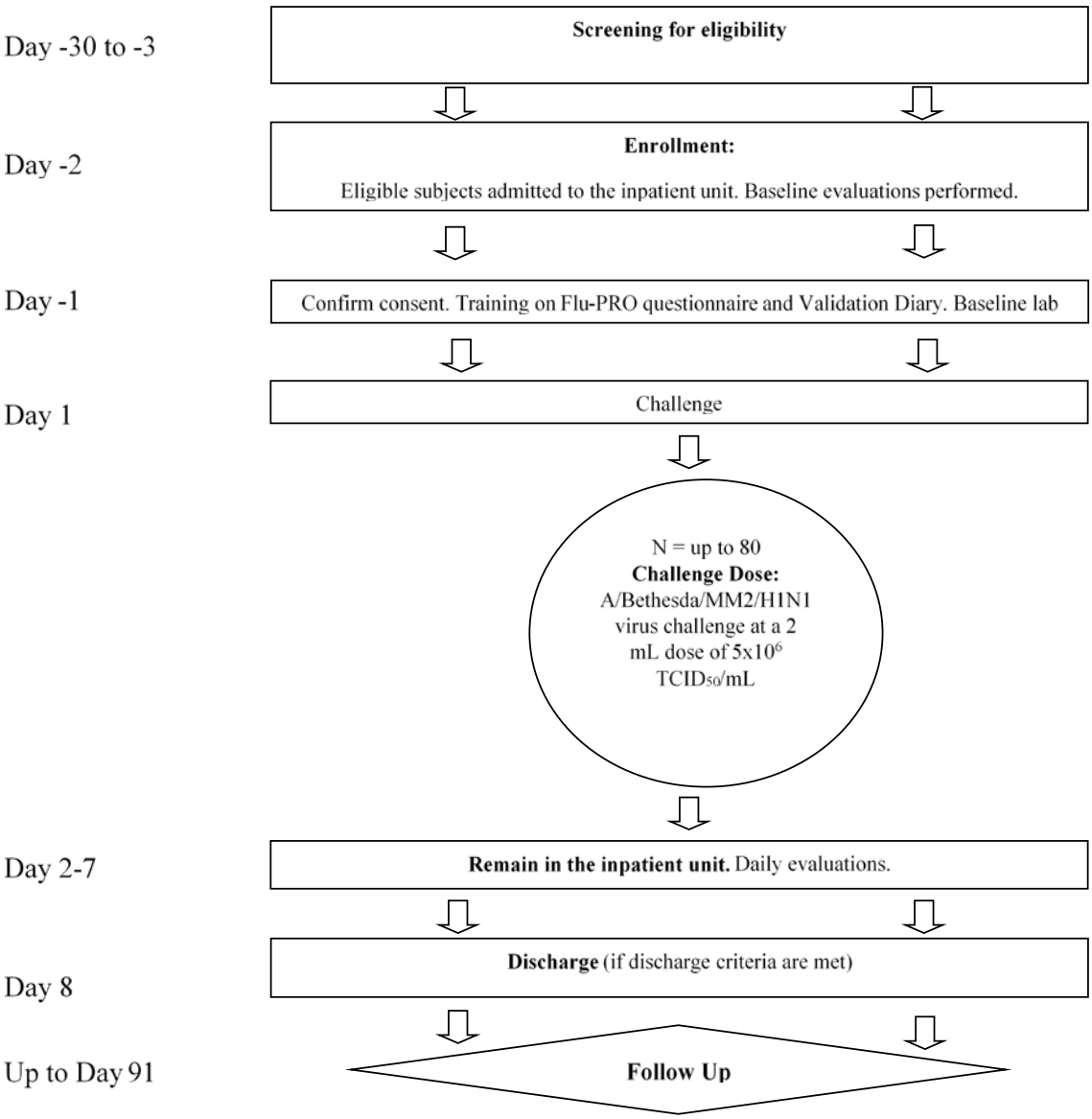

Figure 3: FLU-PRO Survey Instrument and Validation Diary

Participant ID: _____ Participant Initials: _____ Date: ____/_____/_____

FLU-PRO©

People experience the flu in different ways. We would like to know about the symptoms you have been experiencing during the past 24 hours. For each symptom, please mark one box ☐ under the response that best matches your experience. Mark the "Not at all" box, if you did not have that symptom in the past 24 hours.

What time is it? _____ AM / PM (please circle)

Please rate the extent to which you had each symptom during the past 24 hours.

	Not at all	A little bit	Somewhat	Quite a bit	Very much
Runny or dripping nose	☐	☐	☐	☐	☐
Congested or stuffy nose	☐	☐	☐	☐	☐
Sinus pressure	☐	☐	☐	☐	☐
Scratchy or itchy throat	☐	☐	☐	☐	☐
Sore or painful throat	☐	☐	☐	☐	☐
Difficulty swallowing	☐	☐	☐	☐	☐
Teary or watery eyes	☐	☐	☐	☐	☐
Sore or painful eyes	☐	☐	☐	☐	☐
Eyes sensitive to light	☐	☐	☐	☐	☐
Trouble breathing	☐	☐	☐	☐	☐
Chest congestion	☐	☐	☐	☐	☐
Chest tightness	☐	☐	☐	☐	☐
Dry or hacking cough	☐	☐	☐	☐	☐
Wet or loose cough	☐	☐	☐	☐	☐
Felt nauseous (feeling like you wanted to throw-up)	☐	☐	☐	☐	☐
Stomach ache	☐	☐	☐	☐	☐
Felt dizzy	☐	☐	☐	☐	☐
Head congestion	☐	☐	☐	☐	☐
Headache	☐	☐	☐	☐	☐

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Participant ID: _____ Participant Initials: _____ Date: ____/_____/_____

Please rate the extent to which you had each symptom during the past 24 hours.

	Not at all	A little bit	Somewhat	Quite a bit	Very much
Lack of appetite	☐	☐	☐	☐	☐
Sleeping more than usual	☐	☐	☐	☐	☐
Body aches or pains	☐	☐	☐	☐	☐
Weak or tired	☐	☐	☐	☐	☐
Chills or shivering	☐	☐	☐	☐	☐
Felt cold	☐	☐	☐	☐	☐
Felt hot	☐	☐	☐	☐	☐
Sweating	☐	☐	☐	☐	☐

In the past 24 hours, how often have you had any of the following symptoms?

	Never	Rarely	Sometimes	Often	Always
Sneezing	☐	☐	☐	☐	☐
Coughing	☐	☐	☐	☐	☐
Coughed up mucus or phlegm	☐	☐	☐	☐	☐

	0 times	1 time	2 times	3 times	4 or more times
How many times did you vomit?	☐	☐	☐	☐	☐
How many times did you have diarrhea?	☐	☐	☐	☐	☐

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ADDITIONAL DAILY DIARY ITEMS FOR FLU-PRO VALIDATION STUDY

Items to be asked in the daily diary through to Day 14 along with the FLU-PRO items.

1. Did you take any medication for your flu symptoms today? (Please select one response only)
   - ☐1 Yes
   - ☐0 No

2. Do you have asthma, COPD (chronic obstructive pulmonary disease) or both?
   - ☐1 Yes
   - ☐0 No

3. [Only asked if answer to the question above is "yes"]. Did you use any rescue medication today for your asthma or COPD? (Please select one response only)
   - ☐1 Yes
   - ☐0 No

4. Overall, how severe were your flu symptoms today? (Please select one response only)
   - ☐0 No flu symptoms today
   - ☐1 Mild
   - ☐2 Moderate
   - ☐3 Severe
   - ☐4 Very severe

5. Overall, how were your flu symptoms today compared to yesterday? (Please select one response only)
   - ☐1 Much better
   - ☐2 Somewhat better
   - ☐3 A little better
   - ☐4 About the same
   - ☐5 A little worse
   - ☐6 Somewhat worse
   - ☐7 Much worse

6. How much did your flu symptoms interfere with your usual activities today? (Please select one response only)
   - ☐1 Not at all
   - ☐2 A little bit
   - ☐3 Somewhat
   - ☐4 Quite a bit
   - ☐5 Very much

1

7. Have you returned to your usual activities today?
   - ☐1 Yes
   - ☐0 No

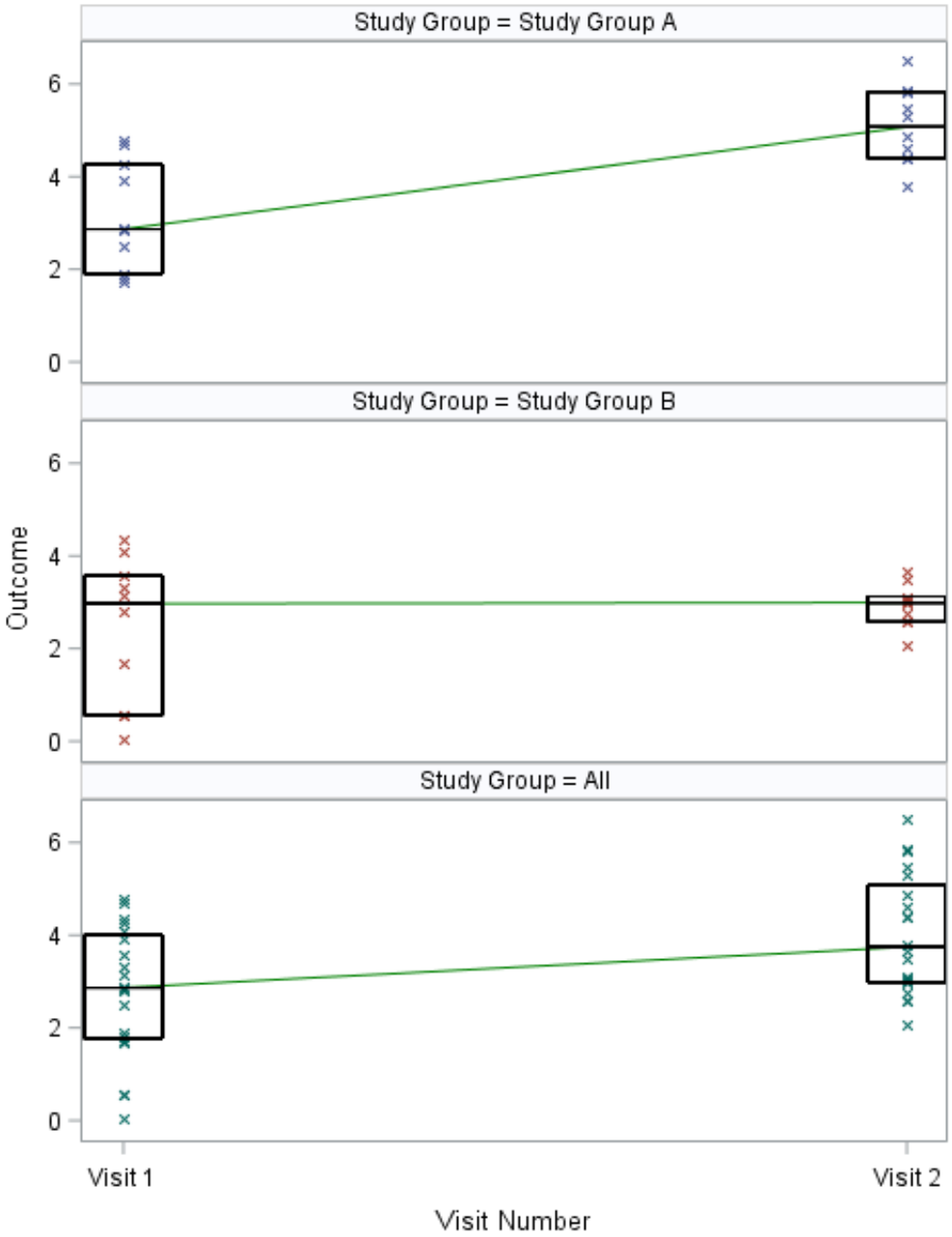
8. In general, how would you rate your physical health today? (Please select one response only)
   - ☐5 Excellent
   - ☐4 Very Good
   - ☐3 Good
   - ☐2 Fair
   - ☐1 Poor

