Official Title: **Protocol VIS410-203, Phase 2b, Multicenter, Randomized, Double-blind, Controlled Study to Evaluate the Efficacy and Safety of Intravenous VIS410 in Addition to Oseltamivir (Tamiflu®) Compared with Oseltamivir Alone in Hospitalized Adults with Influenza A Infection Requiring Oxygen Support**

NCT Number: **NCT03040141**

Document Date: **19 April 2018**

Visterra

Clinical Study Protocol

Title: Phase 2b, Multicenter, Randomized, Double-blind, Controlled Study to Evaluate the Efficacy and Safety of Intravenous VIS410 in Addition to Oseltamivir (Tamiflu®) Compared with Oseltamivir Alone in Hospitalized Adults with Influenza A Infection Requiring Oxygen Support

Product	VIS410
Protocol Number	VIS410-203
EudraCT Number	2016-004009-15
Clinical Phase	2b
Clinical Indication	Influenza A infection
Issue Date (Version)	19 April 2018 (Version 3.0)

Sponsor	Visterra, Inc. 275 Second Avenue, 4th Floor Waltham, MA 02451 United States of America
Sponsor Representative	[redacted]

CONFIDENTIALITY STATEMENT

This study will be conducted in compliance with this protocol, the ICH Note for Guidance on Good Clinical Practice (CPMP/ICH/135/95), and with the applicable regulatory requirement(s).

SIGNATURES

Signature of Sponsor Representative

Title: Phase 2b, Multicenter, Randomized, Double-blind, Controlled Study to Evaluate the Efficacy and Safety of Intravenous VIS410 in Addition to Oseltamivir (Tamiflu®) Compared with Oseltamivir Alone in Hospitalized Adults with Influenza A Infection Requiring Oxygen Support

Name: [redacted]

This Clinical Study Protocol has been reviewed and approved by the Sponsor in order to ensure compliance with Good Clinical Practice.

Signature:
Date: [redacted]

Signature of Investigator

Title: Phase 2b, Multicenter, Randomized, Double-blind, Controlled Study to Evaluate the Efficacy and Safety of Intravenous VIS410 in Addition to Oseltamivir (Tamiflu®) Compared with Oseltamivir Alone in Hospitalized Adults with Influenza A Infection Requiring Oxygen Support

Name:
Affiliation:
Address:

I have read and understood all sections of the protocol entitled, "Phase 2b, Multicenter, Randomized, Double-blind, Controlled Study to Evaluate the Efficacy and Safety of Intravenous VIS410 in Addition to Oseltamivir (Tamiflu®) Compared with Oseltamivir Alone in Hospitalized Adults with Influenza A Infection Requiring Oxygen Support."

I agree to supervise all aspects of the protocol and to conduct the clinical investigation in accordance with the final protocol, the International Conference on Harmonisation Tripartite Guideline: Good Clinical Practice E6 (R1) and all applicable government regulations. I will not make changes to the protocol before consulting with Visterra, Inc., or implement protocol changes without independent ethics committee approval except to eliminate an immediate risk to subjects. I agree to administer study treatment only to subjects under my personal supervision or the supervision of a sub-Investigator.

I will not supply the investigational product to any person not authorized to receive it. Confidentiality will be protected. Subject identity will not be disclosed to third parties or appear in any study reports or publications.

I will not disclose information regarding this clinical investigation or publish results of the investigation without authorization from Visterra, Inc.

Signature:
Date:

SYNOPSIS

Name of Company:	Visterra, Inc.	(for national authority only)
Name of the Finished Product:	Not applicable	
Name of the Active Substance:	VIS410	
Study Title:	Phase 2b, Multicenter, Randomized, Double-blind, Controlled Study to Evaluate the Efficacy and Safety of Intravenous VIS410 in Addition to Oseltamivir (Tamiflu®) Compared with Oseltamivir Alone in Hospitalized Adults with Influenza A Infection Requiring Oxygen Support	
Protocol Number:	VIS410-203	
EudraCT Number:	2016-004009-15	
Clinical Indication:	Influenza A infection	
Clinical Phase:	2b	
Number of Clinical Sites:	Approximately 140 sites worldwide	
Number of Subjects:	Approximately 120	

Study Objectives

Primary Efficacy Objective

- Evaluation of the effect of 2 dose levels of VIS410 + oseltamivir on clinical outcome as assessed by comparison of clinical status ordinal scale Day 7 scores between treatment groups, and between all VIS410 recipients versus placebo.

Primary Safety Objective

- Safety and tolerability of 2 dose levels of a single intravenous (IV) dose of VIS410 when administered in combination with oseltamivir in hospitalized subjects with influenza A infection.

Secondary Objectives

- Among patients requiring supplemental oxygen therapy at the time of enrollment with baseline room air oxygen saturation of ≤92%, time to cessation of oxygen (O_2) support resulting in stable oxygen saturation (SpO_2) by pulse oximetry. Stable SpO_2 is defined as two consecutive SpO_2 values of > 92% on room air that are at least 8 hours apart.
- For any patient requiring supplemental oxygen therapy at the time of enrollment (regardless of oxygen saturation), time to cessation of oxygen support.
- Evaluate the effect of 2 dose levels of VIS410 + oseltamivir vs oseltamivir alone on the following parameters:
  - Viral load in upper respiratory samples
  - Time to clinical response
  - Time to cessation of ventilator support
  - Time to resumption of normal activities
  - All-cause and attributable 14-, 28-, and 56-day mortality
  - Clinical status ordinal scale mean area under the curve for Days 1-7 and Days 1-14 using linear numeric scores for the ordinal categories.
  - Comparison of clinical status ordinal scale scores for selected individual days (ie, Days 3, 4, 5, and 6)
  - Comparison of clinical status ordinal scale scores using modified ordinal scale criteria (ie, pooling of selected severity criteria scores)
  - Comparison of discrete ordinal scale parameters, including days of ventilator support, days in intensive

care, and duration of hospitalization
- Healthcare resource utilization
- Analysis of time to alleviation of signs and symptoms of influenza in the subset of subjects able to complete the Influenza Patient Reported Outcomes (FluPRO) Questionnaire at baseline and post-dose by Kaplan Meier analysis
- Proportion of subjects with new documented bacterial pneumonia/superinfection
- Proportion of subjects with influenza-related complications

- Pharmacokinetics of VIS410 in serum
- Immunogenicity of VIS410
- Emergence of resistance to VIS410 and oseltamivir

Exploratory Objectives

- Evaluate the pharmacokinetics of VIS410 from nasopharyngeal secretions and tracheal aspirate (ventilated subjects only)
- Assess the effects of VIS410 on viral load in tracheal aspirate (ventilated subjects only)
- Assess correlations between virology, safety, VIS410 dose, pharmacokinetics, viral shedding, immunology, signs and symptoms of influenza, and other endpoints
- Assess the anti-influenza immune response

Study Design

This is a Phase 2b, multicenter, randomized, double-blind, controlled study comparing the efficacy and safety of 2 dose levels of VIS410 (2000 and 4000 mg) in combination with oseltamivir vs oseltamivir alone in hospitalized subjects with influenza A infection requiring oxygen support. Subjects will be randomly assigned at a ratio of 1:1:1 using a permuted block randomization method to receive VIS410 at a dose of 2000 mg (low dose) or 4000 mg (high dose) or placebo (0.9% sodium chloride), administered as a single IV infusion over 2 hours. Randomization will be stratified by presence or absence of positive pressure ventilation (PPV) at baseline to ensure an even distribution across treatment arms. All subjects will receive oseltamivir 75 mg twice daily (BID) for 5 days (total of 10 doses) as part of standard of care (SOC). Treatment with oseltamivir may be extended for up to 5 additional days (up to 20 doses of total therapy) if clinical symptoms warrant further treatment as assessed by the Investigator.

Note: based on SOC, subjects may have started oseltamivir therapy prior to randomization.

All subjects will also receive a pretreatment regimen of diphenhydramine 50 mg PO or IV plus either ibuprofen 400 mg PO or acetylsalicylic acid 320-325 mg PO approximately 60 minutes before infusion. Approximately 120 evaluable subjects (40/arm) with confirmed influenza A infection will be treated.

Subjects admitted to the hospital within 5 days of onset of initial symptoms who require supplemental oxygen will undergo a rapid influenza test or a local polymerase chain reaction (PCR) test, fluorescent immunoassay (FIA) test, or enzyme-linked immunosorbent assay (ELISA) test to confirm influenza A infection. Subjects diagnosed with influenza A will undergo the screening procedures to confirm eligibility. Study assessments are outlined in Table 1 (Schedule of Assessments).

Eligible subjects will be randomized to receive either oseltamivir + VIS410 2000 mg, oseltamivir + VIS410 4000 mg, or oseltamivir + placebo (0.9% sodium chloride) at a ratio of 1:1:1 using a permuted block randomization method according to the table below:

Sample Size	Treatment	VIS410 Dose [a]	Oseltamivir Dose [b]	Infusion
n = 40	VIS410 + oseltamivir	2000 mg IV	75 mg administered BID for a minimum of 5 days	Infused over 120 minutes
n = 40	VIS410 + oseltamivir	4000 mg IV		
n = 40	Placebo [c] + oseltamivir	0 mg IV		

[a] Dose administered in 200 mL

[b] Dosage and administration should follow local prescribing information for oseltamivir based on renal function.

[c] 0.9% sodium chloride (200 mL).

Oseltamivir (Tamiflu®) will be provided by the Sponsor. Treatment with oseltamivir may be extended for up to 5 additional days (up to 20 doses of total therapy) if clinical symptoms \warrant further treatment as assessed by the Investigator. Note: based on SOC, subjects may have started oseltamivir therapy prior to randomization.

Study assessments are outlined in Table 1. Subjects discharged from the hospital prior to Day 14 will attend the applicable outpatient follow-up visit(s) (eg, Day 3, Day 5, Day 7, Day 14) per Table 1.

An independent data safety monitoring board (DSMB) will be established to review all available safety data after 30 subjects and again after approximately 70 subjects have completed Day 14 assessments. The assessment of safety will be determined from vital sign measurements, physical examinations, hematology, chemistry and urinalysis laboratory testing, 12-lead electrocardiograms (ECGs), use of concomitant medications, and review of adverse events (AEs). Based on the safety data provided, the DSMB will determine if it is acceptable to continue the study or reduce the dose as appropriate. Dosing will temporarily pause while the DSMB meets if there are 4 treatment-related serious adverse events (SAEs) or 4 severe gastrointestinal (GI) treatment–emergent adverse events (TEAEs) that require intervention, which is defined as requiring IV fluid and medication to decrease the frequency of diarrhea (ie, loperamide).

Additional DSMB reviews can occur throughout the trial as deemed necessary by the DSMB or Sponsor to ensure subject safety and well-being. Study enrollment and dosing will continue while the DSMB evaluates data.

Study Population

Number of Subjects

Approximately 120 evaluable subjects will be enrolled in 3 equal arms: VIS410 2000 mg, VIS410 4000 mg, and placebo.

Inclusion Criteria

Subjects meeting all of the following criteria are eligible to participate in this study:

1. Male and female subjects aged ≥ 18 years. For a country where the legal age of consent is >18 years old, the country requirements should be followed.
2. Test positive for influenza A by rapid antigen test or with another commercially available test on an adequate nasopharyngeal specimen in accordance with the manufacturer's instructions, or an acceptable local test, including PCR, FIA, or ELISA.
3. Onset of influenza symptoms no more than 5 days before VIS410/placebo infusion; symptoms may include cough, dyspnea, sore throat, fever, myalgias, headache, nasal symptoms (rhinorrhea, congestion), fatigue, diarrhea, anorexia, nausea, and vomiting.
4. Requirement for oxygen support including any positive pressure ventilation (PPV).
5. Women of childbearing potential must have a negative pregnancy test within 2 days prior to VIS410/placebo infusion.
6. Women should fulfill one of the following criteria:
   a. Post-menopausal: either amenorrhea ≥ 12 months or follicle stimulating hormone > 40 mIU/mL as documented in their medical history
   b. Surgically sterile; hysterectomy, bilateral oophorectomy, or tubal ligation

c. Women of childbearing potential participating in heterosexual sexual relations must be willing to use adequate contraception from screening until 60 days post-VIS410/placebo infusion (see Section 6.2).

7. Non-vasectomized (or vasectomized less than 6 months prior to dosing) male subjects who have a female partner of childbearing potential must use an effective birth control method (see Section 6.2) when having heterosexual intercourse, from screening until 60 days post-VIS410/placebo infusion.
8. Subject is able and willing to comply with study procedures, as per protocol.
9. Subject, or a legally acceptable representative, is able to understand the purpose and risks of the study and willing to give voluntary written informed consent.

Exclusion Criteria

Subjects meeting any of the following criteria are excluded from participation in this study:

1. Known or suspected intolerance or hypersensitivity to VIS410, oseltamivir, pretreatment medications (diphenhydramine, or to both ibuprofen and acetylsalicylic acid [ASA]), or closely related compounds (eg, other monoclonal antibodies).
2. Subjects who have received VIS410 in the past.
3. Subjects who have a history of receiving monoclonal antibody products within 3 months prior to VIS410/placebo dosing or planned administration of another monoclonal antibody during the study period.
4. Subjects who have taken more than 6 doses of an approved antiviral therapy for influenza within the prior 96 hours (eg, oral oseltamivir, inhaled zanamivir, IV peramivir, or oral ribavirin) between onset of symptoms and VIS410/placebo dosing.
5. Subjects with known co-infection with influenza B or other viral respiratory infections (eg, respiratory syncytial virus [RSV], parainfluenza viruses, respiratory adenoviruses).
6. Subjects with lung transplant or history of severe chronic lung disease, including cystic fibrosis or any condition requiring home oxygen therapy.
7. Subjects on extracorporeal membrane oxygenation at time of randomization.
8. Subjects with end-stage renal disease (ESRD) who are not undergoing hemodialysis.
9. Subjects with active graft-vs-host disease, hematopoietic stem cell transplant within the previous 90 days, or human immunodeficiency virus infection with a CD4 cell count of less than 200 per cubic millimeter.
10. High probability of mortality within 48 hours of randomization as determined by the Investigator.
11. Women who are pregnant, breast-feeding, or considering to become pregnant.
12. Subjects in whom nasopharyngeal swabbing is not possible.
13. Subjects weighing less than 45 kg.
14. Enrollment in any other investigational drug or device study, any disease or vaccine study within 30 days prior to Day 1 or within 5 half-lives of the investigational compound, whichever is longer.
15. Presence of any preexisting illness that, in the opinion of the Investigator, would place the subject at an unreasonably increased risk through participation in this study.
16. Subjects unable to comply with study protocol procedures and study visit schedules for whatever reason.
17. Known or suspected alcohol or drug abuse, that is, abuse of a level that would compromise the safety or cooperation of the subject in the opinion of the Investigator.

Test Product, Dose, Mode of Administration

VIS410 (2000 mg or 4000 mg) will be administered IV over 2 hours as a single 200-mL infusion, followed by a 25-mL (or volume equivalent to IV line volume if greater than 25 mL) saline flush to ensure all product is administered, in addition to oseltamivir (Tamiflu) 75 mg BID for total of 5 days (10 doses). For patients with renal insufficiency, refer to prescribing information for administration of oseltamivir. For patients unable to swallow, oseltamivir capsules can be opened and mixed with liquid for ease of administration.

Reference Product, Dose, Mode of Administration

Placebo (normal saline solution 0.9%) will be administered IV over 2 hours as a single 200-mL infusion, followed by a 25-mL (or volume equivalent IV line volume if greater than 25 mL) saline flush to ensure all product is administered, in addition to oseltamivir (Tamiflu) 75 mg BID for total of 5 days (10 doses). For patients with renal

insufficiency, refer to prescribing information for administration of oseltamivir.

VIS410 and Placebo Preparation

The required amount of VIS410 to be dosed (2000 mg or 4000 mg) will be diluted with normal saline up to a maximum total volume of 200 mL; for placebo subjects, 200 mL of normal saline will be prepared. Length of IV line will ideally be set for maximum volume of 25 mL, so that the 25-mL (or increased volume as noted above) saline flush following administration will ensure all VIS410/placebo has been administered. In the event that the infusion line volume is greater than 25 mL the post-administration saline flush should be increased to match the volume of the infusion line kit.

The VIS410/placebo infusion will be administered IV using a 0.22-µm in-line filter and will be controlled by a volumetric pump. Standard, uniform-length infusion lines will be used whenever possible, and microfilters will be provided by the Sponsor.

The infusion bag will be covered with an opaque sleeve in the pharmacy to maintain the study blind. The VIS410/placebo will be administered IV at a rate of 100 mL/h, over 2 hours. After the dose has been administered, the infusion will be followed by an appropriate volume saline flush as described above. The infusion time may be extended up to 4 hours at the Investigator's discretion based on local infusion site–related symptoms.

All subjects will be given a pretreatment regimen of PO or IV diphenhydramine 50 mg plus either ibuprofen 400 mg PO or acetylsalicylic acid 320-325 mg PO or crushed and given through the nasogastric tube approximately 60 minutes before IV infusion of VIS410/placebo.

Study Duration

The total study duration for each subject (screening through study exit) will be approximately 8 weeks (Day 56).

Study Endpoints

Primary Efficacy Endpoint

- The primary efficacy outcome analysis compares Day 7 clinical status ordinal scale scores between treatment groups, and between all VIS410 recipients versus placebo. Clinical status is measured daily for 14 days using the below seven-level ordinal scale, with the classifications presented from the worst clinical outcome to the best clinical outcome in descending order; for each day, subject status will be classified by the worst clinical outcome for which they qualify.
  - Death
  - ICU stay with mechanical ventilation
  - ICU stay without mechanical ventilation
  - Non-ICU hospitalization with supplemental oxygen
  - Non-ICU hospitalization without supplemental oxygen
  - Discharge with partial resumption of normal activities
  - Discharge with full resumption of normal activities

Primary Safety Endpoint

- The proportion of subjects with AEs and SAEs following administration of VIS410

Secondary Endpoints

The difference between VIS410 + oseltamivir and oseltamivir alone treatment groups in the following endpoints:

- Among patients requiring supplemental oxygen therapy at the time of enrollment with baseline room air oxygen saturation of ≤92%, time to cessation of oxygen (O_2) support resulting in stable oxygen saturation (SpO_2) by pulse oximetry. Stable SpO_2 is defined as two consecutive SpO_2 values of > 92% on room air that are at least 8 hours apart.
- For any patient requiring supplemental oxygen therapy at the time of enrollment (regardless of oxygen saturation), time to cessation of oxygen support
- Peak viral load, viral area under the concentration–time curve (AUC), duration of viral shedding, and time to resolution of viral load from nasopharyngeal swabs by $TCID_{50}$ and qRT-PCR
- Time to clinical response defined as resolution of at least 4 of 5 vital signs:

- Afebrile with core temperature ≤ 37.8°C, without use of antipyretics (oral ≤ 37.2°C)
- Oxygen saturation ≥ 95% on room air without support or a return to pre-infection status, if pre-infection status was < 95%
- Pulse rate ≤ 100/min
- Systolic blood pressure (SBP) ≥ 90 mm/Hg, without vasopressor use
- Respiratory rate ≤24 beats per minute

- Clinical status ordinal scale mean area under the curve for Days 1-7 and Days 1-14 using linear numeric scores for the ordinal categories.
- Comparison of clinical status ordinal scale scores for selected individual days (ie, Days 3, 4, 5, and 6)
- Comparison of clinical status ordinal scale scores using modified ordinal scale criteria (ie, pooling of selected severity criteria scores)
- Comparison of discrete ordinal scale parameters, including days of ventilator support, days in intensive care, and duration of hospitalization
- Number of days to resumption of normal activities
- All-cause and attributable mortality rates at Day 14, 28, and 56
- Total number of days in hospital and/or intensive care unit (ICU) from admission to discharge and rate of rehospitalization due to influenza A relapse/complication
- The incidence, severity, and duration of signs and symptoms of influenza-like illness as assessed by the FluPRO Questionnaire (see Appendix 14.1)
- Analysis of time to alleviation of signs and symptoms of influenza in the subset of subjects able to complete the Influenza Patient Reported Outcomes (FluPRO) Questionnaire at baseline and post-dose by Kaplan Meier analysis
- The percentage of subjects with new bacterial pneumonia/superinfection
- The percentage of subjects with influenza-related complications
- VIS410 population pharmacokinetic (PK) parameters in serum
- Titer of anti-VIS410 antibody positive samples
- Genotypic and/or phenotypic assessment to determine the emergence of VIS410 and oseltamivir-resistant viruses

Exploratory Endpoints

- Population PK parameters of VIS410 from nasopharyngeal secretions
- VIS410 concentration in tracheal aspirates
- The difference in viral load between VIS410 + oseltamivir and oseltamivir alone treatment groups in tracheal aspirate of subjects on mechanical ventilation
- Titer of anti-influenza A antibodies by hemagglutinin inhibition assay (HAI) in serum
- Correlations between serum and/or nasopharyngeal PK with viral load, clinical symptoms, presence of antidrug antibodies (ADAs), safety, and additional endpoints

Statistical Methods

Sample Size

Approximately 120 evaluable subjects will be enrolled. The study is exploratory in nature, and is not powered to demonstrate significant differences between treatment groups in primary or secondary outcome measures. The protocol intent is to collect sufficient information to identify the most appropriate candidate endpoints for subsequent Phase 3 study evaluation from among the primary and secondary endpoints described below. Statistical significance testing will therefore be used to assess the relative strength of evidence of the primary and secondary endpoints, to provide reasonable assurance that the endpoints chosen for a confirmatory Phase 3 trial will elucidate treatment differences between VIS410 plus oseltamivir versus oseltamivir alone.

Efficacy

Efficacy analysis will have two complementary purposes in this Phase 2 study, both designed to aid in the optimal design of a confirmatory Phase 3 trial. First, primary and secondary endpoints will be analyzed for treatment group differences, to determine which endpoints might be most sensitive demonstration of treatment group differences using p-values as an indicator. Second, the relative contribution of the various efficacy endpoints to patient well-being and benefit will be assessed, as well as potential correlations of endpoints such as the influence of peak viral load or number of days on ventilator on the time to resumption of normal activities.

The ordinal scale outcomes will be measured daily from Day 1 (baseline) through Day 14, inclusive, using a seven-level hierarchical scale with the classifications ordered from the worst to the best clinical outcomes (see Section 10.6.1). For use in overall summary statistical presentations, the ordinal categories will be assigned decreasing integer scores, with death a score of 6 and discharge with full resumption of normal activities a score of 0. The primary outcome comparison of Ordinal Scale scores at Day 7 between treatment groups will be evaluated by proportional odds ratio analysis, as implemented by logistic regression, including a test of the proportional odds assumption. For this analysis, the response categories will be ordered from best (Discharge with full resumption of normal activities) to worst (Death). Additional exploratory analyses may be conducted to obtain a more complete understanding of the relationship of treatment to the ordinal response, including exact Mantel-Haenszel tests or partial proportional odds models.

The area under the curve (AUC) over time for a given patient will be calculated as the sum of the maximum ordinal score for each day up through 7 and 14 days. An analysis of treatment group differences will be performed on these per-patient AUC values using analysis of variance, with treatment group and strata as fixed factors. A sensitivity analysis will be performed that excludes the category of death on study, to determine if death as an outcome skews the results; death as an outcome will also be analyzed as an independent secondary endpoint. A secondary analysis of treatment group effect on the difference in proportions of patients with the worst (death) versus the best outcome (discharge from hospital and resumption of normal activities) will also be performed. This analysis does not use scores for the ordinal outcome but does account for ordinality, and is therefore not dependent on the relationship of score to severity of outcome. An exploratory analysis will be performed through an exact categorical analysis of treatment group difference in the ordinal scale results using the worst outcome on a per-patient basis.

Additional ordinal scale outcome assessments will include comparison of total numbers of days at more severe scale values (death, time on ventilator, time in ICU) and proportions of patients with ordinal scale worsening post enrollment. Proportion outcomes will be analyzed by categorical data analysis methods, including Mantel-Haenszel chi-square tests adjusted for strata; additional exploratory subgroup analyses may also be performed, for example, based on various age categories. Time from onset of symptoms to study treatment and the number of doses of oseltamivir (Tamiflu) prior to VIS410 treatment for the current case of influenza may be included as covariates in the analyses.

Secondary and exploratory endpoints will be analyzed for descriptive purposes, with significance levels (p-values) provided to illustrate the strength of evidence for treatment effects, and to provide a basis for the choice of endpoints for further study in a confirmatory study. These analyses will also provide estimates for potentially powering additional efficacy endpoints, and to assist in hierarchical ordering of secondary endpoints to provide alpha-control in confirmatory studies and for labelling purposes.

Time to cessation of O_2 support resulting in a stable SpO_2 will be analyzed using a Cox proportional hazards regression model including data from patients on O_2 support. A P-value (using the Wald statistic) for each VIS410 dose vs placebo will be presented.

The total number of days in the hospital and/or ICU from admission to discharge and the rate of rehospitalization

due to influenza A relapse/reinfection will be summarized descriptively by treatment group.

The probability of time to clinical response (defined as resolution of vital signs) will be calculated via Kaplan-Meier. A significance test (using the log-rank test) for each VIS410 dose vs placebo will be presented. Results will be tabulated and presented graphically as well. The number and percentage of subjects in each treatment group with clinical response will be summarized.

Serum Pharmacokinetics

Serum concentrations will be listed by subject for VIS410 and summary statistics by group will be presented, including means, geometric means, standard deviations, coefficient of variation (CV), medians, and ranges, as appropriate. Summary graphs, including mean concentration-time profiles by group, will also be presented. The serum concentration data will be analyzed by population PK methods using nonlinear mixed effects modeling as implemented in NONMEM or equivalent software. Additional analyses and summaries may be generated as appropriate.

Pharmacokinetics of Nasopharyngeal Secretions and Tracheal Aspirates

Nasopharyngeal secretion and tracheal aspirate concentrations will be listed by subject for VIS410 and summary statistics by group will be reported as described for the serum concentrations. The computed PK parameters will be listed by subject for VIS410. Summary statistics and PK parameters will be presented including means, geometric means, standard deviations, CV, medians, and ranges, as appropriate. Summary graphs, including mean concentration-time profiles by group, will also be presented. The nasopharyngeal concentration data may also be analyzed by population PK methods using nonlinear mixed effects modeling as implemented in NONMEM or equivalent software. Additional analyses and summaries may be generated as appropriate.

Adverse Events

The original terms in the electronic data capture (EDC) system used by Investigators to identify AEs other than symptoms of influenza A will be fully described and coded according to the Medical Dictionary for Regulatory Activities (MedDRA). The reported AEs will be allocated to phases based on their start date. All AEs will be listed. All AEs with onset during the treatment phase (ie, TEAEs) will be summarized.

AEs will be summarized overall and by treatment group and by MedDRA body organ system and preferred term, severity, relatedness, and seriousness.

The difference in proportions of subjects with AEs, TEAEs, hypersensitivity reaction, anaphylactic reaction, AEs of special interest (AESIs), and SAEs between treatment groups will be calculated.

Special attention will be paid to those subjects who died, discontinued the study drug due to an AE, or experienced a severe or serious AE. Summaries, listings, and narratives will be provided, as appropriate.

Injection Site Tolerability

Injection site tolerability is defined as AEs demonstrating injection site irritation or tissue damage. Injection site tolerability will be reported by variable, treatment group, and time point.

Clinical Laboratory Tests

Actual values and changes from baseline of each continuous biochemistry, hematology, and urinalysis test will be evaluated by means of descriptive statistics by assessment time point and by treatment group. For categorical urinalysis tests, frequency tables of actual values will be provided by assessment time point and by treatment group.

Relative changes in clinical laboratory test values compared to values at baseline will be evaluated according to the Division of Microbiology and Infectious Diseases (DMID) Adult Toxicity Table (see Appendix 14.3) or in accordance with the normal ranges of the clinical laboratory (below, within, or above normal range) for parameters for which no toxicity grades are defined. A toxicity grade shift from baseline table of the abnormalities will be provided by assessment time point and by treatment group.

A listing of subjects with any clinical laboratory test result outside the reference ranges will be provided.

Vital Signs

Actual values and changes from baseline of heart rate, respiratory rate, temperature, SBP, and diastolic blood

pressure (DBP) measurements will be evaluated by means of descriptive statistics by assessment time point and by treatment group.

A shift from baseline table of vital sign abnormalities will be provided by assessment time point and by treatment group.

Electrocardiography

The ECG variables that will be analyzed are heart rate, PR interval, QRS interval, and QT interval. Values for QT corrected for heart rate (QTc) will be derived. QTc corrected according to Fridericia (QTcF)[13] will be the primary correction parameter.

Actual values and changes from baseline of ECG variables will be evaluated by means of descriptive statistics by assessment time point and by treatment group.

A shift from baseline table of ECG abnormalities will be provided by assessment time point and by treatment group. For absolute QTcF interval prolongation (> 450, > 480, > 500 ms) and changes from baseline (increase > 30 and > 60 ms), a frequency table by assessment time point and by treatment group will be provided.

Signs and Symptoms of Influenza

Descriptive statistics will be used to compare the duration of symptoms of influenza-like illness by treatment group in the subset of subjects able to complete the FluPRO Questionnaire at baseline and post-dose. Frequency tabulation of the occurrence and severity of each of the subject-reported symptoms of influenza-like illness by assessment time point and by treatment group will be generated. The number and percentage of subjects who were not able to complete the assessment at each visit will be summarized.

Complications of Influenza

Complications of influenza are defined as pneumonia, myocarditis, worsening of chronic bronchitis, sinusitis, otitis or other chronic pulmonary diseases, encephalopathy, and death. Complications of influenza and use of antibiotic therapy for bacterial pneumonia will be reported by variable, treatment group, and time point. Diagnosis of bacterial pneumonia will be based on local clinical practice, usually an assessment of clinical features and any radiographic evidence of bacterial pneumonia (eg, infiltrate) and/or microbiologic evidence of bacterial pneumonia. All-cause and attributable mortality will be evaluated on Day 14 and 28.

Healthcare Resource Utilization

Descriptive statistics will be used to compare the total duration of hospitalization, number of subjects requiring ICU admission post-randomization, overall number of days in the ICU, number of hours on ventilation, rehospitalization due to influenza-like illness, duration of oseltamivir therapy, and number of days to resumption of usual activities between treatment arms. Time to resumption of usual activities will be determined from the visual analog scale (VAS) (scale ranges from 0 to 10, where 0 indicates subject is unable to perform any of his/her usual activities prior to influenza onset, and 10 indicates subject is able to fully perform all usual activities).

Viral Load

One-way ANOVA or Mann-Whitney U test will be used to assess the difference between treatment groups (each VIS410 dose vs placebo) in the viral load AUC based on qRT-PCR and $TCID_{50}$ from nasopharyngeal swabs. Descriptive statistics will be used for viral load data (AUC, time to resolution of viral load, duration of viral shedding, and peak viral load based on qRT-PCR and $TCID_{50}$) from nasopharyngeal swabs as well as tracheal aspirate by treatment group and VIS410 overall vs placebo. Tables and graphs will be generated as appropriate by dose group. Additional exploratory statistical analyses may be conducted as appropriate. Exploratory dose and exposure response will be evaluated using various statistical and graphical approaches as appropriate.

Immunology

Immunological assessments will be summarized by parameter, treatment group, and time point using descriptive statistics:

- Anti-influenza A antibodies by HAI in serum
- ADA titers

Exploratory Pharmacokinetic/Pharmacodynamic Analyses

Various techniques will be used to explore exposure-response relationships and to compare the strength of the relationship between each independent variable (eg, AUC, C_{max}, concentration at specific time point) and the dependent variables (eg, viral AUC, peak viral load, time to cessation of viral shedding, clinical symptoms, and additional endpoints). These techniques may include graphical and statistical methods, including the creation of boxplots, spaghetti plots, histograms, and a variety of linear, nonlinear, or logistic regression techniques and time-to-event methods. If appropriate, continuous independent variables will be evaluated as such, and as categorical variables (grouping subjects into exposure categories).

Emergence of Viral Resistance to VIS410 and Oseltamivir

Viral sensitivity to VIS410 and oseltamivir will be assessed during the study.

SCHEDULE OF ASSESSMENTS

Table 1. Schedule of Assessments

Study Time Point	Screening[1]	Day 1				Day 3	Day 5 (± 1)	Day 7 (± 1)	Day 14 (± 3)	Day 28 (± 3)	Day 56 (± 7)
		Baseline	Predose	0 Hour	End of Infusion						LFU/ET Visit[2]
Screening/Administrative Assessments											
Informed consent	X										
Inclusion/exclusion criteria	X										
Medical history and demographics	X										
Nasopharyngeal swab for rapid flu test[3]	X										
Onset of symptoms interview	X										
Prior medications[4]	X	X									
Randomization[5]		X									
Subject Diary[6]		X—									—X
Safety Assessments [7]											
Supine ECG[8]	X	X[9]			X						
Vital signs[10]	X	X[9]	X		X	X	X	X	X	X	X
SpO_2[11]	X	X[9]	X		X—						—X
Body temperature[12]	X	X[9]	X		X	X	X	X	X	X	X
Physical exam[13]	X										
FluPRO Questionnaire[14]		X—							—X		
Seven-Level ordinal scale[15]		X—							—X		
Pregnancy test[16]	X	X[17]									X
Chemistry, hematology, urinalysis[18,19]		X					X		X	X	X
Serum creatinine[20]	X										
C-reactive protein (CRP)		X					X				
Erythrocyte sedimentation rate (ESR)[20a]		X					X		X		
Chest x-ray / CT scan[21]	X										
Procalcitonin		X									
AEs, SAEs, and AESIs		X—									—X
Concomitant therapy		X—									—X

Study Time Point	Screening[1]	Day 1				Day 3	Day 5 (± 1)	Day 7 (± 1)	Day 14 (± 3)	Day 28 (± 3)	Day 56 (± 7)
		Baseline	Predose	0 Hour	End of Infusion						LFU/ET Visit[2]
Influenza complications		X—									—X
VAS for assessment of resumption of normal activities		X—									—X
Healthcare utilization[22]		X—									—X
Study Agent Administration/Virologic/PK, and Immunology Assessments											
Pretreatment medications 60 minutes (± 5 min) prior to start of infusion[23]			X								
VIS410/placebo infusion[24]				X							
Oseltamivir therapy[25]				X—			—X				
Serum PK		X			X[26]		X		X	X	X
Nasopharyngeal swab[27,28]			X		X[26]	X	X	X	X	X	X
Tracheal aspirate[29]			X		X	X	X	X			
Serum ADA		X								X	X
Serum HAI sample		X								X	

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[1] Screening and baseline activities may be performed on the same day.
[2] Subjects who terminate the study participation early are encouraged to have all follow-up safety assessments done at the time of study termination. All other subjects will return for a final follow-up visit at Day 56 ± 7 days.
[3] Single nasopharyngeal swab will be obtained from one nostril for influenza A rapid test. This screening test is not necessary if the subject has a prior influenza A positive test by Rapid Antigen Test or with another commercially available test including PCR, FIA, or ELISA within the prior 48 hours of screening.
[4] Collect all medications taken, including over-the-counter, within 7 days prior to VIS410/placebo administration.
[5] Randomization will be stratified by presence or absence of positive pressure ventilation (PPV) at baseline to ensure an even distribution across treatment arms.
[6] Subject diary will be provided upon discharge from the hospital and will record daily oseltamivir dosing (as applicable) and will have a daily VAS for assessing resumption of usual activities until either the subject reports that all usual activities (prior to influenza onset) can be performed or Day 56, whichever comes first. In addition, the Influenza Patient Reported Outcomes (FluPRO) Questionnaire will be provided and completed daily up until Day 14 (if applicable).
[7] Additional safety assessments may be performed outside of the defined protocol criteria, at the Investigator's discretion.
[8] Single 12-lead ECG will be performed after a 5-minute rest in supine position.
[9] To be repeated only if screening and baseline visits are more than 24 hours apart.

[10] Vital signs should be measured after 5 minutes of rest in a supine position and include heart rate, respiratory rate, and blood pressure. During hospitalization, vital signs will be measured at the end of the infusion on Day 1 and then BID (preferably at the same time each day) up until Day 14 or discharge from the hospital if this occurs prior to Day 14.

[11] Baseline SpO_2 on room air to be documented, if available. Once randomized, SpO_2 to be measured using pulse oximetry 3 times daily (approximately every 8 hours) at approximately the same time each day until stable. Stable SpO_2 is defined as two consecutive SpO_2 values of > 92% on room air that are at least 8 hours apart.

[12] During hospitalization, body temperature will be recorded at the end of infusion on Day 1 then BID up until Day 14 or discharge from the hospital if discharge occurs prior to Day 14. In cases where obtaining oral temperature is not possible, core temperature will be obtained. While hospitalized, the maximum temperature should be recorded for each 12-hour interval (from 12 AM to 12 PM and from 12 PM to 12 AM).

[13] Additional targeted physical exam may be performed throughout the study as needed at the Investigator's or his/her designee's discretion.

[14] The Influenza Patient Reported Outcomes (FluPRO) Questionnaire (Appendix 14.1) must be completed (ie, self-reported) by the subject at baseline (Day 1) and daily thereafter through Day 14. Subjects who are not able to complete the questionnaire at baseline (Day 1) will be exempt from this assessment throughout the study. The FluPRO will be completed at home for subjects that are discharged from the hospital prior to Day 14.

[15] The ordinal scale will be completed from baseline (Day 1) and daily thereafter through Day 14. For each day, subjects will be classified by the worst clinical outcome for which they qualify.

[16] A urine or serum pregnancy test must be performed at screening locally (ie, by the investigative site) within 2 days prior to dosing. Negative results must be obtained prior to randomization.

[17] A serum pregnancy test will be performed at the central lab for all female subjects of childbearing potential following randomization at baseline

[18] Subjects to be enrolled based on the Investigator's discretion including any local laboratory results per SOC at institution.

[19] See Laboratory Assessments for list of tests to be performed by the Central Lab in Appendix 14.2.

[20] Serum creatinine to be done locally within 24 hours prior to randomization, the result is required for oseltamivir dosing.

[20a] ESR to be performed locally.

[21] A chest x-ray or computed tomography (CT) scan taken per SOC within 72 hours before dosing is acceptable.

[22] Healthcare utilization (total length of hospital stay, length of ICU stay, and rehospitalization) will be captured through the last follow-up visit.

[23] If the subject has any history of delayed gastric emptying, including premenstruation syndrome or menstruation, he or she may receive oral premedications 120 minutes prior to IV VIS410/placebo infusion.

[24] VIS410/placebo will be administered over 2 hours. Infusion time can be extended to up to 4 hours at discretion of the Investigator only if local infusion site–related symptoms occur.

[25] Minimum of 10 doses of oseltamivir treatment (BID for 5 days) is required. Treatment with oseltamivir may be extended for up to 5 additional days (up to 20 doses of total therapy) if clinical symptoms warrant further treatment as assessed by the Investigator. Note: based on SOC, subjects may have started oseltamivir therapy prior to randomization. For patients unable to swallow, oseltamivir capsules can be opened and mixed with liquid for ease of administration.

[26] To be obtained up to 2 hours post-VIS410 administration.

[27] Nasopharyngeal swabs will be obtained for virology and/or PK from both nostrils (1 swab per nostril). The first 50 randomized subjects will have nasopharyngeal swabs collected up to Day 56 (predose, end of infusion, Days 3, 5, 7, 14, 28 and 56); while in the remaining subjects, nasopharyngeal swabs will be obtained up to Day 14 only.

[28] If the subject remains in the hospital on Day 10, additional nasopharyngeal swabs will be obtained on Day 10.

[29] One tracheal aspirate sample for virology and PK will be obtained (ventilated subjects only).