9. Have you returned to your usual health today?
   - ☐1 Yes
   - ☐0 No

14.2.2 Clinical and Immunogenicity Response Figures by Measure and Time Point

Figure 4: Box Plots of Hemagglutination Inhibition Antibody Against A/Bethesda/MM2/H1N1 by Mild-to-Moderate Influenza Disease Status and Study Day, Modified Intent-to-Treat Population

[Implementation Note: The GMT should be displayed in a single image file with separate panels for group status. Do not include an "All Subjects" panel. The x-axis should be labeled "Study Day" with measurements at baseline, and Day 8, 29 & 61. The y-axis should be labeled "Geometric Mean Titer". Include "[Outcome] assessed at any time during challenge period" footnote within image.]



Figures with similar format:

Figure 5: Box Plots of Neuraminidase Inhibition Antibody Against A/Bethesda/MM2/H1N1 by Mild-to-Moderate Influenza Disease Status and Study Day, Modified Intent-to-Treat Population

Figure 6: Box Plots of Microneutralization Against A/Bethesda/MM2/H1N1 by Mild-to-Moderate Influenza Disease Status and Study Day, Modified Intent-to-Treat Population

Figure 7: Box Plots of Hemagglutination Inhibition Antibody Against A/Bethesda/MM2/H1N1 by Viral Shedding Status and Study Day, Modified Intent-to-Treat Population

Figure 8: Box Plots of Neuraminidase Inhibition Antibody Against A/Bethesda/MM2/H1N1 by Viral Shedding Status and Study Day, Modified Intent-to-Treat Population

Figure 9: Box Plots of Microneutralization Against A/Bethesda/MM2/H1N1 by Viral Shedding Status and Study Day, Modified Intent-to-Treat Population

Figure 10: Box Plots of Hemagglutination Inhibition Antibody Against A/Bethesda/MM2/H1N1 by Infection Status and Study Day, Modified Intent-to-Treat Population

Figure 11: Box Plots of Neuraminidase Inhibition Antibody Against A/Bethesda/MM2/H1N1 by Infection Status and Study Day, Modified Intent-to-Treat Population

Figure 12: Box Plots of Microneutralization Against A/Bethesda/MM2/H1N1 by Infection Status and Study Day, Modified Intent-to-Treat Population

Figure 13: Box Plots of Anti-HA IgG ASC per 10^6 PBMC by Infection Status and Study Day, Modified Intent-to-Treat Population

Figure 14: Box Plots of Anti-NA IgG ASC per 10^6 PBMC by Infection Status and Study Day, Modified Intent-to-Treat Population

Figure 15: Box Plots of Anti-HA IgA ASC per 10^6 PBMC by Infection Status and Study Day, Modified Intent-to-Treat Population

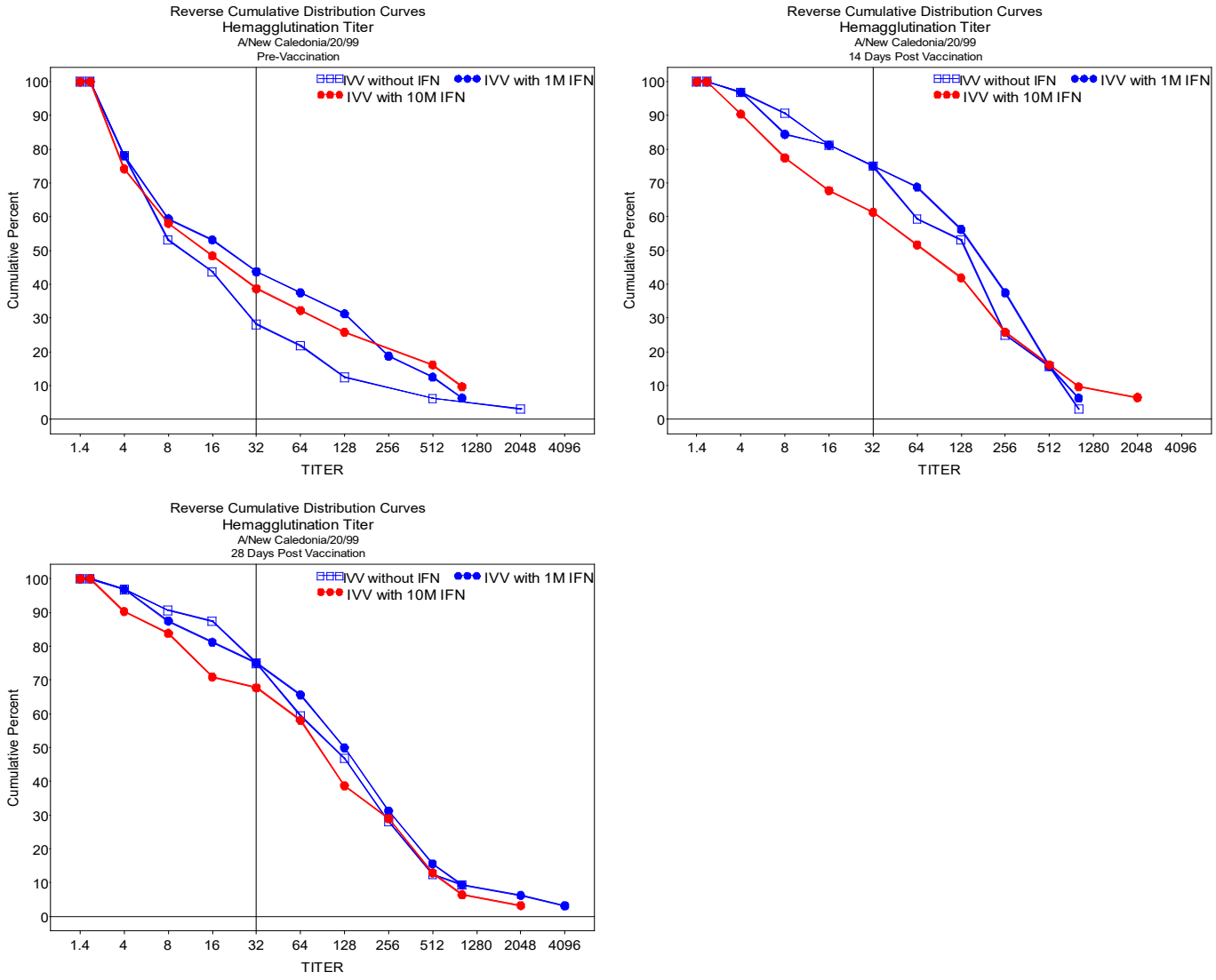
Figure 16: Box Plots of Anti-NA IgA ASC per 10^6 PBMC by Infection Status and Study Day, Modified Intent-to-Treat Population

Figure 17: Box Plots of the Ratio of Anti-HA IgA ASC to Anti-HA IgG ASC by Infection Status and Study Day, Modified Intent-to-Treat Population

Figure 18: Box Plots of the Ratio of Anti-NA IgA ASC to Anti-NA IgG ASC by Infection Status and Study Day, Modified Intent-to-Treat Population

Figure 19: Reverse Cumulative Distribution of Hemagglutination Inhibition Antibody Against A/Bethesda/MM2/H1N1 by Study Day and Mild-to-Moderate Influenza Disease Status, Modified Intent-to-Treat Population

Implementation Note: Each panel with % of subjects on the y-axis and titer on the x-axis and two curves, one for each MMID status. Plot each time point (Baseline, Day 8, Day 29, and Day 61) in a separate panel, with the MMID status in different colors and/or line types.]



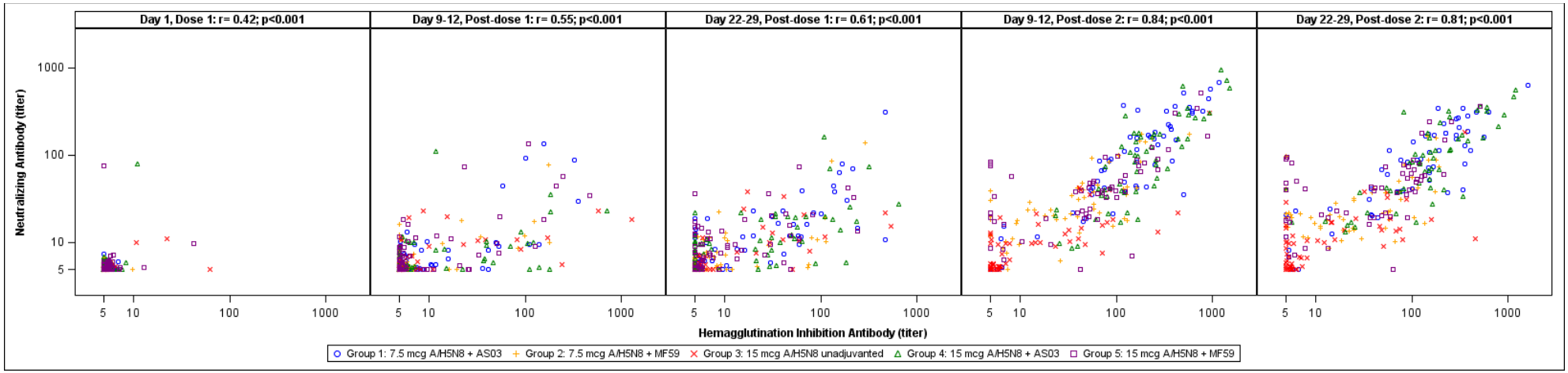
Figures with similar format:

Figure 20: Reverse Cumulative Distribution of Neuraminidase Inhibition Antibody Against A/Bethesda/MM2/H1N1 by Study Day and Mild-to-Moderate Influenza Disease Status, Modified Intent-to-Treat Population

Figure 21: Reverse Cumulative Distribution of Microneutralization Antibody Against A/Bethesda/MM2/H1N1 by Study Day and Mild-to-Moderate Influenza Disease Status, Modified Intent-to-Treat Population

Figure 22: Correlation of Hemagglutination Inhibition and Neuraminidase Inhibition Antibody Against A/Bethesda/MM2/H1N1 by Mild-to-Moderate Influenza Disease Status and Study Day, Modified Intent-to-Treat Population

[Implementation Note: A generic sample figure is shown below. The scatter plots should be presented in a single figure with separate panels for each relevant visit (baseline, and Day 8, 29, and 61). Visit labels should be included in the panel header. Within each panel distinct marker types and colors should be used to indicate the 2 MMID status groups, with MMID status labels included in a legend. The spearman correlation should be calculated over all subjects and annotated within each panel header after the visit label as "r=0.xx, p=0.xx". The x-axis should be labeled by "[Assay 1] Antibody (Titer)" and the y-axis by "[Assay 2] Antibody (Titer)".]



Figures with similar format:

Figure 23: Correlation of Hemagglutination Inhibition and Microneutralization Antibody Against A/Bethesda/MM2/H1N1 by Mild-to-Moderate Influenza Disease and Study Day, Modified Intent-to-Treat Population

Figure 24: Correlation of Neuraminidase Inhibition and Microneutralization Antibody Against A/Bethesda/MM2/H1N1 by Mild-to-Moderate Influenza Disease Study Day, Modified Intent-to-Treat Population

Figure 25: Correlation of Total Viral Shedding and Baseline Antibody Titer by Assay Type, Modified Intent-to-Treat Population

[Implementation Note: The scatter plots should be presented in a single figure with separate panels for Hemagglutination Inhibition, Neuraminidase Inhibition, and Microneutralization Antibody assays. The spearman correlation should be calculated over all subjects and annotated within each panel header after the visit label as "r=0.xx, p=0.xx". The y-axis should be labeled "Log (AUC)" and the x-axis "Baseline Antibody (Titer)".]

Figure 26: Frequency of Positive Viral Shedding Results by Study Day, Safety Population

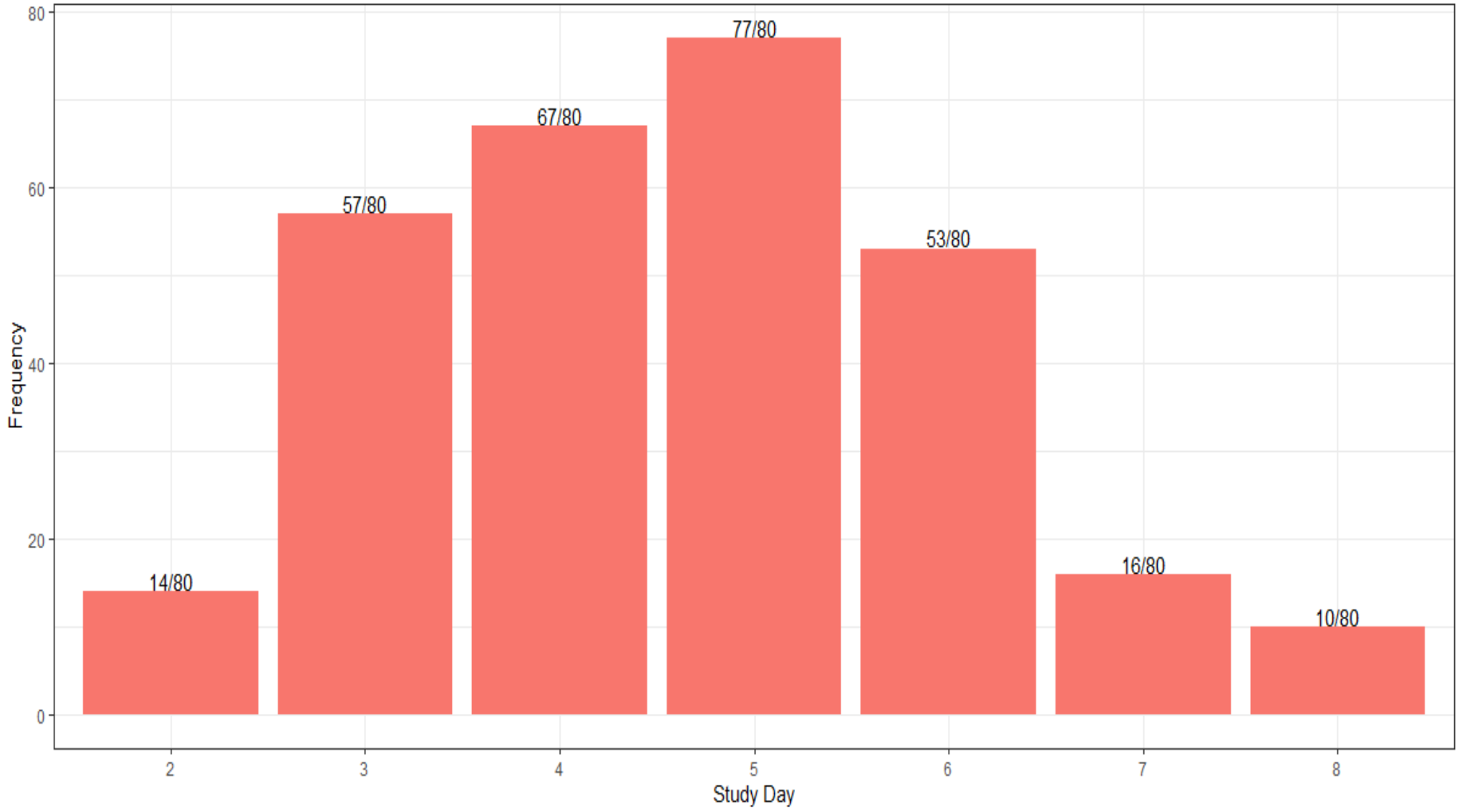
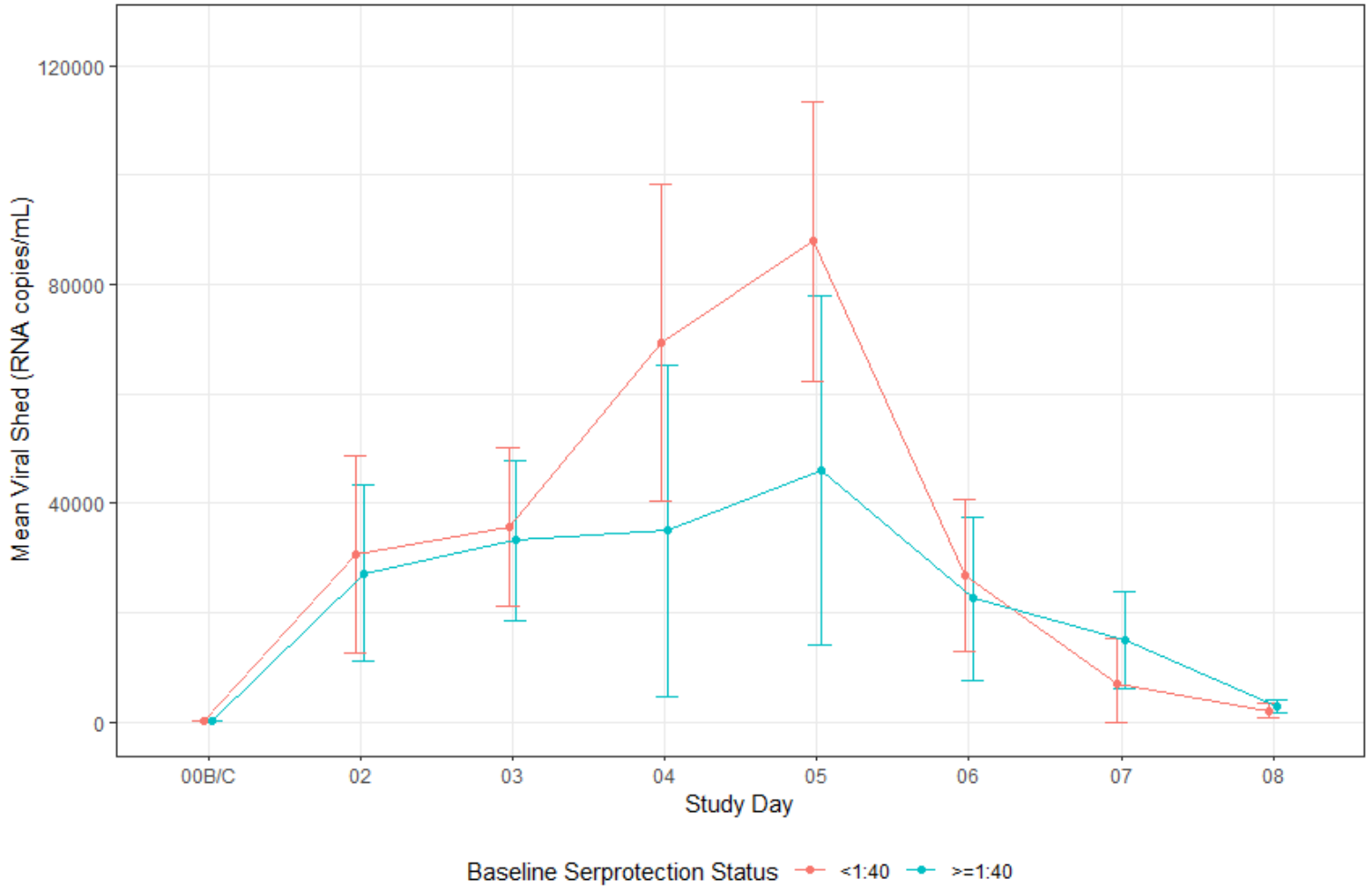


Figure 27: Mean Viral Shedding by Baseline Hemagglutination Inhibition Seroprotection Status, Symptom Status, and Study Day, Safety Population

[Implementation Note: Figure should be paneled by Symptom Status ("Symptomatic" vs "Asymptomatic").]