END OF FOOTNOTES TO SCHEDULE OF ASSESSMENTS TABLE

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LIST OF ABBREVIATIONS AND DEFINITIONS OF TERMS

Abbreviation	Definition
ADA	antidrug antibody
AE	adverse event
AESI	adverse event of special interest
AUC	Area under curve
ASA	acetylsalicylic acid
AUC	area under the concentration–time curve
BID	Latin *bis in die* (two times (twice) daily)
BiPAP	bi-level positive airway pressure
BMI	body mass index; weight in kilograms divided by the square of height in meters
CD-1	cluster of differentiation 1
CIOMS	Council for International Organizations of Medical Science
C_{max}	maximum concentration
CPAP	continuous positive airway pressure
CRO	contract research organization
CT (scan)	computed tomography
CV	coefficient of variation
DBP	diastolic blood pressure
DMID	Division of Microbiology and Infectious Diseases
DSMB	Data Safety Monitoring Board
EC	ethics committee
EC_{50}	half-maximal effective concentration
ECG	electrocardiogram
EDC	electronic data capture (system)
ELISA	enzyme-linked immunosorbent assay
ESRD	end-stage renal disease
ET	end of therapy
FDA	Food and Drug Administration
FIA	fluorescent immunoassay
FluPRO Questionnaire	Influenza Patient Reported Outcomes (FluPRO) Questionnaire
GCP	Good Clinical Practice
GI	gastrointestinal
GLP	Good Laboratory Practice
HAI	hemagglutination inhibition assay
IC_{50}	half-maximal inhibitory concentration
ICF	Informed Consent Form
ICH	International Conference on Harmonisation
ICU	intensive care unit
IEC	independent ethics committee
IgG1	immunoglobulin G1

Abbreviation	Definition
IRB	institutional review board
ITT	intent-to-treat
IV	intravenous
IWRS	Interactive Web Response System
LFU	last follow-up
LSLV	last subject last visit
MedDRA	Medical Dictionary for Regulatory Activities
MITT	modified intent-to-treat
NOAEL	no-observed–adverse-effect level
NONMEM	software used for population pharmacokinetic modeling
NSAID	nonsteroidal anti-inflammatory drug
O_2	oxygen
PCR	polymerase chain reaction
PK	pharmacokinetic
PO	Latin *per os* (by mouth)
PP	per protocol
PPV	positive pressure ventilation
PR interval	an ECG parameter
QRS interval	an ECG parameter
qRT-PCR	quantitative reverse transcriptase polymerase chain reaction
QT interval	an ECG parameter
QTc	an ECG parameter; values for QT corrected for heart rate
QTcF	QTc corrected according to Fridericia
RBC	red blood cell
RSV	respiratory syncytial virus
SAE	serious adverse event
SAP	statistical analysis plan
SBP	systolic blood pressure
SOC	standard of care
SSRE	sample size re-estimation
Study drug	VIS410 or placebo
SUSAR	suspected unexpected serious adverse reaction
$TCID_{50}$	half-maximal tissue culture infective dose
TEAE	treatment-emergent adverse event
VAS	visual analog scale
VIS410	investigational product
WHO	World Health Organization

1.0 INTRODUCTION

1.1 Background Information

Severe influenza disease is a common occurrence each season, especially in high-risk groups, such as young children, older adults, patients with pulmonary conditions, inflammatory conditions, malignancies, and pregnant women.[1, 2] Despite available therapy with neuraminidase inhibitors, including oseltamivir (Tamiflu®), zanamivir (Relenza®), and peramivir (Rapivab®), 10% to 44% of hospitalized patients require intensive care and 25% to 50% of these patients die. It is estimated that as many as 400,000 patients are hospitalized with influenza each year in the United States, with up to 49,000 deaths per year.[3]

The World Health Organization (WHO) has reported incidence rates of 3 to 5 million severe cases and about 250,000 to 500,000 influenza-related deaths annually.[4] The 2009 influenza A pandemic (H1N1) spread rapidly to every continent with more than 399,232 reported cases and 4735 deaths.[5]

The therapeutic use of passive polyclonal antibodies to prevent viral infections, including hepatitis B, varicella, cytomegalovirus, rabies, and respiratory syncytial virus (RSV) has been well established. More recently, monoclonal antibodies for viral infections have been developed, including palivizumab (Synagis®), a Food and Drug Administration (FDA)-licensed treatment for the prevention of RSV infection.

Visterra has developed a novel approach to antibody discovery whereby functionally conserved epitopes are identified based on atomic interaction networks and targeted with rationally engineered human antibodies. Using this approach, VIS410, a broad spectrum human immunoglobulin G1 (IgG1) monoclonal antibody with demonstrated efficacy against both Group 1 (including H1 and H5) and Group 2 (including H3 and H7) influenza A strains, in both treatment and prevention models of influenza, was developed. Visterra intends to develop this product for the treatment of influenza A, specifically in hospitalized patients.

This study will provide the first indications of efficacy, safety, and tolerability of VIS410 in hospitalized subjects with influenza A infection requiring oxygen support. Efficacy will be assessed by comparison of clinical status ordinal Day 7 scores between treatment groups.

1.2 Nonclinical Studies

1.2.1 Pharmacology, Pharmacokinetics, and Metabolism

VIS410 is a human IgG1 monoclonal antibody with a high sequence homology to human germ line sequences. VIS410 broadly neutralizes influenza A strains in vitro, with half-maximal inhibitory concentration values (IC_{50}) of approximately 0.1 to 11 µg/mL. Mechanistic studies indicate that VIS410 inhibits hemagglutinin-mediated cell membrane fusion, thus preventing viral replication.

In vivo studies have demonstrated that VIS410 administered to mice in prevention and treatment models of influenza, at doses between 1 and 20 mg/kg (either 24 hours pre-exposure or 24-72 hours post-exposure) can protect mice challenged with influenza A strains (influenza A/Puerto Rico/8/1934 [H1N1], influenza A/Victoria/3/1975 [H3N2], and influenza A/Vietnam/1203/2004 [H5N1]). Additionally, in vivo studies in ferrets have demonstrated that treatment with VIS410 can reduce the transmission of influenza pH1N1 via respiratory droplets. The serum concentrations of VIS410 associated with prevention of infection in ferrets should be readily achievable in humans also. VIS410 also demonstrates protection against a newly emerging highly pathogenic H7N9 (A/Anhui/1/2013) strain of influenza in a severe Dilute Brown Non-Agouti (DBA) mouse model.[6]

1.2.2 Toxicology

In a cynomolgus monkey Good Laboratory Practice (GLP) toxicity/toxicokinetics study, 4 doses of VIS410 were administered intravenously (IV) over a 28-day period at 5, 50, and 250 mg/kg with no clinically significant findings. Based on this GLP study, the no-observed–adverse-effect level (NOAEL) was considered to be 250 mg/kg and the half-life of VIS410 was determined to be 8 to 9 days.

In the cynomolgus monkey toxicology study, VIS410 was administered as an IV infusion over 10 minutes, and no infusion reactions were observed.

In a mouse GLP toxicity/toxicokinetics study (4 doses of VIS410 IV administered at 5, 50 and 250 mg/kg, infused over 14 days to 384 mice), 14 mice (3.6%) died 1-2 hours after Day 14 dosing in the 5- and 50-mg/kg dose groups. This was likely due to an expected immunogenic response in mice, as VIS410 is a human antibody, and not a toxic response. The NOAEL was considered to be 250 mg/kg.

In vitro tissue cross-reactivity studies of VIS410 with normal human tissues, cynomolgus monkey tissues, and cluster of differentiation 1 (CD-1) mouse tissues showed VIS410 to be broadly cross reactive. VIS410 staining of membrane was limited to cells within the surface/mucosal epithelium in colon, small intestine, and fallopian tube. Of note, staining was not observed in vivo, in tissues from toxicology studies in monkeys or mice when assessed by immunohistochemistry post-sacrifice.

The lack of toxicity in the in vivo cynomolgus monkey and mouse toxicology studies suggests that the cross-reactivity findings were of little toxicological significance.

An in vitro–soluble cytokine release assay performed using human whole blood at VIS410 concentrations that encompass the expected concentrations of VIS410 in human blood when administered as IV doses of 2 to 50 mg/kg, did not stimulate any acute or sustained release of any pro-inflammatory cytokines.

1.3 Clinical Studies

1.3.1 Phase 1 Study (VIS-C001)

In a Phase 1 study (VIS-C001), a total of 30 healthy volunteers received a single, 120-minute IV infusion of 2, 5, 15, 30, or 50 mg/kg of VIS410 (200-mL volume; 6 subjects per cohort) and 11 subjects received a single, 120-minute IV infusion of placebo (sodium chloride 0.9%; 200-mL volume).

The PK and safety data are presented in Section 1.3.1.1 and Section 1.3.1.2.

1.3.1.1 Pharmacokinetic Results

PK profiles of the serum and nasopharyngeal samples demonstrated that VIS410 exposure was approximately proportional to the dose administered with a mean serum $t_{1/2}$ of 12.9 days. This $t_{1/2}$ is within the expected range for the IgG1 molecules. In addition, the 13-day $t_{1/2}$ supports the single dosing of VIS410, as the circulating levels should be sufficiently high during the normal course of an influenza infection in hospitalized patients (5–14 days). The maximum concentration (C_{max}) values in the serum for the 30- and 50-mg/kg cohorts were 980 and 1316 µg/mL, respectively.

Nasopharyngeal swabs were taken from the 15-, 30-, and 50-mg/kg cohorts for upper respiratory PK. The nasopharyngeal PK profiles were approximately dose proportional. Mean nasal C_{max} values at the 30- and 50-mg/kg dose levels were 20.0 and 25.3 µg/mL, respectively.

1.3.1.2 Safety Results

In the Phase 1 study, VIS410 was administered IV over 2 hours. Overall, VIS410 was generally safe and well tolerated at all dose levels studied. TEAEs were reported for 20 of 30 subjects (66.7%) receiving VIS410 and 7 of 11 subjects (63.6%) receiving placebo. There were no drug-related SAEs, no drug-related discontinuations, and no infusion-related reactions, such as anaphylaxis or injection site reactions.

Notable AEs included one unrelated SAE of leukopenia and esophagitis secondary to primary herpes simplex virus-1 infection in 1 subject (VIS410, 30-mg/kg cohort). This SAE resolved, and the subject did well. Another unrelated SAE of acute appendicitis was also reported (placebo cohort).

The most common AEs observed were diarrhea (10 of 41 [24.4%]) and headache (8 of 41 [19.5%]; 5 subjects received VIS410). All diarrheal or loose stool AEs were observed in the VIS410-treated subjects and were usually mild to moderate, started ~2 to 4 hours after dosing, were transient, and resolved spontaneously within 24 hours after dosing with no associated dehydration. There were no associated elevations in alanine aminotransferase, aspartate aminotransferase, or bilirubin parameters.

Mean hematology, serum chemistry, and urinalysis results were within the normal limits, and the mean values and changes from baseline were similar across the treatment groups and placebo.

Overall, mean vital sign measurements observed after dosing were similar to those observed at baseline. Mean changes from baseline were also similar across VIS410 dose levels, and no apparent treatment- or dose-related trends were observed.

The antidrug antibody (ADA) data demonstrated a low level of positive ADA in 4 out of 30 subjects receiving VIS410 (Table 2). The subjects with positive ADA did not demonstrate any impact on their PK profile.

Table 2. Summary ADA Data – Phase 1 Study VIS-C001

Cohort Dose (mg/kg)	No. of Confirmed ADA Positive	Pre-infusion Titer	Confirmed Day	Positive Titer
2	0/6	None detected	NA	None detected
5	3/6	None detected	120	Titer 10
		None detected	14	Titer 10
			120	Titer 40
		None detected	120	Titer 40
15	1/6	None detected	120	Titer 40
30	0/6	None detected	NA	None detected
50	0/6	None detected	NA	None detected

1.3.2 Phase 2a Challenge Study (VIS410-201)

The Phase 2a study is a randomized, double-blind, placebo-controlled study to assess the safety, tolerability and antiviral activity of a single IV dose of VIS410 in healthy subjects after a viral inoculation with influenza A (H1N1) (Study VIS410-201).

The study was conducted at a single site in Antwerpen, Belgium, by SGS Life Sciences. Briefly, 24 hours after inoculation with a 106 tissue culture infective dose (TCID) of naturally attenuated influenza A (H1N1) virus,7 subjects received a single IV administration of VIS410 (2300-mg fixed dose, equivalent of 30 mg/kg) or placebo (7:5 randomization to VIS410:placebo) in Part 1 of the study. Subjects were quarantined 10 days post-inoculation and discharged thereafter.

A total of 46 subjects were treated with VIS410 (n = 33) or placebo (n = 13) across 5 cohorts. Cohorts were performed sequentially and included different pretreatment regimens used to mitigate the gastrointestinal (GI) events observed with VIS410. Preliminary findings from the first 31 subjects are presented in Section 1.3.2.1 and Section 1.3.2.2; the study has now completed enrollment and the data are pending final analysis.

1.3.2.1 Preliminary Efficacy Data

A partially blinded prespecified interim analysis of the first 3 cohorts was conducted to evaluate the safety and efficacy of the 2300-mg VIS410 dose and to determine whether dose escalation was appropriate. The distribution of subjects is presented in Table 3.

Table 3. Distribution of Subjects

Treatment	No. Dosed	Per Protocol [a]	MITT [b]
Placebo	13	9	7
VIS410 (2300 mg)	18	16	13
Total	31	25	20

HAI = hemagglutinin inhibition assay positive seroconversion at Day 14 or 28.

[a] Subjects with HAI > 10 at baseline excluded per inclusion/exclusion criteria.

[b] All randomized subjects who received study drug, met the inclusion criterion of seronegative by HAI (≤ 10) on Day 1, and were infected.

Based on the preliminary results, VIS410 2300-mg demonstrated a statistically significant antiviral effect with a 76% and 91% reduction in nasopharyngeal viral load area under the curve (AUC) by qRT-PCR and $TCID_{50}$, respectively, compared to placebo (Table 4).

Table 4. Viral Load and AUC Data

Viral Measure	Placebo (n = 7)	VIS410 (n = 13)	Reduction (%)	*P* Value [a]
Median viral AUC $TCID_{50}$ (log_{10} x hours/mL)	552	47	91	0.019
Median viral AUC qPCR (log_{10} x hours/mL)	1033	232	76	0.024
Median peak viral load $TCID_{50}$ (log_{10}/mL)	5.0	2.75	2.25	0.009
Median peak viral load qPCR (log_{10}/mL)	7.1	5.6	1.53	0.043

[a] Calculated with Mann-Whitney U Test.

In addition, analysis of the symptoms data demonstrated a trend in respiratory symptom resolution that was consistent with the virology data. Although this study was not powered to detect a statistical difference in symptom relief, a 2-day reduction in resolution of upper respiratory symptoms and AUC was observed in the VIS410 2300-mg group vs placebo.

1.3.2.2 Preliminary Safety Data

In the first cohort, 7 subjects received 2300 mg of VIS410 while 5 subjects received placebo. Moderate to severe cramping, loose stool and/or diarrhea were reported in 6 of the 7 subjects receiving VIS410 and none of the placebo subjects. Most symptoms were observed either during the infusion or within 30 minutes of the end of infusion and resolved within ~12 hours. Given the association with the infusion, these AEs were all considered possibly related to the study drug. In

subsequent cohorts, the addition of a pretreatment regimen containing diphenhydramine mitigated the GI AEs, and VIS410 was generally well tolerated with a reduction in frequency and the severity of GI events from moderate/severe to mild.

As statistically significant efficacy was demonstrated with the 2300-mg dose in the first 3 cohorts, the protocol was amended to optimize the tolerability of VIS410. This analysis was conducted by testing different pretreatment regimens and escalating the dose of VIS410 to 4600 mg in the context of the virus challenge model. This portion of the study (designated as Part 2) was open label; however, virologic data were collected to understand viral response and pharmacokinetics/pharmacodynamics.

Eleven subjects were enrolled in Cohort 4 and were inoculated with H1N1 influenza virus and administered VIS410 2300 mg 24 hours later. Four subjects were pretreated with diphenhydramine 50 mg PO and montelukast 10 mg PO (Group 1); 4 subjects were pretreated with diphenhydramine 50 mg PO and ibuprofen 600 mg PO (Group 2); and 3 subjects were pretreated with diphenhydramine 50 mg IV and montelukast 10 mg PO (Group 3).

Three subjects in Group 1 and 3 subjects in Group 3 experienced transient loose stool that resolved spontaneously. Only 1 of 4 subjects in Group 2 pretreated with diphenhydramine and ibuprofen experienced mild and transient loose stools. Based on these results, it was determined that the regimen of diphenhydramine plus ibuprofen resulted in the best tolerability profile, and this pretreatment regimen was further assessed in 4 subjects at the 4600-mg dose of VIS410 following viral challenge. Overall, 4600 mg of VIS410 following pretreatment was generally well tolerated. None of the 4 subjects reported cramping or any other symptoms associated with severe infusion reactions. Mild and transient loose stools were observed in 3 of the subjects; one event was considered unrelated to study drug. A fourth subject had a transient episode of nausea and vomiting about 8.3 hours after the start of infusion. All events were considered resolved within 24 hours of onset.

1.3.3 Phase 1 Study (VIS410-102)

The Phase 1 study (VIS410-102) is a Phase 1, randomized, placebo-controlled study to evaluate safety, tolerability, and PK of a single 2300-mg and 3800-mg dose of VIS410 following administration of different pretreatment regimens. A total of 83 subjects received study drug [placebo (n = 12), VIS410 2300 mg (n = 59), VIS410 3800 mg (n = 12)].

VIS410 2300 mg was evaluated as a 2-hour infusion (n = 12), 4-hour infusion (n = 12), 2-hour infusion following a pretreatment regimen of diphenhydramine 50 mg PO and ibuprofen 400 mg PO 60 minutes before infusion (n = 12), 2-hour infusion following a pretreatment regimen of cetirizine 10 mg PO 60 minutes before infusion (n = 12), and 2-hour infusion following a pretreatment regimen of diphenhydramine 50 mg PO and acetylsalicylic acid 325 mg PO 60 minutes before a 2-hour infusion (n = 11).

The 3800-mg VIS410 dose was evaluated as a 2-hour infusion following a pretreatment regimen of diphenhydramine 50 mg PO and ibuprofen 400 mg PO 60 minutes before infusion (n = 12). The pretreatment regimens were selected in order to mitigate the GI AEs observed with VIS410. This study is currently ongoing, and a preliminary summary of safety data is presented in Section 1.3.3.1.

1.3.3.1 Preliminary Safety Data

To date there have been no reports of SAEs. One subject at the 3800-mg dose discontinued study drug infusion due to AEs of nausea, vomiting, chills, and hot flashes. All events resolved after study drug discontinuation despite the slow decrease in serum concentration of VIS410. VIS410 was not associated with any signs or symptoms that are consistent with systemic allergic reactions or anaphylaxis. Gastrointestinal events were observed at a higher frequency in the VIS410-treated subjects compared to placebo.

Based on the data from the 2300-mg and 3800-mg dose groups, VIS410 was associated with GI events ranging from mild to severe; however, a 2300-mg dose of VIS410 with a single-dose pretreatment regimen of diphenhydramine and low dose ibuprofen or aspirin was associated with a low rate of GI events (16.7% and 9.1%, respectively). Overall, the most commonly reported events were loose stool, diarrhea, and abdominal cramping. Subjects who received no-pretreatment regimen had the highest number of GI AEs ranging from mild to severe.

The prolonged infusion of VIS410 (over 4 hours) resulted in a larger number of GI events than the 2-hour infusion (41 events in 10 subjects vs 18 events in 6 subjects, respectively), suggesting that GI events will not be mitigated by slowing the infusion rate. Pretreatment regimens of an antihistamine (cetirizine) or an antihistamine (diphenhydramine) plus a prostaglandin inhibitor (ibuprofen or aspirin) mitigated the GI events compared with the no-pretreatment cohorts. The rate (13 GI events in 7 subjects) and severity of events (12 of 13 mild) were slightly lower in the cetirizine group than the no-pretreatment groups.

The lowest rate of GI events with VIS410 was observed in subjects who received the diphenhydramine plus aspirin pretreatment regimen, with only 1 subject reporting a mild episode of loose stool and abdominal pain approximately 22 hours after initiation of study drug administration that resolved spontaneously with no intervention. Another effective pretreatment regimen included diphenhydramine plus ibuprofen at both 2300- and 3800-mg dose, with only 2 subjects in the 2300-mg dose group reporting a mild episode of loose stool 7 to 9 hours after initiation of study drug administration that resolved spontaneously with no intervention; 1 of the 2 subjects reported mild nausea as well. The same pretreatment with a higher dose of 3800 mg, showed a low rate of GI events with 1 subject reporting a mild episode of loose stool approximately 21 hours after start of infusion and another subject reporting mild nausea. Both events resolved spontaneously with no intervention. In addition, 1 subject at the 3800-mg dose discontinued study drug infusion due to AEs of nausea, vomiting, chills, and hot flashes.

To date, no clinically significant changes have been observed in hematology, chemistry, and urinalysis parameters. Specifically, no alterations in electrolytes were observed in the subjects with loose stool or diarrhea.

In summary, the type and timing of GI AEs observed in this study are consistent with prior studies, are mostly mild in intensity, are self-limiting in nature, and resolve following the completion of infusion with no sequelae. These data suggest that a pretreatment regimen is necessary to mitigate the GI events associated with VIS410; however, an antihistamine alone may not be as effective in mitigating the AEs. Addition of a prostaglandin inhibitor/nonsteroidal anti-inflammatory drug (NSAID), such as ibuprofen or aspirin, to an antihistamine was necessary to further reduce the frequency and severity of the GI events. These data were consistent with previous safety data in an influenza virus challenge study where a regimen of diphenhydramine and ibuprofen mitigated the severity and/or frequency of GI AEs in 8 subjects given 2300 mg (n = 4) and 4600 mg of VIS410 (n = 4). For more detailed information on VIS410, refer to the current Investigator's Brochure.[8]

1.4 Overall Rationale for the Study

Visterra is developing VIS410, a fully human IgG1 antibody, as a broad spectrum therapeutic agent against all strains of influenza A virus. New alternative treatments for influenza are urgently needed. Severe influenza disease occurs seasonally in many high-risk groups, such as young children, older adults, patients with pulmonary conditions, inflammatory conditions, malignancies, and pregnant women.[1,2] Current influenza therapies are limited to the neuraminidase inhibitors, including oseltamivir (Tamiflu), zanamivir (Relenza) and peramivir (Rapivab). In spite of these available treatments, 10% to 44% of hospitalized patients require intensive care, and 25% to 50% of these patients die. In the United States, it is estimated that as many as 400,000 patients are hospitalized with influenza each year, with up to 49,000 deaths per year.[3] A comparison of annual mortality rates from infectious disease in the United States further demonstrates the lack of effective interventions against this deadly disease (Table 5).

Table 5. Annual Mortality Rates from Infectious Diseases in the United States

Disease	Approximate No. of Deaths per Year
Influenza (varies by season)	25,000-49,000
Methicillin-resistant *Staphylococcus aureus* (2005)	18,650
Hepatitis C (2007)	15,000
Acquired immune deficiency syndrome (2007)	12,700
Invasive pneumococcal disease from *Streptococcus pneumoniae* (2009)	5,000
Hepatitis B	1,800

Furthermore, certain strains of influenza have comparatively high mortality rates: the 1918 H1N1 Spanish flu strain resulted in deaths of 1% to 3% of the world's population, compared with the 1968 pandemic strain that killed 0.03% of the world's population. In more recent years,

avian H5 and H7 strains have been documented to have 33% to 60% mortality rates despite the use of currently licensed antiviral treatments. Furthermore, the continued emergence of resistance to current antiviral drugs increases the need for new therapeutics.

This study will be used to determine the efficacy and safety of VIS410 in hospitalized subjects with influenza A infection requiring oxygen support, and is an important study for progression of the program toward evaluating VIS410 in severe influenza and those at high risk.

1.5 Risk-Benefit Analysis

In animal studies, single or multiple dose administrations of VIS410 were generally well tolerated at doses of up to 250 mg/kg. In the Phase 1 study VIS-C001 in 41 subjects (see Section1.3.1), all doses studied were generally safe and well tolerated with only mild to moderate loose stool or diarrhea seen at the highest doses administered (30 and 50 mg/kg).

The most common AE was a mild/Grade 1 loose stool or diarrhea that was self-limiting. Based on preliminary data from the Phase 2a challenge study (VIS410-201), administration of 2300 mg of VIS410 in the context of an influenza infection resulted in 6 of 12 subjects having moderate to severe cramping and loose stools that mostly resolved within 10 hours.

Following this initial cohort study, a pretreatment regimen was implemented using antihistamines, including diphenhydramine that resulted in 4 of 9 pretreated subjects experiencing mild stomach discomfort.

An additional open label tolerability study tested several pretreatment regimens, including diphenhydramine and ibuprofen, in the influenza-infected volunteers. Following dosing of 2300 mg of VIS410 in the ibuprofen and diphenhydramine pretreatment group, the subjects tolerated VIS410 well with only 1 of 4 subjects experiencing any GI AE, and this was a mild transient loose stool without any associated cramping.

At 4600 mg of VIS410, pretreatment with ibuprofen and diphenhydramine was also generally well tolerated with no associated cramping. Transient and mild loose stools were reported in 2 of the 4 subjects, and 1 subject also had a transient episode of nausea and vomiting. The AEs observed to date were mostly related GI events, were mild to moderate in intensity, resolved spontaneously with no additional intervention, and decreased in severity and frequency following pretreatment with ibuprofen and diphenhydramine.

The pretreatment regimen of diphenhydramine with an NSAID (ibuprofen or acetylsalicylic acid) was further evaluated in a Phase 1 study and was shown to be safe and well tolerated with minimal GI AEs.

Considering the potential benefit of VIS410 in the treatment of severe influenza A infection and the types and severity of AEs observed to date following a pretreatment regimen of an antihistamine and an NSAID, the benefit outweighs the risk of further evaluation of VIS410 in more severe influenza A infection in hospitalized subjects.

1.5.1 Potential Risks

1.5.1.1 Potential Risks of VIS410 Administration

The potential human toxicity of VIS410 should be evaluated in the context of the human experience in clinical studies. As described in Section 1.3, GI AEs, such as transient loose stool/diarrhea and abdominal cramping, were the most commonly observed AEs in the clinical studies. The frequency and severity of these events were greatly mitigated with administration of a pretreatment regimen consisting of an antihistamine and an NSAID.

In mice treated with 4 doses of VIS410 over 14 days, 14 of 384 animals had severe systemic hypersensitivity (anaphylaxis) with death within 1 to 2 hours after the fourth dose. This was not considered to be a VIS410 specific effect as the NOAEL was 250 mg/kg but rather a consequence of immunological response in mice to a human antibody. ADAs were found in 8 animals at the end of the 28-day recovery period; other human antibodies have elicited ADA in mice without clinical problems in humans.

Administration of VIS410, IV once weekly for 4 weeks in cynomolgus monkeys was well tolerated. All animals survived and there were no abnormal clinical observations suggesting distress to the animals (hypersensitivity-related or other) throughout the duration of the study period (including the administration or recovery periods).

ADAs were elicited in 3 of the 42 (7%) samples that had a reduction in titers that were most likely due to drug exposure. As with the mice, there were no findings suggesting VIS410 toxicity in clinical, laboratory, or pathology examinations.

In an in vitro–soluble cytokine release assay performed using human whole blood, VIS410 did not stimulate any dramatic or pervasive release of any pro-inflammatory cytokine (see Section 1.2).

Pharmacological class effects commonly associated with marketed monoclonal antibody products used for treatment in humans include serious infusion reactions including anaphylaxis (see Section 1.3). VIS410 is to be administered as a single infusion and is directed at an exogenous viral target. As a result, it is considered to be of low probability that other SAEs (such as infection risks reported for approved monoclonal antibodies directed against endogenous human protein targets or immune modulating cytokines often administered as multiple doses over a long period) will be observed with VIS410.

As described in Section 1.3, a single administration of VIS410 to healthy volunteers was safe and well tolerated. The potential exists for subjects to experience cramping or loose stools, but pretreatment with histamine antagonists with an NSAID (ibuprofen or acetylsalicylic acid) substantially ameliorated the GI effects.

Given the potential for acute severe hypersensitivity and other adverse reactions observed with other monoclonal antibody therapies, subjects should be closely monitored for relevant signs and

symptoms following administration of VIS410. VIS410 should only be administered when personnel and therapies are immediately available for the treatment of anaphylaxis and other hypersensitivity reactions.

For more detailed information on VIS410, refer to the current Investigator's Brochure.[8]

1.5.1.2 Potential Risks of Oseltamivir Administration

Oseltamivir (Tamiflu) is indicated for the treatment of uncomplicated influenza infection in patients who have been symptomatic for no more than 2 days.

Oseltamivir is contraindicated in patients with known hypersensitivity to any of the components of the product.

The following reactions have also been reported following treatment with oseltamivir therapy during clinical studies or post-marketing:

- Body as a whole: Swelling of the face or tongue, allergy, anaphylactic/anaphylactoid reactions
- Dermatologic: Dermatitis, rash, eczema, urticaria, erythema multiforme, Stevens-Johnson syndrome, toxic epidermal necrolysis
- Digestive: Hepatitis, liver function tests abnormal
- Cardiac: Arrhythmia
- Gastrointestinal: Nausea, vomiting, GI bleeding, hemorrhagic colitis
- Neurologic: Seizure
- Metabolic: Aggravation of diabetes
- Psychiatric: Delirium, including symptoms such as altered level of consciousness, confusion, abnormal behavior, delusions, hallucinations, agitation, anxiety, nightmares

For more information, refer to the prescribing information for oseltamivir.[9]

1.5.1.3 Potential Risks of Diphenhydramine, Ibuprofen, and Acetylsalicylic Acid Administration

The risk associated with the use of a single over-the-counter dose of diphenhydramine, ibuprofen, and acetylsalicylic acid in this study is considered low.

Many of the adverse reactions due to NSAIDs, such as acetylsalicylic acid or ibuprofen, are dose and duration dependent.

Ibuprofen and acetylsalicylic acid are contraindicated in patients with known hypersensitivity to NSAIDs and acetylsalicylic acid–induced asthma. Ibuprofen is contraindicated for treatment of perioperative pain in the setting of coronary artery bypass graft surgery.

Ibuprofen may cause an increased risk of serious cardiovascular thrombotic events, myocardial infarction, and stroke, which can be fatal. Patients with cardiovascular disease or risk factors for cardiovascular disease may be at greater risk.

NSAIDs cause an increased risk of serious GI AEs, including bleeding, ulceration, and perforation of the stomach or intestines, which can be fatal. Elderly patients are at greater risk for serious GI events.

Long-term administration of NSAIDs has resulted in renal injury. Patients at greatest risk of this reaction are those with impaired renal function, heart failure, liver dysfunction, those taking diuretics and ACE inhibitors, and the elderly. NSAIDs should be avoided in patients with severe renal failure.

NSAIDs can cause serious skin AEs, such as exfoliative dermatitis, Stevens-Johnson syndrome, and toxic epidermal necrolysis.

Acetylsalicylic acid can result in GI side effects, such as stomach pain, heartburn, nausea, vomiting, dyspepsia, and GI bleed. Patients with history of active peptic ulcer disease should avoid acetylsalicylic acid.

The following adverse reactions have been reported following acetylsalicylic acid administration:

- Body as a whole: Fever, hypothermia, thirst
- Cardiovascular: Dysrhythmias, hypotension, tachycardia
- Central nervous system: Agitation, cerebral edema, coma, confusion, dizziness, headache, subdural or intracranial hemorrhage, lethargy, seizures
- Fluid and electrolyte: Dehydration, hyperkalemia, metabolic acidosis, respiratory alkalosis
- Gastrointestinal: Dyspepsia, GI bleeding, ulceration and perforation, nausea, vomiting, transient elevations of hepatic enzymes, hepatitis, Reye syndrome, pancreatitis
- Hematologic: Prolongation of the prothrombin time, disseminated intravascular coagulation, coagulopathy, thrombocytopenia
- Hypersensitivity: Acute anaphylaxis, angioedema, asthma, bronchospasm, laryngeal edema, urticaria
- Musculoskeletal: Rhabdomyolysis
- Metabolism: Hypoglycemia (in children), hyperglycemia
- Reproductive: Prolonged pregnancy and labor, stillbirths, lower birth weight infants, antepartum and postpartum bleeding
- Respiratory: Hyperpnea, pulmonary edema, tachypnea
- Special senses: Hearing loss, tinnitus. Patients with higher frequency hearing loss may have difficulty perceiving tinnitus. In these patients, tinnitus cannot be used as a clinical indicator of salicylism.

- Urogenital: Interstitial nephritis, papillary necrosis, proteinuria, renal insufficiency and failure

For more information, refer to the prescribing information for ibuprofen[10] and acetylsalicylic acid.[11]

Diphenhydramine is contraindicated in patients with hypersensitivity to diphenhydramine hydrochloride and other antihistamines of similar chemical structure. Antihistamines should be used with caution in patients with narrow-angle glaucoma, stenosing peptic ulcer, pyloroduodenal obstruction, symptomatic prostatic hypertrophy, or bladder-neck obstruction.

Diphenhydramine may cause drowsiness, and patients should be warned about engaging in activities requiring mental alertness, such as driving a car or operating appliances, machinery, etc. Diphenhydramine may diminish mental alertness, and is more likely to cause dizziness, sedation, and hypotension in elderly patients.

Diphenhydramine should be used with caution in patients with a history of bronchial asthma, increased intraocular pressure, hyperthyroidism, cardiovascular disease, or hypertension.

The following reactions have also been reported following treatment with diphenhydramine:

- General: Urticaria, drug rash, anaphylactic shock, photosensitivity, excessive perspiration, chills, and dryness of mouth, nose, and throat
- Cardiovascular: Hypotension, headache, palpitations, tachycardia, extrasystoles
- Hematologic: Hemolytic anemia, thrombocytopenia, agranulocytosis
- Central nervous system: Sedation, sleepiness, dizziness, disturbed coordination, fatigue, confusion, restlessness, excitation, nervousness, tremor, irritability, insomnia, euphoria, paresthesia, blurred vision, diplopia, vertigo, tinnitus, acute labyrinthitis, neuritis, convulsions
- Gastrointestinal: Epigastric distress, anorexia, nausea, vomiting, diarrhea, constipation
- Genitourinary: Urinary frequency, difficult urination, urinary retention, early menses
- Respiratory: Thickening of bronchial secretions, tightness of chest or throat and wheezing, nasal stuffiness

For more information, refer to the prescribing information for diphenhydramine.[12]

1.5.2 Potential Benefits

Animals that received VIS410 as either treatment or prophylaxis experienced prolonged survival compared with vehicle/placebo-treated animals following challenge with various strains of influenza A virus.

At this time, the actual benefits of VIS410 for the treatment of influenza patients are unknown. However, data from a recently conducted Phase 2a human challenge study in 31 healthy

volunteers given influenza A demonstrated a statistically significant reduction in viral AUC following treatment with VIS410 compared with placebo with a shorter duration of symptoms (2 days) in the VIS410-treated group compared with the placebo group.

1.5.3 Dose Selection Rationale

A variety of methods were utilized to determine the efficacious dose of VIS410 for the treatment of influenza in humans. These methods included evaluating doses and exposures to ascertain the VIS410 levels that satisfied such in vitro susceptibility measures as neutralization EC_{50} and binding affinity; utilizing mg/kg dose and PK data to bridge the efficacious dose from preclinical species to humans; and performing exploratory PK/PD analyses of serum and nasal exposure vs VIS410 antiviral activity in a human influenza virus challenge study.

VIS410 broadly neutralizes influenza A strains in vitro with EC_{50} values ranging from 0.03 to 64.2 µg/mL (median of 3.15 µg/mL) and binds with high avidity (30 to 380 picomolar) to hemagglutinin across Group 1 and Group 2 subtypes. VIS410 doses of approximately 2300 mg, as used in the human challenge model, will result in C_{max} serum levels of ~873 µg/mL for the duration of viral shedding (well above the median of EC_{50} value of 3.15 µg/mL). Furthermore, in preclinical animal studies, VIS410 demonstrated efficacy at doses of 20 to 30 mg/kg or less against various Group 1 and Group 2 influenza A strains with EC_{50} values ranging from 0.57 to 2.4 µg/mL. Using a standard mg/kg conversion, these doses equate to a 1500- to 2300-mg dose in humans (assuming a 75-kg subject).

In a human influenza virus challenge study, a single dose of 2300 mg demonstrated a significant reduction in viral AUC and peak viral load compared with placebo, suggesting that doses of approximately 2300 mg may be efficacious in naturally occurring influenza infection.

For strains with higher EC_{50}, doses greater than 2300 mg may be necessary to achieve adequate concentration at the primary site of infection. Based on these data, doses of 2000 and 4000 mg were selected for evaluation in this Phase 2b study. A dose of 4000 mg will ensure separation in VIS410 systemic exposures from the 2000-mg dose level to identify potential differences in safety and efficacy between the 2 doses. A dose level of 2000 mg provides a safety margin of approximately 9.4-fold to the nonclinical NOAEL (250 mg/kg), with the 4000-mg dose providing a safety margin of approximately 4.7-fold, assuming a 75-kg subject. In addition, in the Phase 1 study, doses of 2300 and 3800 mg were well tolerated following a pretreatment regimen of diphenhydramine plus ibuprofen or acetylsalicylic acid.

2.0 STUDY OBJECTIVES

2.1 Primary Objectives

2.1.1 Primary Efficacy Objective

- Evaluation of the effect of 2 dose levels of VIS410 + oseltamivir on clinical outcome as assessed by comparison of clinical status ordinal scale Day 7 scores between treatment groups, and between all VIS410 recipients versus placebo.

2.1.2 Primary Safety Objective

- Safety and tolerability of 2 dose levels of a single IV dose of VIS410 when administered in combination with oseltamivir in hospitalized subjects with influenza A infection

2.2 Secondary Objectives

- Among patients requiring supplemental oxygen therapy at the time of enrollment with baseline room air oxygen saturation of ≤ 92%, time to cessation of oxygen (O_2) support resulting in stable oxygen saturation (SpO_2) by pulse oximetry. Stable SpO_2 is defined as two consecutive SpO_2 values of > 92% on room air that are at least 8 hours apart.
- For any patient requiring supplemental oxygen therapy at the time of enrollment (regardless of oxygen saturation), time to cessation of oxygen support.
- Evaluate the effect of VIS410 + oseltamivir vs oseltamivir alone on the following endpoints:
  - Viral load in upper respiratory samples
  - Time to clinical response
  - Time to cessation of ventilator support
  - Time to resumption of normal activities
  - All-cause and attributable Day 14, 28, and 56 mortality
  - Clinical status ordinal scale mean area under the curve for Days 1-7 and Days 1-14 using linear numeric scores for the ordinal categories.
  - Comparison of clinical status ordinal scale scores for selected individual days (ie, Days 3, 4, 5, and 6)
  - Comparison of clinical status ordinal scale scores using modified ordinal scale criteria (ie, pooling of selected severity criteria scores)
  - Comparison of discrete ordinal scale parameters, including days of ventilator support, days in intensive care, and duration of hospitalization
  - Healthcare resource utilization
  - Analysis of time to alleviation of signs and symptoms of influenza in the subset of subjects able to complete the Influenza Patient Reported Outcomes (FluPRO) Questionnaire at baseline and post-dose by Kaplan Meier analysis
  - Proportion of subjects with new documented bacterial pneumonia/superinfection
  - Proportion of subjects with influenza-related complications
- Pharmacokinetics of VIS410 in serum
- Immunogenicity of VIS410

- Emergence of resistance to VIS410 and oseltamivir

2.3 Exploratory Objectives

- Evaluate the pharmacokinetics of VIS410 from nasopharyngeal secretions and tracheal aspirate (ventilated subjects only)
- Assess the effects of VIS410 on viral load in tracheal aspirate (ventilated subjects only)
- Assess correlations between virology, safety, VIS410 dose, pharmacokinetics, viral shedding, immunology, signs and symptoms of influenza, and other endpoints
- Assess the anti-influenza immune response

3.0 STUDY ENDPOINTS

3.1 Primary Endpoints

3.1.1 Primary Efficacy Endpoint

- The primary efficacy outcome analysis compares Day 7 clinical status ordinal scale scores between treatment groups, and between all VIS410 recipients versus placebo. Clinical status is measured daily for 14 days using the below seven-level ordinal scale, with the classifications presented from the worst clinical outcome to the best clinical outcome in descending order; for each day, subject status will be classified by the worst clinical outcome for which they qualify
  - Death
  - ICU stay with mechanical ventilation
  - ICU stay without mechanical ventilation
  - Non-ICU hospitalization with supplemental oxygen
  - Non-ICU hospitalization without supplemental oxygen
  - Discharge with partial resumption of normal activities
  - Discharge with full resumption of normal activities

3.1.2 Primary Safety Endpoint

- The proportion of subjects with AEs and SAEs following administration of VIS410.

3.2 Secondary Endpoints

- Among patients requiring supplemental oxygen therapy at the time of enrollment with baseline room air oxygen saturation of ≤ 92%, time to cessation of oxygen (O_2) support resulting in stable oxygen saturation (SpO_2) by pulse oximetry. Stable SpO_2 is defined as two consecutive SpO_2 values of > 92% on room air that are at least 8 hours apart.
- For any patient requiring supplemental oxygen therapy at the time of enrollment (regardless of oxygen saturation), time to cessation of oxygen support.
- The difference between VIS410 + oseltamivir and oseltamivir alone treatment groups in the following endpoints:
  - Peak viral load, viral AUC, duration of viral shedding, and time to resolution of viral load from nasopharyngeal swabs by $TCID_{50}$ and qRT-PCR
  - Time to clinical response defined as resolution of at least 4 of 5 vital signs
    - Afebrile with core temperature ≤ 37.8°C, without use of antipyretics (oral ≤ 37.2°C)
    - Oxygen saturation ≥ 95% on room air without support or a return to pre-infection status, if pre-infection status was < 95%
    - Pulse rate ≤ 100/min
    - Systolic blood pressure ≥ 90 mm/Hg, without vasopressor use
    - Respiratory rate ≤ 24 beats per minute

- Clinical status ordinal scale mean area under curve for Days 1-7 and Days 1-14, using linear numeric scores for the ordinal categories.
- Comparison of clinical status ordinal scale scores for selected individual days (ie, Days 3, 4, 5, and 6)
- Comparison of clinical status ordinal scale scores using modified ordinal scale criteria (ie, pooling of selected severity criteria scores)
- Comparison of discrete ordinal scale parameters, including days of ventilator support, days in intensive care, and duration of hospitalization
- Number of days to resumption of usual activities
- All-cause and attributable mortality rates at Day 14, 28, and 56
- Total number of days in hospital and/or intensive care unit (ICU) from admission to discharge and rate of rehospitalization due to influenza A relapse/complication
- The incidence, severity, and duration of signs and symptoms of influenza-like illness as assessed by the FluPRO Questionnaire (see Appendix 14.1)
- Analysis of time to alleviation of signs and symptoms of influenza in the subset of subjects able to complete the Influenza Patient Reported Outcomes (FluPRO) Questionnaire at baseline and post-dose by Kaplan Meier analysis
- The percentage of subjects with new bacterial pneumonia/superinfection
- The percentage of subjects with influenza-related complications

- VIS410 population pharmacokinetic (PK) parameters in serum
- Titer of anti-VIS410 antibody positive samples
- Genotypic and/or phenotypic assessment to determine the emergence of VIS410 and oseltamivir-resistant viruses

3.3 Exploratory Endpoints

- Population PK parameters of VIS410 from nasopharyngeal secretions
- VIS410 concentrations in tracheal aspirates
- The difference in viral load between VIS410 + oseltamivir and oseltamivir alone treatment groups in tracheal aspirates of subjects on mechanical ventilation
- Titer of anti-influenza A antibodies by hemagglutinin inhibition assay (HAI) in serum
- Correlations between serum and/or nasopharyngeal PK with viral load, clinical symptoms, presence of ADAs, safety, and additional endpoints

4.0 STUDY DESIGN

4.1 Overview

This is a Phase 2b multicenter, randomized, double-blind, controlled study comparing the efficacy and safety of 2 dose levels of VIS410 (2000 and 4000 mg) in combination with oseltamivir vs oseltamivir alone in hospitalized subjects with influenza A infection requiring oxygen. Subjects will be randomly assigned at a ratio of 1:1:1 using a permuted block randomization method to receive VIS410 at a dose of 2000 mg (low dose) or 4000 mg (high dose) or placebo (0.9% sodium chloride), administered as a single IV infusion over 2 hours.