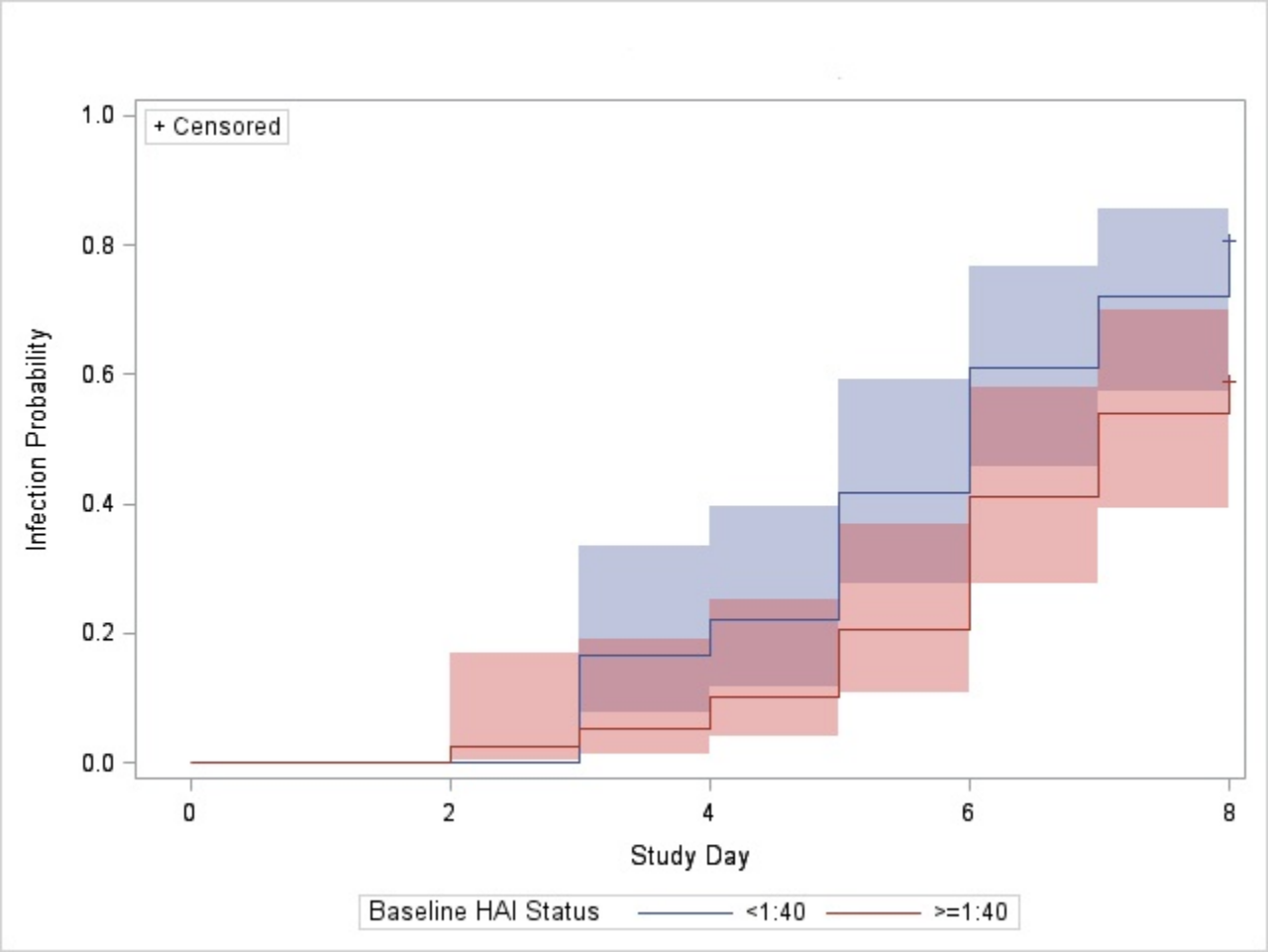


Figures with similar format:

Figure 28: Mean Viral Shedding by Baseline Neuraminidase Inhibition Seroprotection Status, Symptom Status, and Study Day, Safety Population

Figure 29: Mean Viral Shedding by Baseline Microneutralization Seroprotection Status, Symptom Status, and Study Day, Safety Population

Figure 30: Kaplan-Meier Curves for Time to Viral Shedding by Baseline Hemagglutination Inhibition Seroprotection Status, Modified Intent-to-Treat Population

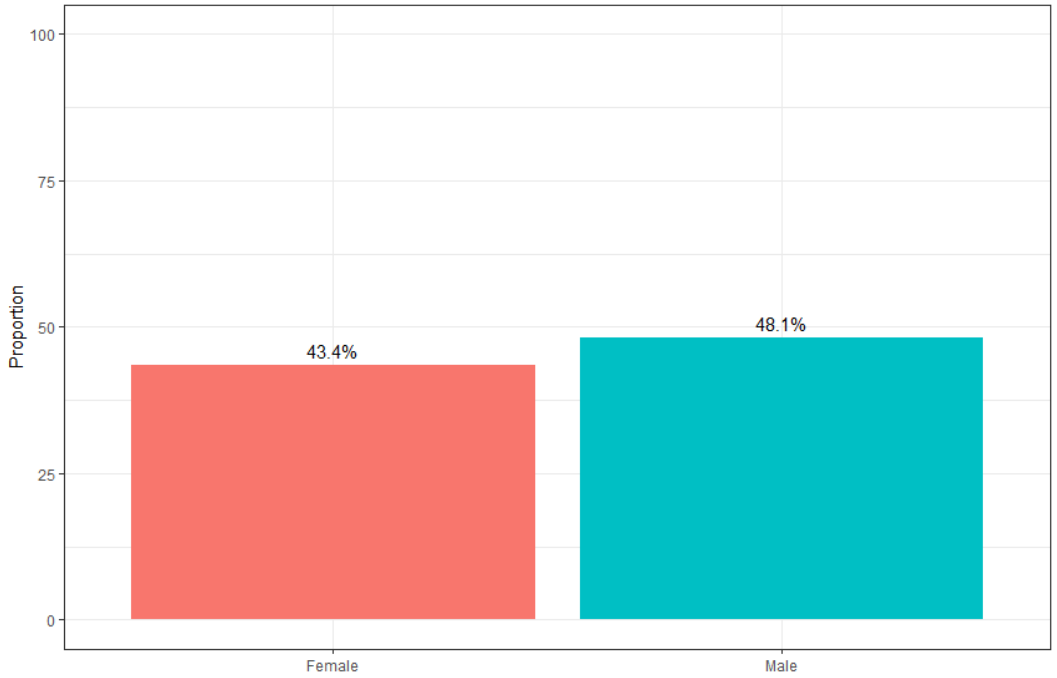


Figures with similar format:

Figure 31: Kaplan-Meier Curves for Time to Viral Shedding by Baseline Neuraminidase Inhibition Seroprotection Status, Modified Intent-to-Treat Population

Figure 32: Kaplan-Meier Curves for Time to Viral Shedding by Baseline Microneutralization Seroprotection Status, Modified Intent-to-Treat Population

Figure 33: Proportion of Subjects with Mild-to-Moderate Influenza Disease by Sex, Modified Intent-to-Treat Population



Figures with similar format:

Figure 34: Proportion of Subjects with Mild-to-Moderate Influenza Disease by Receipt of Prior Seasonal Vaccination, Modified Intent-to-Treat Population

Figure 35: Proportion of Subjects with Mild-to-Moderate Influenza Disease by Age Category, Modified Intent-to-Treat Population

Figure 36: Proportion of Mild-to-Moderate Influenza Disease by Age Category, Sex, and Receipt of Prior Seasonal Vaccination, Safety Population

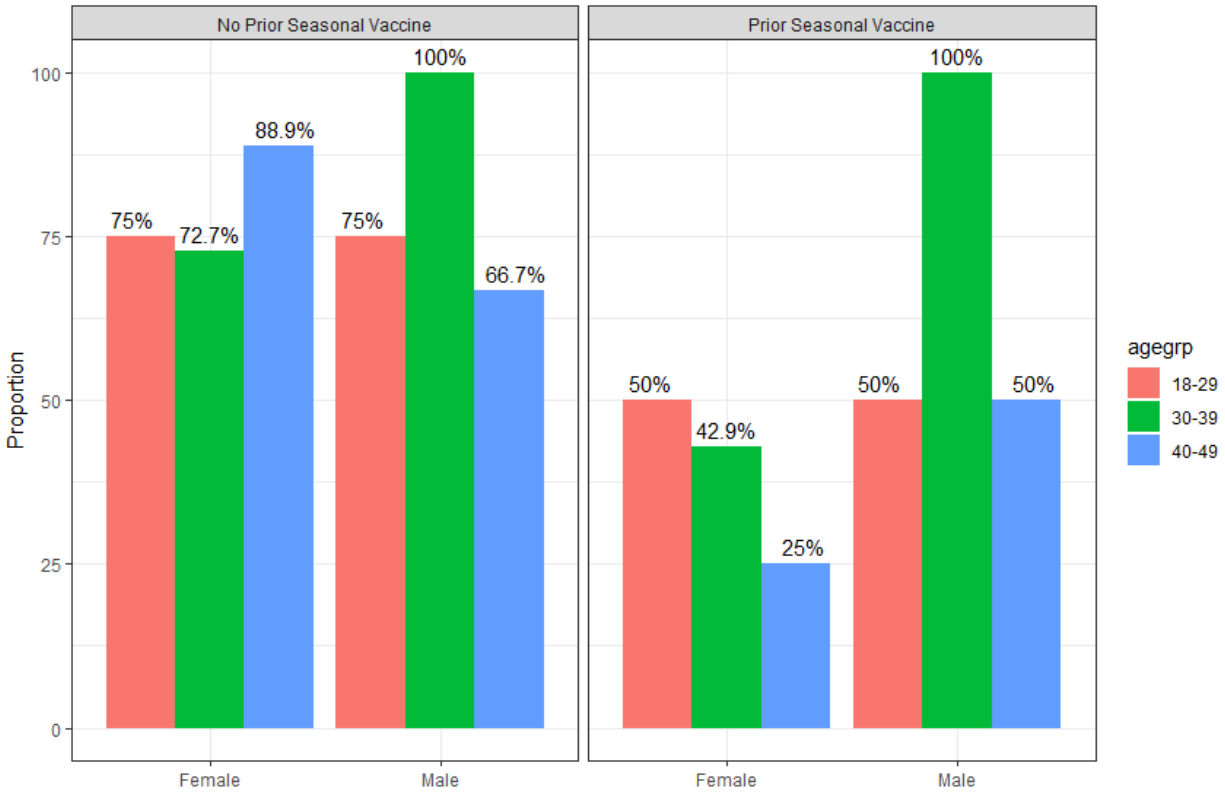
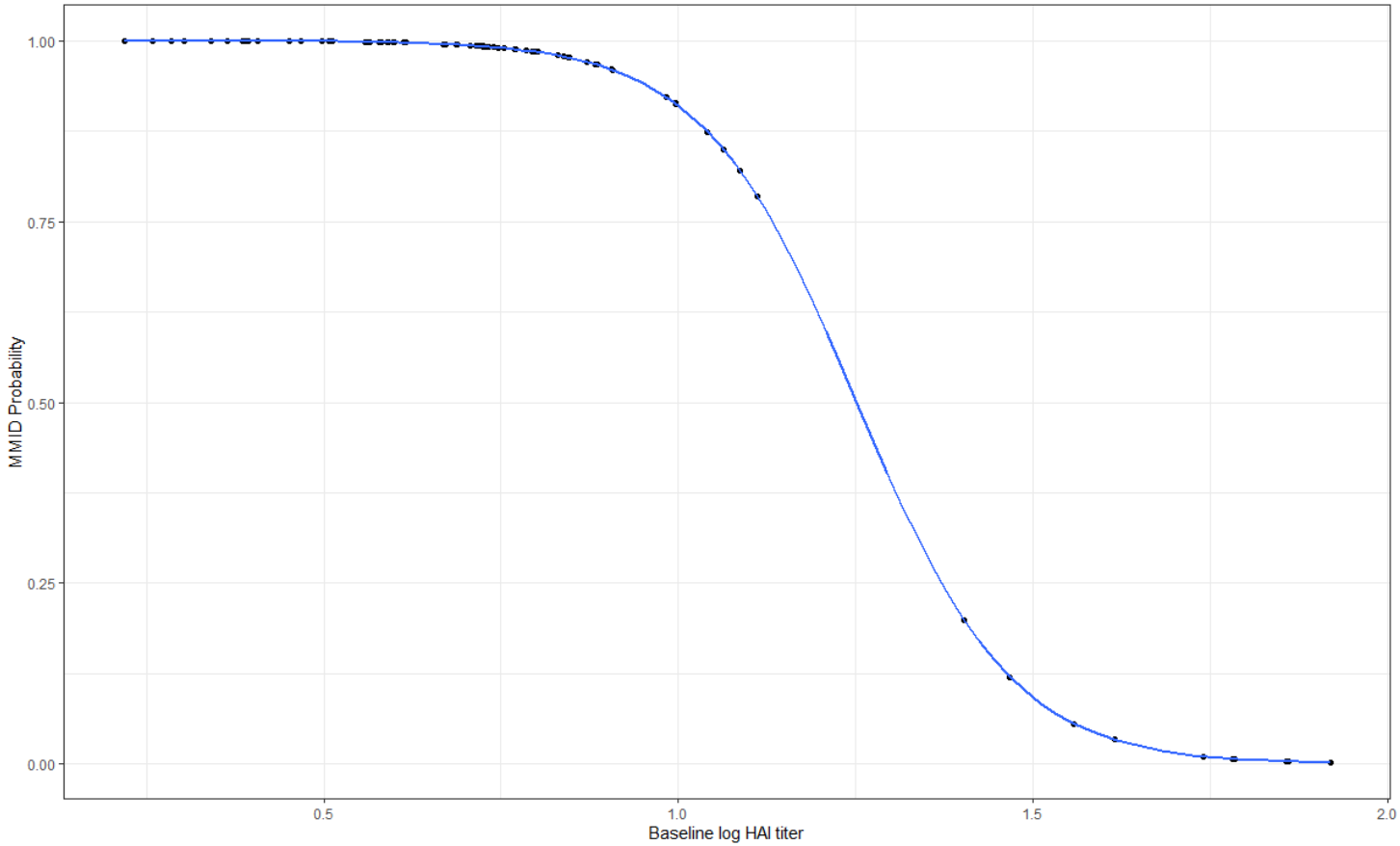


Figure 37: Predicted Probabilities of Mild-to-Moderate Influenza Disease According to Baseline Hemagglutination Inhibition Antibody Against A/Bethesda/MM2/H1N1 – Univariable Model, Modified Intent-to-Treat Population



Figures with similar format:

Figure 38: Predicted Probabilities of Mild-to-Moderate Influenza Disease According to Baseline Neuraminidase Inhibition Antibody Against A/Bethesda/MM2/H1N1 – Univariable Model, Modified Intent-to-Treat Population

Figure 39: Predicted Probabilities of Mild-to-Moderate Influenza Disease According to Baseline Microneutralization Antibody Against A/Bethesda/MM2/H1N1 – Univariable Model, Modified Intent-to-Treat Population

14.3.1.1 Solicited Symptoms

Figure 40: Maximum Extent of FLU-PRO Solicited Symptoms by Study Day Post Challenge, Safety Population

[Implementation Note: The figure can be shaded with the default coloring instead of the patterns. The bar chart should be presented in a single-paneled figure labeled as "All Subjects". Label y-axis "Study Day".]

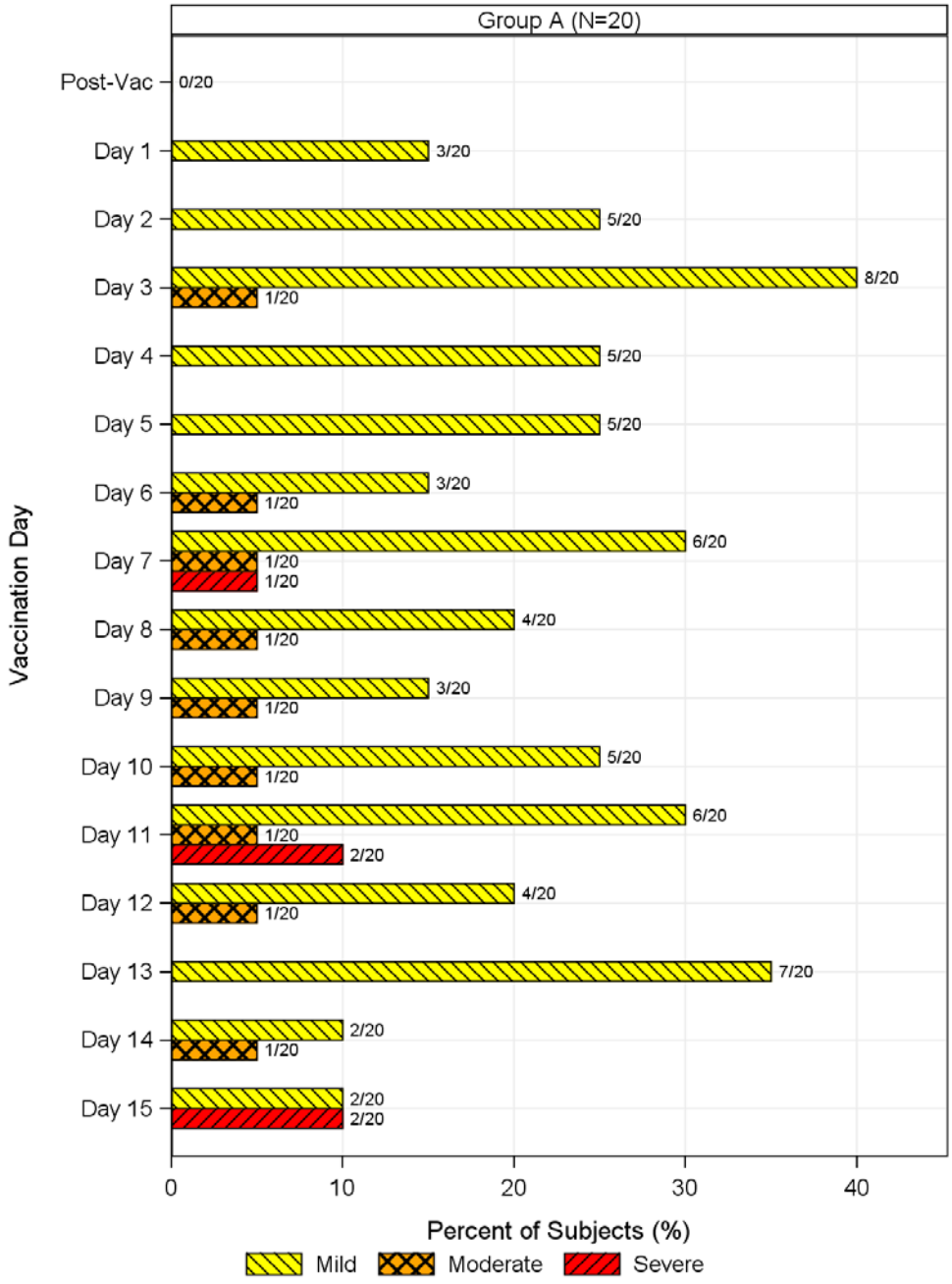
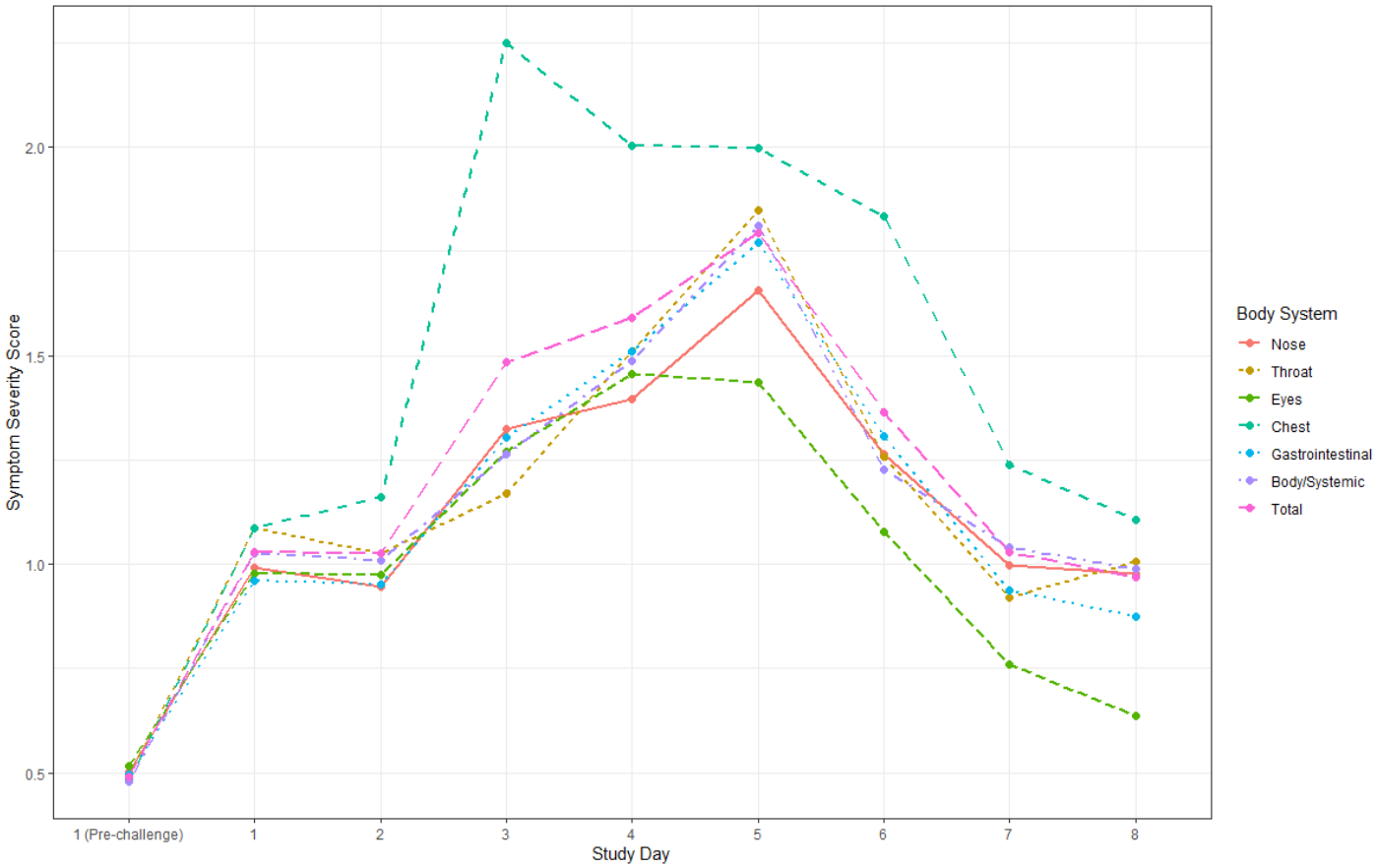