Randomization will be stratified by presence or absence of positive pressure ventilation (PPV) at baseline to ensure an even distribution across treatment arms (see Section 10.2). All subjects will receive oseltamivir 75 mg twice daily (BID) for 5 days (total of 10 doses) as part of standard of care (SOC). All subjects will also receive a pretreatment regimen of diphenhydramine 50 mg PO or IV plus either ibuprofen 400 mg PO or acetylsalicylic acid 320-325 mg PO 60 minutes before VIS410/placebo infusion. Approximately 120 evaluable subjects (40/arm) with confirmed influenza A infection will be treated.

Subjects admitted to the hospital within 5 days of onset of initial symptoms who require supplemental oxygen will undergo a rapid influenza test (supplied by the Sponsor) or a PCR, fluorescent immunoassay (FIA), or enzyme-linked immunosorbent assay (ELISA) test to confirm influenza A infection. Subjects diagnosed with influenza A will undergo the screening procedures to confirm eligibility. Study assessments are outlined in Table 1, Schedule of Assessments.

Eligible subjects will be randomized to receive either oseltamivir + VIS410 2000 mg, oseltamivir + VIS410 4000 mg, or oseltamivir + placebo (0.9% sodium chloride) at a ratio of 1:1:1 using a permuted block randomization method summarized in Table 6.

Table 6. Permuted Block Randomization Method

Subjects	Treatment	VIS410 Dose [a]	Oseltamivir Dose [b]	Infusion
n = 40	VIS410 + oseltamivir	2000 mg IV	75 mg administered BID for a minimum of 5 days	Infused over 120 minutes
n = 40	VIS410 + oseltamivir	4000 mg IV		
n =40	Placebo [c] + oseltamivir	0 mg IV		

[a] Dose administered in 200 mL.

[b] Dosage and administration should follow local prescribing information for oseltamivir based on renal function.

[c] 0.9% sodium chloride (200 mL).

Oseltamivir (Tamiflu) will be provided by the Sponsor. Treatment with oseltamivir may be extended for up to 5 additional days (up to 20 doses of total therapy) if clinical symptoms

warrant further treatment as assessed by the Investigator. Note: based on SOC, subjects may have started oseltamivir therapy prior to randomization.

Study assessments are outlined in Table 1. Subjects discharged from the hospital prior to Day 14 will attend the applicable outpatient follow-up visit(s) (eg, Day 3, Day 5, Day 7, Day 14) per Table 1.

4.2 Study Duration

The total study duration for each subject (screening through study exit) will be approximately 8 weeks (Day 56).

4.3 Data Safety Monitoring Board

An independent data safety monitoring board (DSMB) will be established to review all available safety data after 30 subjects, and subsequently, approximately 70 subjects, have completed study Day 14. The assessment of safety will be determined from vital sign measurements, physical examinations, hematology, chemistry and urinalysis laboratory testing, 12-lead electrocardiograms (ECGs), use of concomitant medications, and review of AEs. Based on the safety data provided, the DSMB will determine if it is acceptable to continue the study or reduce the dose as appropriate.

Additional DSMB reviews can occur throughout the trial as deemed necessary by the DSMB or Sponsor to ensure subject safety and well-being. Study enrollment and dosing will continue while the DSMB evaluates data.

Further details will be described in a separate DSMB charter.

4.4 Discussion of Study Design

This study is a Phase 2b, multicenter, randomized, double-blind study of 2 doses of VIS410 vs placebo in the treatment of hospitalized subjects with influenza A infection requiring oxygen support. VIS410 and placebo will be administered on top of the SOC, ie, oseltamivir.

The dose of oseltamivir (75 mg BID for 5 days) was chosen for this study based on the approved dose in the United States and other regions for the treatment of uncomplicated influenza infection. The dosage regimen of oseltamivir as the SOC in this study serves as a clinically appropriate, ethically acceptable treatment.

The study will be blinded using placebo infusion (as described in Section 7.2) to prevent bias in the assessment of effect. The investigative site personnel, the Sponsor and their representatives involved in the monitoring or conducting the study, and the subjects will all be blinded to the study drug codes.

5.0 STUDY POPULATION

5.1 Number of Subjects

Approximately 120 evaluable subjects will be randomized into 3 treatment arms: VIS410 2000mg, VIS410 4000 mg, and placebo.

5.2 Selection of Study Population

The study population comprises subjects at least 18 years of age with confirmed influenza A infection who satisfy all inclusion and exclusion criteria, and who do not have any conditions, in the opinion of the Investigator, that may place the subject(s) at increased risk, confound study data, or significantly interfere with study participation. Section 5.2.1 and Section 5.2.2, which describe the inclusion and exclusion criteria, respectively.

5.2.1 Inclusion Criteria

Subjects meeting all of the following criteria are eligible to participate in this study:

1. Male and female subjects aged ≥ 18 years. For a country where the legal age of consent is >18 years old, the country requirements should be followed.
2. Test positive for influenza A by rapid antigen test or with another commercially available test on an adequate nasopharyngeal specimen in accordance with the manufacturer's instructions, or an acceptable local test including, PCR, FIA, or ELISA.
3. Onset of influenza symptoms no more than 5 days before VIS410/placebo infusion; symptoms may include cough, dyspnea, sore throat, fever, myalgias, headache, nasal symptoms (rhinorrhea, congestion), fatigue, diarrhea, anorexia, nausea, and vomiting.
4. Requirement for oxygen support including any positive pressure ventilation (PPV).
5. Women of childbearing potential must have a negative pregnancy test within 2 days prior to VIS410/placebo infusion.
6. Women should fulfill one of the following criteria:
    a. Post-menopausal: either amenorrhea ≥ 12 months or follicle stimulating hormone > 40 mIU/mL as documented in their medical history
    b. Surgically sterile; hysterectomy, bilateral oophorectomy, or tubal ligation
    c. Women of childbearing potential participating in heterosexual relations must be willing to use adequate contraception from screening until 60 days post-VIS410/placebo infusion (see Section 6.2).
7. Non-vasectomized (or vasectomized less than 6 months prior to dosing) male subjects who have a female partner of childbearing potential must use an effective birth control method (see Section 6.2) when having heterosexual intercourse, from screening until 60 days post-VIS410/placebo infusion.
8. Subject is able and willing to comply with study procedures, as per protocol.
9. Subject, or a legally acceptable representative, is able to understand the purpose and risks of the study and willing to give voluntary written informed consent.

5.2.2 Exclusion Criteria

Subjects meeting any of the following criteria are excluded from participation in this study:

1. Known or suspected intolerance or hypersensitivity to VIS410, oseltamivir, pretreatment medications (diphenhydramine, or to both ibuprofen and acetylsalicylic acid [ASA]), or closely related compounds (eg, other monoclonal antibodies).
2. Subjects who have received VIS410 in the past.
3. Subjects who have a history of receiving monoclonal antibody products within 3 months prior to VIS410/placebo dosing or planned administration of another monoclonal antibody during the study period.
4. Subjects who have taken more than 6 doses of an approved antiviral therapy for influenza within the prior 96 hours (eg, oral oseltamivir, inhaled zanamivir, IV peramivir, or oral ribavirin) between onset of symptoms and VIS410/placebo dosing.
5. Subjects with known co-infection with influenza B or other viral respiratory infections (eg, RSV, parainfluenza viruses, respiratory adenoviruses).
6. Subjects with lung transplant or history of severe chronic lung disease, including cystic fibrosis or any condition requiring home oxygen therapy.
7. Subjects on extracorporeal membrane oxygenation at time of randomization.
8. Subjects with ESRD who are not undergoing hemodialysis.
9. Subjects with active graft-vs-host disease, hematopoietic stem cell transplant within the previous 90 days, or human immunodeficiency virus infection with a CD4 cell count of less than 200 per cubic millimeter.
10. High probability of mortality within 48 hours of randomization as determined by the Investigator.
11. Women who are pregnant, breast-feeding, or considering to become pregnant.
12. Subjects in whom nasopharyngeal swabbing is not possible.
13. Subjects weighing less than 45 kg.
14. Enrollment in any other investigational drug or device study, any disease or vaccine study within 30 days prior to Day 1 or within 5 half-lives of the investigational compound, whichever is longer.
15. Presence of any preexisting illness that, in the opinion of the Investigator, would place the subject at an unreasonably increased risk through participation in this study.
16. Subjects unable to comply with study protocol procedures and study visit schedules for whatever reason.
17. Known or suspected alcohol or drug abuse, that is, abuse of a level that would compromise the safety or cooperation of the subject in the opinion of the Investigator.

6.0 STUDY ASSESSMENTS

Table 1 summarizes the schedule for study visits and procedures performed at each study visit.

6.1 Prescreening, Screening, and Study Procedures

6.1.1 Prescreening Procedures

Subjects may be asked to sign a Prescreening Consent prior to undergoing a single nasopharyngeal swab (one nostril) for rapid influenza A testing. Subjects will be given a full explanation of the nature of the study and provide full written informed consent before any study-specific assessments or procedures are performed. Note: subjects that tested positive for influenza A within 48 hours prior to screening do not need to be retested.

6.1.2 Screening Procedures

Subjects will be given a full explanation of the nature of the study, provide full written informed consent before any study-specific assessments or procedures are performed, and undergo full screening procedures. Subjects will be assessed for inclusion and exclusion criteria, and those who fulfill all the inclusion and none of the exclusion criteria will be randomized into the study. A screen failure log must be maintained by the Investigator for all entry criteria failures. Table 1 lists the procedures performed at screening. Screening and baseline assessments (ie, Day 1 evaluations and procedures) may be performed on the same day.

6.1.3 Study Procedures

On Day 1, eligibility of the subjects will be confirmed and assessments will be performed as described in Table 1.

All results from the screening procedure needed to evaluate eligibility, including any local clinical laboratory results, must be available prior to randomization on Day 1. All clinical assessments required for the determination of subject eligibility will be performed by the local clinical laboratory, thereby precluding the need to wait for central laboratory data. Any abnormal assessment at the screening visit will be assessed according to its clinical relevance, and if found relevant, the subject will not be included in the study.

Once a subject has satisfied entry criteria, he/she will be assigned a unique identifier. The site's unblinded pharmacist or properly trained designee will access the Interactive Web Response System (IWRS) to obtain the study treatment. Randomization will be stratified by presence or absence of PPV at baseline to ensure balance between the treatment arms. Subjects will be considered randomized when the pharmacist or designee obtains the subject number and treatment assignment from the IWRS.

Subjects may not be randomized into this study more than once. Subjects who have participated in any previous study of VIS410 may not be randomized to this study. Subjects will be

monitored daily while in the hospital up to Day 14 (± 3 days) with an additional visit on Day 28 (± 3 days) and the last follow-up visit on Day 56 (± 7 days). In order to provide some flexibility for the subjects regarding the site visits and to maintain the integrity of the study design, a time window is permitted for the follow-up visits in case of time conflict or unforeseen circumstances. Note: subjects discharged from the hospital prior to Day 14 will attend the applicable outpatient follow-up visit(s) (eg, Day 3, Day 5, Day 7, Day 14).

6.1.4 Unscheduled Visits

Unscheduled visits can be planned at the discretion of the Investigator to obtain additional information to ensure the safety of the subject.

Unscheduled visits may be planned to assess, confirm, and follow up on out-of-range clinical laboratory test, vital sign, or ECG values that determine a subject's eligibility. Findings made during unscheduled visits should be recorded in the EDC system and the source documents.

Subjects will be instructed to contact the trial center if they exhibit worsening of symptoms or a new episode of influenza-like illness, so that they may be monitored and asked to return to the site for additional assessments (safety, nasopharyngeal swabbing) at the Investigator's discretion.

6.1.5 Early Withdrawal Procedures

Subjects should be encouraged to complete all study assessments. However, subjects who terminate the study before completion of the scheduled study procedures for any reason (AE, withdrawal of consent, etc.) should be encouraged to complete the early termination assessments (ie, identical to end of therapy [ET] assessments), including safety, prior to withdrawal, if possible.

6.1.6 Treatment of VIS410 Overdose

Given the absence of previous instances of VIS410 overdosing, clinicians have no specific treatment for the treatment of VIS410 overdose. However, high doses (ie, up to 4600 mg) have been evaluated in clinical study without any incidents of significant toxicity. In the event of VIS410 overdose, treatment administered to the subject(s) should be symptomatic and supportive.

Any overdose in this clinical study, with or without associated AEs, must be reported to the contract research organization (CRO) according to the procedures for SAE reporting as outlined in Section 9.7.1 and Section 9.7.2. All reports of overdose must be documented in the EDC. Any AEs associated with the overdose should be reported on relevant AE/SAE sections in the EDC.

6.2 Pregnancy Safeguards

Pregnancy will be determined by evaluation of β-human chorionic gonadotropin in serum or urine for all women of childbearing potential. Subjects who are pregnant or nursing will be

excluded from the study. During the course of the study drug administration period within the study, any nursing mother(s) or subject(s) with suspected or confirmed pregnancy will be discontinued from study drug therapy but will be encouraged to undergo follow-up for safety monitoring for themselves (ie, the pregnant female) and the baby.

All women of childbearing potential and all male subjects must practice effective contraception from screening until 60 days post-VIS410/placebo infusion. For the purposes of this study, women who do not satisfy at least one of the following criteria listed below (ie, the criteria for defining non-childbearing potential) are considered to be physiologically capable of becoming pregnant and are, therefore, defined as women of childbearing potential.

The criteria for defining women as being of non-childbearing potential are:

- Post-menopausal: ≥ 12 months of natural (spontaneous) amenorrhea, or
- Follicle stimulating hormone > 40 mIU/mL as documented in their medical history, or
- Surgical bilateral oophorectomy with or without hysterectomy, or
- Hysterectomy, or
- Bilateral tubal ligation

Women of childbearing potential and all male subjects participating in heterosexual relations must be willing to practice effective contraception from screening until 60 days post-VIS410/placebo infusion. For the purposes of the study, highly effective contraception is defined as:

- Male vasectomy with negative semen analysis documentation at least 6 months prior to dosing.

OR

- Combination of an established form of hormonal contraception (oral, injected, or implanted) or an intrauterine device or intrauterine system or sponge

Plus one of the following:

- A physical barrier method of contraception with use of a spermicide, such as condom or occlusive cap (diaphragm or cervical/vault caps) with spermicidal foam/gel/film/cream suppository. The use of barrier contraceptives should always be supplemented with the use of a spermicide, unless not available in a country. OR
- Male vasectomy with negative semen analysis documentation less than 6 months prior to dosing. OR
- Complete abstinence can be considered an acceptable method of contraception at the discretion of the Investigator. Periodic abstinences (eg, calendar, ovulation, symptothermal, or post-ovulation methods) and withdrawal are NOT considered acceptable methods of contraception.

The combination of 2 barrier methods, periodic abstinence (eg, calendar, ovulation, symptothermal, or post-ovulation methods), and withdrawal are not considered acceptable methods of contraception.

Male subjects should not donate sperm for at least 60 days after receipt of study product.

Pregnancy reporting is described in Section 9.7.3.

6.3 Procedures for Sample and Data Collection

6.3.1 SpO_2 Oxygen Support

Baseline SpO_2 on room air to be documented, if available. Once randomized, SpO_2 will be measured using pulse oximetry 3 times per day (approximately every 8 hours) at approximately the same time each day until stable according to Table 1. Stable SpO_2 is defined as two consecutive SpO_2 values of > 92% on room air that are at least 8 hours apart.

6.3.2 Viral Load

A nasopharyngeal swab will be collected from each nostril as described in Table 1 and Laboratory Manual. These swabs will be sent to a central virology laboratory for processing into appropriate aliquots for virology and PK analysis. Further details on sample collection, processing, shipment, and storage will be described in the Laboratory Manual.

The Investigator may also perform additional tests in the local laboratory consistent with SOC for the illness being treated.

Note: Virology samples taken from all subjects may be infectious and will be classified as "diagnostic specimens" for dispatch purposes.

6.3.3 Time to Cessation of Ventilator Support

Start and stop time for mechanical ventilation will be documented in EDC.

6.3.4 Resumption of Normal Activities

Subjects will complete a visual analog scale (VAS) daily from Day 1 (baseline) until either the subject reports that all usual activities (prior to influenza onset) can be performed or Day 56, whichever comes first. The VAS scale ranges from 0 to 10, where 0 indicates subject is unable to perform usual activities at all, and 10 indicates subject is able to perform all usual activities fully)

6.3.5 Health Resource Utilization

The following parameters for health resource utilization will be documented on an ongoing basis during the study:

- Hospital admission and discharge date
- ICU admission and discharge date
- Rehospitalization and reason for rehospitalization up to the last follow-up visit (Day 56 ± 7 days)

6.3.6 Signs and Symptoms of Influenza

The Influenza Patient-Reported Outcome (FluPRO) Questionnaire (see 14.1) must be completed (ie, self-reported) by the subject at baseline (Day 1) and post-dose as defined in Table 1. Each subject will be evaluated at baseline to determine if subject is able to independently report his/her influenza signs and symptoms. Subjects who are not able to complete the baseline FluPRO Questionnaire are exempt from this assessment throughout the study. The subject's inability to self-report his/her symptoms of influenza at baseline should be documented in the EDC.

6.3.7 Serum Pharmacokinetics

Throughout the study, 5-mL venous blood samples will be collected for analysis of VIS410 in serum, according to the time points defined in Table 1. The exact date and time of blood sampling and of administration of the study drug must be recorded in the EDC system. Serum samples for determination of the concentration of VIS410 will be analyzed under the responsibility of the Sponsor. Further procedures for sample collection, shipment, processing, and storage will be described in the Laboratory Manual.

6.3.8 Viral Resistance

Samples for virologic and PK analysis will be collected as described in Table 1 and the Laboratory Manual. These samples will be sent to a central virology laboratory for processing into appropriate aliquots for virology and PK analysis, including resistance testing.

Further procedures for sample collection, processing, shipment, and storage will be described in the Laboratory Manual.

Note: Nasopharyngeal and tracheal samples taken from all subjects may be infectious and will be classified as "diagnostic specimens" for dispatch purposes.

6.3.9 Clinical Outcomes per Seven-Level Ordinal Scale

The ordinal scale outcomes will be measured daily from Day 1 (baseline) through Day 14 using the following seven-level hierarchical scale with the classifications presented from the worst clinical outcome to the best clinical outcome in descending order.

For each day, subjects will be classified by the worst clinical outcome for which they qualify. For example, a patient with both ventilation and oxygen on the hospital floor on Day 1 will be classified as a mechanical ventilation patient for Day 1.

- Death
- ICU stay with mechanical ventilation
- ICU stay without mechanical ventilation
- Non-ICU hospitalization with supplemental oxygen
- Non-ICU hospitalization without supplemental oxygen
- Discharge with partial resumption of normal activities
- Discharge with full resumption of normal activities

6.3.10 Pharmacokinetics of Nasopharyngeal Samples

A nasopharyngeal swab will be collected from each nostril as described in Table 1 and Laboratory Manual. These swabs will be sent to a central virology laboratory for processing into appropriate aliquots for virology and PK analysis. Further details on sample collection, processing, shipment, and storage will be described in the Laboratory Manual.

Note: Nasopharyngeal samples taken from all subjects may be infectious and will be classified as "diagnostic specimens" for dispatch purposes.

6.3.11 Pharmacokinetics and Viral Load in Tracheal Aspirate

Tracheal aspirate will be obtained in subjects on mechanical ventilation, at the time points listed in Table 1. The samples will be sent to a central virology laboratory for processing into appropriate aliquots for virology and PK analysis. Samples for determination of the concentration of VIS410 will be analyzed under the responsibility of the Sponsor. Further details on sample collection, processing, shipment, and storage will be described in the Laboratory Manual.

Note: Tracheal samples taken from all subjects may be infectious and will be classified as "diagnostic specimens" for dispatch purposes.

6.3.12 Immunology

The immunology parameters under evaluation are the detection of ADA titers and anti-influenza A antibody titers by HAI.

Blood samples for immunology assessments will be collected according to the time points defined in Table 1. Blood samples will be collected by venipuncture or via indwelling cannula in the forearm into standard serum separator tubes. The exact date and time of blood sampling must be recorded in the EDC system. Further procedures for sample collection, shipment, processing, and storage will be described in the Laboratory Manual.

6.3.12.1 Antidrug Antibody Response

Anti-VIS410 antibody titers will be determined by Visterra or designee. These samples may be stored to assess anti-VIS410 antibodies with neutralizing capabilities in accordance with local regulations.

6.3.12.2 Anti-Influenza Antibody Response

Anti-influenza A antibody titers will be determined by Visterra or designee.

6.4 Safety of Total Blood Sampling Volume

The total volume of blood that will be drawn from each subject will be approximately 178 mL over the course of the study. In order to obtain additional information to ensure a subject's safety, additional blood samples (up to 50 mL) may be taken at the discretion of the Investigator.

6.5 Safety Evaluations

The safety assessment in this study will be based on AEs, clinical laboratory tests, vital signs, electrocardiography, and physical examination, as described in the following sections.

6.5.1 Adverse Events

Adverse events (AEs) will be monitored continuously from dosing until the last study-related activity. At regular intervals during the study, subjects will be asked non-leading questions to determine the occurrence of any AEs. All AEs reported spontaneously during the course of the study will be recorded as well.

Subjects will be observed during infusion and for at least 2 hours after the completion of administration of VIS410/placebo to allow monitoring for possible hypersensitivity reactions, anaphylactic reactions, or other AEs that occur during this time period.

All AEs will be recorded in the EDC system. For detailed definitions and reporting procedures of AEs (see Section 9.7).

6.5.2 Injection Site Tolerability

Injection site tolerability is defined as AEs demonstrating significant local injection site irritation or tissue damage.

6.5.3 Complications of Influenza

Complications of influenza are defined as pneumonia, myocarditis, worsening of chronic bronchitis, sinusitis, otitis or other chronic pulmonary diseases, encephalopathy, and death. Complications of influenza and use of antibiotic therapy for bacterial pneumonia will be reported by variable, treatment group, and time point. Diagnosis of bacterial pneumonia will be based on

local clinical practice, usually an assessment of clinical features and any radiographic evidence of bacterial pneumonia (eg, infiltrate) and/or microbiologic evidence of bacterial pneumonia. All-cause and attributable mortality will be evaluated on Day 14 and Day 28.

6.5.4 Clinical Laboratory Tests

Blood samples will be collected by venipuncture or via indwelling cannula at the time points indicated in Table 1.

Appendix 14.2 lists the biochemistry, hematology, and coagulation tests that will be performed by the central laboratory on the safety blood samples. Note: subjects to be enrolled based on the Investigator's discretion including any local laboratory results per SOC at institution.

Creatine kinase-MB, creatinine kinase, and troponin are to be measured by the central laboratory in the event of a subject having chest pain. Subjects with chest pain should be managed per SOC.

In the event of a subject exhibiting the signs and symptoms of an anaphylactic reaction, when possible at the time of the event, a 5-ml serum sample should be collected for further assessment (ie, tryptase, chymase) to be measured by the central laboratory.

A midstream urine sample will be collected for the central laboratory for urinalysis by dipstick, flow cytometry, and microscopic examination. Appendix 14.2 lists the urinalysis parameters that will be assessed.

A serum pregnancy test will be performed by the central laboratory for all time points indicated in Table 1. In addition, a negative pregnancy test (serum or urine) result within 2 days prior to dosing must be available from the local laboratory prior to randomization.

For oseltamivir dosing, creatinine clearance (Cockcroft-Gault Equation) must be calculated using the serum creatinine value from the local laboratory prior to randomization.

The Investigator must review the laboratory report, document this review, and record any change occurring during the study he/she considers to be clinically relevant in the EDC system. Laboratory values outside the normal range will be flagged, and their clinical relevance will be assessed by the Investigator.

6.5.5 Vital Signs and Body Temperature

Vital sign parameters will be assessed after 5 minutes in supine position at the time points indicated in Table 1. The vital sign parameters that will be assessed are supine SBP, DBP, heart rate, and respiratory rate.

Any change from baseline in vital sign values occurring during the study that is considered to be clinically relevant by the Investigator should be recorded as an AE.

Oral body temperature will be recorded at the end of infusion then BID while in the hospital and then throughout the study according to the study procedures outlined in Table 1. In cases where obtaining oral temperature is not possible, core temperature (eg, tympanic, axillary) will be obtained.

While the patient is hospitalized, the maximum temperature should be recorded for each 12-hour interval (from 12 AM to 12 PM and from 12 PM to 12 AM). Fever is defined as a core body temperature ≥ 38°C.

6.5.6 Electrocardiography

Single, 12-lead ECGs will be performed after a 5-minute rest at the time points indicated in Table 1. The ECG parameters assessed are heart rate, PR interval, QRS interval, and QT interval. Values for QT corrected for heart rate (QTc) will be derived. QTc corrected according to Fridericia (QTcF) will be the primary correction parameter.[13] QTc parameters can be calculated subsequently if not immediately available at the site.

Any change from baseline in ECG values occurring during the study that is considered to be clinically relevant by the Investigator should be recorded in the EDC system.

6.5.7 Physical Examination

A complete physical examination will be performed at the time points indicated in Table 1. Physical examination at screening will include height and weight. To obtain the actual body weight, subjects must be weighed at screening. The height should be measured barefoot, if possible. The screening physical examination will be complete; thereafter, targeted physical examinations may be performed at the discretion of the Investigator.

A complete physical examination includes examination of body systems (including, but not limited to, general appearance, skin, neck, eyes, ears, nose, throat, breast, lungs, heart, abdomen, back, lymph nodes, extremities, and nervous system).

The targeted physical examination should be focused, at the Investigator's discretion, based on the subject's condition and circumstances. The targeted physical examination should note any changes in the subject's condition (body systems) since the last assessment and does not preclude examination of any of the body systems as clinically indicated.

Any change in physical examination occurring during the study that is considered to be clinically relevant by the Investigator should be recorded as an AE.

6.5.8 Radiological Assessment

A baseline chest x-ray or CT scan will be performed for the assessment of pneumonia. However, a chest x-ray or CT scan performed as part of routine SOC within 72 hours before randomization will be acceptable. The results of any such studies will be recorded in the EDC.

6.6 Appropriateness of Measurements

The assessments that will be made in this study are standard, and are generally recognized as reliable, accurate, and relevant.

7.0 STUDY TREATMENTS

7.1 Test Product, Dose, Mode of Administration

The Investigator must ensure that the investigational product will be used only in accordance with the protocol. It is forbidden to use investigational drug material for purposes other than as defined in this protocol.

VIS410 (2000 mg or 4000 mg) will be administered IV over 2 hours as a single, 200-mL infusion, followed by a 25-mL (or volume equivalent to length of IV line) saline flush to ensure all product is administered, in addition to oseltamivir (Tamiflu) 75 mg BID for total of 5 days (10 doses). Treatment with oseltamivir may be extended for up to 5 additional days (up to 20 doses of total therapy) if clinical symptoms warrant further treatment as assessed by the Investigator. Note: based on SOC, subjects may have started oseltamivir therapy prior to randomization. No dose adjustment is necessary for VIS410 based on renal or hepatic impairment. For patients with renal insufficiency, refer to prescribing information for administration of oseltamivir.

VIS410 is a colorless to slightly yellow, clear to opalescent solution, essentially free of particles. VIS410 is formulated at a concentration of 25 mg/mL in 40-mM citrate-sodium phosphate, 150-mM sodium chloride, and 0.025% polysorbate 80.

The investigational product will be provided by the Sponsor. VIS410 will be supplied in Type I 20-mL glass vials containing a nominal 20-mL solution. A copy of the certificate of analysis of the investigational product will be sent to the clinical center.

The investigational drug product is manufactured by Lyophilization Services of New England in accordance with Good Manufacturing Practice as required by the current Good Clinical Practice (GCP). Manufacturing, packaging, and labeling of the investigational product, VIS410, is conducted under the responsibility of the Sponsor. The study drug will be labeled according to local law and regulatory requirements. Specific dilution procedures for VIS410 will be described in detail in the Pharmacy Manual.

Placebo will be a normal saline solution (0.9%) and will be prepared by the pharmacist.

7.2 Reference Product, Dose, Mode of Administration

Placebo (normal saline solution 0.9%) will be administered IV over 2 hours as a single 200-mL infusion, followed by a 25-mL (or volume equivalent to volume of IV line) saline flush to ensure all product is administered, in addition to oseltamivir (Tamiflu) 75 mg BID for total of 5 days (10 doses). Treatment with oseltamivir may be extended for up to 5 additional days (up to 20 doses of total therapy) if clinical symptoms warrant further treatment as assessed by the Investigator. For patients with renal insufficiency, refer to prescribing information for administration of oseltamivir. Note: based on SOC, subjects may have started oseltamivir therapy prior to randomization.

7.3 Instructions for Preparation, Use, and Administration

The required amount of VIS410 to be dosed (2000 mg or 4000 mg) will be diluted with normal saline up to a total volume of 200 mL. For placebo subjects, 200 mL of normal saline will be prepared. This infusion will be followed by a 25-mL (or if IV line volume is >25 mL, an equivalent volume) saline flush. The saline flush following administration will ensure all VIS410/placebo has been administered.

The study infusion will be administered IV using a 0.22-µm in-line filter and will be controlled by a volumetric pump. Standard, infusion lines will be used, and microfilters will be provided by the Sponsor as appropriate.

The infusion bag will be covered with an opaque sleeve in the pharmacy to maintain the study blind. The VIS410/placebo will be administered IV at a rate of 100 mL/h, over 2 hours.

When 200 mL has been administered, the line will be flushed with normal saline. The infusion time may be longer at the Investigator's discretion based only on local infusion site–related symptoms up to a maximum of 4 hours.

Refer to the Pharmacy Manual for directions on storage, handling, stability data, preparation, and use. For more detailed information on VIS410, refer to the current Investigator's Brochure.[8]

7.4 The Use of Oseltamivir as Standard of Care

All subjects will receive oseltamivir 75 mg BID for 5 days as part of SOC. Treatment with oseltamivir may be extended for up to 5 additional days (up to 20 doses of total therapy) if clinical symptoms warrant further treatment as assessed by the Investigator. Oseltamivir (Tamiflu) will be provided by the Sponsor. Note: based on SOC, subjects may have started oseltamivir therapy prior to randomization.

For patients unable to swallow, oseltamivir capsules can be opened and mixed with liquid for ease of administration.

Dose adjustment is recommended for patients with a serum creatinine clearance of 60 mL/min or less[9] (see Table 7).

Table 7. Recommended Dosage Modifications for Treatment of Influenza in Adults with Renal Impairment or End-Stage Renal Disease on Dialysis [a]

Renal Impairment (Creatinine Clearance)	Recommended Treatment Regimen [b]
Mild (> 60-90 mL/minute)	75 mg BID for 5 days
Moderate (> 30-60 mL/minute)	30 mg BID for 5 days
Severe (> 10-30 mL/minute)	30 mg once daily for 5 days
ESRD patients on hemodialysis	30 mg immediately and then 30 mg after every hemodialysis cycle (treatment duration not to exceed 5 days)
ESRD patients on continuous ambulatory peritoneal dialysis [c] (≤10 mL/minute)	A single 30-mg dose administered immediately
ESRD patients **not** on dialysis	Tamiflu is not recommended

ESRD = end-stage renal disease.

[a] Creatinine clearance to be calculated using Cockcroft-Gault formula.

[b] Capsules or oral suspension can be used for 30-mg dosing.

[c] Data derived from studies in continuous ambulatory peritoneal dialysis patients.

For patients with renal insufficiency, refer to prescribing information for administration of oseltamivir.[9]

The dose of oseltamivir (75 mg BID for 5 days) was chosen for this study based on the approved dose in the United States and other regions for the treatment of uncomplicated influenza infection. The dosage regimen of oseltamivir as the SOC in this study serves as a clinically appropriate, ethically acceptable treatment.

7.5 Pretreatment Regimen

All subjects will be given a pretreatment regimen of PO or IV diphenhydramine 50 mg plus either ibuprofen 400 mg or acetylsalicylic acid 320-325 mg PO or crushed and given through the nasogastric tube approximately 60 minutes before IV infusion of VIS410/placebo.

If the subject has any history of delayed gastric emptying, including premenstruation syndrome or menstruation, the subject may receive premedications 120 minutes prior to IV infusion of VIS410/placebo.

Refer to the Pharmacy Manual for directions on storage, handling, stability data, and preparation and use of diphenhydramine, ibuprofen, and acetylsalicylic acid.

For more information, refer to the prescribing information for ibuprofen[10] and acetylsalicylic acid.[11]

7.6 Other Medications Administered in This Study

All prescription and over-the-counter medications that the subject received from 7 days prior to study drug and throughout the study (up to the LFU evaluation) will be documented in the EDC.

Herbal, nutritional, and dietary supplements received by the subject within 7 days before randomization will be documented in the EDC only if medically indicated.

Administration of concomitant medications must be reported in the appropriate section of the EDC along with dates of administration and reasons for use. Generic names for medications should be used, if possible.

Refer to the Pharmacy Manual for directions on storage, handling, stability data, and preparation and use of oseltamivir.

For more information, refer to the prescribing information for oseltamivir.[9]

7.7 Treatment Compliance

Investigative product will be administered by study staff, and all drug administration data will be reported in the EDC and the medical records of each subject.

The number of oseltamivir doses administered while the subject is in the hospital will be documented by the Investigator or his/her designee. In case of discharge while on oseltamivir therapy, subjects will document the number of doses taken at home and any missed doses; subjects self-administering oseltamivir at home will bring the pack(s) to each study visit so study staff may count and record the number of pills taken. Any unused capsules must be returned to the study site.

7.8 Storage and Drug Accountability

The Investigator (or designee) is responsible for the safe storage of all study drugs assigned to the clinical site. The investigational product should be stored in a locked, secure storage facility with access limited to those individuals authorized to dispense the investigational product, and maintained within the appropriate ranges of temperature. All study drugs must be stored in the original packaging and as specified at delivery.

The study investigational product be stored between 2°C and 8°C and should be protected from light during storage at the clinical site.

For storage of oseltamivir (Tamiflu), diphenhydramine, ibuprofen, and acetylsalicylic acid, refer to the manufacturer’s prescribing information. Also refer to the Pharmacy Manual for directions on storage, handling, stability data, and preparation and use of diphenhydramine, ibuprofen, and acetylsalicylic acid.

Regular temperature recordings of the study drug storage room at the clinical site should be performed. In case a deviation in the storage conditions should occur, the site must not further dispense the affected study drug; instead, the Sponsor should be notified and will confirm whether or not the study drug may be further dispensed.

The Investigator is responsible for ensuring that all study drugs received at the clinical site are inventoried and accounted for throughout the study.

Study drugs should be dispensed under the supervision of the Investigator, a qualified member of the investigational staff, or by a hospital/clinic pharmacist. The Investigator must maintain accurate records demonstrating date and amount of drugs administered to whom and by whom. Study drugs will be supplied only to subjects participating in the study.

The monitor responsible for drug accountability will periodically check the supplies of study drugs held by the Investigator or pharmacist to ensure accountability and appropriate storage conditions for all the study drugs held at the site.

Unused study drugs must be available for verification by the Sponsor's monitor responsible for drug accountability during on-site monitoring visits. Any discrepancies between returned and expected returned study drugs should be explained.

After the last visit of the last subject in the study (LSLV), any used and unused investigational product will be returned to the Sponsor or, with the Sponsor's written permission, destroyed at the investigative site per the standard operating procedures.

Hazardous materials, such as used needles and syringes, should be disposed of immediately in a safe manner and therefore will not be retained for drug accountability purposes.

8.0 STUDY METHODS AND PROCEDURES

Table 1 summarizes the schedule for study visits and procedures performed at each study visit.

8.1 Prior & Concomitant Therapy

All medications taken within 7 days prior to signing of the informed consent form will be documented in the EDC. All therapies other than the study drug administered from signing the informed consent form until the last study visit must be recorded in the EDC system (name of the drug, dosage, route, and dates of administration).

8.1.1 Permitted Prior Therapy

Subjects are allowed to receive up to 6 doses of an approved anti-influenza therapy within the prior 96 hours (ie, oral oseltamivir, inhaled zanamivir, or oral ribavirin) between onset of symptoms and VIS410/placebo dosing.

8.1.2 Permitted Concomitant Therapies

Use of contraception or other concomitant therapy for management of subjects' other underlying medical conditions is permissible during the study.

All other concomitant medications necessary for the health and well-being of the subject will be permitted. In addition, any other treatment (not explicitly excluded) which is considered necessary for the subject's welfare may be given at the discretion of the Investigator.

8.2 Prohibited Prior & Concomitant Medications

8.2.1 Prohibited Prior Medications

Per Exclusion Criterion 2 (Section 5.2.2), subjects are not allowed to receive monoclonal antibody products within 3 months prior to VIS410/placebo dosing.

Per Exclusion Criterion 3 (Section 5.2.2), subjects who have taken more than 6 doses of an approved anti-influenza therapy within the prior 96 hours (ie, oral oseltamivir [Tamiflu], inhaled zanamivir [Relenza], or oral ribavirin) between onset of symptoms and VIS410/placebo dosing will be excluded from the study.

8.2.2 Prohibited Concomitant Medications

Per Exclusion Criterion 2 (Section 5.2.2), subjects will not be allowed to receive additional monoclonal antibody products during the study period. Following randomization, use of other antiviral therapy for treatment of influenza A infection will not be permitted. Excluded antiviral medications include but are not limited to rimantadine, amantadine, peramivir, zanamivir, and laninamivir.

The use of symptom-modifying drugs, such as NSAIDs, antihistamines, or pseudoephedrine, are not specifically excluded; however, their use outside of pretreatment should be discouraged by the Investigator, and paracetamol/acetaminophen should be encouraged as a replacement medication.

8.3 Schedule of Examination Procedures by Study Visit

The study comprises up to 8 study visits over approximately 8 weeks. Subjects will attend the clinic at every study visit.

AEs will be recorded from dosing to the subject's last visit.

Table 1 summarizes the schedule for study visits and procedures performed at each study visit.

8.3.1 Sequence of Assessments at a Single Visit

If the following assessments are to be performed at the same study visit, the FluPRO Questionnaire should be completed first, when possible:

- FluPRO Questionnaire
- ECG
- Blood sampling
- Nasopharyngeal swab

8.3.2 Day of Treatment Procedures (Day 1)

On Day 1, eligibility of the subjects will be confirmed and assessments will be performed as described in Table 1.

All results from the screening procedures required to evaluate eligibility must be available prior to randomization on Day 1. Any abnormal assessment at the screening will be assessed according to its clinical relevance, and if found relevant, the subject will not be included in the study.

Once a subject has fulfilled the entry criteria, he/she will be assigned a unique identifier. The site's unblinded pharmacist or properly trained designee will randomize the subject using the IWRS.

On Day 1, all subjects will receive the pretreatment regimen prior to receiving VIS410/placebo.

8.3.3 Post-Treatment Procedures (Day 3 to 56)

Subjects will be monitored daily while in the hospital (up to Day 14) with an additional visit on Day 28 (± 3 days), and the last follow-up visit on Day 56 (± 7 days) per the procedures and

schedule listed in Table 1. Note: subjects discharged from the hospital prior to Day 14 will attend the applicable outpatient follow-up visit(s) (eg, Day 3, Day 5, Day 7, Day 14) per Table 1.

In order to provide some flexibility for the subjects regarding the site visits and to maintain the integrity of the study design, a time window is permitted for the follow-up visits in case of time conflict or unforeseen circumstances.

8.4 Subject Diary

A subject diary will be provided upon discharge from the hospital and will record the following:

- Daily oseltamivir dosing to be completed for as long as the subject continues to take oseltamivir
- Daily VAS for assessing resumption of usual activities to be completed up until either the subject reports that all pre-influenza usual activities can be performed or Day 56, whichever comes first
- Daily Influenza Patient Reported Outcomes (FluPRO) Questionnaire to be completed until Day 14, if applicable

9.0 ADVERSE EVENTS

9.1 Definitions

9.1.1 Adverse Event

An AE is any untoward medical occurrence in a patient administered a medicinal (investigational or non-investigational) product in a clinical study. An AE does not necessarily have a causal relationship with the treatment. An AE can therefore be any unfavorable and unintended sign (including an abnormal finding), symptom, or disease temporally associated with the use of a medicinal (investigational or non-investigational) product, whether or not related to that medicinal (investigational or non-investigational) product.