Figure 41: Mean Symptom Severity Score by Body System and Study Day, Safety Population

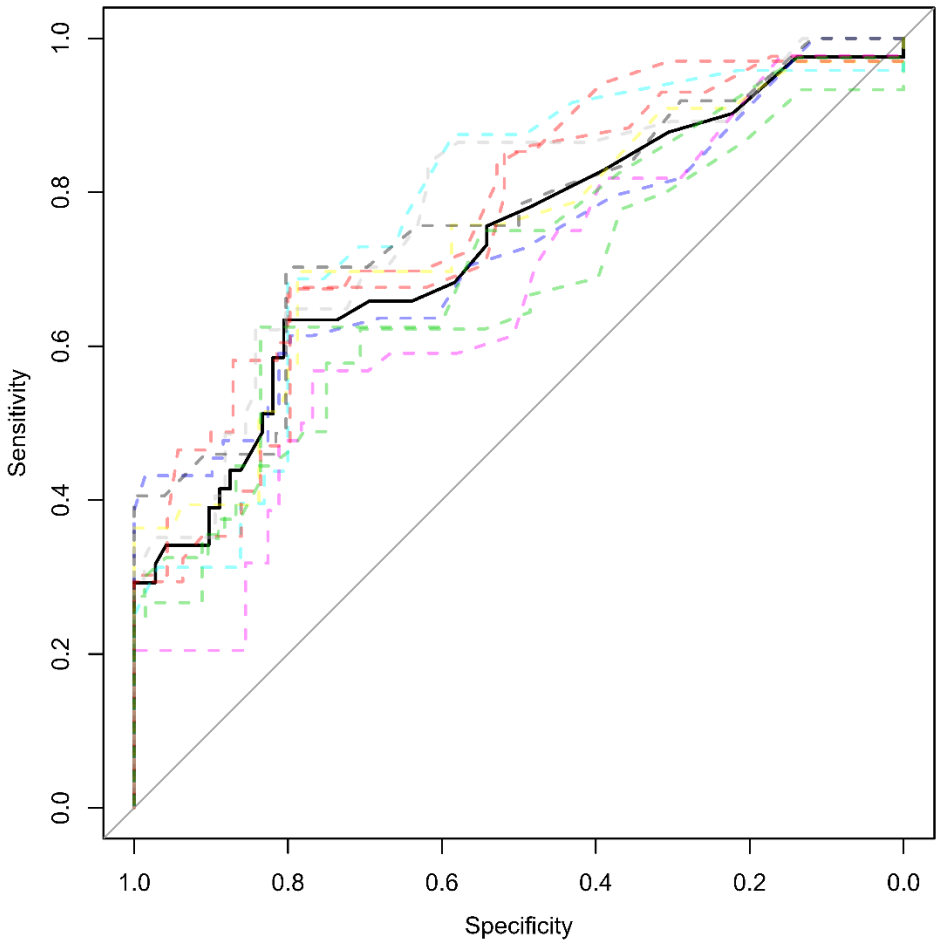


Figures with similar format:

Figure 42: Mean Symptom Severity Score by Body System, Study Day and Viral Shedding Status, Safety Population

[Implementation Note: Figure should be paneled by Viral Shedding Status as of Study Day 8 (Positive versus Negative).]

Figure 43: Influenza Symptom Scoring Algorithm – Predictive Accuracy Across 10-Fold Cross-Validation, Modified Intent-to-Treat Population



Note: Each dotted line indicates the performance for one iteration of the 10-fold CV, and the solid line gives the internal performance of the mean model on the entire dataset.

14.3.1.2 Unsolicited Adverse Events

Figure 44: Incidence of Unsolicited Adverse Events by MedDRA System Organ Class and Severity, Safety Population

[Implementation Note: The SOCs should be sorted in descending incidence, with Any SOC at the top. The figure can be shaded with the default coloring instead of the patterns. The bar chart should be presented in a single-paneled figure labeled as "All Subjects".]

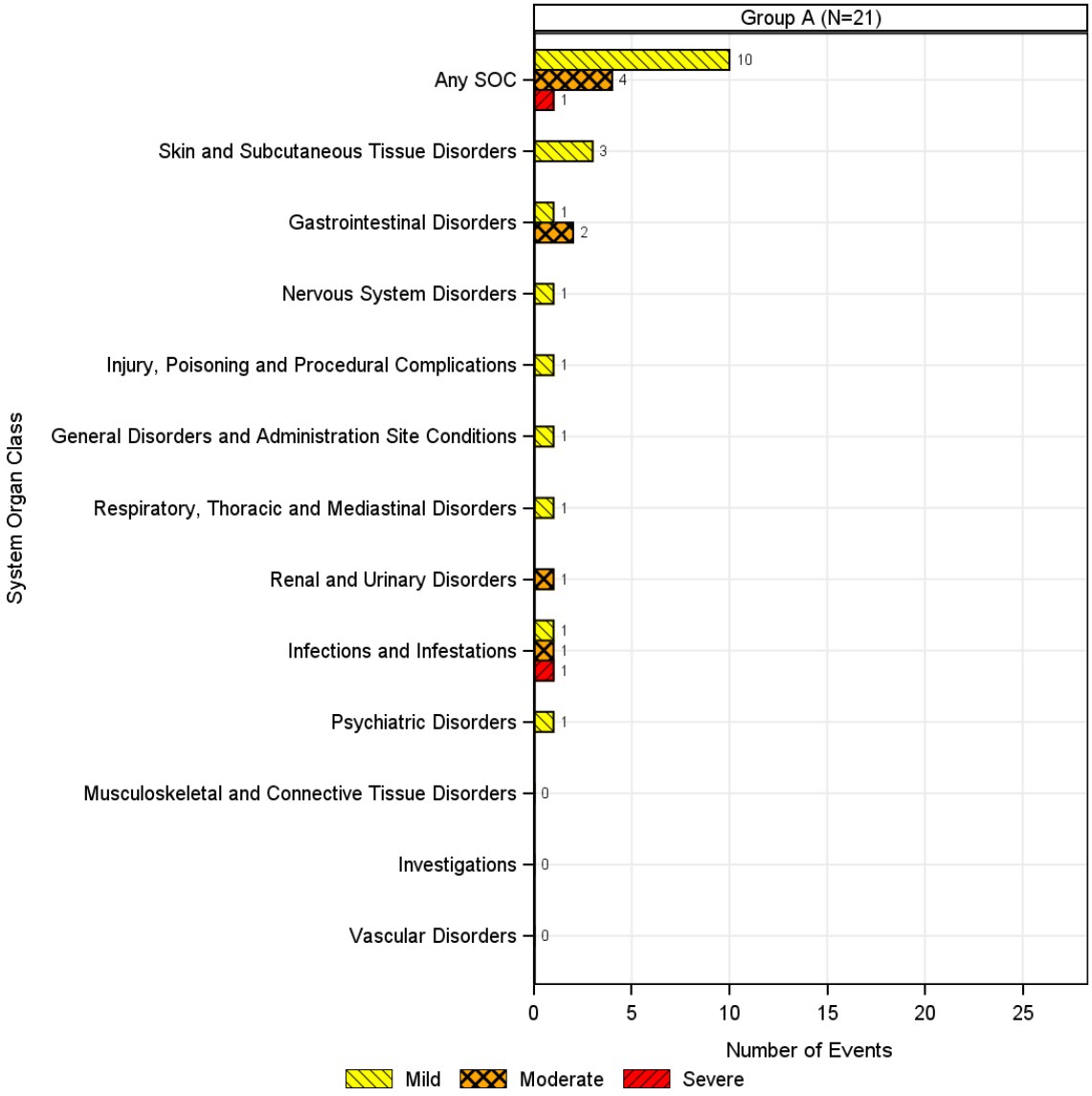


Figure 45: Frequency of Unsolicited Adverse Events by MedDRA System Organ Class and Relationship to Study Challenge, Safety Population

[Implementation Note: A Generic figure is shown below. The bar chart should be presented in a single-paneled figure labeled as "All Subjects". This figure includes serious and non-serious unsolicited adverse events.]