This includes any occurrence that is new in onset or aggravated in severity or frequency from the baseline condition, or abnormal results of diagnostic procedures, including clinical laboratory test abnormalities.

9.1.2 Signs and Symptoms of Influenza vs Adverse Events

The solicited and unsolicited signs and symptoms of influenza will not be reported as AEs as these constitute an endpoint of the study and will be recorded as such.

9.1.3 Adverse Event of Special Interest

An adverse event of special interest (AESI; serious or non-serious) is one of scientific and medical concern specific to the Sponsor's product or program, for which ongoing monitoring by the Sponsor may be warranted. Such an event should be entered into EDC in a timely manner and if applicable, reported as an SAE. Depending on the nature of the event, rapid communication by the trial Sponsor to other parties (eg, regulators) might also be warranted (based on CIOMS VI).

For VIS410, the AESIs include the following:

- Abdominal cramping
- Diarrhea/Loose stool
- Nausea
- Vomiting
- Pruritus
- Rash
- Hypotension
- Throat tightening
- Trouble breathing or wheezing

9.1.4 Serious Adverse Events

A serious adverse event (SAE) is any untoward medical occurrence that at any dose meets any of the following conditions:

- Results in death.
- Is life-threatening, ie, the patient was at risk of death at the time of the event (eg, ventricular fibrillation and anaphylaxis). The term does not refer to an event which hypothetically might have caused death if it were more severe.
- Requires inpatient hospitalization or prolongation of existing inpatient hospitalization. Hospitalization refers to an overnight admission into hospital for the purpose of investigating and/or treating the AE. Hospitalization for an elective procedure, or routinely scheduled treatment for a preexisting condition that has not worsened, is not an SAE.
- Results in persistent or significant disability/incapacity, ie, causing substantial disruption of the patient's ability to conduct normal life.
- Is a congenital anomaly/birth defect.
- Is medically significant, ie, may not be immediately life-threatening or result in death or hospitalization but may jeopardize the patient's health or may require intervention to prevent one of the above outcomes. Examples of such events are intensive treatment in an emergency room or at home for allergic bronchospasm, blood dyscrasias, or convulsions that do not result in hospitalization or development of drug dependency or drug abuse.

9.1.5 Discontinuation Due to an Adverse Event

Study drug may be discontinued due to an AE at the discretion of the subject and/or the Investigator. Subjects in whom study drug is discontinued will undergo the early withdrawal procedures listed in Section 6.1.5.

9.1.6 Unlisted (Unexpected) Adverse Events

An AE is considered unlisted if the nature or severity is not consistent with the applicable product reference safety information (Investigator's Brochure).[8]

9.1.7 Suspected Unexpected Serious Adverse Reactions

Suspected unexpected serious adverse reactions (SUSARs) are SAEs that are unexpected and judged by the Investigator or Sponsor to be related to the study treatment administered.

The Sponsor or designee will report SUSARs and other applicable SAEs to the appropriate regulatory authorities, central ethics committees (ECs)/institutional review boards (IRBs)/ and Investigators as required, according to local law.

9.1.8 Treatment-Emergent Adverse Event

A treatment-emergent adverse event (TEAE) is defined as any event not present prior to the initiation of the treatments or any event already present that worsens in either intensity or frequency following exposure to the treatments.

9.2 Grading of Adverse Event Intensity

Each AE must be graded on a 4-point scale (Grades 1-4) of increasing intensity according to the Division of Microbiology and Infectious Diseases (DMID) Adult Toxicity Table presented in Appendix 14.3.

Criteria in the DMID table are generally grouped by body system, ie, hematology, chemistries, enzymes, urinalysis, cardiovascular, respiratory, GI, neurological, musculoskeletal, skin, and systemic.

For abnormalities not specifically listed in the DMID Table (Appendix 14.3), a guide for estimating severity grade (mild, moderate, severe, life-threatening) is provided:

- Grade 1: Mild transient or mild discomfort (< 48 hours); no medical intervention/therapy required
- Grade 2: Moderate/mild to moderate limitation in activity—some assistance may be needed; no or minimal medical intervention/therapy required
- Grade 3: Severe/marked limitation in activity, some assistance usually required; medical intervention/therapy required, hospitalizations possible
- Grade 4: Life-threatening; Extreme limitation in activity, significant assistance required; significant medical intervention/therapy required, hospitalization or hospice care probable

Note: The semi-colon within the description of the grade indicates ‘and’.

Any clinical event deemed by the clinician to be serious or life-threatening should be considered an SAE.

9.3 Causality Assessment

The following binary choice will be used by the Investigator to describe the causality assessment with the test treatment:

- Reasonable possibility: There is evidence to suggest a causal relationship between the test treatment and the AE (eg, AE is uncommon and known to be strongly associated with drug exposure or is uncommon in the study population, but not commonly associated with drug exposure).
- No reasonable possibility: There is no evidence to suggest a causal relationship between the test treatment and the AE.

9.4 Action Taken Regarding Investigational Product

The action taken toward the study drug must be described as one of the following:

- Permanently discontinued
- Stopped temporarily
- Modified infusion rate
- No action Taken
- Not applicable

9.5 Outcome

The outcome of each AE must be rated as one of the following:

- Recovered/resolved
- Recovering/resolving
- Not recovered/not resolved
- Recovered with sequelae/resolved with sequelae
- Fatal
- Unknown

9.6 Recording Adverse Events

All (S)AEs occurring during the clinical investigation must be documented in the EDC system from the time of dosing with any study treatment through Day 56.

Whenever possible, diagnoses should be given when signs and symptoms are due to a common etiology (eg, skin erythema, induration, and edema should be reported as “cellulitis”). Investigators must record their opinion concerning the relationship of the (S)AE to the study drug in the EDC system. All measures required for (S)AE management must be recorded in the source documents and reported according to the Sponsor’s instructions.

All (S)AEs occurring at any time during the study (including the follow-up period) will be followed by the Investigator until satisfactory resolution (eg, value back to baseline value) or stabilization or until final database lock. If necessary, in order to obtain additional information to ensure safety to the subject, additional blood and urine samples may be taken at the discretion of the Investigator. Certain long-term AEs related to therapy cannot be followed until resolution within the setting of this study. In these cases, follow-up will be the responsibility of the treating physician.

9.7 Reporting Procedures

9.7.1 Reporting Serious Adverse Events

All SAEs, irrespective of the circumstances or suspected cause, must be reported on a Serious Adverse Event Form by the Investigator to the Sponsor or designee within 24 hours of their knowledge of the event, preferably by email.

Contact details for reporting SAEs:

The Serious Adverse Event Form should include a clearly written narrative describing signs, symptoms, and treatment of the event, diagnostic procedures, as well as any relevant laboratory data and any sequelae, in order to allow a complete medical assessment of the case and independent determination of the possible causality.

Follow-up and outcomes should be reported for all subjects that experience an SAE.

It is critical that the information provided on the Serious Adverse Event Form matches the information recorded in the EDC system for the same event. In addition, the same information is to be recorded in the source documents.

Copies of additional reports and documents should be sent when requested and applicable. Follow-up reports relative to the subject's subsequent course must be submitted to the CRO until the event has subsided or, in case of permanent impairment, until the condition stabilizes.

9.7.2 Reporting SAEs to Competent Authorities/Ethics Committees

Visterra or designee is responsible for appropriate reporting of AEs to the regulatory authorities. Visterra or designee will also report to the Investigator all SAEs that are unlisted (unexpected) and associated with the use of the study drug. The Investigator must report these events to the appropriate independent ethics committee/institutional review board (IEC/IRB) that approved the protocol, unless otherwise required and documented by the IEC/IRB.

Visterra or designee will be responsible for unblinding and submitting SUSARs involving VIS410/placebo to the applicable regulatory authorities according to International Conference on Harmonisation (ICH) guidelines. In addition, Visterra or designee will be responsible for the submission of safety letters to the central IEC/IRB and to participating Investigators of all SUSARs involving VIS410 according to applicable regulations. For clinical sites that use a local

IEC/IRB, it is the responsibility of the Investigator to promptly notify the local IEC/IRB of all unexpected serious adverse drug reactions involving risk to human subjects.

After termination of the clinical study (determined as LSLV), any unexpected safety issue that changes the risk-benefit analysis and is likely to have an impact on the subjects who have participated in it, will be reported by Visterra or designee as soon as possible to the competent authority(ies) concerned together with proposed actions.

9.7.3 Reporting a Pregnancy

Subjects should not become pregnant during the study.

Pregnancy is not an AE; however, the Investigator must report any pregnancy which occurs in a female subject or the female partner of a male subject up to 60 days after last dose of VIS410/placebo by emailing the Pregnancy Notification Form to the Sponsor or designee within 24 hours of the study site staff becoming aware of the pregnancy. The Investigator will also follow up with the subject to determine the outcome of the pregnancy and any fetal or neonatal sequelae will be obtained and documented. The Investigator or study site staff must report the outcome of the pregnancy to the Sponsor or designee.

Contact details for reporting Pregnancy:

[redacted]

Congenital abnormalities and birth defects in the offspring of male or female subjects should be reported as an SAE if conception occurred during the study treatment period or in the 60-day period after the last dose of VIS410/placebo.

10.0 STATISTICAL METHODS

Further details of the statistical methodology, in addition to those described below, will be described in the statistical analysis plan (SAP). The SAP will be prepared and finalized before database lock and analysis of the data. Additional efficacy analyses including, but not limited to, subgroup efficacy analysis will be specified in the SAP.

10.1 Determination of Sample Size

The study is exploratory in nature, and is not powered to demonstrate significant differences between treatment groups in primary or secondary outcome measures. The protocol intent is to collect sufficient information to identify the most appropriate candidate endpoints for subsequent Phase 3 study evaluation from among the primary and secondary endpoints described below. Statistical significance testing will therefore be used to assess the relative strength of evidence of the primary and secondary endpoints, to provide reasonable assurance that the endpoints chosen for a confirmatory Phase 3 trial will elucidate treatment differences between VIS410 plus oseltamivir versus oseltamivir alone.

10.2 Randomization and Blinding

An IWRS will be used to allocate the randomized treatments to subjects with stratification by presence or absence of positive pressure ventilation (PPV) at baseline. Pharmacists must obtain the status of the subject relative to PPV at baseline via query, and this data must be input into the IWRS prior to randomization. The randomized treatment assignment will be transferred electronically for integration with the clinical study data at the appropriate time.

PPV includes any respiratory assistance using a mechanical ventilation device and can be either invasive or noninvasive ventilation. Invasive PPV includes intubation with endotracheal tube with mechanical ventilation (most common) or tracheostomy with mechanical ventilation. Noninvasive PPV includes use of facemask, nasal plugs, or nasal mask with mechanical ventilation. The devices are named according to the type of mechanical ventilation given continuous positive airway pressure (CPAP) or bi-level positive airway pressure (BiPAP).

Randomization will be used to avoid bias in the assignment of subjects to treatment, to increase the likelihood that known and unknown subject attributes (eg, demographic and baseline characteristics) are evenly balanced across treatment arms.

The maintenance of the study blind is critical for an unbiased assessment of the safety and efficacy of the study drug. All study staff that evaluate subjects and render decisions regarding subject care will remain blinded to the study treatment each subject receives.

The designated study site pharmacist or designee will remain unblinded. Also, unblinded clinical research associates will handle study drug accountability.

The study will be conducted in a double-blind manner. The CRO, site study personnel, and Visterra personnel will not be aware of which treatment (VIS410 or placebo) the subjects have been given. Subjects will not be aware of which treatment they have been administered.

Subjects will be randomized 1:1:1 to receive VIS410 2000 mg, VIS410 4000 mg, or placebo over a 2-hour infusion with pretreatment regimen of PO or IV diphenhydramine 50 mg plus either ibuprofen 400 mg PO or acetylsalicylic acid 320-325 mg PO approximately 60 minutes before IV infusion of VIS410/placebo.

Allocation of each subject to a given treatment sequence will be described in a randomization schedule prepared by Visterra or designee. The randomization will be balanced using randomly permuted blocks across the treatment groups. Based on this randomization code, the site's unblinded pharmacist or trained designee will prepare and dispense VIS410/placebo.

The randomization schedule will not be available to the subjects, Investigators, blinded monitors, or employees of the clinical center involved in the management of the study before unblinding of the data, unless in case of emergency.

The Sponsor's clinical team will also be blinded during the study, as they will not have direct access to the randomization schedule. The CRO personnel performing data management and statistical activities will receive a copy of the randomization schedule during database lock. Other team members will not have access to any data that could lead to unblinding.

Unblinding of the individual subject's treatment (via IWRS) by the Investigator should be limited to medical emergencies or urgent clinical situations in which knowledge of the subject's study treatment is necessary for clinical management. In such cases, the Investigator is encouraged to contact the Medical Monitor to discuss and agree to the need for unblinding to occur. In situations in which the Investigator has tried, but is unable to reach the Medical Monitor, they should use their best judgment, based on the nature and urgency of the clinical situation, and may proceed with unblinding without having successfully reached and discussed the situation with the Medical Monitor.

Once a subject's treatment assignment has been unblinded, the Medical Monitor should be notified within 24 hours of unblinding of the treatment, without revealing the study treatment. Information relating to unblinding (eg, reason and date) shall be clearly recorded in the subject's study file, as part of relevant standard operating procedures. In addition, the Investigator should consider whether the clinical event prompting unblinding should be considered an SAE, according to the regulatory definitions or criteria for SAEs, and if so, submit an SAE report to Visterra or designee. If the code is broken by the Investigator or by someone of his/her staff, the subject must be withdrawn from the study and must be followed as appropriate.

Visterra or designee will also unblind any SAE reports that are serious, unexpected, and considered to be related to investigational product, in accordance with safety reporting guidance and regulations. If the code is broken by the Sponsor for safety reporting purposes, the subject

may remain in the study. To maintain study blinding, the infusion bag will be covered by an opaque sleeve in the pharmacy.

10.3 Analysis Populations

The following analysis populations will be defined for the study:

- **Intent-to-treat (ITT) population** – All subjects randomized to treatment. Subjects in the ITT population will be analyzed based on the treatment to which they were randomized, irrespective of what they actually received.
- **Safety population** – All ITT subjects who received IV VIS410/placebo. Subjects in the safety population will be analyzed based on the actual treatment they received, irrespective of the treatment to which they were randomized.
- **Modified intent-to-treat (MITT)** – All subjects of the safety population that have an assessment of O_2 support after randomization and are confirmed influenza A positive. Subjects in the MITT population will be analyzed based on the treatment to which they were randomized, irrespective of what they actually received. This population was selected for the analysis of the primary endpoint in order to maintain the benefits of randomization and avoid the bias associated with the non-random loss of the participants.
- **Per protocol (PP) population** – All MITT subjects who adhere to relevant study procedures and have an outcome assessment. Further specific details defining the analyses to be performed on this population will be described in the SAP.
- **PK population (PK)** – All subjects in the safety population that have at least one result that can be used in the PK summaries.

10.4 Initial Characteristics of the Subject Sample

Summary statistics will be provided per treatment group for demographic (eg, age, height, weight, body mass index [BMI], race, gender) and other initial subject characteristics (eg, medical history, concomitant diseases) will be provided per treatment group and for the total group. The ITT population will be used for the summaries. Enrollment, protocol deviations, and discontinuations from the study will be summarized by treatment group. Protocol deviations are defined as any notable variation from the protocol, including enrollment of a subject who did not meet all inclusion and exclusion criteria, and failure to perform the assessments and procedures within the required time frame.

Summary statistics will be provided to show subject exposure to pretreatment medications, subject exposure to VIS410/placebo/oseltamivir, and inadvertent administration of VIS410/placebo to which subjects were not randomized by treatment group.

Prior and concomitant medications will be coded using the WHO Drug Dictionary.

10.5 Handling of Missing Data

Procedures for handling missing data will be defined in the Statistical Analysis Plan, and may include last observation carried forward or an interpolation method.

10.6 Efficacy Analyses

Efficacy analyses will be performed using the MITT population. Efficacy analysis will also be performed in the PP population to demonstrate consistency with the primary analysis population.

10.6.1 Day 7 Ordinal Scale Status

The ordinal scale outcomes will be measured daily from Day 1 (baseline) through Day 14 using the seven-level hierarchical scale with the classifications presented in Table 8. The clinical outcomes therein are ordered from the worst clinical outcome to the best clinical outcome in descending order. For each day, subjects will be classified by the worst clinical outcome for which they qualify.

The number and percentage of subjects in each treatment group with each classification will be summarized for each day from Day 1 through Day 14, inclusive.

Table 8. Hierarchical Seven-Level Ordinal Scale for Clinical Outcomes

Clinical Parameter
Death
ICU stay with mechanical ventilation
ICU stay without mechanical ventilation
Non-ICU hospitalization with supplemental oxygen
Non-ICU hospitalization without supplemental oxygen
Discharge with partial resumption of normal activities
Discharge with full resumption of normal activities

Note: Clinical outcomes listed from worst clinical outcome to best clinical outcome in descending order.

10.6.2 Time to Cessation of O_2 Support

Time to cessation of O_2 support resulting in a stable SpO_2will be analyzed using a Cox model. Time from onset of symptoms to VIS410 treatment and the number of doses of oseltamivir (Tamiflu) prior to VIS410 treatment for the current case of influenza will be included as a covariate in the analysis. A *P*-value (using the Wald statistic) for each VIS410 dose vs placebo will be presented.

Secondarily, subgroup analyses will be performed to describe the time to cessation of O_2 for subgroups, such as use of positive pressure ventilation, use of endotracheal intubation, time from onset of symptom to VIS410 therapy, number of oseltamivir doses prior to VIS410, elderly, and

underlying lung disease. Clinical and virologic endpoints will also be evaluated by influenza A subtypes.

10.6.3 Time to Clinical Response

Time to clinical response is defined as resolution of 4 of 5 vital signs that will be determined upon physical examination. Clinical response is defined as:

- Afebrile with core temperature ≤ 37.8°C, without use of antipyretics (oral ≤ 37.2°C)
- Respiratory rate ≤ 24 beats per minute
- Oxygen saturation ≥ 95% on room air without support or a return to pre-infection status, if pre-infection status was < 95%
- Pulse rate ≤ 100/min
- SBP ≥ 90 mm/Hg, without vasopressor use

The probability of time to clinical response (defined as resolution of vital signs) will be calculated via Kaplan-Meier. A significance test (using the log-rank test) for each VIS410 dose vs placebo will be presented. The number and percentage of subjects in each treatment group with clinical response will be summarized. Results will be tabulated and presented graphically as well.

10.6.4 Time to Cessation of Ventilator Support

The probability of time to cessation of ventilator support will be calculated via Kaplan-Meier. A P-value (using the log-rank test) for each VIS410 dose vs placebo will be presented. Results will be tabulated and presented graphically as well.

10.6.5 Healthcare Resource Utilization

Descriptive statistics will be used to compare the total number of days in the hospital and/or ICU from admission to discharge, number of subjects requiring ICU admission post-randomization, overall number of days in the ICU, number of hours on ventilation, rehospitalization due to influenza A relapse/reinfection, the total number of days of oseltamivir therapy, and the total number of days to resumption of usual activities by treatment group.

Time (number of days) to resumption of usual activities will be determined from the VAS (scale ranged from 0 to 10, where 0 indicates subject is unable to perform any of his/her usual activities prior to influenza onset, and 10 indicates subject is able to fully perform all usual activities).

This evaluation will be performed using the MITT population.

10.6.6 Signs and Symptoms of Influenza

Descriptive statistics will be used to compare the duration of symptoms of influenza-like illness in the subset of subjects able to complete the FluPRO Questionnaire at baseline and post-dose by

treatment group. The number and percentage of subjects who were not able to complete the assessment at each visit will be summarized.

Frequency tabulation of the occurrence and severity of each subject-reported symptom of influenza-like illness (via FluPRO Questionnaire) will be summarized by assessment time point and by treatment group. Time to resolution of symptoms will be evaluated by Kaplan Meier analysis.

Analyses of the signs and symptoms of influenza will be conducted only on the subset of the MITT population who had baseline FluPRO Questionnaire assessments. Additional populations may be analyzed as described in the SAP.

10.7 Safety Analyses

Safety analyses will be performed on the safety population.

10.7.1 Adverse Events

The original terms in the electronic data capture (EDC) system used by Investigators to identify AEs other than symptoms of influenza A will be fully described and coded according to the Medical Dictionary for Regulatory Activities (MedDRA). The reported AEs will be allocated to phases based on their start date. All AEs will be listed. All AEs with onset during the treatment phase (ie, treatment-emergent AEs [TEAEs]) will be summarized.

AEs will be summarized overall and by treatment group and by MedDRA body organ system and preferred term, severity, relatedness, and seriousness.

The difference in proportions of subjects with AEs, TEAEs, hypersensitivity reaction, anaphylactic reaction, AESIs, and SAEs between treatment groups will be calculated.

Special attention will be paid to those subjects who died, discontinued due to an AE, or experienced a severe or serious AE. Summaries, listings, and narratives will be provided, as appropriate.

10.7.2 Local Injection Site Tolerability

Injection site tolerability is defined as AEs demonstrating local injection site irritation or tissue damage. Injection site tolerability will be reported by variable, treatment group, and time point.

10.7.3 Complications of Influenza

Complications of influenza are defined as pneumonia, myocarditis, worsening of chronic bronchitis, sinusitis, otitis or other chronic pulmonary diseases, encephalopathy, and death. Complications of influenza and use of antibiotic therapy for bacterial pneumonia will be reported by variable, treatment group, and time point. Diagnosis of bacterial pneumonia will be based on local clinical practice, usually an assessment of clinical features and any radiographic evidence

of bacterial pneumonia (eg, infiltrate) and/or microbiologic evidence of bacterial pneumonia. All-cause and attributable mortality will be evaluated on Days 14 and 28.

10.7.4 Clinical Laboratory Tests

Actual values and changes from baseline of each continuous biochemistry, hematology, and urinalysis test will be evaluated by means of descriptive statistics by assessment time point and by treatment group. For categorical urinalysis tests, frequency tables of actual values will be provided by assessment time point and by treatment group.

Relative changes in clinical laboratory test values compared to values at baseline will be evaluated according to the DMID table (see Appendix 14.3) or in accordance with the normal ranges of the clinical laboratory (below, within, or above normal range) for parameters for which no toxicity grades are defined. A toxicity grade shift from baseline table of the abnormalities will be provided by assessment time point and by treatment group.

A listing of subjects with any clinical laboratory test result outside the reference ranges will be provided.

10.7.5 Vital Signs

Actual values and changes from baseline of heart rate, respiratory rate, temperature, SBP, and DBP measurements will be evaluated by means of descriptive statistics by assessment time point and by treatment group.

A shift from baseline table of vital sign abnormalities will be provided by assessment time point and by treatment group.

10.7.6 Electrocardiography

The ECG variables that will be analyzed are heart rate, PR interval, QRS interval, and QT interval. Values for QT corrected for heart rate (QTc) will be derived. QTcF will be the primary correction parameter.[13]

Actual values and changes from baseline of ECG variables will be evaluated by means of descriptive statistics by assessment time point and by treatment group.

A shift from baseline table of ECG abnormalities will be provided by assessment time point and by treatment group. For absolute QTcF interval prolongation (> 450, > 480, > 500 ms) and changes from baseline (increase > 30 and > 60 ms), a frequency table by assessment time point and by treatment group will be provided.

10.8 Pharmacokinetic/Pharmacodynamic Analyses

10.8.1 Serum Pharmacokinetics

Serum concentrations will be listed by subject for VIS410 and summary statistics by group will be presented, including means, geometric means, standard deviations, coefficient of variation (CV), medians, and ranges, as appropriate. Summary graphs, including mean concentration-time profiles by group, will also be presented. The serum concentration data will be analyzed by population PK methods using nonlinear mixed effects modeling as implemented in NONMEM or equivalent software. Population PK analysis will be performed to describe the time course of serum concentrations of VIS410. The influence of covariates on PK parameters will be investigated, if necessary and appropriate. If necessary, the data may be pooled with data from previous studies. Additional analyses and summaries may be generated as appropriate.

Results of population PK or PK/PD analyses may be reported outside the clinical study report.

10.8.2 Pharmacokinetics of Nasopharyngeal Secretions and Tracheal Aspirate

Nasopharyngeal swabs will be obtained from both nostrils (1 swab per nostril). The first 50 randomized subjects will have nasopharyngeal swabs collected up to Day 56 (predose, end of infusion, Days 3, 5, 7, 14, 28 and 56); while in the remaining subjects, nasopharyngeal swabs will be obtained up to Day 14 only. If the subject remains in the hospital on Day 10, then additional nasopharyngeal swabs will be obtained on Day 10. The VIS410 concentrations in the nasopharyngeal secretions and tracheal aspirate will be listed by subject, and summary statistics by group will be reported as described for the serum concentrations. The computed PK parameters will be listed by subject for VIS410. Summary statistics and PK parameters will be presented, including means, geometric means, standard deviations, CV, medians, and ranges, as appropriate. Summary graphs, including mean concentration-time profiles by group, will also be presented. The nasopharyngeal concentration data may also be analyzed by population PK methods using nonlinear mixed effects modeling as implemented in NONMEM or equivalent software. If necessary, the data may be pooled with data from previous studies. Additional analyses and summaries may be generated as appropriate.

Results of population PK or PK/PD analyses may be reported outside the clinical study report.

10.8.3 Exploratory Pharmacokinetic/Pharmacodynamic Analyses

Various techniques will be used to explore exposure-response relationships and to compare the strength of the relationship between each independent variable (eg, AUC, C_{max}, concentration at specific time point) and the dependent variables (eg, viral AUC, peak viral load, time to cessation of viral shedding, clinical symptoms, and additional endpoints). These techniques may include graphical and statistical methods, including the creation of boxplots, spaghetti plots, histograms, and a variety of linear, nonlinear, or logistic regression techniques and time-to-event methods. If appropriate, continuous independent variables will be evaluated as such, and as categorical variables (grouping subjects into exposure categories).

Results of the PK/PD analyses may be reported outside the clinical study report.

10.9 Viral Load

Student t-test or Mann-Whitney U test will be used to assess the difference between treatment groups in AUC based on qRT-PCR and $TCID_{50}$ from nasopharyngeal swabs and tracheal aspirate, when appropriate (ie, intubated patients). Descriptive statistics will be used for viral load data (AUC, time to resolution of viral load, duration of viral shedding, and peak viral load based on qRT-PCR and $TCID_{50}$) from nasopharyngeal swabs as well as tracheal aspirate by treatment group and VIS410 overall vs placebo. Tables and graphs will be generated as appropriate for the MITT and PP populations by dose group. Additional exploratory statistical analyses may be conducted as appropriate. Exploratory dose and exposure response will be evaluated using various statistical and graphical approaches as appropriate.

10.10 Immunological Analyses

Immunological assessments will be summarized for the MITT population by parameter, treatment group, and time point using descriptive statistics:

- Anti-influenza A antibodies by HAI in serum
- ADA titers

10.11 Viral Resistance

Viral sensitivity to VIS410 and oseltamivir will be assessed during the study.

11.0 STUDY TERMINATION AND COMPLETION

11.1 Study Completion

A subject will be considered to have completed the study if he or she has completed the last follow-up visit (Day 56 ± 7 days).

11.2 Study Drug Discontinuation

Subjects may be discontinued from the study drug administration in the event of:

- A severe AE or SAE
- A positive pregnancy test of the subject, or if the subject/partner is non-compliant with the contraception requirements (see Section 6.2)
- Development of a medical condition that requires concomitant treatment with a prohibited therapy (see Section 8.2)

Such subjects should be encouraged to continue study participation by undergoing all safety evaluations until the scheduled time of study completion, as appropriate.

11.3 Subject Withdrawal from Study

Subjects have the right to withdraw from the study at any time for any reason, including personal reasons. A subject can withdraw without giving a reason. The Investigator should however try to find out why a subject has withdrawn from the study and document the reason for withdrawal.

Subjects must be withdrawn from the study in the event of:

- Withdrawal of informed consent
- Failure of the subject to comply with the protocol requirements or to cooperate with the Investigator
- For safety reasons, it being in the best interest of the subject that he/she be withdrawn, in the Investigator's opinion

In the event that a subject is withdrawn from the study, the study monitor and Sponsor should be informed. In case of withdrawal due to an SAE (for details on AE reporting see Section 9.7), the Sponsor should be notified within 24 hours. In case of withdrawal for other reasons, the Sponsor should be notified within 2 days of the event.

If there is a medical reason for withdrawal, the subject will remain under the supervision of the Investigator until satisfactory health has returned.

Subjects who are withdrawn from the study prior to completion of the scheduled study procedures for any reason (AE, withdrawal of consent, etc.) should be invited to complete the assessments as much as possible. As long as the subject consents, all relevant assessments of the

day on which the subject withdrew from the study should be completed, at least those related to safety. In case of an AE, the appropriate follow-up will be performed.

VIS410/placebo assigned to a withdrawn subject must not be assigned to another subject. Subjects withdrawn from the study will not be replaced.

11.4 Stopping Rules or Discontinuation Criteria

The study will be overseen by a DSMB (see Section 4.3). Based upon any safety assessments and after mutual agreement with the Investigator (or designee) and the Sponsor, the study may be temporarily or permanently halted. Study enrollment and dosing will continue while the DSMB evaluates data.

Dosing will temporarily pause while the DSMB meets if:

- A total of 4 treatment-related *serious* adverse events (SAEs) occurred, or
- A total of 4 *severe* GI TEAEs *requiring intervention* occurred. Intervention is defined as requiring IV fluid and medication to decrease the frequency of diarrhea (ie, loperamide).

11.5 Protocol Compliance

In accordance with ICH E6 (R1) Guideline for GCP, the Investigator should not implement any deviation from or changes of the protocol without agreement by the Sponsor (or designee) and documented approval from the IEC/IRBs of a protocol amendment, except where necessary to eliminate an immediate hazard(s) to study subjects, or when the change(s) involves only logistical or administrative aspects of the study (eg, change in monitor[s], change of telephone number[s]).

The protocol must be read thoroughly, and the instructions must be followed. However, exceptions will be made in emergency situations when the protection, safety, and well-being of the subject requires immediate intervention based on the judgment of the Investigator or a responsible, appropriately trained, and credentialed professional(s) designated by the Investigator as a sub-Investigator.

In the event of a significant protocol deviation due to an emergency, accident, or error, the Investigator (or designee) must contact the Medical Monitor at the earliest possible time. This allows for an early joint decision to be made as to whether or not the subject should continue in the study. The Investigator, the Sponsor (or designee), and the Medical Monitor will document this decision.

All protocol deviations will be documented in a database.

12.0 ETHICAL AND ADMINISTRATIVE CONSIDERATIONS

12.1 Ethical Conduct of the Study

12.1.1 Study-Specific Design Considerations

Potential subjects will be fully informed of the risks and requirements of the study, and during the study, subjects will be provided with any new information that may affect their decision to continue participation. Subjects will be informed that their consent to participate in the study is voluntary and that they may withdraw at any time without the need to provide a reason and without penalty or loss of benefits to which they would otherwise be entitled.

Only subjects who are fully able to understand the risks, benefits, and potential AEs of the study, and provide their consent voluntarily will be enrolled.

12.1.2 Subject Informed Consent

Each subject or a legally acceptable representative must give written consent according to local requirements after the nature of the study has been fully explained. The consent form must be signed before performance of any study-related activity. The consent form that is used must be approved by both the Sponsor and by the reviewing IEC/IRB. The informed consent should be in accordance with principles that originated in the Declaration of Helsinki, current ICH and GCP guidelines, applicable regulatory requirements, and Sponsor policy.

Before enrolling potential subjects in the study, the Investigator or an authorized member of the investigational staff must explain to the subject the aims, methods, reasonably anticipated benefits, and potential hazards of the study, and any discomfort that participation in the study may entail. Subjects will be told that the Investigator will maintain a subject identification register for the purposes of long-term follow up if needed and that their records may be accessed by health authorities and authorized Sponsor staff without violating the confidentiality of the subject, to the extent permitted by the applicable law(s) or regulations. By signing the Informed Consent Form (ICF) the subject is authorizing such access and agrees to allow his/her study physician to recontact the subject for the purpose of obtaining consent for additional safety evaluations, if needed.

The language used in the oral and written information about the study, including the ICF, should be nontechnical and practical and should be understandable to the subject or the subject’s legal representative. The subject will be given sufficient time to read the ICF and the opportunity to ask questions. After this explanation and before entry into the study, consent should be appropriately recorded by means of the subject's personally dated signature. After having obtained the consent, a copy of the ICF must be given to the subject.

If the subject is unable to read or write, an impartial witness should be present for the entire informed consent process (which includes reading and explaining all written information) and

should personally date and sign the ICF after the oral consent of the subject is obtained, if permitted by local law.

12.1.3 Independent Ethics Committee or Institutional Review Board

An independent ethics committee (IEC) or institutional review board (IRB) should safeguard the rights, safety, and well-being of all study subjects. Special attention should be paid to studies that may include vulnerable subjects.

Before the start of the study, the Investigator (or Sponsor where required) will provide the IEC/IRB with current and complete copies of the following documents:

- Final protocol and, if applicable, amendments
- Sponsor-approved ICF (and any updates) and any other written materials to be provided to the subjects
- Sponsor-approved subject recruiting materials
- Investigator's Brochure (or equivalent information) and addenda
- Available safety information
- Information on compensation for study-related injuries or payment to subjects for participation in the study, if applicable
- Investigator's current curriculum vitae or other documentation evidencing qualifications (unless not required, as documented by IEC/IRB)
- Information regarding funding, name of the Sponsor, institutional affiliations, other potential conflicts of interest, and incentives for subjects
- Any other documents that the IEC/IRB may require to fulfill its obligation
- This study will be undertaken only after the IEC/IRB has given full written approval of the final protocol and amendments (if any), the ICF(s) and updates (if any), applicable recruiting materials, and any other written information to be provided to the subjects, and the Sponsor has received a copy of this approval. This approval letter must be dated and must clearly identify the IEC/IRB and the documents being approved.
- During the study, the Investigator (or Sponsor where required) will send the following documents and updates to the IEC/IRB for their review and approval, where appropriate:
- Protocol amendments
- Revision(s) to ICF and any other written materials to be provided to subjects
- If applicable, new or revised subject recruiting materials approved by the Sponsor
- Revisions to compensation for study-related injuries or payment to subjects for participation in the study, if applicable
- Investigator's Brochure addenda or new edition(s)
- Summaries of the status of the study at intervals stipulated in guidelines of the IEC/IRB (at least annually)
- Reports of AEs that are serious, unlisted, and associated with the investigational drug

- New information that may adversely affect the safety of the subjects or the conduct of the study
- Deviations from or changes to the protocol to eliminate immediate hazards to the subjects
- Report of deaths of subjects under the Investigator's care
- Notification if a new Investigator is responsible for the study at the site
- Development Safety Update Report, Short Term Study-Specific Safety Summary and Line Listings, where applicable
- Any other requirements of the IEC/IRB

For all protocol amendments (excluding the ones that are purely administrative, with no consequences for subjects, data, or study conduct), the amendment and applicable ICF revisions must be submitted promptly to the IEC/IRB for review and approval before implementation of the change(s), except when necessary to eliminate an immediate hazard to the study subjects. If a deviation from or a change to the protocol was implemented to eliminate an immediate hazard to study subjects, then the implemented deviation or change, the reasons for it, and, if appropriate, the protocol amendment should be submitted to the IEC/IRB as soon as possible.

The Investigator (or Sponsor where required) will notify the IEC/IRB about the study completion after the LSLV.

12.1.4 Protection of Subject Data

The collection and processing of personal data from subjects enrolled in this study will be limited to those data that are necessary to investigate the safety, quality, and utility of the investigational study drug(s) used in this study.

These data must be collected and processed with adequate precautions to ensure confidentiality and compliance with applicable data privacy protection laws and regulations. Appropriate technical and organizational measures to protect the personal data against unauthorized disclosures or access, accidental or unlawful destruction, or accidental loss or alteration must be put in place. Sponsor personnel whose responsibilities require access to personal data agree to keep the identity of study subjects confidential.

The informed consent obtained from the subject includes explicit consent for the processing of personal data and for the Investigator to allow direct access to his or her original medical records for study-related monitoring, audit, IEC/IRB review, and regulatory inspection. This consent also addresses the transfer of the data to other entities and to other countries.

12.2 Investigator Responsibilities

The Investigator(s) should be qualified by education, training, and experience to assume responsibility for the proper conduct of the study, should meet all the qualifications specified by the applicable regulatory requirement(s), and should provide evidence of such qualifications

through up-to-date curriculum vitae and/or other relevant documentation requested by the Sponsor, the IEC/IRB, and/or the regulatory authority(ies).

The Investigator is responsible for ensuring that the clinical study is performed in accordance with the protocol, current ICH guidelines on GCP, and applicable regulatory and country-specific requirements.

Good Clinical Practice is an international ethical and scientific quality standard for designing, conducting, recording, and reporting studies that involve the participation of human subjects. Compliance with this standard provides public assurance that the rights, safety, and well-being of study subjects are protected, consistent with the principles that originated in the Declaration of Helsinki, and that the clinical study data are credible.

12.3 Administration

12.3.1 Protocol Amendments

Neither the Investigator nor the Sponsor will modify this protocol without a formal amendment. All protocol amendments must be issued by the Sponsor, and signed and dated by the Investigator. Protocol amendments must not be implemented without prior IEC/IRB approval, or when the relevant competent authority has raised any grounds for non-acceptance, except when necessary to eliminate immediate hazards to the subjects, in which case an amendment must be promptly submitted to the IEC/IRB and relevant competent authority. Documentation of amendment approval by the Investigator and IEC/IRB must be provided to the Sponsor or its designee. When the change(s) involve(s) only logistic or administrative aspects of the study, the IRB (and IEC where required) only needs to be notified.

12.3.2 Subject Identification, Enrollment, and Screening

The Investigator agrees to complete a subject identification and enrollment record to permit easy identification of each subject during and after the study. This document will be reviewed by the Sponsor site contact for completeness.

The subject identification and enrollment record will be treated as confidential and will be filed by the Investigator in the study file. To ensure subject confidentiality, no copy will be made. All reports and communications relating to the study will identify subjects by initials and/or assigned number only.

The Investigator must also maintain a subject-screening record, which reports all subjects who were seen to determine eligibility for inclusion in the study.

12.3.3 Source Documentation

The EDC system is an electronic data capturing and information management system that will also serve as the data management system for this study. The system combines all aspects of

source data capturing with process control and clinical study management. All clinical and laboratory data, except those which are paper based, will be collected in the EDC system. The responsible study monitor will check data at the monitoring visits to the clinical study site. The Investigator will ensure that the data collected are accurate, complete, and legible. Data will be monitored within the EDC system by the study monitor who has only reading rights. Any changes required following monitoring will be made by site personnel or the Investigator and will be documented with a full audit trail within the EDC system.

At a minimum, source documentation must be available for the following: subject identification, eligibility, and study identification; date of informed consent; dates of visits; results of safety and efficacy parameters as required by the protocol; record of all AEs; follow up of AEs; concomitant medication; drug receipt/dispensing/return records; study drug administration information; laboratory printouts (if not available digitally); date of study completion; and reason for early discontinuation of study drug or withdrawal from the study, if applicable.

It is recommended that the author of an entry in the (e-)source documents be identifiable.

Source data may be directly captured from devices, transferred from third parties (eg, laboratory data), or entered manually into the EDC system in use at the clinical center. In such case, the majority of the source data will only be available electronically. The remainder of the data, captured initially on paper, may be entered retrospectively into the EDC system.

Following the ICH/GCP guidelines, direct access to (e-)source documentation (medical records) must be allowed.

12.3.4 Case Report Form Completion

All source data, except those that are paper based, will be collected directly into the EDC system. Paper-based sources will be manually transcribed in the EDC system. All data captured in the EDC system will be transferred to the clinical database electronically.

12.4 Monitoring and Quality Assurance

The monitoring of the study will be conducted under the responsibility of the Sponsor by the CRO.

The monitor will perform on-site or remote monitoring visits as frequently as necessary. The monitor will record dates of the on-site visits in a study center visit log that will be kept at the site. At these visits, the monitor will compare the data captured in the EDC system for completeness and accuracy and perform source data verification of any data that have been captured as paper-based sources or entered in the system later on. The nature and location of all source documents will be identified to ensure that all sources of original data required to complete the EDC system are known to the Sponsor and investigational staff and are accessible for verification by the Sponsor site contact(s). If electronic records are maintained at the investigational site, the method of verification must be discussed with the investigational staff.

Direct access to (e-)source documentation (medical records) must be allowed at all times for the purpose of verifying that the data recorded in the EDC system are consistent with the original (e-)source data. Findings from this review of captured data will be discussed with the investigational staff. During on-site monitoring visits (notified and agreed upfront with the investigational staff), the relevant investigational staff will be available, the (e-)source documentation will be accessible, and a suitable environment will be provided for review of study-related documents. The monitor will meet with the Investigator on a regular basis during the study to provide feedback on the study conduct.

12.4.1 Data Management

Data management of the study will be the responsibility of the Sponsor and will be conducted by the CRO.

After the data are released by the Investigator and the monitor has reviewed the data for completeness and accuracy, the data will be uploaded into the clinical database to perform cleaning activities. Only the data of randomized subjects will be captured in the clinical database.

Computerized data cleaning checks will be used in addition to manual review, including listings review, to check for discrepancies and to ensure the consistency and completeness of the data. Queries emerging during data cleaning will be generated by the clinical data manager in the EDC system. The Investigator or his designee will answer the queries and update the source data, if needed. Any changes required are to be documented with a full audit trail within the EDC system.

An interim lock of the database will occur at the time of an interim analysis.

The final clinical database will be locked as soon as it is considered clean. Only authorized and well-documented updates to the study data are possible after final database lock. The locked final database is used in the final statistical analysis for study reporting. Measures will be undertaken to protect subject data handed over by the Investigator to the Data Management Department and during inspections against disclosure to unauthorized third parties. Subject confidentiality will be maintained at all times.

12.4.2 Data Quality Assurance

The accuracy and reliability of the study data will be assured by the selection of qualified Investigators and appropriate study centers, review of protocol procedures with the Investigator and associated personnel prior to the study, and by periodic monitoring visits by the Sponsor or designee.

Written instructions will be provided for the collection, preparation, and shipment of samples.

The Sponsor or its designee will review the EDC system for accuracy and completeness during (on-site) monitoring visits and after transmission to the Sponsor; any discrepancies will be

resolved with the Investigator or designee, as appropriate. After upload of the data into the clinical study database, they will be verified for accuracy.

In accordance with Good Clinical Research Practice Guidelines and Recommendations, the Sponsor will be entitled to audit the facilities used in the clinical and laboratory parts of the study, as well as to access all the data files pertaining to the study. Similar procedures may also be conducted by agents of any regulatory body, either as part of a national GCP compliance program or to review the results of this study in support of a regulatory submission. The Investigator should immediately notify the Sponsor if they have been contacted by a regulatory agency concerning an upcoming inspection.

12.4.3 On-Site Audits

Investigators and institutions involved in the study will permit study-related monitoring, audits, IRB review, and regulatory inspections by providing direct access to all study records. In the event of an audit, the Investigator agrees to allow the Sponsor, representatives of the Sponsor, or a government or regulatory agency access to all study records.

The Investigator should promptly notify the Sponsor of any audits scheduled by any regulatory authorities and promptly forward copies of any audit reports received to the Sponsor.

Similar procedures may also be conducted by agents of any regulatory body, either as part of a national GCP compliance program or to review the results of this study in support of a regulatory submission. The Investigator should immediately notify the Sponsor if they have been contacted by a regulatory agency concerning an upcoming inspection.

12.5 Study Termination

The Sponsor reserves the right to terminate the study at any time. In case of an early termination of the study for safety reasons, or temporary halt by the Sponsor, the IEC/IRB should be notified within 15 calendar days, including a detailed written explanation of the reasons for the termination/halt.

The end-of-study declaration will be submitted to the regulatory authorities and IEC after the complete study has ended. This notification will also be submitted within 90 days of the end of the study.

12.5.1 Record Retention

In compliance with the ICH/GCP guidelines, the Investigator/institution will maintain an archived copy of the EDC data and all paper source documents that support the data collected from each subject, as well as all study documents as specified in ICH/GCP Section 8, Essential Documents for the Conduct of a Clinical Trial, and all study documents as specified by the applicable regulatory requirement(s). The Investigator/institution will take measures to prevent accidental or premature destruction of these documents.

Essential documents must be retained until at least 2 years after the last approval of a marketing application in an ICH region and until there are no pending or contemplated marketing applications in an ICH region or until at least 2 years have elapsed since the formal discontinuation of clinical development of the investigational product. These documents will be retained for a longer period if required by the applicable regulatory requirements or by an agreement with the Sponsor. It is the responsibility of the Sponsor to inform the Investigator/institution as to when these documents no longer need to be retained.

If the responsible Investigator retires, relocates, or for other reasons withdraws from the responsibility of keeping the study records, custody must be transferred to a person who will accept the responsibility. The Sponsor must be notified in writing of the name and address of the new custodian. Under no circumstance shall the Investigator relocate or dispose of any study documents before having obtained written approval from the Sponsor.

If it becomes necessary for the Sponsor or the appropriate regulatory authority to review any documentation relating to this study, the Investigator must permit access to such reports.

12.5.2 Use of Information and Publication

All information, including but not limited to information regarding VIS410 or the Sponsor's operations (eg, patent application, formulas, manufacturing processes, basic scientific data, prior clinical data, formulation information) supplied by the Sponsor to the Investigator and not previously published, and any data generated as a result of this study are considered confidential and remain the sole property of the Sponsor. The Investigator agrees to maintain this information in confidence, to use this information only to accomplish this study, and not to use it for other purposes without the Sponsor's prior written consent.

The Investigator(s) must not submit any part of the data from this protocol for publication without the prior consent of Visterra, Inc.

The Investigator understands that the information developed in this clinical study will be used by the Sponsor in connection with the continued development of VIS410, and thus may be disclosed as required to other clinical Investigators or regulatory agencies. To permit the information derived from the clinical studies to be used, the Investigator is obligated to provide the Sponsor with all data obtained in the study.

The results of the study will be reported in a clinical study report written by the CRO under responsibility of the Sponsor and will contain EDC system data from all investigational sites that participated in the study. Recruitment performance or specific expertise related to the nature and the key assessment parameters of the study will be used to determine a coordinating Investigator.

Clinical narratives will be written for the following events:

- All deaths (irrespective of drug relationship)

- All other SAEs during treatment with the study drug
- All discontinuations of the study due to AEs related to the study drug
- At the discretion of the team and after statistical analysis of the data, certain discontinuations not related to AEs or treatment failure, ie, related to lost to follow-up or withdrawal of consent (irrespective of treatment group)
- Any events of special interest explicitly requested by the regulatory agencies

A summary of this final report will be provided to the Investigators, to the applicable regulatory authorities, and IECs/IRBs, if required by the applicable regulatory requirements, within 1 year of the end of the study (LSLV).

The Sponsor shall have the right to publish such data and information without approval from the Investigator.

Individual site publications are not expected, as individual sites may not recruit enough subjects to enable detailed publications; therefore, the results of this study will be reported in total.

If an Investigator wishes to publish information from the study, a copy of the manuscript must be provided to the Sponsor for review at least 60 days before submission for publication or presentation. Expedited reviews will be arranged for abstracts, poster presentations, or other materials. If requested by the Sponsor in writing, the Investigator will withhold such publication for up to an additional 60 days to allow for filing of a patent application. In the event that issues arise regarding scientific integrity or regulatory compliance, the Sponsor will review these issues with the Investigator. The Sponsor will not mandate modifications to scientific content and does not have the right to suppress information. Authorship of publications resulting from this study will be based on the guidelines on authorship, such as those described in the Uniform Requirements for Manuscripts Submitted to Biomedical Journals, which state that the named authors must have made a significant contribution to the design of the study or analysis and interpretation of the data, provided critical review of the paper, and given final approval of the final version.

12.5.3 Registration of Clinical Studies and Disclosure of Results

The Sponsor or designee will register and/or disclose the existence of and the results of clinical studies as required by law.

12.5.4 Confidentiality

All study documents are provided by the Sponsor in confidence to the Investigator and his/her appointed staff. None of this material may be disclosed to any party not directly involved in the study without Sponsor's written permission.

The Investigator must assure that subjects' anonymity will be maintained. The Investigator will keep a separate list with at least the initials, the subjects' study numbers, names, addresses, and

telephone numbers. The Investigator will maintain this for the longest period of time allowed by his/her own institution and, in any case, until further communication from the Sponsor.

13.0 REFERENCES

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14.0 APPENDICES

14.1 Influenza Patient Reported Outcomes Questionnaire

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

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	☐	☐	☐	☐	☐
Lack of appetite	☐	☐	☐	☐	☐
Sleeping more than usual	☐	☐	☐	☐	☐

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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
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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14.2 Laboratory Assessments

Urinalysis	Hematology	Chemistry
Dipstick: • Specific gravity • pH • Glucose • Protein • Blood • Ketones • Bilirubin • Urobilinogen • Nitrite • Leukocyte esterase Urine sedimentation count: • Erythrocytes (RBC) • Leukocytes (WBC) • Epithelial cells	Hemoglobin	Albumin
	Hematocrit	Alkaline phosphate
	Red blood cells (RBC)	Alanine amino transferase
	White blood cells (WBC) with differential	Aspartate amino transferase
		Bicarbonate
	Lymphocytes	Total bilirubin
	Monocytes	Direct bilirubin [b]
	Neutrophils [a]	Blood urea nitrogen (or urea)
	Eosinophils	Calcium
	Basophils	Chloride
	Platelets	Creatinine
		Glucose [c]
		Lactate dehydrogenase
		Phosphate, inorganic
		Potassium
		Total protein
Microscopy: • Crystals • Casts • Bacteria		Sodium
		Creatine kinase-MB [d], Creatinine kinase [d], Troponin [d], Tryptase [e], Chymase [e]
	Other Assessments	**Coagulation**
Antibody screening test (immunoassay), rapid Any positive rapid test result should be confirmed according to local guidelines	Urine pregnancy test Erythrocyte Sedimentation Rate (ESR) C-Reactive Protein (CRP)	Partial thromboplastin time Activated partial thromboplastin time

[a] If immature neutrophils are detected, the sample is to be flagged and a blood slide for microscopic analysis will be made. If Bands are detected in the microscopic analysis, then a result will be provided.

[b] Assay if total bilirubin is above normal range.

[c] Baseline only.

[d] Only measure if subject has chest pain.

[e] In the event of a subject exhibiting the signs and symptoms of an anaphylactic reaction, when possible at the time of the event, a 5-mL serum sample should be collected for further assessment (ie tryptase, chymase).

14.3 Division of Microbiology and Infectious Diseases Adult Toxicity Table

The DMID Adult Toxicity Table appears on the following pages.

ABBREVIATIONS: Abbreviations utilized in the Table:

ULN = Upper Limit of Normal	LLN = Lower Limit of Normal
R_x = Therapy	Req = Required
Mod = Moderate	IV = Intravenous
ADL = Activities of Daily Living	Dec = Decreased

ESTIMATING SEVERITY GRADE

For abnormalities NOT found elsewhere in the Toxicity Tables use the scale below to estimate grade of severity:

GRADE 1 **Mild** Transient or mild discomfort (< 48 hours); no medical intervention/therapy required

GRADE 2 **Moderate** Mild to moderate limitation in activity - some assistance may be needed; no or minimal medical intervention/therapy required

GRADE 3 **Severe** Marked limitation in activity, some assistance usually required; medical intervention/therapy required, hospitalizations possible

GRADE 4 **Life-threatening** Extreme limitation in activity, significant assistance required; significant medical intervention/therapy required, hospitalization or hospice care probable

SERIOUS OR LIFE-THREATENING AEs

ANY clinical event deemed by the clinician to be serious or life-threatening should be considered a grade 4 event. Clinical events considered to be serious or life-threatening include, but are not limited to: seizures, coma, tetany, diabetic ketoacidosis, disseminated intravascular coagulation, diffuse petechiae, paralysis, acute psychosis, severe depression.

COMMENTS REGARDING THE USE OF THESE TABLES

Standardized and commonly used toxicity tables (Division of AIDS, NCI's Common Toxicity Criteria, and World Health Organization (WHO)) have been adapted for use by the DMID and modified to better meet the needs of participants in DMID trials.

For parameters not included in the following Toxicity Tables, sites should refer to the "Guide for Estimating Severity Grade" located above.

Criteria are generally grouped by body system.

Some protocols may have additional protocol specific grading criteria, which will supersede the use of these tables for specified criteria.

HEMATOLOGY				
	Grade 1	**Grade 2**	**Grade 3**	**Grade 4**
Hemoglobin	9.5 - 10.5 g/dL	8.0 - 9.4gm/dL	6.5 - 7.9 g/dL	< 6.5 g/dL
Absolute Neutrophil Count	1000-1500/ mm3	750-999/ mm3	500-749/ mm^3	<500/ mm^3
Platelets	75,000-99,999/ mm^3	50,000-74,999/ mm^3	20,000-49,999/ mm^3	<20,000/ mm^3
WBCs	11,000-13,000/mm^3	13,000-15,000/mm^3	15,000-30,000/ mm^3	>30,000 or <1,000/mm^3
% Polymorphonuclear Leucocytes + Band Cells	> 80%	90 – 95%	>95%	----------
Abnormal Fibrinogen	Low: 100-200 mg/dL High: 400-600 mg/dL	Low: <100 mg/dL High: >600 mg/dL	Low: < 50 mg/dL ----------	Fibrinogen associated with gross bleeding or with disseminated coagulation
Fibrin Split Product	20-40 mcg/ mL	41-50 mcg/ mL	51-60 mcg/ mL	> 60 mcg/ mL
Prothrombin Time (PT)	1.01 - 1.25 x ULN	1.26-1.5 x ULN	1.51 -3.0 x ULN	>3 x ULN
Activated Partial Thromboplastin (APPT)	1.01 -1.66 x ULN	1.67 - 2.33 x ULN	2.34 - 3 x ULN	> 3 x ULN
Methemoglobin	5.0 - 9.9%	10.0 - 14.9%	15.0 - 19.9%	> 20.0%

CHEMISTRIES				
	Grade 1	**Grade 2**	**Grade 3**	**Grade 4**
Hyponatremia	130-135 mEq/ L	123-129 mEq/ L	116-122 mEq/ L	< 116 mEq/ L or abnormal sodium *with* mental status changes or seizures
Hypernatremia	146-150 mEq/ L	151-157 mEq/ L	158-165 mEq/ L	> 165 mEq/ L or abnormal sodium *with* mental status changes or seizures
Hypokalemia	3.0 - 3.4 mEq/ L	2.5 - 2.9 mEq/ L	2.0 - 2.4 mEq/ L or intensive replacement therapy or hospitalization required	< 2.0 mEq/ L or abnormal potassium *with* paresis, ileus or life-threatening arrhythmia
Hyperkalemia	5.6 - 6.0 mEq/ L	6.1 - 6.5 mEq/ L	6.6 - 7.0 mEq/L	> 7.0 mEq/ L or abnormal potassium *with* life-threatening arrhythmia
Hypoglycemia	55-64 mg/dL	40-54 mg/dL	30-39 mg/dL	<30 mg/dL or abnormal glucose *with* mental status changes or coma
Hyperglycemia (nonfasting and no prior diabetes)	116 - 160 mg/dL	161- 250 mg/dL	251 - 500 mg/dL	> 500 mg/dL or abnormal glucose *with* ketoacidosis or seizures
Hypocalcemia (corrected for albumin)	8.4 - 7.8 mg/dL	7.7 - 7.0 mg/dL	6.9 - 6.1 mg/dL	< 6.1 mg/dL or abnormal calcium *with* life threatening arrhythmia or tetany

CHEMISTRIES (continued)				
	Grade 1	**Grade 2**	**Grade 3**	**Grade 4**
Hypercalcemia (correct for albumin)	10.6 - 11.5 mg/dL	11.6-12.5 mg/dL	12.6 - 13.5 mg/dL	> 13.5 mg/dL or abnormal calcium *with* life threatening arrhythmia
Hypomagnesemia	1.4 - 1.2 mEq/ L	1.1 - 0.9 mEq/ L	0.8 - 0.6 mEq/ L	< 0.6 mEq/ L or abnormal magnesium *with* life-threatening arrhythmia
Hypophosphatemia	2.0 - 2.4 mg/dL	1.5 -1.9 mg/dL or replacement Rx required	1.0 -1.4 mg/dL intensive therapy or hospitalization required	< 1.0 mg/dL or abnormal phosphate *with* life-threatening arrhythmia
Hyperbilirubinemia (when accompanied by any increase in other liver function test)	1.1 - <1.25 x ULN	1.25 - <1.5 x ULN	1.5 – 1.75 x ULN	> 1.75 x ULN
Hyperbilirubinemia (when other liver function are in the normal range)	1.1 - <1.5 x ULN	1.5 - <2.0 x ULN	2.0 – 3.0 x ULN	> 3.0 x ULN
BUN	1.25 - 2.5 x ULN	2.6 - 5 x ULN	5.1 - 10 x ULN	> 10 x ULN
Hyperuricemia (uric acid)	7.5 – 10.0 mg/dL	10.1 – 12.0 mg/dL	12.1 – 15.0 mg/dL	>15.0 mg/dL
Creatinine	1.1 - 1.5 x ULN	1.6 - 3.0 x ULN	3.1 - 6 x ULN	> 6 x ULN or dialysis required

ENZYMES				
	Grade 1	**Grade 2**	**Grade 3**	**Grade 4**
AST (SGOT)	1.1 - <2.0 x ULN	2.0 – <3.0 x ULN	3.0 – 8.0 x ULN	> 8 x ULN
ALT (SGPT)	1.1 - <2.0 x ULN	2.0 – <3.0 x ULN	3.0 – 8.0 x ULN	> 8 x ULN
GGT	1.1 - <2.0 x ULN	2.0 – <3.0 x ULN	3.0 – 8.0 x ULN	> 8 x ULN
Alkaline Phosphatase	1.1 - <2.0 x ULN	2.0 – <3.0 x ULN	3.0 – 8.0 x ULN	> 8 x ULN
Amylase	1.1 - 1.5 x ULN	1.6 - 2.0 x ULN	2.1 - 5.0 x ULN	> 5.1 x ULN
Lipase	1.1 - 1.5 x ULN	1.6 - 2.0 x ULN	2.1 - 5.0 x ULN	> 5.1 x ULN

URINALYSIS				
	Grade 1	**Grade 2**	**Grade 3**	**Grade 4**
Proteinuria	1+ or 200 mg - 1 g loss/day	2-3+ or 1- 2 g loss/day	4+ or 2-3.5 g loss/day	nephrotic syndrome or >3.5 g loss/day
Hematuria	microscopic only <10 RBC/HPF	gross, no clots >10 RBC/HPF	gross, with or without clots, OR red blood cell casts	obstructive or required transfusion

CARDIOVASCULAR				
	Grade 1	**Grade 2**	**Grade 3**	**Grade 4**
Cardiac Rhythm		asymptomatic, transient signs, no Rx required	recurrent/persistent; symptomatic Rx required	unstable dysrythmia; hospitalization and treatment required
Hypertension	transient increase >20 mm/ Hg; no treatment	recurrent, chronic increase > 20mm/ Hg. /treatment required	acute treatment required; outpatient treatment or hospitalization possible	end organ damage or hospitalization required
Hypotension	transient orthostatic hypotension with heart rate increased by <20 beat/min or decreased by <10 mm Hg systolic BP, No treatment required	symptoms due to orthostatic hypotension or BP decreased by <20 mm Hg systolic; correctable with oral flu id treatment	requires IV fluids; no hospitalization required	mean arterial pressure <60mm/ Hg or end organ damage or shock; requires hospitalization and vasopressor treatment
Pericarditis	minimal effusion	mild/ moderate asymptomatic effusion, no treatment	symptomatic effusion; pain; EKG changes	tamponade; pericardiocentesis or surgery required
Hemorrhage, Blood Loss	microscopic/occult	mild, no transfusion	gross blood loss; 1-2 units transfused	massive blood loss; > 3 units transfused

RESPIRATORY				
	Grade 1	**Grade 2**	**Grade 3**	**Grade 4**
Cough	transient- no treatment	persistent cough; treatment responsive	Paroxysmal cough; uncontrolled with treatment	----------
Bronchospasm, Acute	transient; no treatment; 70% - 80% FEV_1 of peak flow	Requires treatment; normalizes with bronchodilator; FEV_1 50%-70% (of peak flow)	no normalization with bronchodilator; FEV_1 25% - 50% of peak flow; or retractions present	cyanosis: FEV_1 < 25% of peak flow or intubation necessary
Dyspnea	dyspnea on exertion	dyspnea with normal activity	dyspnea at rest	dyspnea requiring Oxygen therapy

GASTROINTESTINAL				
	Grade 1	**Grade 2**	**Grade 3**	**Grade 4**
Nausea	mild or transient; maintains reasonable intake	moderate discomfort; intake decreased significantly; some activity limited	no significant intake; requires IV flu ids	hospitalization required;
Vomiting	1 episode in 24 hours	2-5 episodes in 24 hours	>6 episodes in 24 hours or needing IV fluids	physiologic consequences requiring hospitalization or requiring parenteral nutrition
Constipation	requiring stool softener or dietary modification	Requiring laxatives	Obstipation requiring manual evacuation or enema	obstruction or toxic megacolon
Diarrhea	mild or transient; 3-4 loose stools/day or mild diarrhea last <1 week	moderate or persistent; 5-7 loose stools/day or diarrhea lasting >1 week	>7 loose stools/day or bloody diarrhea; or orthostatic hypotension or electrolyte imbalance or >2 L IV fluids required	hypotensive shock or physiologic consequences requiring hospitalization
Oral Discomfort/Dysphagia	mild discomfort; no difficulty swallowing	some limits on eating/drinking	eating/talking very limited; unable to swallow solid foods	unable to drink fluids; requires IV fluids

NEUROLOGICAL				
	Grade 1	Grade 2	Grade 3	Grade 4
Neuro-Cerebellar	slight incoordination dysdiadochokinesis	intention tremor, dysmetria, slurred speech; nystagmus	locomotor ataxia	incapacitated
Psychiatric	mild anxiety or depression	moderate anxiety or depression; therapy required; change in normal routine	severe mood changes requiring therapy; or suicidal ideation; or aggressive ideation	acute psychosis requiring hospitalization; or suicidal gesture/attempt or hallucinations
Muscle Strength	subjective weakness no objective symptoms/ signs	mild objective signs/symptoms no decrease in function	objective weakness function limited	paralysis
Paresthesia (burning, tingling, etc.)	mild discomfort; no treatment required	moderate discomfort; non-narcotic analgesia required	severe discomfort; or narcotic analgesia required with symptomatic improvement	incapacitating; or not responsive to narcotic analgesia
Neuro-sensory	mild impairment in sensation (decreased sensation, e.g., vibratory, pinprick, hot/cold in great toes) in focal area or symmetrical distribution; or change in taste, smell, vision and/or hearing	Moderate impairment (mod decreased sensation, e.g., vibratory, pinprick, hot/cold to ankles) and/or joint position or mild impairment that is not symmetrical	severe impairment (decreased or loss of sensation to knees or wrists) or loss of sensation of at least mod degree in multiple different body areas (i.e., upper and lower extremities)	sensory loss involves limbs and trunk; paralysis; or seizures

MUSCULOSKELETEAL				
	Grade 1	**Grade 2**	**Grade 3**	**Grade 4**
Arthralgia (joint pain)	mild pain not interfering with function	moderate pain, analgesics and/or pain interfering with function but not with activities of daily living	severe pain; pain and/or analgesics interfering with activities of daily living	disabling pain
Arthritis	mild pain with inflammation, erythema or joint swelling – but not interfering with function	moderate pain with inflammation, erythema or joint swelling – interfering with function, but not with activities of daily living	severe pain with inflammation, erythema or joint swelling –and interfering with activities of daily living	permanent and/or disabling joint destruction
Myalgia	myalgia with no limitation of activity	Muscle tenderness (at other than injection site) or with moderate impairment of activity	severe muscle tenderness with marked impairment of activity	Frank myonecrosis

SKIN				
	Grade 1	**Grade 2**	**Grade 3**	**Grade 4**
Mucocutaneous	erythema; pruritus	diffuse, maculopapular rash, dry desquamation	vesiculation or mist desquamation or ulceration	Exfoliative dermatitis, mucous membrane involvement or erythema, mutiforme or suspected Stevens-Johnson or necrosis requiring surgery
Induration	< 15mm	15-30 mm	>30mm	
Erythema	< 15mm	15-30 mm	>30mm	
Edema	< 15mm	15-30 mm	>30mm	
Rash at Injection Site	< 15mm	15-30 mm	>30mm	
Pruritus	slight itching at injection site	moderate itching at injection extremity	itching over entire body	

SYSTEMIC				
	Grade 1	**Grade 2**	**Grade 3**	**Grade 4**
Allergic Reaction	pruritus without rash	localized urticaria	generalized urticaria; angioedema	anaphylaxis
Headache	mild, no treatment required	transient, moderate; treatment required	severe; responds to initial narcotic therapy	intractable; requires repeated narcotic therapy
Fever: oral	37.7 - 38.5 C or 100.0 - 101.5 F	38.6 - 39.5 C or 101.6 - 102.9 F	39.6 - 40.5 C or 103 - 105 F	> 40 C or > 105 F
Fatigue	normal activity reduced < 48 hours	normal activity decreased 25-50% > 48 hours	normal activity decreased > 50% can't work	unable to care for self

Visterra

PROTOCOL AMENDMENT – SUMMARY OF CHANGES

Phase 2b, Multicenter, Randomized, Double-blind, Controlled Study to Evaluate the Efficacy and Safety of Intravenous VIS410 in Addition to Oseltamivir (Tamiflu®) Compared With Oseltamivir Alone in Hospitalized Adults With Influenza A Infection Requiring Oxygen Support

Product	VIS410
Protocol Number	VIS410-203
EudraCT Number	2016-004009-15
Clinical Phase	2b
Clinical Indication	Influenza A infection

Sponsor	Visterra, Inc. One Kendall Square, Suite B3301 Cambridge, MA 02139 United States of America
Sponsor Representative	[redacted]

ORIGINAL PROTOCOL DATE: **21 October 2016**

AMENDMENT 1 DATE: **20 June 2017**

AMENDMENT 2 DATE: **19 April 2018**

**SUMMARY OF CHANGES TO PROTOCOL VIS410-203
FROM VERSION 2.0 TO VERSION 3.0**

Substantive changes to the protocol for Amendment 3 and their location within the protocol are noted below. Additions are marked as red underlined text and deletions are marked as red strikethrough text. Administrative, stylistic and formatting changes that do not alter the conduct of the study are not summarized in this document.

Location in Protocol	**Changes**	**Rationale**
Cover Page; Signature of Sponsor Representative	Issue Date (Version): ~~20 June 2017~~ (Version ~~2~~.0) 19 April 2018 (Version 3.0) Sponsor Representative [redacted]	Updated date and version number. Sponsor representative updated.
Cover page; Sponsor Address	Visterra, Inc. ~~One Kendall Square, Suite B3301~~ 275 Second Avenue, 4th Floor ~~Cambridge~~ Waltham, MA ~~02139~~02451 United States of America	Company moved location
Synopsis: Number of Clinical Sites	Approximately ~~180~~ 140 sites worldwide	Reduction in number of sites
Synopsis: Number of Subjects	Approximately ~~390~~ 120	Decreasing sample size from 390 to 120 subjects to enable completion in 2 influenza seasons.
Synopsis and Section 2.1.1 *Primary Efficacy Objective*	• ~~Evaluate~~ Evaluation of the effect of 2 dose levels of VIS410 + oseltamivir on ~~the time to normalization of respiratory function compared to oseltamivir alone~~ clinical outcome as assessed by comparison of clinical status ordinal scale Day 7 scores between treatment groups, and between all VIS410 recipients versus placebo.	Utility of ordinal scale observed in prior trials of this size.

Synopsis and Section 2.2 *Secondary Objectives*	• Among patients requiring supplemental oxygen therapy at the time of enrollment with baseline room air oxygen saturation of ≤ 92%, time to cessation of oxygen (O_2) support resulting in stable oxygen saturation (SpO_2) by pulse oximetry. Stable SpO_2 is defined as two consecutive SpO_2 values of > 92% on room air that are at least 8 hours apart.
	• For any patient requiring supplemental oxygen therapy at the time of enrollment (regardless of oxygen saturation), time to cessation of oxygen support.
	• Evaluate the effect of 2 dose levels of VIS410 + oseltamivir vs oseltamivir alone on the following parameters:
	  • Viral load in upper respiratory samples
	  • Time to clinical response
	  • Time to cessation of ventilator support
	  • Time to resumption of normal activities
	  • All-cause and attributable 14-, ~~and~~ 28-, and 56-day mortality
	  • Clinical status ordinal scale mean area under the
		Time to cessation of oxygen support changed from primary endpoint to secondary. Some patients may be on oxygen with O_2 saturation > 92% at baseline.
		
		Added ordinal scale parameters to be assessed

Location in Protocol	Changes	Rationale
	curve for Days 1-7 and Days 1-14 using linear numeric scores for the ordinal categories. • Comparison of clinical status ordinal scale scores for selected individual days (i.e., Days 3, 4, 5, and 6) • Comparison of clinical status ordinal scale scores using modified ordinal scale criteria (i.e. pooling of selected severity criteria scores) • Comparison of discrete ordinal scale parameters, including days of ventilator support, days in intensive care, and duration of hospitalization • Healthcare resource utilization • Analysis of time ~~Time~~ to alleviation of ~~clinical~~ signs and symptoms of influenza in the subset of subjects able to complete the Influenza Patient Reported Outcomes (FluPRO) Questionnaire at baseline and post-dose by Kaplan Meier analysis • Proportion of subjects with new documented bacterial pneumonia/superinfection • Proportion of subjects with influenza-related complications • Pharmacokinetics of VIS410 in serum • Immunogenicity of VIS410 • Emergence of resistance to VIS410 and oseltamivir	
Synopsis and Section 2.3 *Exploratory Objectives*	~~• Evaluate the effect of VIS410 + oseltamivir vs oseltamivir alone on clinical outcomes as measured by a seven level ordinal scale~~ • Evaluate the pharmacokinetics of VIS410 from nasopharyngeal secretions and tracheal aspirate (ventilated subjects only) • Assess the effects of VIS410 on viral load in tracheal aspirate (ventilated subjects only) • Assess correlations between virology, safety, VIS410 dose, pharmacokinetics, viral shedding, immunology, signs and symptoms of influenza, and other endpoints • Assess the anti-influenza immune response	Removed as components are now primary and secondary endpoints
Synopsis	**Study Design** This is a Phase 2b, multicenter, randomized, double-blind, controlled study comparing the efficacy and safety of 2 dose levels of VIS410 (2000 and 4000 mg) in combination with oseltamivir vs oseltamivir alone in hospitalized subjects with influenza A infection requiring oxygen support. Subjects will be randomly assigned at a ratio of 1:1:1 using a permuted block randomization method to receive VIS410 at a dose of 2000 mg (low dose) or 4000 mg (high dose) or placebo (0.9% sodium chloride), administered as a single IV infusion over 2 hours. Randomization will be stratified by presence or absence of positive pressure ventilation (PPV) at baseline to ensure an even distribution across treatment arms. All subjects will receive oseltamivir 75 mg twice daily (BID) for 5 days (total of 10 doses) as part of standard of care (SOC). Treatment with oseltamivir may be extended for	Decreasing sample size from 390 to 120 subjects to enable completion in 2 influenza seasons. Table in section also updated to reflect 40 subjects per arm.

Location in Protocol	Changes	Rationale
	up to 5 additional days (up to 20 doses of total therapy) if clinical symptoms warrant further treatment as assessed by the Investigator. Note: based on SOC, subjects may have started oseltamivir therapy prior to randomization. All subjects will also receive a pretreatment regimen of diphenhydramine 50 mg PO or IV plus either ibuprofen 400 mg PO or acetylsalicylic acid 320-325 mg PO approximately 60 minutes before infusion. Approximately ~~390~~ 120 evaluable subjects (~~130~~ 40/arm) with confirmed influenza A infection will be treated. Subjects admitted to the hospital within 5 days of onset of initial symptoms who require supplemental oxygen will undergo a rapid influenza test or a local polymerase chain reaction (PCR) test, fluorescent immunoassay (FIA) test, or enzyme-linked immunosorbent assay (ELISA) test to confirm influenza A infection. Subjects diagnosed with influenza A will undergo the screening procedures to confirm eligibility. Study assessments are outlined in **Error! eference source not found.** (Schedule of Assessments). Eligible subjects will be randomized to receive either oseltamivir + VIS410 2000 mg, oseltamivir + VIS410 4000 mg, or oseltamivir + placebo (0.9% sodium chloride) at a ratio of 1:1:1 using a permuted block randomization method according to the table below: Oseltamivir (Tamiflu®) will be provided by the Sponsor. Treatment with oseltamivir may be extended for up to 5 additional days (up to 20 doses of total therapy) if clinical symptoms \warrant further treatment as assessed by the Investigator. Note: based on SOC, subjects may have started oseltamivir therapy prior to randomization. Study assessments are outlined in **Error! Reference source ot found.**. Subjects discharged from the hospital prior to Day 14 will attend the applicable outpatient follow-up visit(s) (eg, Day 3, Day 5, Day 7, Day 14) per **Error! Reference source not found.**. An independent data safety monitoring board (DSMB) will be established to review all available safety data after 30 subjects ~~as well as when~~ and again after ~~approximately 120~~ 70 ~~subjects~~ have completed ~~study~~ Day 14 assessments. The assessment of safety will be determined from vital sign measurements, physical examinations, hematology, chemistry and urinalysis laboratory testing, 12-lead electrocardiograms (ECGs), use of concomitant medications, and review of adverse events (AEs). Based on the safety data provided, the DSMB will determine if it is acceptable to continue the study or reduce the dose as appropriate. Dosing will temporarily pause while the DSMB meets if there are 4 treatment-related serious adverse events (SAEs) or 4 severe gastrointestinal (GI) treatment–emergent adverse events (TEAEs) that require intervention, which is defined as requiring IV fluid and medication to decrease the frequency of diarrhea (ie, loperamide). Additional DSMB reviews can occur throughout the trial as deemed necessary by the DSMB or Sponsor to ensure subject safety and well-being. Study enrollment and dosing will continue while the DSMB evaluates data.	Performing second DSMB review after completion of Northern Hemisphere season. No need for DSMB review after 120 subjects; final analysis of data will be performed.

Location in Protocol	Changes	Rationale
	~~In addition, following 50% enrollment (195 subjects), an interim analysis may be conducted, by an unblinded third party, to assess if one of the VIS410 treatment arms can be terminated early for futility. A prespecified sample size reanalysis may also be conducted based on the observed effect size. The interim analysis may also test the primary efficacy objective to assess if the primary efficacy endpoint has been met. An alpha spending function will be designed in order to determine the amount of alpha that will be spent at the interim analysis. The effect size for this study was estimated without the benefit of any prior randomized controlled trial. If the VIS410 effect size proves to be greater than anticipated, then it is possible that the primary study endpoint will be met by the time the interim efficacy analysis is performed. In the event the interim analysis reveals that the primary efficacy endpoint has been satisfied, this Phase 2 study could be terminated for efficacy~~.	Removed interim analysis as sample size decreased to 120 subjects; final analysis to be performed upon completion of enrollment.
Synopsis and Section 5.1 *Number of Subjects*	Approximately ~~390~~ 120 evaluable subjects will be enrolled in 3 equal arms: VIS410 2000 mg, VIS410 4000 mg, and placebo.	Sample size decreased.
Synopsis and Section 5.2.1 *Inclusion Criteria*	9. Subject, or a legally acceptable ~~authorized~~ representative, is able to understand the purpose and risks of the study and willing to give voluntary written informed consent.	Change requested by an Ethics Committee
Synopsis	**Test Product, Dose, Mode of Administration** VIS410 (2000 mg or 4000 mg) will be administered IV over 2 hours as a single 200-mL infusion, followed by a 25-mL (or volume equivalent to IV line volume if greater than 25 mL) saline flush to ensure all product is administered, in addition to oseltamivir (Tamiflu) 75 mg BID for total of 5 days (10 doses). For patients with renal insufficiency, refer to prescribing information for administration of oseltamivir. For patients unable to swallow, oseltamivir capsules can be opened and mixed with liquid for ease of administration. **Reference Product, Dose, Mode of Administration** Placebo (normal saline solution 0.9%) will be administered IV over 2 hours as a single 200-mL infusion, followed by a 25-mL (or volume equivalent IV line volume if greater than 25 mL) saline flush to ensure all product is administered, in addition to oseltamivir (Tamiflu) 75 mg BID for total of 5 days (10 doses). For patients with renal insufficiency, refer to prescribing information for administration of oseltamivir. **VIS410 and Placebo Preparation** The required amount of VIS410 to be dosed (2000 mg or 4000 mg) will be diluted with normal saline up to a maximum total volume of 200 mL; for placebo subjects, 200 mL of normal saline will be prepared. Length of IV line will ideally be set for maximum volume of 25 mL, so that the 25-mL (or increased volume as noted above) saline flush following administration will ensure all VIS410/placebo has been administered. In the event that the infusion line volume is greater than 25 mL the post-administration saline flush should be increased to match the volume of the infusion line kit.	Clarification to ensure all study product administered. Some sites use infusion lines with hold-up volumes of greater than 25 mL.

Location in Protocol	Changes	Rationale
	The VIS410/placebo infusion will be administered IV using a 0.22-µm in-line filter and will be controlled by a volumetric pump. Standard, uniform-length infusion lines will be used whenever possible, and microfilters will be provided by the Sponsor. The infusion bag will be covered with an opaque sleeve in the pharmacy to maintain the study blind. The VIS410/placebo will be administered IV at a rate of 100 mL/h, over 2 hours. After ~~200 mL of~~ the ~~diluted~~ dose has been administered, the infusion will be ~~stopped,~~ followed by ~~a 25 mL~~ an appropriate volume saline flush as described above. The infusion time may be extended up to 4 hours at the Investigator's discretion based on local infusion site–related symptoms. All subjects will be given a pretreatment regimen of PO or IV diphenhydramine 50 mg plus either ibuprofen 400 mg PO or acetylsalicylic acid 320-325 mg PO or crushed and given through the nasogastric tube approximately 60 minutes before IV infusion of VIS410/placebo.	
Synopsis and Section 3.1.1 *Primary Efficacy Endpoint*	~~• Time to cessation of oxygen (O_2) support resulting in stable oxygen saturation (SpO_2) by pulse oximetry. Stable SpO_2 is defined as two consecutive SpO_2 values of > 92% on room air that are at least 8 hours apart.~~ • The primary efficacy outcome analysis compares Day 7 clinical status ordinal scale scores between treatment groups, and between all VIS410 recipients versus placebo. Clinical status is measured daily for 14 days using the below seven-level ordinal scale, with the classifications presented from the worst clinical outcome to the best clinical outcome in descending order; for each day, subject status will be classified by the worst clinical outcome for which they qualify. ▪ Death ▪ ICU stay with mechanical ventilation ▪ ICU stay without mechanical ventilation ▪ Non-ICU hospitalization with supplemental oxygen ▪ Non-ICU hospitalization without supplemental oxygen ▪ Discharge with partial resumption of normal activities ▪ Discharge with full resumption of normal activities	Primary objective changed.
Synopsis and Section 3.2 *Secondary Endpoints*	The difference between VIS410 + oseltamivir and oseltamivir alone treatment groups in the following endpoints: • Among patients requiring supplemental oxygen therapy at the time of enrollment with baseline room air oxygen saturation of ≤ 92%, time to cessation of oxygen (O_2) support resulting in stable oxygen saturation (SpO_2) by pulse oximetry. Stable SpO_2 is defined as two consecutive SpO_2 values of > 92% on room air that are at least 8 hours apart.	Moved from primary objective to secondary

Location in Protocol	Changes	Rationale

	• For any patient requiring supplemental oxygen therapy at the time of enrollment (regardless of oxygen saturation), time to cessation of oxygen support
	• Peak viral load, viral area under the concentration–time curve (AUC), duration of viral shedding, and time to resolution of viral load from nasopharyngeal swabs by $TCID_{50}$ and qRT-PCR
	• Time to clinical response defined as resolution of at least 4 of 5 vital signs:
	  • Afebrile with core temperature ≤ 37.8°C, without use of antipyretics (oral ≤ 37.2°C)
	  • Oxygen saturation ≥ 95% on room air without support or a return to pre-infection status, if pre-infection status was < 95%
	  • Pulse rate ≤ 100/min
	  • Systolic blood pressure (SBP) ≥ 90 mm/Hg, without vasopressor use
	  • Respiratory rate ≤24 beats per minute
	• ~~Total number of days on ventilation~~
	• Clinical status ordinal scale mean area under the curve for Days 1-7 and Days 1-14 using linear numeric scores for the ordinal categories.
	• Comparison of clinical status ordinal scale scores for selected individual days (i.e., Days 3, 4, 5, and 6)
	• Comparison of clinical status ordinal scale scores using modified ordinal scale criteria (i.e. pooling of selected severity criteria scores)
	• Comparison of discrete ordinal scale parameters, including days of ventilator support, days in intensive care, and duration of hospitalization
	• Number of days to resumption of normal activities
	• All-cause and attributable mortality rates at Day 14, ~~and~~ 28, and 56
	• Total number of days in hospital and/or intensive care unit (ICU) from admission to discharge and rate of rehospitalization due to influenza A relapse/complication
	• The incidence, severity, and duration of signs and symptoms of influenza-like illness as assessed by the FluPRO Questionnaire (see Appendix **Error! eference source not found.**)
	• Analysis of time to alleviation of signs and symptoms of influenza in the subset of subjects able to complete the Influenza Patient Reported Outcomes (FluPRO) Questionnaire at baseline and post-dose by Kaplan Meier analysis
	• The percentage of subjects with new bacterial pneumonia/superinfection
	• The percentage of subjects with influenza-related complications
	• VIS410 population pharmacokinetic (PK) parameters in
		Deleted as covered by next two bullets addressing ordinal scale assessments

Location in Protocol	Changes	Rationale
	serum • Titer of anti-VIS410 antibody positive samples • Genotypic and/or phenotypic assessment to determine the emergence of VIS410 and oseltamivir-resistant viruses	
Synopsis and Section 3.3 *Exploratory Endpoints*	~~• Clinical outcome will be measured daily for 14 days using a seven-level ordinal scale; this seven-level ordinal scale is defined as:~~ ~~• Death~~ ~~• ICU stay with mechanical ventilation~~ ~~• ICU stay without mechanical ventilation~~ ~~• Non-ICU hospitalization with supplemental oxygen~~ ~~• Non-ICU hospitalization without supplemental oxygen~~ ~~• Discharge with partial resumption of normal activities~~ ~~• Discharge with full resumption of normal activities~~ • Population PK parameters of VIS410 from nasopharyngeal secretions • VIS410 concentration in tracheal aspirates • The difference in viral load between VIS410 + oseltamivir and oseltamivir alone treatment groups in tracheal aspirate of subjects on mechanical ventilation • Titer of anti-influenza A antibodies by hemagglutinin inhibition assay (HAI) in serum • Correlations between serum and/or nasopharyngeal PK with viral load, clinical symptoms, presence of antidrug antibodies (ADAs), safety, and additional endpoints	Now covered under primary efficacy and secondary endpoints
Synopsis	**Statistical Methods** *Sample Size* Approximately 120 ~~390~~ evaluable subjects will be enrolled. The study is exploratory in nature, and is not powered to demonstrate significant differences between treatment groups in primary or secondary outcome measures. The protocol intent is to collect sufficient information to identify the most appropriate candidate endpoints for subsequent Phase 3 study evaluation from among the primary and secondary endpoints described below. Statistical significance testing will therefore be used to assess the relative strength of evidence of the primary and secondary endpoints, to provide reasonable assurance that the endpoints chosen for a confirmatory Phase 3 trial will elucidate treatment differences between VIS410 plus oseltamivir versus oseltamivir alone. ~~Using a log-rank test, a sample size of 130 influenza A-infected subjects per treatment group will provide 80% power to detect a 1.5-day difference (5 days for oseltamivir alone and 3.5 days for VIS410 plus oseltamivir) in the median time to normalization of respiratory function (cessation of O_2 support) for VIS410 relative to placebo. A two-sided alpha of 0.05 was used for the calculation.~~ *Efficacy* Efficacy analysis will have two complementary purposes in this Phase 2 study, both designed to aid in the optimal design	Analysis changed due to the decrease in sample size.