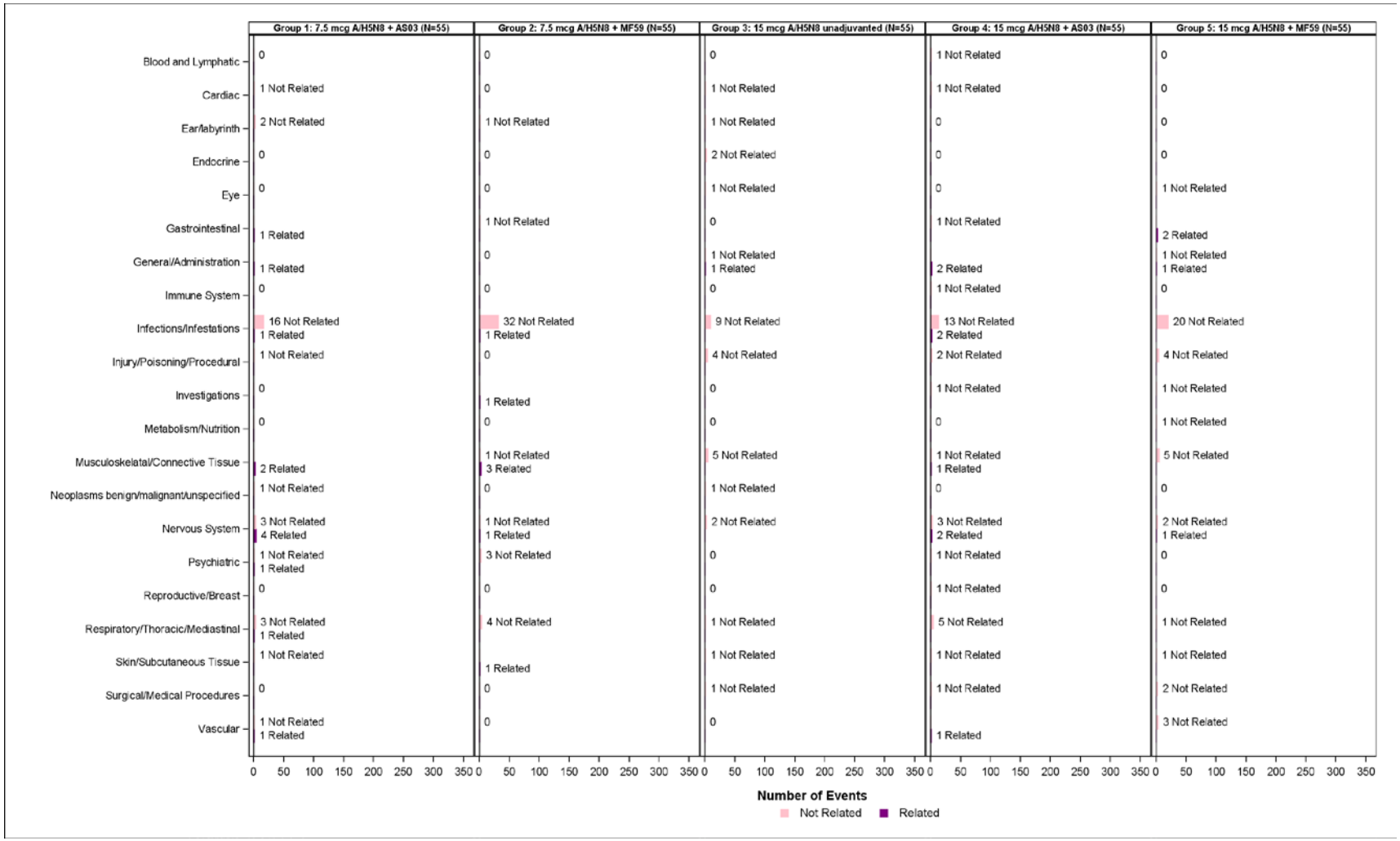
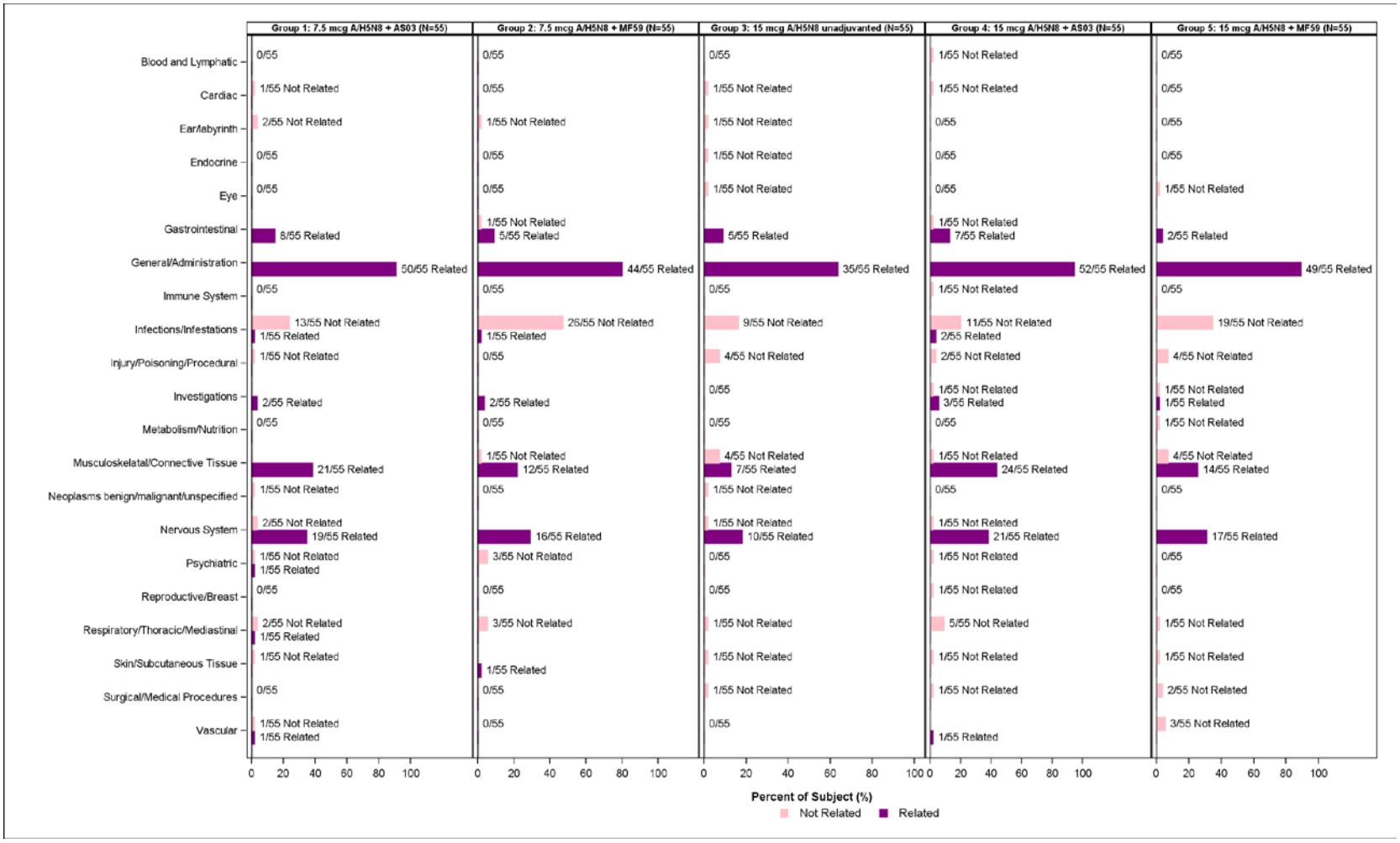


Figure 46: Incidence of Unsolicited Adverse Events by MedDRA System Organ Class and Relationship to Study Challenge, Safety Population

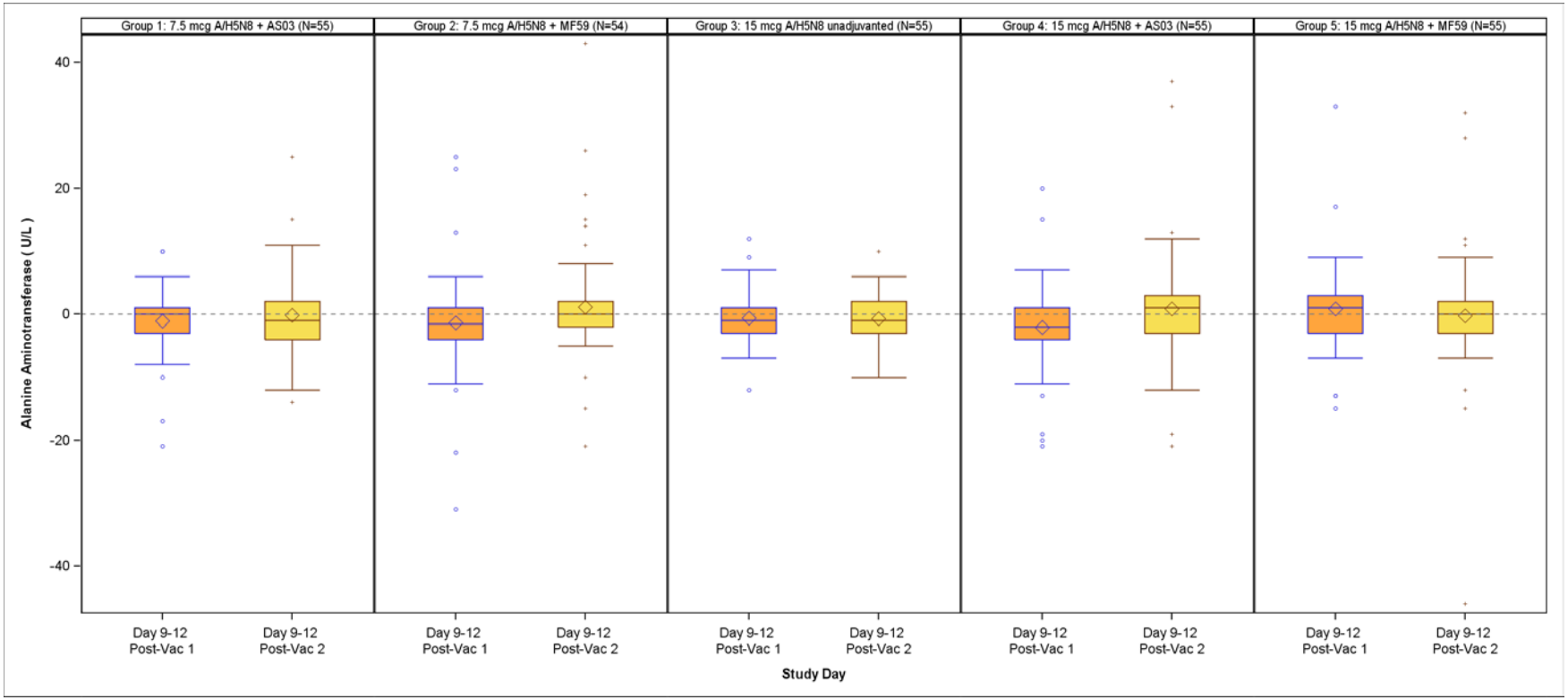
[Implementation Note: A Generic figure is shown below. The bar chart should be presented in a single-paneled figure labeled as "All Subjects". This figure includes serious and non-serious unsolicited adverse events.]