Location in Protocol	Changes	Rationale
	of a confirmatory Phase 3 trial. First, primary and secondary endpoints will be analyzed for treatment group differences, to determine which endpoints might be most sensitive demonstration of treatment group differences using p-values as an indicator, Second, the relative contribution of the various efficacy endpoints to patient well-being and benefit will be assessed, as well as potential correlations of endpoints such as the influence of peak viral load or number of days on ventilator on the time to resumption of normal activities. The ordinal scale outcomes will be measured daily from Day 1 (baseline) through Day 14, inclusive, using a seven-level hierarchical scale with the classifications ordered from the worst to the best clinical outcomes (see Section 10.6.1). For use in overall summary statistical presentations, the ordinal categories will be assigned decreasing integer scores, with death a score of 6 and discharge with full resumption of normal activities a score of 0. The primary outcome comparison of Ordinal Scale scores at Day 7 between treatment groups will be evaluated by proportional odds ratio analysis, as implemented by logistic regression, including a test of the proportional odds assumption. For this analysis, the response categories will be ordered from best (Discharge with full resumption of normal activities) to worst (Death). Additional exploratory analyses may be conducted to obtain a more complete understanding of the relationship of treatment to the ordinal response, including exact Mantel-Haenszel tests or partial proportional odds models. The area under the curve (AUC) over time for a given patient will be calculated as the sum of the maximum ordinal score for each day up through 7 and 14 days. An analysis of treatment group differences will be performed on these per-patient AUC values using analysis of variance, with treatment group and strata as fixed factors. A sensitivity analysis will be performed that excludes the category of death on study, to determine if death as an outcome skews the results; death as an outcome will also be analyzed as an independent secondary endpoint. A secondary analysis of treatment group effect on the difference in proportions of patients with the worst (death) versus the best outcome (discharge from hospital and resumption of normal activities) will also be performed. This analysis does not use scores for the ordinal outcome but does account for ordinality, and is therefore not dependent on the relationship of score to severity of outcome. An exploratory analysis will be performed through an exact categorical analysis of treatment group difference in the ordinal scale results using the worst outcome on a per-patient basis. Additional ordinal scale outcome assessments will include comparison of total numbers of days at more severe scale values (death, time on ventilator, time in ICU) and proportions of patients with ordinal scale worsening post enrollment. Proportion outcomes will be analyzed by categorical data analysis methods, including Mantel-Haenszel chi-square tests adjusted for strata; additional exploratory subgroup analyses may also be performed, for example, based on various age categories. Time from onset	

Location in Protocol	Changes	Rationale
	of symptoms to study treatment and the number of doses of oseltamivir (Tamiflu) prior to VIS410 treatment for the current case of influenza may be included as covariates in the analyses. Secondary and exploratory endpoints will be analyzed for descriptive purposes, with significance levels (p-values) provided to illustrate the strength of evidence for treatment effects, and to provide a basis for the choice of endpoints for further study in a confirmatory study. These analyses will also provide estimates for potentially powering additional efficacy endpoints, and to assist in hierarchical ordering of secondary endpoints to provide alpha-control in confirmatory studies and for labelling purposes. Time to cessation of O_2 support resulting in a stable SpO_2 will be analyzed using a Cox proportional hazards regression model including data from patients on O_2 support. ~~Time from onset of symptoms to VIS410 treatment and number of doses of oseltamivir (Tamiflu) prior to VIS410 treatment for the current case of influenza will be included as a covariate of the analysis~~. A P-value (using the Wald statistic) for each VIS410 dose vs placebo will be presented. The total number of days in the hospital and/or ICU from admission to discharge and the rate of rehospitalization due to influenza A relapse/reinfection will be summarized descriptively by treatment group. The probability of time to clinical response (defined as resolution of vital signs) will be calculated via Kaplan-Meier. A ~~P-value~~ significance test (using the log-rank test) for each VIS410 dose vs placebo will be presented. Results will be tabulated and presented graphically as well. The number and percentage of subjects in each treatment group with clinical response will be summarized. ~~The ordinal scale outcomes will be measured daily from Day 1 (baseline) through Day 14, inclusive, using a seven-level hierarchical scale with the classifications ordered from the worst to the best clinical outcomes (see **Section 0**). The number and percentage of subjects in each treatment group with each classification will be summarized for each day from Day 1 through Day 14, inclusive.~~ ~~*Interim Analyses of Efficacy*~~ ~~An interim analysis of efficacy may be performed by an unblinded third party after 50% of subjects have been enrolled (n = 195) and have an assessment of the primary endpoint (time to normalization of respiratory function). This interim analysis may be conducted to assess if one of the active VIS410 treatment arms will need to be terminated for futility. The interim analysis may also test the primary efficacy objective to assess if the primary efficacy endpoint has been met early. A prespecified sample size reanalysis will also be conducted based on the observed effect size. The DSMB and the unblinded statistician will review the interim analysis results to provide their recommendation on discontinuation of one of the VIS410 treatment arms, if needed, based on pre-specified criteria as outlined in the SAP. The interim analysis may also review the following~~	No interim analysis to be performed due to the decrease in sample size.

Location in Protocol	Changes	Rationale
	~~key secondary endpoints, when available, to assist in the decision of discontinuing a VIS410 treatment arm:~~ ~~• Time to clinical resolution of vital signs~~ ~~• Total number of hours on oxygen support or PPV~~ ~~• Viral load from nasopharyngeal swabs~~ ~~A sample size re-estimation (SSRE) will also be performed at this point in order to determine if the current sample size is sufficient based on the observed effect size in the study. The unblinded statisticians will work directly with the Sponsor on the SSRE. The details of SSRE and individuals with access to the information will be outlined in a separate plan.~~ ///// *Complications of Influenza* Complications of influenza are defined as pneumonia, myocarditis, worsening of chronic bronchitis, sinusitis, otitis, or other chronic pulmonary diseases, encephalopathy, and death. Complications of influenza and use of antibiotic therapy for bacterial pneumonia will be reported by variable, treatment group, and time point. Diagnosis of bacterial pneumonia will be based on local clinical practice, usually an assessment of clinical features and any radiographic evidence of bacterial pneumonia (eg, infiltrate) and/or microbiologic evidence of bacterial pneumonia. All-cause and attributable mortality will be evaluated on Day 14 and 28.	
Table 1 *Schedule of Assessments*	Table: Erythrocyte sedimentation rate (ESR)[20a] Footnotes: [20a] ESR to be performed locally.	Clarification.
List of Abbreviations	AUC – Area under curve	New abbreviation
Section 1.1 *Background Information*	Severe influenza disease is a common occurrence each season, especially in high-risk groups, such as young children, older adults, patients with pulmonary conditions, inflammatory conditions, malignancies, and pregnant women.[i, ii] Despite available therapy with neuraminidase inhibitors, including oseltamivir (Tamiflu®), zanamivir (Relenza®), and peramivir (Rapivab®), 10% to 44% of hospitalized patients require intensive care and 25% to 50% of these patients die. It is estimated that as many as 400,000 patients are hospitalized with influenza each year in the United States, with up to 49,000 deaths per year.[iii] The World Health Organization (WHO) has reported incidence rates of 3 to 5 million severe cases and about 250,000 to 500,000 influenza-related deaths annually.[iv] The 2009 influenza A pandemic (H1N1) spread rapidly to every continent with more than 399,232 reported cases and 4735 deaths.[v] The therapeutic use of passive polyclonal antibodies to prevent viral infections, including hepatitis B, varicella, cytomegalovirus, rabies, and respiratory syncytial virus (RSV) has been well established. More recently, monoclonal antibodies for viral infections have been developed,	

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	including palivizumab (Synagis®), a Food and Drug Administration (FDA)-licensed treatment for the prevention of RSV infection. Visterra has developed a novel approach to antibody discovery whereby functionally conserved epitopes are identified based on atomic interaction networks and targeted with rationally engineered human antibodies. Using this approach, VIS410, a broad spectrum human immunoglobulin G1 (IgG1) monoclonal antibody with demonstrated efficacy against both Group 1 (including H1 and H5) and Group 2 (including H3 and H7) influenza A strains, in both treatment and prevention models of influenza, was developed. Visterra intends to develop this product for the treatment of influenza A, specifically in hospitalized patients. This study will provide the first indications of efficacy, safety, and tolerability of VIS410 in hospitalized subjects with influenza A infection requiring oxygen support. Efficacy will be ~~measured by time to cessation of oxygen support~~ assessed by comparison of clinical status ordinal Day 7 scores between treatment groups.	
Section 4.1 *Overview*	This is a Phase 2b multicenter, randomized, double-blind, controlled study comparing the efficacy and safety of 2 dose levels of VIS410 (2000 and 4000 mg) in combination with oseltamivir vs oseltamivir alone in hospitalized subjects with influenza A infection requiring oxygen. Subjects will be randomly assigned at a ratio of 1:1:1 using a permuted block randomization method to receive VIS410 at a dose of 2000 mg (low dose) or 4000 mg (high dose) or placebo (0.9% sodium chloride), administered as a single IV infusion over 2 hours. Randomization will be stratified by presence or absence of positive pressure ventilation (PPV) at baseline to ensure an even distribution across treatment arms (see Section **Error! eference source not found.**). All subjects will receive oseltamivir 75 mg twice daily (BID) for 5 days (total of 10 doses) as part of standard of care (SOC). All subjects will also receive a pretreatment regimen of diphenhydramine 50 mg PO or IV plus either ibuprofen 400 mg PO or acetylsalicylic acid 320-325 mg PO 60 minutes before VIS410/placebo infusion. Approximately ~~390~~120 evaluable subjects (~~130~~40/arm) with confirmed influenza A infection will be treated. Subjects admitted to the hospital within 5 days of onset of initial symptoms who require supplemental oxygen will undergo a rapid influenza test (supplied by the Sponsor) or a PCR, fluorescent immunoassay (FIA), or enzyme-linked immunosorbent assay (ELISA) test to confirm influenza A infection. Subjects diagnosed with influenza A will undergo the screening procedures to confirm eligibility. Study assessments are outlined in **Error! Reference source not ound.**, Schedule of Assessments. Eligible subjects will be randomized to receive either oseltamivir + VIS410 2000 mg, oseltamivir + VIS410 4000 mg, or oseltamivir + placebo (0.9% sodium chloride) at a ratio of 1:1:1 using a permuted block randomization method	Decreasing sample size from 390 to 120 subjects to enable completion in 2 influenza seasons. Table 6 also updated to reflect 40 subjects per arm.

Location in Protocol	Changes	Rationale
	summarized in **Error! Reference source not found.**. Oseltamivir (Tamiflu) will be provided by the Sponsor. Treatment with oseltamivir may be extended for up to 5 additional days (up to 20 doses of total therapy) if clinical symptoms warrant further treatment as assessed by the Investigator. Note: based on SOC, subjects may have started oseltamivir therapy prior to randomization. ~~In addition, following 50% enrollment (195 subjects), an interim analysis may be conducted, by an unblinded third party, to assess if one of the VIS410 treatment arms can be terminated early for futility. A prespecified sample size reanalysis may also be conducted based on the observed effect size. The interim analysis may also test the primary efficacy objective to assess if the primary efficacy endpoint has been met. An alpha spending function will be designed in order to determine the amount of alpha that will be spent at the interim analysis. The effect size for this study was estimated without the benefit of any prior randomized controlled trial. If the VIS410 effect size proves to be greater than anticipated, then it is possible that the primary study endpoint will be met by the time the interim efficacy analysis is performed. In the event the interim analysis reveals that the primary efficacy endpoint has been satisfied, this Phase 2 study could be terminated for efficacy~~.	Removed interim analysis as sample size decreased to 120 subjects; final analysis to be performed upon completion of enrollment.
Section 4.3 *Data Safety Monitoring Board*	An independent data safety monitoring board (DSMB) will be established to review all available safety data after 30 subjects, ~~as well as when~~ and subsequently, approximately ~~120~~ 70 subjects, have completed study Day 14~~.~~ The assessment of safety will be determined from vital sign measurements, physical examinations, hematology, chemistry and urinalysis laboratory testing, 12-lead electrocardiograms (ECGs), use of concomitant medications, and review of AEs. Based on the safety data provided, the DSMB will determine if it is acceptable to continue the study or reduce the dose as appropriate. Additional DSMB reviews can occur throughout the trial as deemed necessary by the DSMB or Sponsor to ensure subject safety and well-being. Study enrollment and dosing will continue while the DSMB evaluates data. Further details will be described in a separate DSMB charter.	Sample size decreased therefore second DSMB review not required.
Section 5.2.1 *Inclusion Criteria*	9. Subject, or a legally acceptable ~~authorized~~ representative, is able to understand the purpose and risks of the study and willing to give voluntary written informed consent.	Change requested by an Ethics Committee
Section 6.1.3 *Study Procedures*	On Day 1, eligibility of the subjects will be confirmed and assessments will be performed as described in **Error! eference source not found.**. All results from the screening procedure needed to evaluate eligibility, including any local clinical laboratory results, must be available prior to randomization on Day 1. All clinical assessments required for the determination of subject eligibility will be performed by the local clinical laboratory, thereby precluding the need to wait for central laboratory data. Any abnormal assessment at the screening visit will be assessed according to its clinical relevance, and if found relevant, the subject will not be included in the study.	Clarification

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	Once a subject has satisfied entry criteria, he/she will be assigned a unique identifier. The site's unblinded pharmacist or properly trained designee will ~~randomize the subject using the~~ access the Interactive Web Response System (IWRS) to obtain the study treatment. Randomization will be stratified by presence or absence of PPV at baseline to ensure balance between the treatment arms. Subjects will be considered randomized when the pharmacist or designee obtains the subject number and treatment assignment from the IWRS. Subjects may not be randomized into this study more than once. Subjects who have participated in any previous study of VIS410 may not be randomized to this study. Subjects will be monitored daily while in the hospital up to Day 14 (± 3 days) with an additional visit on Day 28 (± 3 days) and the last follow-up visit on Day 56 (± 7 days). In order to provide some flexibility for the subjects regarding the site visits and to maintain the integrity of the study design, a time window is permitted for the follow-up visits in case of time conflict or unforeseen circumstances. Note: subjects discharged from the hospital prior to Day 14 will attend the applicable outpatient follow-up visit(s) (eg, Day 3, Day 5, Day 7, Day 14).	
Section 6.2 *Pregnancy Safeguards*	Pregnancy will be determined by evaluation of β-human chorionic gonadotropin in serum or urine for all women of childbearing potential. Subjects who are pregnant or nursing will be excluded from the study. During the course of the study drug administration period within the study, any nursing mother(s) or subject(s) with suspected or confirmed pregnancy will be discontinued from study drug therapy but will be encouraged to undergo follow-up for safety monitoring for themselves (ie, the pregnant female) and the baby. All women of childbearing potential and all male subjects must practice effective contraception from screening until 60 days post-VIS410/placebo infusion. For the purposes of this study, women who do not satisfy at least one of the following criteria listed below (ie, the criteria for defining non-childbearing potential) are considered to be physiologically capable of becoming pregnant and are, therefore, defined as women of childbearing potential. The criteria for defining women as being of non-childbearing potential are: • Post-menopausal: ≥ 12 months of natural (spontaneous) amenorrhea, or • Follicle stimulating hormone > 40 mIU/mL as documented in their medical history, or • Surgical bilateral oophorectomy with or without hysterectomy, or • Hysterectomy, or • Bilateral tubal ligation Women of childbearing potential and all male subjects participating in heterosexual relations must be willing to practice effective contraception from screening until 60 days post-VIS410/placebo infusion. For the purposes of the study,	

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	highly effective contraception is defined as: • Male vasectomy with negative semen analysis documentation at least 6 months prior to dosing. OR • Combination of an established form of hormonal contraception (oral, injected, or implanted) or an intrauterine device or intrauterine system or sponge Plus one of the following: • A physical barrier method of contraception with use of a spermicide, such as condom or occlusive cap (diaphragm or cervical/vault caps) with spermicidal foam/gel/film/cream suppository. The use of barrier contraceptives should always be supplemented with the use of a spermicide, unless not available in a country. OR • Male vasectomy with negative semen analysis documentation less than 6 months prior to dosing. OR • Complete abstinence can be considered an acceptable method of contraception at the discretion of the Investigator. Periodic abstinences (eg, calendar, ovulation, symptothermal, or post-ovulation methods) and withdrawal are NOT considered acceptable methods of contraception. The combination of 2 barrier methods, periodic abstinence (eg, calendar, ovulation, symptothermal, or post-ovulation methods), and withdrawal are not considered acceptable methods of contraception. Male subjects should not donate sperm for at least 60 days after receipt of study product. Pregnancy reporting is described in Section **Error! eference source not found.**.	Sponge is an acceptable method of contraception Added requirement.
Section 6.5.2 *Injection Site Tolerability*	Injection site tolerability is defined as AEs demonstrating significant local injection site irritation or tissue damage. ~~Injection site tolerability will be reported by variable, treatment group, and time point.~~	Sentence deleted as belongs in SAP
Section 6.5.3 *Complications of Influenza*	Complications of influenza are defined as pneumonia, myocarditis, worsening of chronic bronchitis, sinusitis, otitis, or other chronic pulmonary diseases, encephalopathy, and death. Complications of influenza and use of antibiotic therapy for bacterial pneumonia will be reported by variable, treatment group, and time point. Diagnosis of bacterial pneumonia will be based on local clinical practice, usually an assessment of clinical features and any radiographic evidence of bacterial pneumonia (eg, infiltrate) and/or microbiologic evidence of bacterial pneumonia. All-cause and attributable mortality will be evaluated on Day 14 and Day 28.	
Section 7.1 *Test product, Dose,*	The Investigator must ensure that the investigational product will be used only in accordance with the protocol. It is	Clarification as some sites use lines with greater than 25

Location in Protocol	Changes	Rationale
Mode of Administration	forbidden to use investigational drug material for purposes other than as defined in this protocol. VIS410 (2000 mg or 4000 mg) will be administered IV over 2 hours as a single, 200-mL infusion, followed by a 25-mL (or volume equivalent to length of IV line) saline flush to ensure all product is administered, in addition to oseltamivir (Tamiflu) 75 mg BID for total of 5 days (10 doses). Treatment with oseltamivir may be extended for up to 5 additional days (up to 20 doses of total therapy) if clinical symptoms warrant further treatment as assessed by the Investigator. Note: based on SOC, subjects may have started oseltamivir therapy prior to randomization. No dose adjustment is necessary for VIS410 based on renal or hepatic impairment. For patients with renal insufficiency, refer to prescribing information for administration of oseltamivir. VIS410 is a colorless to slightly yellow, clear to opalescent solution, essentially free of particles. VIS410 is formulated at a concentration of 25 mg/mL in 40-mM citrate-sodium phosphate, 150-mM sodium chloride, and 0.025% polysorbate 80. The investigational product will be provided by the Sponsor. VIS410 will be supplied in Type I 20-mL glass vials containing a nominal 20-mL solution. A copy of the certificate of analysis of the investigational product will be sent to the clinical center. The investigational drug product is manufactured by Lyophilization Services of New England in accordance with Good Manufacturing Practice as required by the current Good Clinical Practice (GCP). Manufacturing, packaging, and labeling of the investigational product, VIS410, is conducted under the responsibility of the Sponsor. The study drug will be labeled according to local law and regulatory requirements. Specific dilution procedures for VIS410 will be described in detail in the Pharmacy Manual. Placebo will be a normal saline solution (0.9%) and will be prepared by the pharmacist.	mL hold-up volume
Section 7.2 *Reference Product, Dose, Mode of Administration*	Placebo (normal saline solution 0.9%) will be administered IV over 2 hours as a single 200-mL infusion, followed by a 25-mL (or volume equivalent to length of IV line) saline flush to ensure all product is administered, in addition to oseltamivir (Tamiflu) 75 mg BID for total of 5 days (10 doses). Treatment with oseltamivir may be extended for up to 5 additional days (up to 20 doses of total therapy) if clinical symptoms warrant further treatment as assessed by the Investigator. For patients with renal insufficiency, refer to prescribing information for administration of oseltamivir. Note: based on SOC, subjects may have started oseltamivir therapy prior to randomization.	Clarification as some sites use lines with greater than 25 mL hold-up volume
Section 7.3 *Instructions for Preparation, Use and Administration*	The required amount of VIS410 to be dosed (2000 mg or 4000 mg) will be diluted with normal saline up to a total volume of 200 mL. For placebo subjects, 200 mL of normal saline will be prepared. This infusion will be followed by a 25-mL ~~saline flush. Length of~~ (or if IV line ~~will be set for maximum~~ volume ~~of~~ is >25 mL, ~~so that The 25 mL~~ an equivalent volume) saline flush. The saline flush following	Clarification to ensure all study product administered. Some sites use infusion lines with hold-up volumes of greater than 25 mL.

Location in Protocol	Changes	Rationale
	administration will ensure all VIS410/placebo has been administered. The study infusion will be administered IV using a 0.22-µm in-line filter and will be controlled by a volumetric pump. Standard, ~~uniform length~~ infusion lines will be used, and microfilters will be provided by the Sponsor as appropriate. The infusion bag will be covered with an opaque sleeve in the pharmacy to maintain the study blind. The VIS410/placebo will be administered IV at a rate of 100 mL/h, over 2 hours. When 200 mL has been administered, the line will be flushed with ~~25 mL of~~ normal saline. The infusion time may be longer at the Investigator's discretion based only on local infusion site–related symptoms up to a maximum of 4 hours. Refer to the Pharmacy Manual for directions on storage, handling, stability data, preparation, and use. For more detailed information on VIS410, refer to the current Investigator's Brochure.**Error! Bookmark not defined.**	
Section 8.3.1 *Sequence of Assessments at a Single Visit*	If the following assessments are to be performed at the same study visit, ~~then the order of assessments should be as follows~~ the FluPRO Questionnaire should be completed first, when possible: • FluPRO Questionnaire • ECG • Blood sampling • Nasopharyngeal swab	Clarification
Section 9.2 *Grading of Adverse Event Intensity*	Each AE must be graded on a ~~4~~-point scale (Grades 1-~~3~~ 4) of increasing intensity according to the Division of Microbiology and Infectious Diseases (DMID) Adult Toxicity Table presented in Appendix **Error! Reference ource not found.**. Criteria in the DMID table are generally grouped by body system, ie, hematology, chemistries, enzymes, urinalysis, cardiovascular, respiratory, GI, neurological, musculoskeletal, skin, and systemic. For abnormalities not specifically listed in the DMID Table (Appendix **Error! Reference source not found.**), a guide or estimating severity grade (mild, moderate, severe, life-threatening) is provided: • Grade 1: Mild transient or mild discomfort (< 48 hours); no medical intervention/therapy required • Grade 2: Moderate/mild to moderate limitation in activity—some assistance may be needed; no or minimal medical intervention/therapy required • Grade 3: Severe/marked limitation in activity, some assistance usually required; medical intervention/therapy required, hospitalizations possible • Grade 4: Life-threatening: Extreme limitation in activity, significant assistance required; significant medical intervention/therapy required, hospitalization or hospice care probable **Note:** The semi-colon within the description of the grade indicates 'and'. Any clinical event deemed by the clinician to be serious or	Correction; DMID scale is 1-4

Location in Protocol	Changes	Rationale
	life-threatening should be considered an SAE.	
Section 10.1 *Determination of Sample Size*	The study is exploratory in nature, and is not powered to demonstrate significant differences between treatment groups in primary or secondary outcome measures. The protocol intent is to collect sufficient information to identify the most appropriate candidate endpoints for subsequent Phase 3 study evaluation from among the primary and secondary endpoints described below. Statistical significance testing will therefore be used to assess the relative strength of evidence of the primary and secondary endpoints, to provide reasonable assurance that the endpoints chosen for a confirmatory Phase 3 trial will elucidate treatment differences between VIS410 plus oseltamivir versus oseltamivir alone. ~~The primary efficacy objective of this study is to evaluate the effect of VIS410 + oseltamivir on the time to normalization of respiratory function compared to oseltamivir alone in the modified intent-to-treat (MITT) population.~~ ~~Using a log-rank test, a sample size of 130 influenza A-infected subjects per treatment group will provide 80% power to detect a 1.5-day difference (5 days for oseltamivir alone and 3.5 days for VIS410 plus oseltamivir) in the median time to cessation of O_2 support for VIS410 relative to placebo. A two-sided alpha of 0.05 was used for the calculation.~~ ~~Approximately 390 evaluable subjects with confirmed influenza A infection will be enrolled in this study.~~	New paragraph due to decreased sample size
Section 10.2 *Randomization and Blinding*	An IWRS will be used to allocate the randomized treatments to subjects with stratification by presence or absence of positive pressure ventilation (PPV) at baseline. Pharmacists must obtain the status of the subject relative to PPV at baseline via query, and this data must be input into the IWRS prior to randomization. The randomized treatment assignment will be transferred electronically for integration with the clinical study data at the appropriate time. PPV includes any respiratory assistance using a mechanical ventilation device and can be either invasive or noninvasive ventilation. Invasive PPV includes intubation with endotracheal tube with mechanical ventilation (most common) or tracheostomy with mechanical ventilation. Noninvasive PPV includes use of facemask, nasal plugs, or nasal mask with mechanical ventilation. The devices are named according to the type of mechanical ventilation given continuous positive airway pressure (CPAP) or bi-level positive airway pressure (BiPAP). Randomization will be used to avoid bias in the assignment of subjects to treatment, to increase the likelihood that known and unknown subject attributes (eg, demographic and baseline characteristics) are evenly balanced across treatment arms. The maintenance of the study blind is critical for an unbiased assessment of the safety and efficacy of the study drug. All study staff that evaluate subjects and render decisions regarding subject care will remain blinded to the study treatment each subject receives.	Explanation

Location in Protocol	Changes	Rationale
	The designated study site pharmacist or designee will remain unblinded. Also, unblinded clinical research associates will handle study drug accountability. The study will be conducted in a double-blind manner. The CRO, site study personnel, and Visterra personnel will not be aware of which treatment (VIS410 or placebo) the subjects have been given. Subjects will not be aware of which treatment they have been administered. Subjects will be randomized 1:1:1 to receive VIS410 2000 mg, VIS410 4000 mg, or placebo over a 2-hour infusion with pretreatment regimen of PO or IV diphenhydramine 50 mg plus either ibuprofen 400 mg PO or acetylsalicylic acid 320-325 mg PO approximately 60 minutes before IV infusion of VIS410/placebo. Allocation of each subject to a given treatment sequence will be described in a randomization schedule prepared by Visterra or designee. The randomization will be balanced using randomly permuted blocks across the treatment groups. Based on this randomization code, the site's unblinded pharmacist or trained designee will prepare and dispense VIS410/placebo. ~~If the interim analysis determines that one of the VIS410 treatment arms can be terminated early for futility, then a second randomization schedule with a unique set of randomization numbers will be created for all subsequent subject enrollment and randomization. As with the initial randomization procedure, randomization will be stratified by presence or absence of PPV at baseline to ensure an even distribution across treatment arms.~~ The randomization schedule will not be available to the subjects, Investigators, blinded monitors, or employees of the clinical center involved in the management of the study before unblinding of the data, unless in case of emergency. The Sponsor's clinical team will also be blinded during the study, as they will not have direct access to the randomization schedule. ~~The Sponsor medical representative(s) may be partially unblinded during the interim analyses. A separate interim analysis plan will provide details on access to unblinded data and the level of unblinding.~~ The CRO personnel performing data management and statistical activities will receive a copy of the randomization schedule during database lock. Other team members will not have access to any data that could lead to unblinding. Unblinding of the individual subject's treatment (via IWRS) by the Investigator should be limited to medical emergencies or urgent clinical situations in which knowledge of the subject's study treatment is necessary for clinical management. In such cases, the Investigator is encouraged to contact the Medical Monitor to discuss and agree to the need for unblinding to occur. In situations in which the Investigator has tried, but is unable to reach the Medical Monitor, they should use their best judgment, based on the nature and urgency of the clinical situation, and may proceed with unblinding without having successfully reached and	Due to decrease in sample size an interim analysis is not going to be performed.

Location in Protocol	Changes	Rationale
	discussed the situation with the Medical Monitor. Once a subject's treatment assignment has been unblinded, the Medical Monitor should be notified within 24 hours of unblinding of the treatment, without revealing the study treatment. Information relating to unblinding (eg, reason and date) shall be clearly recorded in the subject's study file, as part of relevant standard operating procedures. In addition, the Investigator should consider whether the clinical event prompting unblinding should be considered an SAE, according to the regulatory definitions or criteria for SAEs, and if so, submit an SAE report to Visterra or designee. If the code is broken by the Investigator or by someone of his/her staff, the subject must be withdrawn from the study and must be followed as appropriate. Visterra or designee will also unblind any SAE reports that are serious, unexpected, and considered to be related to investigational product, in accordance with safety reporting guidance and regulations. If the code is broken by the Sponsor for safety reporting purposes, the subject may remain in the study. To maintain study blinding, the infusion bag will be covered by an opaque sleeve in the pharmacy.	
Section 10.5 *Handling of Missing Data*	~~No imputations~~ Procedures for handling missing data will be ~~performed~~ defined in the Statistical Analysis Plan, and may include last observation carried forward or an interpolation method.	Clarification
Section 10.6 *Interim Analysis*	~~An interim analysis of efficacy may be performed by an unblinded third party after 50% of subjects have been enrolled (n = 195) and have an assessment of the primary endpoint (time to normalization of respiratory function). This interim analysis may be conducted to assess if one of the active VIS410 treatment arms will need to be terminated for futility. Using the methods of Lan and DeMets an alpha spending function will be created and if the test statistic for one of the active VIS410 treatment arms is less than the lower critical point then the VIS410 treatment arm may be terminated for futility. The interim analysis may also test the primary efficacy objective to assess if the primary efficacy endpoint has been met early.~~ ~~The effect size for this study was estimated without the benefit of any prior randomized, controlled trial. If the VIS410 effect size proves to be greater than anticipated, then it is possible that the primary study endpoint will be met by the time the interim efficacy analysis is performed. In the event the interim analysis reveals that the primary efficacy endpoint has been satisfied, this Phase 2 study could be terminated for efficacy.~~ ~~The DSMB and the unblinded statistician will review the interim analysis results to provide their recommendation on discontinuation of one of the VIS410 treatment arms, if needed, based on pre-specified criteria as outlined in the SAP. The interim analysis may also review the following key secondary endpoints, when available, to assist in the decision of discontinuing a VIS410 treatment arm:~~ ~~• Time to clinical resolution of vital signs~~ ~~• Total number of hours on oxygen support or PPV~~	Due to decrease in sample size an interim analysis is not going to be performed.

Location in Protocol	Changes	Rationale
	~~• Viral load from nasopharyngeal swabs~~ ~~A sample size re-estimation (SSRE) will also be performed at this point in order to determine if the current sample size is sufficient based on the observed effect size in the study. The unblinded statisticians will work directly with the Sponsor on the SSRE. The details of SSRE and individuals with access to the information will be outlined in a separate plan. The MITT population will be used for the interim analysis.~~	
Section 10.7 *Efficacy Analyses*	Section number changed to 10.6 **Efficacy Analyses** Efficacy analyses will be performed using the MITT population. Efficacy analysis will also be performed in the PP population to demonstrate consistency with the primary analysis population. **Day 7 Ordinal Scale Status** The ordinal scale outcomes will be measured daily from Day 1 (baseline) through Day 14 using the seven-level hierarchical scale with the classifications presented in Table 8~~Table 2~~. The clinical outcomes therein are ordered from the worst clinical outcome to the best clinical outcome in descending order. For each day, subjects will be classified by the worst clinical outcome for which they qualify. The number and percentage of subjects in each treatment group with each classification will be summarized for each day from Day 1 through Day 14, inclusive. **Table 1. Hierarchical Seven-Level Ordinal Scale for Clinical Outcomes** **Clinical Parameter** Death ICU stay with mechanical ventilation ICU stay without mechanical ventilation Non-ICU hospitalization with supplemental oxygen Non-ICU hospitalization without supplemental oxygen Discharge with partial resumption of normal activities Discharge with full resumption of normal activities **Note:** Clinical outcomes listed from worst clinical outcome to best clinical outcome in descending order. **Time to Cessation of O_2 Support** Time to cessation of O_2 support resulting in a stable SpO_2will be analyzed using a Cox model. Time from onset of symptoms to VIS410 treatment and the number of doses of oseltamivir (Tamiflu) prior to VIS410 treatment for the current case of influenza will be included as a covariate in the analysis. A *P*-value (using the Wald statistic) for each VIS410 dose vs placebo will be presented. Secondarily, subgroup analyses will be performed to describe the time to cessation of O_2 for subgroups, such as use of positive pressure ventilation, use of endotracheal intubation, time from onset of symptom to VIS410 therapy,	Renumbered due to deletion of previous section 10.6 Moved this section to the top of the efficacy analyses as a component of the ordinal scale is now the primary objective/primary endpoint

Location in Protocol	Changes	Rationale
	number of oseltamivir doses prior to VIS410, elderly, and underlying lung disease. Clinical and virologic endpoints will also be evaluated by influenza A subtypes. **Time to Clinical Response** Time to clinical response is defined as resolution of 4 of 5 vital signs that will be determined upon physical examination. Clinical response is defined as: • Afebrile with core temperature ≤ 37.8°C, without use of antipyretics (oral ≤ 37.2°C) • Respiratory rate ≤ 24 beats per minute • Oxygen saturation ≥ 95% on room air without support or a return to pre-infection status, if pre-infection status was < 95% • Pulse rate ≤ 100/min • SBP ≥ 90 mm/Hg, without vasopressor use The probability of time to clinical response (defined as resolution of vital signs) will be calculated via Kaplan-Meier. A ~~P-value~~ significance test (using the log-rank test) for each VIS410 dose vs placebo will be presented. The number and percentage of subjects in each treatment group with clinical response will be summarized. Results will be tabulated and presented graphically as well. **Time to Cessation of Ventilator Support** The probability of time to cessation of ventilator support will be calculated via Kaplan-Meier. A P-value (using the log-rank test) for each VIS410 dose vs placebo will be presented. Results will be tabulated and presented graphically as well. **Healthcare Resource Utilization** Descriptive statistics will be used to compare the total number of days in the hospital and/or ICU from admission to discharge, number of subjects requiring ICU admission post-randomization, overall number of days in the ICU, number of hours on ventilation, rehospitalization due to influenza A relapse/reinfection, the total number of days of oseltamivir therapy, and the total number of days to resumption of usual activities by treatment group. Time (number of days) to resumption of usual activities will be determined from the VAS (scale ranged from 0 to 10, where 0 indicates subject is unable to perform any of his/her usual activities prior to influenza onset, and 10 indicates subject is able to fully perform all usual activities). This evaluation will be performed using the MITT population. **Signs and Symptoms of Influenza** Descriptive statistics will be used to compare the duration of symptoms of influenza-like illness in the subset of subjects able to complete the FluPRO Questionnaire at baseline and post-dose by treatment group. The number and percentage of subjects who were not able to complete the assessment at each visit will be summarized. Frequency tabulation of the occurrence and severity of each subject-reported symptom of influenza-like illness (via FluPRO Questionnaire) will be summarized by assessment	

Location in Protocol	Changes	Rationale
	time point and by treatment group. Time to resolution of symptoms will be evaluated by Kaplan Meier analysis. Analyses of the signs and symptoms of influenza will be conducted only on the subset of the MITT population who had baseline FluPRO Questionnaire assessments. Additional populations may be analyzed as described in the SAP. ~~**Clinical Outcome by Seven-Level Ordinal Scale**~~ ~~The ordinal scale outcomes will be measured daily from Day 1 (baseline) through Day 14 using the seven-level hierarchical scale with the classifications presented in **Table 2**. The clinical outcomes therein are ordered from the worst clinical outcome to the best clinical outcome in descending order. For each day, subjects will be classified by the worst clinical outcome for which they qualify.~~ ~~The number and percentage of subjects in each treatment group with each classification will be summarized for each day from Day 1 through Day 14, inclusive.~~ ~~**Table 2. Hierarchical Seven-Level Ordinal Scale for Clinical Outcomes**~~ ~~**Clinical Parameter**~~ ~~Death~~ ~~ICU stay with mechanical ventilation~~ ~~ICU stay without mechanical ventilation~~ ~~Non-ICU hospitalization with supplemental oxygen~~ ~~Non-ICU hospitalization without supplemental oxygen~~ ~~Discharge with partial resumption of normal activities~~ ~~Discharge with full resumption of normal activities~~ ~~**Note:** Clinical outcomes listed from worst clinical outcome to best clinical outcome in descending order.~~	Moved this section to the top of the efficacy analyses as a component of the ordinal scale is now the primary objective/primary endpoint.
Section 10.8.3 *Complications of Influenza*	Now Section 10.7.3 Complications of influenza are defined as pneumonia, myocarditis, worsening of chronic bronchitis, sinusitis, otitis or other chronic pulmonary diseases, encephalopathy, and death. Complications of influenza and use of antibiotic therapy for bacterial pneumonia will be reported by variable, treatment group, and time point. Diagnosis of bacterial pneumonia will be based on local clinical practice, usually an assessment of clinical features and any radiographic evidence of bacterial pneumonia (eg, infiltrate) and/or microbiologic evidence of bacterial pneumonia. All-cause and attributable mortality will be evaluated on Days 14 and 28.	Clarification
Sections 10.8 - 10.12	Renumbered sections due to deletion of original section 10.6; now 10.7 - 10.11	Correction
Section 12.1.2 *Subject Informed Consent*	Each subject or a legally ~~authorized~~ acceptable representative must give written consent according to local requirements after the nature of the study has been fully explained. The consent form must be signed before performance of any study-related activity. The consent form that is used must be approved by both the Sponsor and by the reviewing IEC/IRB. The informed consent should be in	Change requested by an Ethics Committee

Location in Protocol	Changes	Rationale
	accordance with principles that originated in the Declaration of Helsinki, current ICH and GCP guidelines, applicable regulatory requirements, and Sponsor policy. Before enrolling potential subjects in the study, the Investigator or an authorized member of the investigational staff must explain to the subject the aims, methods, reasonably anticipated benefits, and potential hazards of the study, and any discomfort that participation in the study may entail. Subjects will be told that the Investigator will maintain a subject identification register for the purposes of long-term follow up if needed and that their records may be accessed by health authorities and authorized Sponsor staff without violating the confidentiality of the subject, to the extent permitted by the applicable law(s) or regulations. By signing the Informed Consent Form (ICF) the subject is authorizing such access and agrees to allow his/her study physician to recontact the subject for the purpose of obtaining consent for additional safety evaluations, if needed. The language used in the oral and written information about the study, including the ICF, should be nontechnical and practical and should be understandable to the subject or the subject's legal representative. The subject will be given sufficient time to read the ICF and the opportunity to ask questions. After this explanation and before entry into the study, consent should be appropriately recorded by means of the subject's personally dated signature. After having obtained the consent, a copy of the ICF must be given to the subject. If the subject is unable to read or write, an impartial witness should be present for the entire informed consent process (which includes reading and explaining all written information) and should personally date and sign the ICF after the oral consent of the subject is obtained, if permitted by local law.	
Section 14.2 *Laboratory Assessments*	**Other Assessments** Urine pregnancy test Erythrocyte Sedimentation Rate (ESR) C-Reactive Protein (CRP)	Added to complete the assays being performed

[i] Treanor JJ, Hayden FG, Vrooman PS, et al. Efficacy and safety of the oral neuraminidase inhibitor oseltamivir in treating acute influenza: a randomized controlled trial. US Oral Neuraminidase Study Group. *JAMA* 2000;283(8):1016–24.
[ii] Schanzer DL, Langley JM, Tam TW. Co-morbidities associated with influenza attributed mortality, 1994-2000, Canada. *Vaccine* 2008;26(36):4697–703.
[iii] CDC. Estimates of deaths associated with seasonal influenza US, 1976-2007. MMWR. 2012;59(33):1057–62.
[iv] WHO. Seasonal Influenza Fact sheet N°211. 2014. http://www.who.int/mediacentre/factsheets/fs211/en. Accessed 10 Mar 2014.
[v] World Health Organization (WHO). Pandemic (H1N1) 2009-update 70. http://www.who.int/csr/don/2009_10_16/en/index.html. Accessed 25 October 2009.

Visterra

PROTOCOL AMENDMENT – SUMMARY OF CHANGES

Phase 2b, Multicenter, Randomized, Double-blind, Controlled Study to Evaluate the Efficacy and Safety of Intravenous VIS410 in Addition to Oseltamivir (Tamiflu®) Compared With Oseltamivir Alone in Hospitalized Adults With Influenza A Infection Requiring Oxygen Support

Product	VIS410
Protocol Number	VIS410-203
EudraCT Number	2016-004009-15
Clinical Phase	2b
Clinical Indication	Influenza A infection

Sponsor	Visterra, Inc. One Kendall Square, Suite B3301 Cambridge, MA 02139 United States of America
Sponsor Representative	[redacted]

ORIGINAL PROTOCOL DATE: **21 October 2016**

AMENDMENT 1 DATE: **20 June 2017**

SUMMARY OF CHANGES TO PROTOCOL VIS410-203 FROM VERSION 1.0 TO VERSION 2.0

Substantive changes to the protocol for Amendment 1 and their location within the protocol are noted below. Additions are marked as red underlined text and deletions are marked as red strikethrough text. Administrative, stylistic and formatting changes that do not alter the conduct of the study are not summarized in this document.

<table>
<tr><th>Location in Protocol</th><th>Changes</th><th>Rationale</th></tr>
<tr><td>Cover Page;
Signature of Sponsor Representative</td><td>Issue Date (Version): ~~21 October 2016~~ (Version ~~1~~.0)
20 June 2017 (Version 2.0)

Sponsor Representative [redacted]</td><td>Updated date and version number.

Sponsor representative updated.</td></tr>
<tr><td>Protocol History</td><td>~~Protocol History~~
~~Visterra, Inc. VIS410-203~~<table><tr><th>~~Document~~</th><th>~~Issue Date~~</th><th>~~Amendment Type~~</th><th>~~Comments~~</th></tr><tr><td>~~Clinical Study Protocol~~</td><td>~~21 October 2016 (Version 1)~~</td><td>~~-~~</td><td>~~This document~~</td></tr></table></td><td>Deleted because changes will be summarized in a separate Summary of Changes document (this file).</td></tr>
<tr><td>Synopsis and Section 2.2 Secondary Objectives</td><td>• Evaluate the effect of VIS410 + oseltamivir vs oseltamivir alone on the following endpoints:
▪ Viral ~~titer~~ load in upper respiratory samples
▪ Time to clinical response
▪ Time to cessation of ventilator support
▪ Time to resumption of normal activities
▪ All-cause and attributable 14- and 28-day mortality
▪ Healthcare resource utilization
▪ Time to alleviation of clinical symptoms of influenza in subset of subjects able to complete the FluPRO Questionnaire at baseline and post-dose
▪ Proportion of subjects with new documented bacterial pneumonia/superinfection
▪ Proportion of subjects with influenza-related complications</td><td>Viral load is a more accurate term to represent the test being conducted.</td></tr>
<tr><td>Synopsis;
Section 2.3 Exploratory Objectives;
Section 3.3 Exploratory Endpoints;
Section 6.3.12.3 Serum Cytokines</td><td>• Evaluate the effect of VIS410 + oseltamivir vs oseltamivir alone on clinical outcomes as measured by a seven-level ordinal scale
• Evaluate the pharmacokinetics of VIS410 from nasopharyngeal secretions and tracheal aspirate (ventilated subjects only)
• Assess the effects of VIS410 on viral ~~titer~~ load in tracheal aspirate (ventilated subjects only)
• Assess correlations between virology, safety, VIS410 dose, pharmacokinetics, viral shedding, ~~immunogenicity~~ immunology, signs and symptoms of influenza, and other endpoints
• Assess the anti-influenza immune response
• ~~Evaluate serum cytokine profiles~~</td><td>Viral load and immunology are more accurate terms.

Exploratory cytokine objective removed from protocol.</td></tr>
</table>

Location in Protocol	Changes	Rationale
Synopsis; Section 4.0 *Study Design;* Section 4.3 *Data Safety Monitoring Board;* Section 6.1.3 *Study Procedures;* Section 7.5 *Pretreatment;* Section 8.3.3 *Post-Treatment Procedures (Day 3 to 56);* Section 11.4 *Stopping Rules or Discontinuation Criteria*	This is a Phase 2b multicenter, randomized, double-blind, controlled study comparing the efficacy and safety of 2 dose levels of VIS410 (2000 and 4000 mg) in combination with oseltamivir vs oseltamivir alone in hospitalized ~~severely ill~~ subjects with influenza A infection requiring oxygen. Subjects will be randomly assigned at a ratio of 1:1:1 using a permuted block randomization method to receive VIS410 at a dose of 2000 mg (low dose) or 4000 mg (high dose) or placebo (0.9% sodium chloride), administered as a single IV infusion over 2 hours. Randomization will be stratified by presence or absence of positive pressure ventilation (PPV) at baseline to ensure an even distribution across treatment arms (see Section 10.2). All subjects will receive oseltamivir 75 mg twice daily (BID) for 5 days (total of 10 doses) as part of standard of care (SOC). Treatment with oseltamivir may be extended for up to 5 additional days (up to 20 doses of total therapy) if clinical symptoms warrant further treatment as assessed by the Investigator. Note: based on SOC, subjects may have started oseltamivir therapy prior to randomization. All subjects will also receive a pretreatment regimen of diphenhydramine 50 mg PO or IV plus either ibuprofen 400 mg PO or acetylsalicylic acid 320-325 mg PO 60 minutes before VIS410/placebo infusion. Approximately 390 evaluable subjects (130/arm) with confirmed influenza A infection will be treated. Subjects admitted to the hospital within 5 days of onset of initial symptoms who require supplemental oxygen ~~of at least 40% (4 L/min) and/or hypoxemia defined as SpO_2 of less than 90%~~ will undergo a rapid influenza test (supplied by the Sponsor) or a PCR test, fluorescent immunoassay (FIA) test, or enzyme-linked immunosorbent assay (ELISA) test to confirm influenza A infection. Subjects diagnosed with influenza A will undergo the screening procedures to confirm eligibility. Study assessments are outlined in Table 1, Schedule of Assessments. Eligible subjects will be randomized to receive either oseltamivir + VIS410 2000 mg, oseltamivir + VIS410 4000 mg, or oseltamivir + placebo (0.9% sodium chloride) at a ratio of 1:1:1 using a permuted block randomization method summarized in Table 6. **(Randomization table, unchanged)** Oseltamivir (Tamiflu®) will be provided by the Sponsor. Treatment with oseltamivir may be extended for up to 5 additional days (up to 20 doses of total therapy) if clinical symptoms ~~are not resolved by Day 5 or~~ warrant further treatment as assessed by the Investigator. Note: based on SOC, subjects may have started oseltamivir therapy prior to randomization. Study assessments are outlined in Table 1. Subjects discharged from ~~will be monitored daily while in~~ the hospital prior ~~up~~ to Day 14 will attend ~~(± 3 days) with additional visits on Day 28 (± 3 days) and~~ the applicable outpatient ~~last~~ follow-up visit(s) (eg, Day 3, Day 5, ~~on~~ Day~~56 (±~~ 7~~days). Study assessments are outlined in~~, Day 14) per Table 1 ~~(Schedule of Assessments)~~.	Based on change to Inclusion Criteria regarding oxygen requirement, it is more accurate to define subjects as hospitalized, not severely ill. Clarified that subjects will receive either ibuprofen or acetylsalicylic acid, not both. Dose for acetylsalicylic acid changed to comply with standard dose in various countries. Inclusion Criteria regarding oxygen requirement has been modified to allow participation of less severe infections, hence subjects no longer require a specific concentration or amount of oxygen support to be eligible for the study. Clarified that subjects may have started oseltamivir treatment prior to enrollment in the study; total minimum and maximum doses the same for all subjects regardless of when a subject began oseltamivir. Clarified that subjects discharged from the hospital will attend follow-up visits; the number of outpatient follow-up visits are dependent on when the subject is discharged from the hospital. Defined GI TEAEs that require intervention. Updated statistical language related to the error rate and the interim analysis. The DSMB will review safety data after

Location in Protocol	Changes	Rationale
	An independent data safety monitoring board (DSMB) will be established to review all available safety data after 30 subjects as well as when ~~and~~ approximately 120 subjects have completed study Day 14 ~~7~~. The assessment of safety will be determined from vital sign measurements, physical examinations, hematology, chemistry and urinalysis laboratory testing, 12-lead electrocardiograms (ECGs), use of concomitant medications, and review of adverse events (AEs). Based on the safety data provided, the DSMB will determine if it is acceptable to continue the study or reduce the dose as appropriate. Dosing will temporarily pause while the DSMB meets if there are 4 treatment-related serious adverse events (SAEs) or 4 severe gastrointestinal (GI) treatment–emergent adverse events (TEAEs) that require intervention, which is defined as requiring IV fluid and medication to decrease the frequency of diarrhea (ie, loperamide). Additional DSMB reviews can occur throughout the trial as deemed necessary by the DSMB or Sponsor to ensure subject safety and well-being. Study enrollment and dosing will continue while the DSMB evaluates data. In addition, following 50% enrollment (195 subjects), an interim analysis may ~~will~~ be conducted, by an unblinded third party, to assess if one of the VIS410 treatment arms can be terminated early for futility. A prespecified sample size reanalysis may also be conducted based on the observed effect size. The interim analysis may ~~will~~ also test the primary efficacy objective to assess if the primary efficacy endpoint has been met. An alpha spending function will be designed in order to determine the amount of alpha that will be spent at the interim analysis. The effect size for this study was estimated without the benefit of any prior randomized controlled trial. If the VIS410 effect size proves to be greater than anticipated, then it is possible that the primary study endpoint will be met by the time the interim efficacy analysis is performed. In the event the interim analysis reveals that the primary efficacy endpoint has been satisfied, this Phase 2 study could be terminated for efficacy.	approximately 30 subjects and 120 subjects complete Day 14 to allow more safety data to be reviewed.
Synopsis and Section 5.2.1 *Inclusion Criteria*	**Only revised list items shown:** 4. Requirement for oxygen support including any positive pressure ventilation (PPV). ~~4. Need for supplemental oxygen of at least 40% (4 L/min) and/or hypoxemia defined as SpO2 of less than 90%~~	Inclusion Criteria regarding oxygen requirement has been modified to allow participation of less severe infections, hence subjects no longer require a specific concentration or amount of oxygen support to be eligible for the study.
Synopsis and Section 5.2.2 *Exclusion Criteria*	**Only revised list items shown:** 2. Subjects who have received VIS410 in the past ~~History of receiving monoclonal antibody products (including VIS410) within 3 months prior to VIS410/placebo dosing or planned administration during the study period~~. 3. Subjects who have a h~~i~~story of receiving monoclonal antibody products within 3 months prior to VIS410/placebo dosing or planned administration of	Clarified that subjects are not eligible for the study if they have received VIS410 in the past. Clarified exclusion criteria of subjects that have Influenza B or another viral

Location in Protocol	Changes	Rationale
	another monoclonal antibody during the study period. 5. Subjects with known co-infection with influenza B or other viral respiratory infections (eg, respiratory syncytial virus [RSV], parainfluenza viruses, respiratory adenoviruses).~~infection~~ 6. Subjects with lung transplant or history of severe chronic lung disease, including cystic fibrosis or any condition requiring ~~> 2 L/minute of~~ home oxygen therapy.~~(ie, severe chronic obstructive pulmonary disease [COPD], pulmonary fibrosis)~~ 7. Subjects on extracorporeal membrane oxygenation ~~(ECMO)~~ at time of randomization. ~~8. Hospitalization for > 48 hours prior to randomization~~ 8. Subjects with end-stage renal disease (ESRD) who are not undergoing hemodialysis.	respiratory infection. Clarified exclusion criteria of cystic fibroses subjects; due to changes in patient population , subjects that require oxygen therapy at home are not eligible for the study. Subjects hospitalized before enrollment in the study are eligible as long as the onset of influenza symptoms are no more than 5 days before treatment with VIS410/ placebo (Inclusion Criteria 3). For consistency with oseltamivir prescribing information, subjects with ESRD who are not undergoing hemodialysis are excluded from the study.
Synopsis; Section 7.1 *Test Product, Dose, Mode of Administration;* Section 7.2 *Reference Product, Dose, Mode of Administration*	**¶ 2 only:** VIS410 (2000 mg or 4000 mg) will be administered IV over 2 hours as a single, 200-mL infusion, followed by a 25-mL saline flush to ensure all product is administered, in addition to oseltamivir (Tamiflu) 75 mg BID for total of 5 days (10 doses). Treatment with oseltamivir may be extended for up to 5 additional days (up to 20 doses of total therapy) if clinical symptoms ~~are not resolved by Day 5 and/or~~ warrant further treatment as assessed by the Investigator. Note: based on SOC, subjects may have started oseltamivir therapy prior to randomization. No dose adjustment is necessary for VIS410 based on renal or hepatic impairment. For patients with renal insufficiency, r~~R~~efer to prescribing information for administration of oseltamivir ~~in patients with renal insufficiency~~.	Clarified that subjects are required to be treated with oseltamivir for a minimum of 5 days but that treatment may be extended up to 10 days. Clarified that subjects may have started oseltamivir treatment prior to enrollment in the study; the total minimum and maximum doses are the same for all subjects regardless of when a subject began oseltamivir.
Synopsis and Section 7.3 *Instructions for Preparation, Use, and Administration*	The infusion bag will be covered with an opaque sleeve in the pharmacy to maintain the study blind. The VIS410/placebo will be administered IV at a rate of 100 mL/h, over 2 hours. After 200 mL of the diluted dose has been administered, the infusion will be stopped, followed by a 25-mL saline flush. The infusion time may be extended up to 4 hours at the Investigator's discretion based on local infusion site–related symptoms.~~ VIS410 or placebo will be administered within 48 hours of being prepared by the study pharmacist.~~ When 200 mL has been administered, the line will be flushed with 25 mL of normal saline. The infusion time may be longer at the Investigator's discretion based only on local infusion site–related symptoms up to a maximum of 4 hours.	The window for administration of VIS410/placebo after preparation by the pharmacist has been deleted and will be noted in the Pharmacy Manual.

Location in Protocol	Changes	Rationale
	~~VIS410 or placebo will be administered within 48 hours of being prepared by the study pharmacist. After preparation, the VIS410/placebo infusion bag should be stored at room temperature.~~	
Synopsis and Section 3.1.1 *Primary Efficacy Endpoint*	Time to cessation of O_2 support resulting in ~~a~~ stable SpO_2 by pulse oximetry. Stable SpO_2 is defined as two consecutive SpO_2 values of > 92% on room air that are~~> 95% for~~ at least ~~8~~6 hours apart. ~~on room air, return to baseline respiratory status, or hospital discharge with no need for additional O_2 support, whichever occurs first.~~	Time to cessation of O_2 support redefined based on update to Inclusion Criteria regarding oxygen requirement.
Synopsis and Section 3.2 *Secondary Endpoints*	**Only revised bullet with its sub-bullets shown:** • Time to clinical response defined as resolution of at least 4 of 5 vital signs ▪ Afebrile with core temperature ≤ 37.8°C, without use of antipyretics (oral ≤ 37.2°C) ▪ Oxygen saturation ≥~~>~~ 95% on room air without support or a return to pre-infection status, if pre-infection status was < 95% ▪ Pulse rate ≤ 100/min ▪ Systolic blood pressure ~~(SBP)~~ ≥ 90 mm/Hg, without vasopressor use ▪ Respiratory rate ≤~~<~~ 24 beats per minute	Specified that temperature endpoint specific to core temperature; also defined equivalent oral temperature. Updated definition of time to clinical response for oxygen saturation and respiratory rate.
Section 6.1.1 *Prescreening Procedures*	Subjects may be asked to sign a Prescreening Consent prior to undergoing a single nasopharyngeal swab (one nostril) for rapid influenza A testing. Subjects will be given a full explanation of the nature of the study and provide full written informed consent before any study-specific assessments or procedures are performed. Note: subjects that tested positive for influenza A within 48 hours prior to screening do not need to be retested. ~~This screening test is not necessary if the subject has a prior influenza A positive test by RAT or with another commercially available test including PCR, FIA, or ELISA within the prior 48 hours of screening.~~	Clarified that a pre-screening consent to perform rapid influenza testing is optional as this may depend on country and site requirements.
Section 6.2 *Pregnancy Safeguards*	Pregnancy will be determined by evaluation of β-human chorionic gonadotropin in serum or urine for all women of childbearing potential. Subjects who are pregnant or nursing will be excluded from the study. During the course of the study drug administration period within the study, any nursing mother(s) or subject(s) with suspected or confirmed pregnancy will be discontinued from study drug therapy but will be encouraged to undergo follow-up for safety monitoring for themselves (ie, the pregnant female) and the baby. All women of childbearing potential and all male subjects must practice effective contraception from screening until 60 days post-VIS410/placebo infusion. For the purposes of this study, women who do not satisfy at least one of the following criteria listed below (ie, the criteria for defining non-childbearing potential) are considered to be physiologically capable of becoming pregnant and are, therefore, defined as women of childbearing potential. The criteria for defining women as being of non-childbearing potential are:	Clarified that any pregnancy should be reported up until 60 days post VIS410/placebo dosing. Clarified the contraception requirements for women of childbearing potential and all male subjects.

Location in Protocol	Changes	Rationale
	• Post-menopausal: ≥ 12 months of natural (spontaneous) amenorrhea, or • ~~Six weeks after surgical bilateral oophorectomy with or without hysterectomy, or~~ • Follicle stimulating hormone > 40 mIU/mL as documented in their medical history, or • Surgical bilateral oophorectomy with or without hysterectomy, or • Hysterectomy, or • Bilateral tubal ligation Women of childbearing potential and all male subjects participating in heterosexual relations must be willing to practice effective contraception from screening until 60 days post-VIS410/placebo infusion. For the purposes of the study, highly effective contraception is defined as: • Male vasectomy with negative semen analysis documentation at least 6 months prior to dosing. OR • Combination of an established form of hormonal contraception (oral, injected, or implanted) or an intrauterine device or intrauterine system Plus one of the following: ~~And~~ • A physical barrier method of contraception with use of a spermicide, such as condom or occlusive cap (diaphragm or cervical/vault caps) with spermicidal foam/gel/film/cream suppository. The use of barrier contraceptives should always be supplemented with the use of a spermicide, unless not available in a country. OR • Male vasectomy with negative semen analysis documentation less than ~~The use of contraception does not apply if the male partner has been vasectomized at least~~ 6 months prior to dosing. OR • Complete abstinence can be considered an acceptable method of contraception at the discretion of the Investigator. Periodic abstinences (eg, calendar, ovulation, symptothermal, or post-ovulation methods) and withdrawal are NOT considered acceptable methods of contraception. The combination of 2 barrier methods, periodic abstinence (eg, calendar, ovulation, symptothermal, or post-ovulation methods), and withdrawal are not considered acceptable methods of contraception. Pregnancy reporting is described in Section 9.7.3.	
Section 6.3.1 *SpO_2 Oxygen Support*	Baseline SpO_2 on room air to be documented, if available. Once randomized, SpO_2 will be measured using pulse oximetry 3 times per day (approximately every ~~preferably~~ 8 hours ~~apart~~) at approximately the same time each day until stable according to Table 1. Stable ~~the schedule of assessment.~~ SpO_2 is defined as two consecutive SpO_2 values of > 92% ~~will be measured using pulse oximetry until > 95%~~	Clarified time points for obtaining SpO_2 measurements and definition of stable SpO_2.

Location in Protocol	Changes	Rationale
	~~for at least 6 hours~~ on room air that are at least 8 hours apart.	
Synopsis; Section 6.3.2; Section 6.3.11; Section 10.10	**Section subheading titles only** 6.3.2 *Viral Load ~~Titer~~* 6.3.11 Pharmacokinetics and Viral Load ~~Titer~~ in Tracheal Aspirate 10.10 *Viral Load ~~Titer~~*	Section heading only revised.
Section 6.3.4 *Resumption of Normal Activities*	Subjects will complete a visual analog scale (VAS) daily from Day 1 (baseline) until either the subject reports that all usual activities (prior to influenza onset) can be performed or Day 56, whichever comes first. The VAS scale ranges ~~scale ranged~~ from 0 to 10, where 0 indicates subject is unable to perform usual activities at all, and 10 indicates subject is able to perform all usual~~) according to the time points defined in~~ activities fully)	Added language to require a subject to complete VAS from baseline until the subject reports they are able to perform all usual activities they were performing prior to influenza onset.
Section 6.3.8 *Viral Resistance*	Samples for virologic and PK analysis ~~A nasopharyngeal swab~~ will be collected ~~from each nostril~~ as described in Table 1 and the laboratory manual. These samples ~~swabs~~ will be sent to a central virology laboratory for processing into appropriate aliquots for virology and PK analysis, including resistance testing. Further procedures for sample collection, processing, shipment, and storage will be described in the laboratory manual. Note**:** Nasopharyngeal and tracheal samples taken from all subjects may be infectious and will be classified as "diagnostic specimens" for dispatch purposes.	Updated language to more accurately describe virologic analysis which includes PK analysis and resistance testing.
Section 6.3.12 *Immunology ~~Immunogenicity~~*	**¶ 1 only:** The immunology ~~immunogenicity~~ parameters under evaluation are the detection of ADA titers and anti-influenza A antibody titers by ~~hemagglutination inhibition assay (~~HAI~~), and serum cytokine concentrations~~.	Updated language to more accurately describe immunology evaluation. Exploratory cytokine objective removed from protocol.
Section 6.5.3 *Complications of Influenza;* Section 10.8.3 *Complications of Influenza*	Complications of influenza are defined as pneumonia, myocarditis, worsening of chronic bronchitis, sinusitis, otitis, other chronic pulmonary diseases, encephalopathy, and death. Complications of influenza and use of antibiotic therapy for bacterial pneumonia will be reported by variable, treatment group, and time point. Diagnosis of bacterial pneumonia will be based on local clinical practice, usually an assessment of clinical features and any radiographic evidence of bacterial pneumonia (eg, infiltrate) and/or microbiologic evidence of bacterial pneumonia. All-cause and attributable mortality will be evaluated on Day 14 and Day 28.	Clarified that diagnosis of bacterial pneumonia will be based on local clinical practice.
Section 6.5.4 *Clinical Laboratory Tests*	Blood samples will be collected by venipuncture or via indwelling cannula at the time points indicated in Table 1. ~~Standard laboratory tests will be performed by a central or local laboratory.~~ Appendix 14.2 lists the biochemistry, hematology, and coagulation~~, and serology~~ tests that will be performed by the central laboratory on the safety blood samples. Note: subjects to be enrolled based on the Investigator's discretion including any local laboratory results per SOC at institution.	Updated language to state that the required baseline laboratory tests will be sent to the central laboratory for consistency of data for trial analysis. However, for enrollment, Investigators should use local laboratory testing for pregnancy and serum creatinine. Subject's

Location in Protocol	Changes	Rationale
	Creatine kinase-MB, creatinine kinase, and troponin are to be measured by the central laboratory in the event of a subject having chest pain. Subjects with chest pain should be managed per SOC. In the event of a subject exhibiting the signs and symptoms of an anaphylactic reaction, when possible at the time of the event, a 5-ml serum sample should be collected for further assessment (ie, tryptase, chymase) to be measured by the central laboratory. A midstream urine sample will be collected for the central laboratory for urinalysis by dipstick, flow cytometry, and microscopic examination. Appendix 14.2 lists the urinalysis parameters that will be assessed. A serum ~~Serum or urine~~ pregnancy tests will be performed by ~~at~~ the central laboratory for all time points indicated in Table 1. In addition, a negative Pregnancy test (serum or urine) result within 2 days prior to dosing ~~results~~ must be available from the local laboratory prior to randomization. ~~A negative result is required for randomization.~~ For oseltamivir dosing, creatinine clearance (Cockcroft-Gault Equation) must be calculated using the serum creatinine value from the local laboratory prior to randomization. The Investigator must review the laboratory report, document this review, and record any change occurring during the study he/she considers to be clinically relevant in the EDC system. Laboratory values outside the normal range will be flagged, and their clinical relevance will be assessed by the Investigator.	overall health for participation in the trial should be based on institutional practices and local laboratory results. Clarified that laboratory tests required in the case of a subject experiencing chest pain will be sent to the central laboratory for analysis while these subjects should be managed based on institutional practices and Investigator discretion. Clarified that all pregnancy testing will be performed at the central laboratory but that a negative pregnancy test performed by the local laboratory is required within 2 days of dosing. Specified that creatinine clearance should be calculated by using the Cockcroft-Gault equation.
Section 6.5.5 *Vital Signs and Body Temperature*	Vital sign parameters will be assessed after 5 minutes in supine position at the time points indicated in Table 1. The vital sign parameters that will be assessed are supine SBP, DBP, heart rate, and respiratory rate. Any change from baseline in vital sign values occurring during the study that is considered to be clinically relevant by the Investigator should be recorded as an AE ~~adverse event~~. Oral body temperature will be recorded at the end of infusion then BID while in the hospital and then throughout the study according to the study procedures outlined in Table 1. In cases where obtaining oral temperature is not possible, core temperature (eg, tympanic, axillary) will be obtained. While the patient is hospitalized, the maximum temperature should be recorded for each 12-hour interval (from 12 AM to 12 PM and from 12 PM to 12 AM). Fever is defined as a core body temperature ≥ 38°C.	Provided examples of core temperature methods. Clarified that fever is defined based on core temperature.
Section 6.5.8 *Radiological Assessment*	A baseline chest x-ray or CT scan will be performed for the assessment of pneumonia. However, a chest x-ray or CT scan performed as part of routine SOC ~~standard of care~~ within 72 ~~48~~ hours before randomization will be acceptable. The results of any such studies will be recorded in the EDC.	The window has been expanded to allow use of local tests for diagnosis of pneumonia and minimize unnecessary tests for the purpose of the study. The 72-hour window will allow detection of pneumonia at

Location in Protocol	Changes	Rationale
		baseline, if present.
Section 7.4 *The Use of Oseltamivir as Standard of Care*	All subjects will receive oseltamivir 75 mg BID for 5 days as part of ~~standard of care (~~SOC~~).~~ Treatment with oseltamivir may be extended for up to 5 additional days (up to 20 doses of total therapy) if clinical symptoms ~~are not resolved by Day 5 or~~ warrant further treatment as assessed by the Investigator. Oseltamivir (Tamiflu) will be provided by the Sponsor. Note: based on SOC, subjects may have started oseltamivir therapy prior to randomization. For patients unable to swallow, oseltamivir capsules can be opened and mixed with liquid for ease of administration. Dose adjustment is recommended for patients with a serum creatinine clearance of 60 mL/min or less[9] (see Table 7).	Clarified that subjects may have started oseltamivir treatment prior to enrollment in the study. Added guidelines on administration of oseltamivir in subjects unable to swallow in compliance with Tamiflu product information.
Section 7.7 *Treatment Compliance*	**¶ 2 only:** The number of oseltamivir doses administered while the subject is in the hospital will be documented by the Investigator or his/her designee. In case of discharge while on oseltamivir therapy, subjects will document the number of doses taken at home and any missed doses; subjects self-administering oseltamivir at home will bring the pack(s) ~~pill bottle~~ to each study visit so study staff may count and record the number of pills taken. Any unused capsules must be returned to the study site.	Clarified that oseltamivir will be provided in packs, not pill bottles.
Section 8.2.1 *Prohibited Prior Medications*	**¶ 1 only:** Per Exclusion Criterion 3 (Section 5.2.2) subjects are not allowed to receive monoclonal antibody products ~~(including VIS410)~~ within 3 months prior to VIS410/placebo dosing.	Removed incorrect reference to VIS410.
Section 8.2.2 *Prohibited Concomitant Medications*	**¶ 1 only:** Per Exclusion Criterion 2 (Section 5.2.2), subjects will not be allowed to receive additional monoclonal antibody products during the study period. Following randomization, use of other antiviral therapy for treatment of influenza A infection will not be permitted. Excluded antiviral medications include but are not limited to rimantadine, amantadine, peramivir, zanamivir, and laninamivir.	Clarified language since VIS410 is a monoclonal antibody.
Section 8.4 *Subject Diary*	A subject diary will be provided upon discharge from the hospital and will record the following: • Daily oseltamivir dosing to be completed for as long as the subject continues to take oseltamivir • Daily VAS for assessing resumption of usual activities to be completed up until either the subject reports that all pre-influenza usual activities can be performed or Day 56, whichever comes first • Daily Influenza Patient Reported Outcomes (FluPRO) Questionnaire to be completed until Day 14, if applicable	Added specific list of what will be included in the subject diary.
Section 9.1.3 *Adverse Event of Special Interest*	For VIS410, the AESIs include the following: • Abdominal cramping • Diarrhea/Loose stool • Nausea • ~~and~~ vomiting	Updated AESI list to include single episodes of loose stool.

Location in Protocol	Changes	Rationale
	• Pruritus • Rash • Hypotension • Throat tightening • Trouble breathing or wheezing	
Section 9.4 *Action Taken Regarding Investigational Product*	The action taken toward the study drug must be described as one of the following: • Permanently discontinued • Stopped temporarily • Modified infusion rate • No action Taken • Not applicable	Updated to match what is captured in the database if an AE occurs and include options in case AE occurs after end of infusion or did not result in a change with regards to VIS410 infusion.
Section 9.6 *Recording Adverse Events*	**¶ 1 only:** All (S)AEs occurring during the clinical investigation must be documented in the EDC system from the time of dosing with any study treatment through Day 56.	Clarified timeframe when AEs are captured.
Section 9.7.1 *Reporting Serious Adverse Events*	All SAEs, irrespective of the circumstances or suspected cause, must be reported on a Serious Adverse Event Form by the Investigator to the Sponsor or designee within 24 hours of their knowledge of the event, preferably by email. ~~fax. Other means of transmission can be decided when fax is not possible (email).~~ **Contact details for reporting SAEs:** [redacted]	Updated to specify that SAEs should be sent via email as preferred method and included additional contact numbers for reporting.
Section 9.7.3 *Reporting a Pregnancy*	Subjects should not become pregnant during the study. Pregnancy is not an AE; however, the Investigator must report any pregnancy which occurs in a female subject or the female partner of a male subject up to 60 ~~56~~ days after last dose of VIS410/placebo by emailing ~~faxing~~ the pregnancy notification form to the Sponsor or designee within 24 hours of the study site staff becoming aware of the pregnancy. The Investigator will also follow up with the subject to determine the outcome of the pregnancy and any fetal or neonatal sequelae will be obtained and documented. The Investigator or study site staff must report the outcome of the pregnancy to the Sponsor or designee. **Contact details for reporting Pregnancy:** [redacted]	Changed pregnancy reporting to 60 days for consistency with prior studies. Updated to specify that pregnancy should be reported on the pregnancy notification form and sent via email.

Location in Protocol	Changes	Rationale
	Congenital abnormalities and birth defects in the offspring of male or female subjects should be reported as an SAE if conception occurred during the study treatment period or in the 60 ~~56~~-day period after the last dose of VIS410/placebo.	
Section 10.1 *Determination of Sample Size*	**¶ 2 only:** Using a log-rank test, a sample size of 130 influenza A–infected subjects per treatment group will provide 80% power to detect a 1.5-day difference (5 days for oseltamivir alone and 3.5 days for VIS410 plus oseltamivir) in the median time to cessation of O_2 support ~~resulting in a stable SpO_2 > 95% for at least 6 hours on room air, return to baseline respiratory status, or hospital discharge with no need for additional O_2 support, whichever occurs first~~ for VIS410 relative to placebo. A two-sided alpha of 0.05 was used for the calculation.	Section updated to maintain consistency with prior changes.
Section 10.6 *Interim Analysis*	An ~~The first~~ interim analysis of efficacy may ~~will~~ be performed by an unblinded third party after 50% of subjects have been enrolled (n = 195) and have an assessment of the primary endpoint (time to normalization of respiratory function). This interim analysis may ~~will~~ be conducted to assess if one of the active VIS410 treatment arms will need to be terminated for futility. Using the methods of Lan and DeMets an alpha spending function will be created and if the test statistic for one of the active VIS410 treatment arms is less than the lower critical point then the VIS410 treatment arm may be terminated for futility. The interim analysis may ~~The interim analysis will~~ also test the primary efficacy objective to assess if the primary efficacy endpoint has been met early. The effect size for this study was estimated without the benefit of any prior randomized, controlled trial. If the VIS410 effect size proves to be greater than anticipated, then it is possible that the primary study endpoint will be met by the time the interim efficacy analysis is performed. In the event the interim analysis reveals that the primary efficacy endpoint has been satisfied, this Phase 2 study could be terminated for efficacy. The DSMB and the unblinded statistician will review the interim analysis results to provide their recommendation on discontinuation of one of the VIS410 treatment arms, if needed, based on pre-specified criteria as outlined in the SAP. The interim analysis may ~~will~~ also review the following key secondary endpoints, when available, to assist in the decision of discontinuing a VIS410 treatment arm: • Time to clinical resolution of vital signs • Total number of hours on oxygen support or PPV • Viral load ~~titer~~ from nasopharyngeal swabs A sample size re-estimation (SSRE) will also be performed at this point in order to determine if the current sample size is sufficient based on the observed effect size in the study. The unblinded statisticians will work directly with the Sponsor on the SSRE. The details of SSRE and individuals with access to the information will be out	Clarified that an interim analysis may (or may not) be conducted due to rate of enrollment. Updated statistical methods and language to include more details related to the error rate and the interim analysis.

Location in Protocol	Changes	Rationale
Section 10.7.1 *Time to Cessation of O_2 Support*	**¶ 1 only:** Time to cessation of O_2 support resulting in a stable SpO_2 will ~~≥ 95% for at least 6 hours, return to baseline respiratory status, or hospital discharge with no need for O_2 support, whichever occurs first, will~~ be analyzed using a Cox model. Time from onset of symptoms to VIS410 treatment and the number of doses of oseltamivir (Tamiflu) prior to VIS410 treatment for the current case of influenza will be included as a covariate in the analysis. A *P*-value (using the Wald statistic) for each VIS410 dose vs placebo will be presented.	Section updated to maintain consistency with prior changes. Clarified the statistical method planned to determine P-value.
Section 10.7.2 *Time to Clinical Response;* Section 10.7.3 *Time to Cessation of Ventilator Support*	Time to clinical response is defined as resolution of 4 of 5 vital signs that will be determined upon physical examination. Clinical response is defined as: • Afebrile with core temperature ≤ 37.8°C, without use of antipyretics (oral ≤ 37.2°C) • Respiratory rate ~~<~~ 24 beats per minute • Oxygen saturation ~~>~~ 95% on room air without support or a return to pre-infection status, if pre-infection status was < 95% • Pulse rate ≤ 100/min • SBP ≥ 90 mm/Hg, without vasopressor use The probability of time to clinical response (defined as resolution of vital signs) will be calculated via Kaplan-Meier. A P-value (using the log-rank test) for each VIS410 dose vs placebo will be presented. The number and percentage of subjects in each treatment group with clinical response will be summarized. Results will be tabulated and presented graphically as well.	Section updated to maintain consistency with prior changes. Added details on statistical method planned to determine P-value.
Section 10.7.4 *Healthcare Resource Utilization*	**¶ 2 only:** Time (number of days) to resumption of usual activities will be determined from the VAS ~~visual analog scale~~ (scale ranged from 0 to 10, where 0 indicates subject is unable to perform any of his/her usual activities prior to influenza onset ~~at all~~, and 10 indicates subject is able to fully perform all usual ~~return to his/her normal~~ activities ~~fully~~).	Added language to clarify that a subject needs to complete VAS from baseline until the subject reports they are able to perform all usual activities they were performing prior to influenza onset.
Section 10.9.2 *Pharmacokinetics of Nasopharyngeal Secretions and Tracheal Aspirate*	Nasopharyngeal swabs will be obtained from both nostrils (1 swab per nostril). The first 50 randomized subjects will have nasopharyngeal swabs collected up to Day 56 (predose, end of infusion, Days 3, 5, 7, 14, 28 and 56); while in the remaining subjects, nasopharyngeal swabs will be obtained up to Day 14 only. If the subject remains in the hospital on Day 10, then additional nasopharyngeal swabs will be obtained on Day 10. The VIS410 concentrations in the nasopharyngeal secretions and tracheal aspirate will be listed by subject, and summary statistics by group will be reported as described for the serum concentrations. The computed PK parameters will be listed by subject for VIS410. Summary statistics and PK parameters will be presented, including means, geometric means, standard deviations, CV, medians, and ranges, as appropriate. Summary graphs, including mean concentration-time profiles by group, will also be presented. The nasopharyngeal concentration data may also be analyzed by population PK ~~pharmacokinetic~~ methods using nonlinear	Updated the planned PK analysis on nasopharyngeal swabs. to minimize the number of nasopharyngeal swabs in this study, the full nasal PK profile (Day 56) will be determined based on the first 50 subjects only. The remainder of the subjects will only have nasal PK along with nasal virology up to Day 14. PK analysis will be extrapolated beyond Day 14in these subjects based on population PK analysis.

Location in Protocol	Changes	Rationale
	mixed effects modeling as implemented in NONMEM or equivalent software. If necessary, the data may be pooled with data from previous studies. Additional analyses and summaries may be generated as appropriate. Results of population PK or PK/PD analyses may be reported outside the clinical study report.	
Section 10.11 *Immunological Analyses*	Immunological assessments will be summarized for the MITT population by parameter, treatment group, and time point using descriptive statistics: • Anti-influenza A antibodies by HAI in serum • ADA titers • ~~Concentrations of cytokines in serum~~	Updated language to more accurately describe all immunological analyses in one section.
Section 10.12 *Viral Resistance*	Viral sensitivity to VIS410 and oseltamivir will be assessed ~~over time~~ during the study. ~~This evaluation will be performed using the safety population~~	Clarified that viral resistance will be assessed. Analysis population will be defined in the SAP.
Appendix 14.1	Footnote to Questionnaire Copyright© 2014, 2015, 2016 Leidos Biomedical Research, Inc. All rights reserved. ~~Research Tool in Development: Do NOT copy or distribute~~	Added appropriate copyright information.
Schedule of Assessments	Visit window of ± 1 day added to Day 5 and Day 7	Visit window added to allow flexibility for subjects discharged from the hospital prior to Day 5.

Visterra

PROTOCOL AMENDMENT – SUMMARY OF CHANGES

Phase 2b, Multicenter, Randomized, Double-blind, Controlled Study to Evaluate the Efficacy and Safety of Intravenous VIS410 in Addition to Oseltamivir (Tamiflu®) Compared With Oseltamivir Alone in Hospitalized Adults With Influenza A Infection Requiring Oxygen Support

Product	VIS410
Protocol Number	VIS410-203
EudraCT Number	2016-004009-15
Clinical Phase	2b
Clinical Indication	Influenza A infection

Sponsor	Visterra, Inc. One Kendall Square, Suite B3301 Cambridge, MA 02139 United States of America
Sponsor Representative	[redacted]

ORIGINAL PROTOCOL DATE: **21 October 2016**

AMENDMENT 1 DATE: **3 July 2017**

AMENDMENT 2 DATE: **19 April 2018**

**SUMMARY OF CHANGES TO PROTOCOL VIS410-203
FROM VERSION 2.1 TO VERSION 3.1**

Substantive changes to the protocol for Amendment 3.1 and their location within the protocol are noted below. Additions are marked as red underlined text and deletions are marked as red strikethrough text. Administrative, stylistic and formatting changes that do not alter the conduct of the study are not summarized in this document.

Location in Protocol	Changes	Rationale
Cover Page; Signature of Sponsor Representative	Issue Date (Version): ~~3 July 2017 (Version 2.1 South Africa)~~ 19 April 2018 (Version 3.1 - South Africa) Sponsor Representative [redacted]	Updated date and version number. Sponsor representative updated.
Cover page; Sponsor Address	Visterra, Inc. ~~One Kendall Square, Suite B3301~~ 275 Second Avenue, 4th Floor ~~Cambridge~~ Waltham, MA ~~02139~~02451 United States of America	Company moved location
Synopsis: Number of Clinical Sites	Approximately ~~180~~ 140 sites worldwide	Reduction in number of sites
Synopsis: Number of Subjects	Approximately ~~390~~ 120	Decreasing sample size from 390 to 120 subjects to enable completion in 2 influenza seasons.
Synopsis and Section 2.1.1 *Primary Efficacy Objective*	• ~~Evaluate~~ Evaluation of the effect of 2 dose levels of VIS410 + oseltamivir on ~~the time to normalization of respiratory function compared to oseltamivir alone~~ clinical outcome as assessed by comparison of clinical status ordinal scale Day 7 scores between treatment groups, and between all VIS410 recipients versus placebo.	Utility of ordinal scale observed in prior trials of this size.