14.3.5 Displays of Laboratory Results

Figure 47: Laboratory Results by Scheduled Visits: Mean Changes from Baseline by Laboratory Parameter – Biochemistry Parameters, Safety Population

[Implementation Note: Plot should be generated in a single image file with panels for each parameter: Alanine Transaminase and Creatinine for biochemistry parameters, then White Blood Cells, Absolute lymphocyte count, Hemoglobin, and Platelets for hematology parameters. Box-plots should be plotted for the baseline change at each post-challenge study day (Day 2, 4, and 8). Y-axis should be labeled "[Parameter] Change from baseline ([units]).]



Figures with similar format:

Figure 48: Laboratory Results: Mean Changes from Baseline by Study Day and Study Arm – Hematology Parameters, Safety Population

APPENDIX 3. LISTINGS MOCK-UPS

LISTINGS

Listing 1: Listing of Subjects Receiving Investigational Product

(not included in SAP, but this is a placeholder for the CSR)

16.2 Database Listings by Subject

16.2.1 Discontinued Subjects

Listing 2: Subjects Discharged Early or Terminated Early

Subject ID	Category	Reason for Early Discharge or Early Termination	Study Day
	Early Termination		
	Early Discharge		

16.2.2 Protocol Deviations

Listing 3: Subject-Specific Protocol Deviations

Subject ID	DV Number	Deviation	Deviation Category	Study Day	Reason for Deviation	Deviation Resulted in AE?	Deviation Resulted in Subject Termination?	Deviation Affected Product Stability?	Deviation Resolution	Comments

Listing 4: Non-Subject-Specific Protocol Deviations

Site	Start Date	Deviation	End Date	Reason for Deviation	Deviation Resulted in Subject Termination?	Deviation Affected Product Stability?	Deviation Category	Deviation Resolution	Comments

16.2.3 Subjects Excluded from the Efficacy Analysis

Listing 5: Subjects Excluded from Analysis Populations

Subject ID	Analyses in which Subject is Included	Analyses from which Subject is Excluded	Results Available?	Reason Subject Excluded
	[e.g., Safety, mITT, PP]	[e.g., Safety, mITT, PP, Day x]		

Note: "Yes" in the "Results available" column indicates that available data were removed from the analysis. "No" indicates that no data were available for inclusion in the analysis.

16.2.4 Demographic Data

Listing 6: Demographic Data

Subject ID	Sex	Age at Enrollment (years)	Ethnicity	Race	BMI	Seasonal Influenza Vaccine Received (2018-2019)	Non-Seasonal Influenza Vaccine Receipt (Subtype)
						No	Yes (A/H1N1)
						Yes	

Listing 7: Pre-Existing and Concurrent Medical Conditions

Subject ID	MH Number	Medical History Term	Condition Start Day	Condition End Day	MedDRA System Organ Class	MedDRA Preferred Term

16.2.5 Compliance and/or Drug Concentration Data (if available)

Not applicable for this study.

16.2.6 Clinical and Immunogenicity Response Data

Listing 8: Individual Efficacy/Immunogenicity Response Data - Hemagglutination Inhibition against A/Bethesda/MM2/H1N1

Subject ID	Planned Study Day	Actual Study Day	Titer Replicate 1	Titer Replicate 2	Titer Replicate 3	Titer Replicate 4	Titer Replicate 5

Listings with similar format:

Listing 9: Individual Immunogenicity Response Data – Neuraminidase Inhibition against A/Bethesda/MM2/H1N1

Listing 10: Individual Immunogenicity Response Data – Microneutralization against A/Bethesda/MM2/H1N1

Listing 11: Nasopharyngeal (NP) Swab Results - Influenza

[Implementation Note: This listing includes all viral shedding NP swab assessments. Results should be indicated as "Positive" or "Negative" for Qualitative Multiplex assays. Quantitative RT-PCR results should be included in parentheses after the numeric result, e.g., xxx.x (Positive/Negative).]

Subject ID	Planned Study Day	Actual Study Day	Quantitative RT-PCR Result (RNA copies/mL)[a]	Qualitative Multiplex Virus Assay				
				Result 1 (Subtype)	Result 2 (Subtype)	Result 3 (Subtype)	Result 4 (Subtype)	Result 5 (Subtype)

[a] Positive is defined as greater than or equal to the lower limit of detection (LOD). The limit of detection (LOD) is X copies per reaction or Y copies per mL.

Listing 12: Solicited Mild-to-Moderate Influenza Disease Symptoms with Severity Scores of 3 or 4

Subject ID	Study Day	Assessment[a]	Symptom	Severity Score

[a] FP = Symptoms reported by subject on the FLU-PRO Survey Instrument and Validation Diary; LB = Clinical laboratory assessments used in the MMID definition.

16.2.7 Adverse Events

Listing 13: Unsolicited Adverse Events

Adverse Event	No. of Days Post Associated Dose (Duration)	Severity	SAE?	Relationship to Study Challenge	In Not Related, Alternative Etiology	Subject Discontinued Due to AE	Outcome	MedDRA System Organ Class	MedDRA Preferred Term
Comments: [Verbatim comments from the clinical site are displayed here]									
Subject ID: , AE Number:									
Comments: [Verbatim comments from the clinical site are displayed here]									
Note: For additional details about SAEs, refer to "Listing of Serious Adverse Events" table.									

16.2.8 Individual Laboratory Measurements

Listing 14: Clinical Laboratory Results – Chemistry

Subject ID	Planned Study Day	Actual Study Day	Sex	Age (years)	Laboratory Parameter (Units)	Result (Severity Grade)	Reference Range Low	Reference Range High

Listing 15: Clinical Laboratory Results – Hematology

Subject ID	Planned Study Day	Actual Study Day	Sex	Age (years)	Laboratory Parameter (Units)	Result (Severity Grade)	Reference Range Low	Reference Range High

16.2.9 Vital Signs and Physical Exam Findings

Listing 16: Vital Signs

[Implementation Note: This listing includes all vital sign assessments, scheduled and unscheduled. The severity should be included in parentheses after the result for abnormal assessments, e.g., 100.7 (Mild).]

Subject ID	Planned Study Day	Actual Study Day	Assessment Time	Temperature (°C)	Systolic Blood Pressure (mmHg)	Diastolic Blood Pressure (mmHg)	Heart Rate (beats/min)	Respiratory Rate (breaths/min)	Saturation of Peripheral Oxygen (%)	Weight (kg)	Height (cm)

Listing 17: Physical Exam Findings

Subject ID	Planned Study Day	Actual Study Day	Body System	Abnormal Finding	Reported as an AE? (AE Description; Number)	Reported as a Solicited Symptom?	Appearance of Illness	Abnormal Tympanic Membrane Appearance	Presence of Pharyngeal Erythema	Abnormal Lung Auscultation	Cough Present During Exam

Listing 18: Electrocardiogram Findings

Subject ID	Planned Study Day	Actual Study Day	Study Day Relative to Challenge	Time (24- hour clock)	Position of Subject	Overall Result	If Abnormal, Was the ECG Clinically Significant?	Comments

16.2.10 Concomitant Medications

Listing 19: Concomitant Medications

Subject ID	CM Number	Medication	Medication Start Day	Medication End Day	Indication	Taken for an AE? (AE Description; Number)	Taken for a condition on Medical History? (MH Description; Number)	ATC Level 1 (ATC Level 2)

16.2.11 Pregnancy Reports

Listing 20: 16.2.11.1: Pregnancy Reports – Maternal Information

Subject ID	Pregnancy Number	Study Day Corresponding to Estimated Date of Conception	Source of Maternal Information	Pregnancy Status	Mother's Pre-Pregnancy BMI	Mother's Weight Gain During Pregnancy (LB)	Tobacco, Alcohol, or Drug Use During Pregnancy?	Medications During Pregnancy?	Maternal Complications During Pregnancy?	Maternal Complications During Labor, Delivery, or Post-Partum?

Note: Maternal Complications are included in the Adverse Event listing. Medications taken during pregnancy are included in the Concomitant Medications Listing.

Listing 21: 16.2.11.2: Pregnancy Reports – Gravida and Para

			Live Births												
Subject ID	Pregnancy Number	Gravida	Extremely PB[a]	Very Early PB[a]	Early PB[a]	Late PB[a]	Early TB[b]	Full TB[b]	Late TB[b]	Post TB[b]	Still Births	Spontaneous Abortion/ Miscarriage	Elective Abortions	Therapeutic Abortions	Major Congenital Anomaly with Previous Pregnancy?

Note: Gravida includes the current pregnancy, para events do not.

[a] Preterm Birth

[b] Term Birth

Listing 22: 16.2.11.3: Pregnancy Reports – Live Birth Outcomes

Subject ID	Pregnancy Number	Fetus Number	Pregnancy Outcome (for this Fetus)	Fetal Distress During Labor and Delivery?	Delivery Method	Gestational Age at Live Birth (weeks)	Size for Gestational Age	Apgar Score, 1 minute	Apgar Score, 5 minutes	Cord pH	Congenital Anomalies?	Illnesses/ Hospitalizations within 1 Month of Birth?
Note: Congenital Anomalies are included in the Adverse Event listing.												

Listing 23: 16.2.11.4: Pregnancy Reports – Still Birth Outcomes

Subject ID	Pregnancy Number	Fetus Number	Pregnancy Outcome (for this Fetus)	Fetal Distress During Labor and Delivery?	Delivery Method	Gestational Age at Still Birth (weeks)	Size for Gestational Age	Cord pH	Congenital Anomalies?	Autopsy Performed?	If Autopsy, Etiology for Still Birth Identified?

Listing 24: 16.2.11.5: Pregnancy Reports – Spontaneous, Elective, or Therapeutic Abortion Outcomes

Subject ID	Pregnancy Number	Fetus Number	Pregnancy Outcome (for this Fetus)	Gestational Age at Termination (weeks)	Abnormality in Product of Conception?	Reason for Therapeutic Abortion