Synopsis and Section 2.2 *Secondary Objectives*	• Among patients requiring supplemental oxygen therapy at the time of enrollment with baseline room air oxygen saturation of ≤ 92%, time to cessation of oxygen (O_2) support resulting in stable oxygen saturation (SpO_2) by pulse oximetry. Stable SpO_2 is defined as two consecutive SpO_2 values of > 92% on room air that are at least 8 hours apart.
	• For any patient requiring supplemental oxygen therapy at the time of enrollment (regardless of oxygen saturation), time to cessation of oxygen support.
	• Evaluate the effect of 2 dose levels of VIS410 + oseltamivir vs oseltamivir alone on the following parameters:
	  • Viral load in upper respiratory samples
	  • Time to clinical response
	  • Time to cessation of ventilator support
	  • Time to resumption of normal activities
	  • All-cause and attributable 14-, ~~and~~ 28-, and 56-day mortality
		Time to cessation of oxygen support changed from primary endpoint to secondary. Some patients may be on oxygen with O_2 saturation > 92% at baseline.
		
		Added ordinal scale parameters to be assessed

Location in Protocol	Changes	Rationale
	• Clinical status ordinal scale mean area under the curve for Days 1-7 and Days 1-14 using linear numeric scores for the ordinal categories. • Comparison of clinical status ordinal scale scores for selected individual days (i.e., Days 3, 4, 5, and 6) • Comparison of clinical status ordinal scale scores using modified ordinal scale criteria (i.e. pooling of selected severity criteria scores) • Comparison of discrete ordinal scale parameters, including days of ventilator support, days in intensive care, and duration of hospitalization • Healthcare resource utilization • Analysis of time ~~Time~~ to alleviation of ~~clinical~~ signs and symptoms of influenza in the subset of subjects able to complete the Influenza Patient Reported Outcomes (FluPRO) Questionnaire at baseline and post-dose by Kaplan Meier analysis • Proportion of subjects with new documented bacterial pneumonia/superinfection • Proportion of subjects with influenza-related complications • Pharmacokinetics of VIS410 in serum • Immunogenicity of VIS410 • Emergence of resistance to VIS410 and oseltamivir	
Synopsis and Section 2.3 *Exploratory Objectives*	~~• Evaluate the effect of VIS410 + oseltamivir vs oseltamivir alone on clinical outcomes as measured by a seven level ordinal scale~~ • Evaluate the pharmacokinetics of VIS410 from nasopharyngeal secretions and tracheal aspirate (ventilated subjects only) • Assess the effects of VIS410 on viral load in tracheal aspirate (ventilated subjects only) • Assess correlations between virology, safety, VIS410 dose, pharmacokinetics, viral shedding, immunology, signs and symptoms of influenza, and other endpoints • Assess the anti-influenza immune response	Removed as components are now primary and secondary endpoints
Synopsis	**Study Design** This is a Phase 2b, multicenter, randomized, double-blind, controlled study comparing the efficacy and safety of 2 dose levels of VIS410 (2000 and 4000 mg) in combination with oseltamivir vs oseltamivir alone in hospitalized subjects with influenza A infection requiring oxygen support. Subjects will be randomly assigned at a ratio of 1:1:1 using a permuted block randomization method to receive VIS410 at a dose of 2000 mg (low dose) or 4000 mg (high dose) or placebo (0.9% sodium chloride), administered as a single IV infusion over 2 hours. Randomization will be stratified by presence or absence of positive pressure ventilation (PPV) at baseline to ensure an even distribution across treatment arms. All subjects will receive oseltamivir 75 mg twice daily (BID) for 5 days	Decreasing sample size from 390 to 120 subjects to enable completion in 2 influenza seasons. Table in section also updated to reflect 40 subjects per arm.

Location in Protocol	Changes	Rationale
	(total of 10 doses) as part of standard of care (SOC). Treatment with oseltamivir may be extended for up to 5 additional days (up to 20 doses of total therapy) if clinical symptoms warrant further treatment as assessed by the Investigator. Note: based on SOC, subjects may have started oseltamivir therapy prior to randomization. All subjects will also receive a pretreatment regimen of diphenhydramine 50 mg PO or IV plus either ibuprofen 400 mg PO or acetylsalicylic acid 320-325 mg PO approximately 60 minutes before infusion. Approximately ~~390~~ 120 evaluable subjects (~~130~~ 40/arm) with confirmed influenza A infection will be treated. Subjects admitted to the hospital within 5 days of onset of initial symptoms who require supplemental oxygen will undergo a rapid influenza test or a local polymerase chain reaction (PCR) test, fluorescent immunoassay (FIA) test, or enzyme-linked immunosorbent assay (ELISA) test to confirm influenza A infection. Subjects diagnosed with influenza A will undergo the screening procedures to confirm eligibility. Study assessments are outlined in **Error! Reference source not found.** (Schedule of Assessments). Eligible subjects will be randomized to receive either oseltamivir + VIS410 2000 mg, oseltamivir + VIS410 4000 mg, or oseltamivir + placebo (0.9% sodium chloride) at a ratio of 1:1:1 using a permuted block randomization method according to the table below: Oseltamivir (Tamiflu®) will be provided by the Sponsor. Treatment with oseltamivir may be extended for up to 5 additional days (up to 20 doses of total therapy) if clinical symptoms \warrant further treatment as assessed by the Investigator. Note: based on SOC, subjects may have started oseltamivir therapy prior to randomization. Study assessments are outlined in **Error! Reference source not found.**. Subjects discharged from the hospital prior to Day 14 will attend the applicable outpatient follow-up visit(s) (eg, Day 3, Day 5, Day 7, Day 14) per **Error! Reference source not found.**. An independent data safety monitoring board (DSMB) will be established to review all available safety data after 30 subjects ~~as well as when~~ and again after ~~approximately 120~~ 70 ~~subjects~~ have completed ~~study~~ Day 14 assessments. The assessment of safety will be determined from vital sign measurements, physical examinations, hematology, chemistry and urinalysis laboratory testing, 12-lead electrocardiograms (ECGs), use of concomitant medications, and review of adverse events (AEs). Based on the safety data provided, the DSMB will determine if it is acceptable to continue the study or reduce the dose as appropriate. Dosing will temporarily pause while the DSMB meets if there are 4 treatment-related serious adverse events (SAEs) or 4 severe gastrointestinal (GI) treatment–emergent adverse events (TEAEs) that require intervention, which is defined as requiring IV fluid and medication to decrease the frequency of diarrhea (ie, loperamide).	Performing second DSMB review after completion of Northern Hemisphere season. No need for DSMB review after 120 subjects; final analysis of data will be performed.

Location in Protocol	Changes	Rationale
	Additional DSMB reviews can occur throughout the trial as deemed necessary by the DSMB or Sponsor to ensure subject safety and well-being. Study enrollment and dosing will continue while the DSMB evaluates data. ~~In addition, following 50% enrollment (195 subjects), an interim analysis may be conducted, by an unblinded third party, to assess if one of the VIS410 treatment arms can be terminated early for futility. A prespecified sample size reanalysis may also be conducted based on the observed effect size. The interim analysis may also test the primary efficacy objective to assess if the primary efficacy endpoint has been met. An alpha spending function will be designed in order to determine the amount of alpha that will be spent at the interim analysis. The effect size for this study was estimated without the benefit of any prior randomized controlled trial. If the VIS410 effect size proves to be greater than anticipated, then it is possible that the primary study endpoint will be met by the time the interim efficacy analysis is performed. In the event the interim analysis reveals that the primary efficacy endpoint has been satisfied, this Phase 2 study could be terminated for efficacy~~.	Removed interim analysis as sample size decreased to 120 subjects; final analysis to be performed upon completion of enrollment.
Synopsis and Section 5.1 *Number of Subjects*	Approximately ~~390~~ 120 evaluable subjects will be enrolled in 3 equal arms: VIS410 2000 mg, VIS410 4000 mg, and placebo.	Sample size decreased.
Synopsis and Section 5.2.1 *Inclusion Criteria*	9. Subject, or a legally acceptable ~~authorized~~ representative, is able to understand the purpose and risks of the study and willing to give voluntary written informed consent.	Change requested by an Ethics Committee
Synopsis	**Test Product, Dose, Mode of Administration** VIS410 (2000 mg or 4000 mg) will be administered IV over 2 hours as a single 200-mL infusion, followed by a 25-mL (or volume equivalent to IV line volume if greater than 25 mL) saline flush to ensure all product is administered, in addition to oseltamivir (Tamiflu) 75 mg BID for total of 5 days (10 doses). For patients with renal insufficiency, refer to prescribing information for administration of oseltamivir. For patients unable to swallow, oseltamivir capsules can be opened and mixed with liquid for ease of administration. **Reference Product, Dose, Mode of Administration** Placebo (normal saline solution 0.9%) will be administered IV over 2 hours as a single 200-mL infusion, followed by a 25-mL (or volume equivalent IV line volume if greater than 25 mL) saline flush to ensure all product is administered, in addition to oseltamivir (Tamiflu) 75 mg BID for total of 5 days (10 doses). For patients with renal insufficiency, refer to prescribing information for administration of oseltamivir. **VIS410 and Placebo Preparation** The required amount of VIS410 to be dosed (2000 mg or 4000 mg) will be diluted with normal saline up to a maximum total volume of 200 mL; for placebo subjects, 200 mL of normal saline will be prepared. Length of IV	Clarification to ensure all study product administered. Some sites use infusion lines with hold-up volumes of greater than 25 mL.

Location in Protocol	Changes	Rationale
	line will ideally be set for maximum volume of 25 mL, so that the 25-mL (or increased volume as noted above) saline flush following administration will ensure all VIS410/placebo has been administered. In the event that the infusion line volume is greater than 25 mL the post-administration saline flush should be increased to match the volume of the infusion line kit. The VIS410/placebo infusion will be administered IV using a 0.22-µm in-line filter and will be controlled by a volumetric pump. Standard, uniform-length infusion lines will be used whenever possible, and microfilters will be provided by the Sponsor. The infusion bag will be covered with an opaque sleeve in the pharmacy to maintain the study blind. The VIS410/placebo will be administered IV at a rate of 100 mL/h, over 2 hours. After ~~200 mL of~~ the ~~diluted~~ dose has been administered, the infusion will be ~~stopped,~~ followed by ~~a 25 mL~~ an appropriate volume saline flush as described above. The infusion time may be extended up to 4 hours at the Investigator's discretion based on local infusion site–related symptoms. All subjects will be given a pretreatment regimen of PO or IV diphenhydramine 50 mg plus either ibuprofen 400 mg PO or acetylsalicylic acid 320-325 mg PO or crushed and given through the nasogastric tube approximately 60 minutes before IV infusion of VIS410/placebo.	
Synopsis and Section 3.1.1 *Primary Efficacy Endpoint*	~~• Time to cessation of oxygen (O_2) support resulting in stable oxygen saturation (SpO_2) by pulse oximetry. Stable SpO_2 is defined as two consecutive SpO_2 values of > 92% on room air that are at least 8 hours apart.~~ • The primary efficacy outcome analysis compares Day 7 clinical status ordinal scale scores between treatment groups, and between all VIS410 recipients versus placebo. Clinical status is measured daily for 14 days using the below seven-level ordinal scale, with the classifications presented from the worst clinical outcome to the best clinical outcome in descending order; for each day, subject status will be classified by the worst clinical outcome for which they qualify. ▪ Death ▪ ICU stay with mechanical ventilation ▪ ICU stay without mechanical ventilation ▪ Non-ICU hospitalization with supplemental oxygen ▪ Non-ICU hospitalization without supplemental oxygen ▪ Discharge with partial resumption of normal activities ▪ Discharge with full resumption of normal activities	Primary objective changed.
Synopsis and Section 3.2 *Secondary Endpoints*	The difference between VIS410 + oseltamivir and oseltamivir alone treatment groups in the following endpoints:	

Location in Protocol	Changes	Rationale

	• Among patients requiring supplemental oxygen therapy at the time of enrollment with baseline room air oxygen saturation of ≤ 92%, time to cessation of oxygen (O_2) support resulting in stable oxygen saturation (SpO_2) by pulse oximetry. Stable SpO_2 is defined as two consecutive SpO_2 values of > 92% on room air that are at least 8 hours apart.
	• For any patient requiring supplemental oxygen therapy at the time of enrollment (regardless of oxygen saturation), time to cessation of oxygen support
	• Peak viral load, viral area under the concentration–time curve (AUC), duration of viral shedding, and time to resolution of viral load from nasopharyngeal swabs by $TCID_{50}$ and qRT-PCR
	• Time to clinical response defined as resolution of at least 4 of 5 vital signs:
	  • Afebrile with core temperature ≤ 37.8°C, without use of antipyretics (oral ≤ 37.2°C)
	  • Oxygen saturation ≥ 95% on room air without support or a return to pre-infection status, if pre-infection status was < 95%
	  • Pulse rate ≤ 100/min
	  • Systolic blood pressure (SBP) ≥ 90 mm/Hg, without vasopressor use
	  • Respiratory rate ≤24 beats per minute
	• ~~Total number of days on ventilation~~
	• Clinical status ordinal scale mean area under the curve for Days 1-7 and Days 1-14 using linear numeric scores for the ordinal categories.
	• Comparison of clinical status ordinal scale scores for selected individual days (i.e., Days 3, 4, 5, and 6)
	• Comparison of clinical status ordinal scale scores using modified ordinal scale criteria (i.e. pooling of selected severity criteria scores)
	• Comparison of discrete ordinal scale parameters, including days of ventilator support, days in intensive care, and duration of hospitalization
	• Number of days to resumption of normal activities
	• All-cause and attributable mortality rates at Day 14, ~~and~~ 28, and 56
	• Total number of days in hospital and/or intensive care unit (ICU) from admission to discharge and rate of rehospitalization due to influenza A relapse/complication
	• The incidence, severity, and duration of signs and symptoms of influenza-like illness as assessed by the FluPRO Questionnaire (see Appendix **Error! Reference source not found.**)
	• Analysis of time to alleviation of signs and symptoms of influenza in the subset of subjects able to complete the Influenza Patient Reported Outcomes (FluPRO)
		Moved from primary objective to secondary
		
		Deleted as covered by next two bullets addressing ordinal scale assessments

Location in Protocol	Changes	Rationale
	Questionnaire at baseline and post-dose by Kaplan Meier analysis • The percentage of subjects with new bacterial pneumonia/superinfection • The percentage of subjects with influenza-related complications • VIS410 population pharmacokinetic (PK) parameters in serum • Titer of anti-VIS410 antibody positive samples • Genotypic and/or phenotypic assessment to determine the emergence of VIS410 and oseltamivir-resistant viruses	
Synopsis and Section 3.3 *Exploratory Endpoints*	~~• Clinical outcome will be measured daily for 14 days using a seven-level ordinal scale; this seven-level ordinal scale is defined as:~~ ~~• Death~~ ~~• ICU stay with mechanical ventilation~~ ~~• ICU stay without mechanical ventilation~~ ~~• Non-ICU hospitalization with supplemental oxygen~~ ~~• Non-ICU hospitalization without supplemental oxygen~~ ~~• Discharge with partial resumption of normal activities~~ ~~• Discharge with full resumption of normal activities~~ • Population PK parameters of VIS410 from nasopharyngeal secretions • VIS410 concentration in tracheal aspirates • The difference in viral load between VIS410 + oseltamivir and oseltamivir alone treatment groups in tracheal aspirate of subjects on mechanical ventilation • Titer of anti-influenza A antibodies by hemagglutinin inhibition assay (HAI) in serum • Correlations between serum and/or nasopharyngeal PK with viral load, clinical symptoms, presence of antidrug antibodies (ADAs), safety, and additional endpoints	Now covered under primary efficacy and secondary endpoints
Synopsis	**Statistical Methods** *Sample Size* Approximately 120 ~~390~~ evaluable subjects will be enrolled. The study is exploratory in nature, and is not powered to demonstrate significant differences between treatment groups in primary or secondary outcome measures. The protocol intent is to collect sufficient information to identify the most appropriate candidate endpoints for subsequent Phase 3 study evaluation from among the primary and secondary endpoints described below. Statistical significance testing will therefore be used to assess the relative strength of evidence of the primary and secondary endpoints, to provide reasonable assurance that the endpoints chosen for a confirmatory Phase 3 trial will elucidate treatment differences between VIS410 plus oseltamivir versus oseltamivir alone. ~~Using a log-rank test, a sample size of 130 influenza A-infected subjects per~~	Analysis changed due to the decrease in sample size.

Location in Protocol	Changes	Rationale
	~~treatment group will provide 80% power to detect a 1.5-day difference (5 days for oseltamivir alone and 3.5 days for VIS410 plus oseltamivir) in the median time to normalization of respiratory function (cessation of O_2 support) for VIS410 relative to placebo. A two-sided alpha of 0.05 was used for the calculation.~~ *Efficacy* Efficacy analysis will have two complementary purposes in this Phase 2 study, both designed to aid in the optimal design of a confirmatory Phase 3 trial. First, primary and secondary endpoints will be analyzed for treatment group differences, to determine which endpoints might be most sensitive demonstration of treatment group differences using p-values as an indicator, Second, the relative contribution of the various efficacy endpoints to patient well-being and benefit will be assessed, as well as potential correlations of endpoints such as the influence of peak viral load or number of days on ventilator on the time to resumption of normal activities. The ordinal scale outcomes will be measured daily from Day 1 (baseline) through Day 14, inclusive, using a seven-level hierarchical scale with the classifications ordered from the worst to the best clinical outcomes (see Section 10.6.1). For use in overall summary statistical presentations, the ordinal categories will be assigned decreasing integer scores, with death a score of 6 and discharge with full resumption of normal activities a score of 0. The primary outcome comparison of Ordinal Scale scores at Day 7 between treatment groups will be evaluated by proportional odds ratio analysis, as implemented by logistic regression, including a test of the proportional odds assumption. For this analysis, the response categories will be ordered from best (Discharge with full resumption of normal activities) to worst (Death). Additional exploratory analyses may be conducted to obtain a more complete understanding of the relationship of treatment to the ordinal response, including exact Mantel-Haenszel tests or partial proportional odds models. The area under the curve (AUC) over time for a given patient will be calculated as the sum of the maximum ordinal score for each day up through 7 and 14 days. An analysis of treatment group differences will be performed on these per-patient AUC values using analysis of variance, with treatment group and strata as fixed factors. A sensitivity analysis will be performed that excludes the category of death on study, to determine if death as an outcome skews the results; death as an outcome will also be analyzed as an independent secondary endpoint. A secondary analysis of treatment group effect on the difference in proportions of patients with the worst (death) versus the best outcome (discharge from hospital and resumption of normal activities) will also be performed. This analysis does not use scores for the ordinal outcome but does account for ordinality, and is therefore not dependent on the relationship of score to severity of outcome. An exploratory analysis will be performed	

Location in Protocol	Changes	Rationale
	through an exact categorical analysis of treatment group difference in the ordinal scale results using the worst outcome on a per-patient basis. Additional ordinal scale outcome assessments will include comparison of total numbers of days at more severe scale values (death, time on ventilator, time in ICU) and proportions of patients with ordinal scale worsening post enrollment. Proportion outcomes will be analyzed by categorical data analysis methods, including Mantel-Haenszel chi-square tests adjusted for strata; additional exploratory subgroup analyses may also be performed, for example, based on various age categories. Time from onset of symptoms to study treatment and the number of doses of oseltamivir (Tamiflu) prior to VIS410 treatment for the current case of influenza may be included as covariates in the analyses. Secondary and exploratory endpoints will be analyzed for descriptive purposes, with significance levels (p-values) provided to illustrate the strength of evidence for treatment effects, and to provide a basis for the choice of endpoints for further study in a confirmatory study. These analyses will also provide estimates for potentially powering additional efficacy endpoints, and to assist in hierarchical ordering of secondary endpoints to provide alpha-control in confirmatory studies and for labelling purposes. Time to cessation of O_2 support resulting in a stable SpO_2 will be analyzed using a Cox proportional hazards regression model including data from patients on O_2 support. ~~Time from onset of symptoms to VIS410 treatment and number of doses of oseltamivir (Tamiflu) prior to VIS410 treatment for the current case of influenza will be included as a covariate of the analysis~~. A P-value (using the Wald statistic) for each VIS410 dose vs placebo will be presented. The total number of days in the hospital and/or ICU from admission to discharge and the rate of rehospitalization due to influenza A relapse/reinfection will be summarized descriptively by treatment group. The probability of time to clinical response (defined as resolution of vital signs) will be calculated via Kaplan-Meier. A ~~P-value~~ significance test (using the log-rank test) for each VIS410 dose vs placebo will be presented. Results will be tabulated and presented graphically as well. The number and percentage of subjects in each treatment group with clinical response will be summarized. ~~The ordinal scale outcomes will be measured daily from Day 1 (baseline) through Day 14, inclusive, using a seven-level hierarchical scale with the classifications ordered from the worst to the best clinical outcomes (see **Section 0**). The number and percentage of subjects in each treatment group with each classification will be summarized for each day from Day 1 through Day 14, inclusive.~~ ~~*Interim Analyses of Efficacy*~~	

Location in Protocol	Changes	Rationale
	~~An interim analysis of efficacy may be performed by an unblinded third party after 50% of subjects have been enrolled (n = 195) and have an assessment of the primary endpoint (time to normalization of respiratory function). This interim analysis may be conducted to assess if one of the active VIS410 treatment arms will need to be terminated for futility. The interim analysis may also test the primary efficacy objective to assess if the primary efficacy endpoint has been met early. A prespecified sample size reanalysis will also be conducted based on the observed effect size. The DSMB and the unblinded statistician will review the interim analysis results to provide their recommendation on discontinuation of one of the VIS410 treatment arms, if needed, based on pre-specified criteria as outlined in the SAP. The interim analysis may also review the following key secondary endpoints, when available, to assist in the decision of discontinuing a VIS410 treatment arm:~~ ~~• Time to clinical resolution of vital signs~~ ~~• Total number of hours on oxygen support or PPV~~ ~~• Viral load from nasopharyngeal swabs~~ ~~A sample size re-estimation (SSRE) will also be performed at this point in order to determine if the current sample size is sufficient based on the observed effect size in the study. The unblinded statisticians will work directly with the Sponsor on the SSRE. The details of SSRE and individuals with access to the information will be outlined in a separate plan.~~ ///// *Complications of Influenza* Complications of influenza are defined as pneumonia, myocarditis, worsening of chronic bronchitis, sinusitis, otitis, or other chronic pulmonary diseases, encephalopathy, and death. Complications of influenza and use of antibiotic therapy for bacterial pneumonia will be reported by variable, treatment group, and time point. Diagnosis of bacterial pneumonia will be based on local clinical practice, usually an assessment of clinical features and any radiographic evidence of bacterial pneumonia (eg, infiltrate) and/or microbiologic evidence of bacterial pneumonia. All-cause and attributable mortality will be evaluated on Day 14 and 28.	No interim analysis to be performed due to the decrease in sample size.
Table 1 *Schedule of Assessments*	Table: Erythrocyte sedimentation rate (ESR)[20a] Footnotes: [20a] ESR to be performed locally.	Clarification.
List of Abbreviations	AUC – Area under curve	New abbreviation
Section 1.1 *Background Information*	Severe influenza disease is a common occurrence each season, especially in high-risk groups, such as young children, older adults, patients with pulmonary conditions, inflammatory conditions, malignancies, and pregnant women.[i, ii] Despite available therapy with neuraminidase inhibitors, including oseltamivir (Tamiflu®), zanamivir (Relenza®), and peramivir (Rapivab®), 10% to 44% of	

Location in Protocol	Changes	Rationale
	hospitalized patients require intensive care and 25% to 50% of these patients die. It is estimated that as many as 400,000 patients are hospitalized with influenza each year in the United States, with up to 49,000 deaths per year.[iii] The World Health Organization (WHO) has reported incidence rates of 3 to 5 million severe cases and about 250,000 to 500,000 influenza-related deaths annually.[iv] The 2009 influenza A pandemic (H1N1) spread rapidly to every continent with more than 399,232 reported cases and 4735 deaths.[v] The therapeutic use of passive polyclonal antibodies to prevent viral infections, including hepatitis B, varicella, cytomegalovirus, rabies, and respiratory syncytial virus (RSV) has been well established. More recently, monoclonal antibodies for viral infections have been developed, including palivizumab (Synagis®), a Food and Drug Administration (FDA)-licensed treatment for the prevention of RSV infection. Visterra has developed a novel approach to antibody discovery whereby functionally conserved epitopes are identified based on atomic interaction networks and targeted with rationally engineered human antibodies. Using this approach, VIS410, a broad spectrum human immunoglobulin G1 (IgG1) monoclonal antibody with demonstrated efficacy against both Group 1 (including H1 and H5) and Group 2 (including H3 and H7) influenza A strains, in both treatment and prevention models of influenza, was developed. Visterra intends to develop this product for the treatment of influenza A, specifically in hospitalized patients. This study will provide the first indications of efficacy, safety, and tolerability of VIS410 in hospitalized subjects with influenza A infection requiring oxygen support. Efficacy will be ~~measured by time to cessation of oxygen support~~ assessed by comparison of clinical status ordinal Day 7 scores between treatment groups.	
Section 4.1 *Overview*	This is a Phase 2b multicenter, randomized, double-blind, controlled study comparing the efficacy and safety of 2 dose levels of VIS410 (2000 and 4000 mg) in combination with oseltamivir vs oseltamivir alone in hospitalized subjects with influenza A infection requiring oxygen. Subjects will be randomly assigned at a ratio of 1:1:1 using a permuted block randomization method to receive VIS410 at a dose of 2000 mg (low dose) or 4000 mg (high dose) or placebo (0.9% sodium chloride), administered as a single IV infusion over 2 hours. Randomization will be stratified by presence or absence of positive pressure ventilation (PPV) at baseline to ensure an even distribution across treatment arms (see Section **Error! Reference source not found.**). All subjects will receive oseltamivir 75 mg twice daily (BID) for 5 days (total of 10 doses) as part of standard of care (SOC). All subjects will also receive a pretreatment regimen of diphenhydramine 50 mg PO or IV plus either ibuprofen 400 mg PO or acetylsalicylic acid 320-325 mg PO 60 minutes before VIS410/placebo infusion. Approximately	Decreasing sample size from 390 to 120 subjects to enable completion in 2 influenza seasons. Table 6 also updated to reflect 40 subjects per arm.

Location in Protocol	Changes	Rationale
	~~390~~120 evaluable subjects (~~1304~~0/arm) with confirmed influenza A infection will be treated. Subjects admitted to the hospital within 5 days of onset of initial symptoms who require supplemental oxygen will undergo a rapid influenza test (supplied by the Sponsor) or a PCR, fluorescent immunoassay (FIA), or enzyme-linked immunosorbent assay (ELISA) test to confirm influenza A infection. Subjects diagnosed with influenza A will undergo the screening procedures to confirm eligibility. Study assessments are outlined in **Error! Reference source not found.**, Schedule of Assessments. Eligible subjects will be randomized to receive either oseltamivir + VIS410 2000 mg, oseltamivir + VIS410 4000 mg, or oseltamivir + placebo (0.9% sodium chloride) at a ratio of 1:1:1 using a permuted block randomization method summarized in **Error! Reference source not found.**. Oseltamivir (Tamiflu) will be provided by the Sponsor. Treatment with oseltamivir may be extended for up to 5 additional days (up to 20 doses of total therapy) if clinical symptoms warrant further treatment as assessed by the Investigator. Note: based on SOC, subjects may have started oseltamivir therapy prior to randomization. ~~In addition, following 50% enrollment (195 subjects), an interim analysis may be conducted, by an unblinded third party, to assess if one of the VIS410 treatment arms can be terminated early for futility. A prespecified sample size reanalysis may also be conducted based on the observed effect size. The interim analysis may also test the primary efficacy objective to assess if the primary efficacy endpoint has been met. An alpha spending function will be designed in order to determine the amount of alpha that will be spent at the interim analysis. The effect size for this study was estimated without the benefit of any prior randomized controlled trial. If the VIS410 effect size proves to be greater than anticipated, then it is possible that the primary study endpoint will be met by the time the interim efficacy analysis is performed. In the event the interim analysis reveals that the primary efficacy endpoint has been satisfied, this Phase 2 study could be terminated for efficacy~~.	Removed interim analysis as sample size decreased to 120 subjects; final analysis to be performed upon completion of enrollment.
Section 4.3 *Data Safety Monitoring Board*	An independent data safety monitoring board (DSMB) will be established to review all available safety data after 30 subjects, ~~as well as when~~ and subsequently, approximately ~~120~~ 70 subjects, have completed study Day 14~~.~~ The assessment of safety will be determined from vital sign measurements, physical examinations, hematology, chemistry and urinalysis laboratory testing, 12-lead electrocardiograms (ECGs), use of concomitant medications, and review of AEs. Based on the safety data provided, the DSMB will determine if it is acceptable to continue the study or reduce the dose as appropriate. Additional DSMB reviews can occur throughout the trial as deemed necessary by the DSMB or Sponsor to ensure subject safety and well-being. Study enrollment and dosing will continue while the DSMB evaluates data.	Sample size decreased therefore second DSMB review not required.

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	Further details will be described in a separate DSMB charter.	
Section 5.2.1 *Inclusion Criteria*	9. Subject, or a legally acceptable ~~authorized~~ representative, is able to understand the purpose and risks of the study and willing to give voluntary written informed consent.	Change requested by an Ethics Committee
Section 6.1.3 *Study Procedures*	On Day 1, eligibility of the subjects will be confirmed and assessments will be performed as described in **Error! Reference source not found.**. All results from the screening procedure needed to evaluate eligibility, including any local clinical laboratory results, must be available prior to randomization on Day 1. All clinical assessments required for the determination of subject eligibility will be performed by the local clinical laboratory, thereby precluding the need to wait for central laboratory data. Any abnormal assessment at the screening visit will be assessed according to its clinical relevance, and if found relevant, the subject will not be included in the study. Once a subject has satisfied entry criteria, he/she will be assigned a unique identifier. The site's unblinded pharmacist or properly trained designee will ~~randomize the subject using the~~ access the Interactive Web Response System (IWRS) to obtain the study treatment. Randomization will be stratified by presence or absence of PPV at baseline to ensure balance between the treatment arms. Subjects will be considered randomized when the pharmacist or designee obtains the subject number and treatment assignment from the IWRS. Subjects may not be randomized into this study more than once. Subjects who have participated in any previous study of VIS410 may not be randomized to this study. Subjects will be monitored daily while in the hospital up to Day 14 (± 3 days) with an additional visit on Day 28 (± 3 days) and the last follow-up visit on Day 56 (± 7 days). In order to provide some flexibility for the subjects regarding the site visits and to maintain the integrity of the study design, a time window is permitted for the follow-up visits in case of time conflict or unforeseen circumstances. Note: subjects discharged from the hospital prior to Day 14 will attend the applicable outpatient follow-up visit(s) (eg, Day 3, Day 5, Day 7, Day 14).	Clarification
Section 6.2 *Pregnancy Safeguards*	Pregnancy will be determined by evaluation of β-human chorionic gonadotropin in serum or urine for all women of childbearing potential. Subjects who are pregnant or nursing will be excluded from the study. During the course of the study drug administration period within the study, any nursing mother(s) or subject(s) with suspected or confirmed pregnancy will be discontinued from study drug therapy but will be encouraged to undergo follow-up for safety monitoring for themselves (ie, the pregnant female) and the baby. All women of childbearing potential and all male subjects must practice effective contraception from screening until 60 days post-VIS410/placebo infusion. For the purposes of	

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	this study, women who do not satisfy at least one of the following criteria listed below (ie, the criteria for defining non-childbearing potential) are considered to be physiologically capable of becoming pregnant and are, therefore, defined as women of childbearing potential. The criteria for defining women as being of non-childbearing potential are: • Post-menopausal: ≥ 12 months of natural (spontaneous) amenorrhea, or • Follicle stimulating hormone > 40 mIU/mL as documented in their medical history, or • Surgical bilateral oophorectomy with or without hysterectomy, or • Hysterectomy, or • Bilateral tubal ligation Women of childbearing potential and all male subjects participating in heterosexual relations must be willing to practice effective contraception from screening until 60 days post-VIS410/placebo infusion. For the purposes of the study, highly effective contraception is defined as: • Male vasectomy with negative semen analysis documentation at least 6 months prior to dosing. OR • Combination of an established form of hormonal contraception (oral, injected, or implanted) or an intrauterine device or intrauterine system or sponge Plus one of the following: • A physical barrier method of contraception with use of a spermicide, such as condom or occlusive cap (diaphragm or cervical/vault caps) with spermicidal foam/gel/film/cream suppository. The use of barrier contraceptives should always be supplemented with the use of a spermicide, unless not available in a country. OR • Male vasectomy with negative semen analysis documentation less than 6 months prior to dosing. OR • Complete abstinence can be considered an acceptable method of contraception at the discretion of the Investigator. Periodic abstinences (eg, calendar, ovulation, symptothermal, or post-ovulation methods) and withdrawal are NOT considered acceptable methods of contraception. The combination of 2 barrier methods, periodic abstinence (eg, calendar, ovulation, symptothermal, or post-ovulation methods), and withdrawal are not considered acceptable methods of contraception.	Sponge is an acceptable method of contraception Added requirement.

Location in Protocol	Changes	Rationale
	Male subjects should not donate sperm for at least 60 days after receipt of study product. Pregnancy reporting is described in Section **Error! Reference source not found.**.	
Section 6.5.2 *Injection Site Tolerability*	Injection site tolerability is defined as AEs demonstrating significant local injection site irritation or tissue damage. ~~Injection site tolerability will be reported by variable, treatment group, and time point.~~	Sentence deleted as belongs in SAP
Section 6.5.3 *Complications of Influenza*	Complications of influenza are defined as pneumonia, myocarditis, worsening of chronic bronchitis, sinusitis, otitis, or other chronic pulmonary diseases, encephalopathy, and death. Complications of influenza and use of antibiotic therapy for bacterial pneumonia will be reported by variable, treatment group, and time point. Diagnosis of bacterial pneumonia will be based on local clinical practice, usually an assessment of clinical features and any radiographic evidence of bacterial pneumonia (eg, infiltrate) and/or microbiologic evidence of bacterial pneumonia. All-cause and attributable mortality will be evaluated on Day 14 and Day 28.	
Section 7.1 *Test product, Dose, Mode of Administration*	The Investigator must ensure that the investigational product will be used only in accordance with the protocol. It is forbidden to use investigational drug material for purposes other than as defined in this protocol. VIS410 (2000 mg or 4000 mg) will be administered IV over 2 hours as a single, 200-mL infusion, followed by a 25-mL (or volume equivalent to length of IV line) saline flush to ensure all product is administered, in addition to oseltamivir (Tamiflu) 75 mg BID for total of 5 days (10 doses). Treatment with oseltamivir may be extended for up to 5 additional days (up to 20 doses of total therapy) if clinical symptoms warrant further treatment as assessed by the Investigator. Note: based on SOC, subjects may have started oseltamivir therapy prior to randomization. No dose adjustment is necessary for VIS410 based on renal or hepatic impairment. For patients with renal insufficiency, refer to prescribing information for administration of oseltamivir. VIS410 is a colorless to slightly yellow, clear to opalescent solution, essentially free of particles. VIS410 is formulated at a concentration of 25 mg/mL in 40-mM citrate-sodium phosphate, 150-mM sodium chloride, and 0.025% polysorbate 80. The investigational product will be provided by the Sponsor. VIS410 will be supplied in Type I 20-mL glass vials containing a nominal 20-mL solution. A copy of the certificate of analysis of the investigational product will be sent to the clinical center. The investigational drug product is manufactured by Lyophilization Services of New England in accordance with Good Manufacturing Practice as required by the current Good Clinical Practice (GCP). Manufacturing, packaging, and labeling of the investigational product, VIS410, is conducted under the responsibility of the Sponsor. The study drug will be labeled according to local	Clarification as some sites use lines with greater than 25 mL hold-up volume

Location in Protocol	Changes	Rationale
	law and regulatory requirements. Specific dilution procedures for VIS410 will be described in detail in the Pharmacy Manual. Placebo will be a normal saline solution (0.9%) and will be prepared by the pharmacist.	
Section 7.2 *Reference Product, Dose, Mode of Administration*	Placebo (normal saline solution 0.9%) will be administered IV over 2 hours as a single 200-mL infusion, followed by a 25-mL (or volume equivalent to length of IV line) saline flush to ensure all product is administered, in addition to oseltamivir (Tamiflu) 75 mg BID for total of 5 days (10 doses). Treatment with oseltamivir may be extended for up to 5 additional days (up to 20 doses of total therapy) if clinical symptoms warrant further treatment as assessed by the Investigator. For patients with renal insufficiency, refer to prescribing information for administration of oseltamivir. Note: based on SOC, subjects may have started oseltamivir therapy prior to randomization.	Clarification as some sites use lines with greater than 25 mL hold-up volume
Section 7.3 *Instructions for Preparation, Use and Administration*	The required amount of VIS410 to be dosed (2000 mg or 4000 mg) will be diluted with normal saline up to a total volume of 200 mL. For placebo subjects, 200 mL of normal saline will be prepared. This infusion will be followed by a 25-mL ~~saline flush. Length of~~ (or if IV line ~~will be set for maximum~~ volume ~~of~~ is >25 mL, ~~so that The 25 mL~~ an equivalent volume) saline flush. The saline flush following administration will ensure all VIS410/placebo has been administered. The study infusion will be administered IV using a 0.22-µm in-line filter and will be controlled by a volumetric pump. Standard, ~~uniform length~~ infusion lines will be used, and microfilters will be provided by the Sponsor as appropriate. The infusion bag will be covered with an opaque sleeve in the pharmacy to maintain the study blind. The VIS410/placebo will be administered IV at a rate of 100 mL/h, over 2 hours. When 200 mL has been administered, the line will be flushed with ~~25 mL of~~ normal saline. The infusion time may be longer at the Investigator's discretion based only on local infusion site–related symptoms up to a maximum of 4 hours. Refer to the Pharmacy Manual for directions on storage, handling, stability data, preparation, and use. For more detailed information on VIS410, refer to the current Investigator's Brochure.**Error! Bookmark not defined.**	Clarification to ensure all study product administered. Some sites use infusion lines with hold-up volumes of greater than 25 mL.
Section 8.3.1 *Sequence of Assessments at a Single Visit*	If the following assessments are to be performed at the same study visit, ~~then the order of assessments should be as follows~~ the FluPRO Questionnaire should be completed first, when possible: • FluPRO Questionnaire • ECG • Blood sampling • Nasopharyngeal swab	Clarification
Section 9.2 *Grading of*	Each AE must be graded on a 4-point scale (Grades 1-~~3~~ 4) of increasing intensity according to the Division of	Correction; DMID scale is 1-4

Location in Protocol	Changes	Rationale
Adverse Event Intensity	Microbiology and Infectious Diseases (DMID) Adult Toxicity Table presented in Appendix **Error! Reference source not found.**. Criteria in the DMID table are generally grouped by body system, ie, hematology, chemistries, enzymes, urinalysis, cardiovascular, respiratory, GI, neurological, musculoskeletal, skin, and systemic. For abnormalities not specifically listed in the DMID Table (Appendix **Error! Reference source not found.**), a guide for estimating severity grade (mild, moderate, severe, life-threatening) is provided: • Grade 1: Mild transient or mild discomfort (< 48 hours); no medical intervention/therapy required • Grade 2: Moderate/mild to moderate limitation in activity—some assistance may be needed; no or minimal medical intervention/therapy required • Grade 3: Severe/marked limitation in activity, some assistance usually required; medical intervention/therapy required, hospitalizations possible • Grade 4: Life-threatening: Extreme limitation in activity, significant assistance required; significant medical intervention/therapy required, hospitalization or hospice care probable **Note:** The semi-colon within the description of the grade indicates 'and'. Any clinical event deemed by the clinician to be serious or life-threatening should be considered an SAE.	
Section 10.1 *Determination of Sample Size*	The study is exploratory in nature, and is not powered to demonstrate significant differences between treatment groups in primary or secondary outcome measures. The protocol intent is to collect sufficient information to identify the most appropriate candidate endpoints for subsequent Phase 3 study evaluation from among the primary and secondary endpoints described below. Statistical significance testing will therefore be used to assess the relative strength of evidence of the primary and secondary endpoints, to provide reasonable assurance that the endpoints chosen for a confirmatory Phase 3 trial will elucidate treatment differences between VIS410 plus oseltamivir versus oseltamivir alone. ~~The primary efficacy objective of this study is to evaluate the effect of VIS410 + oseltamivir on the time to normalization of respiratory function compared to oseltamivir alone in the modified intent-to-treat (MITT) population.~~ ~~Using a log-rank test, a sample size of 130 influenza A-infected subjects per treatment group will provide 80% power to detect a 1.5-day difference (5 days for oseltamivir alone and 3.5 days for VIS410 plus oseltamivir) in the median time to cessation of O_2 support for VIS410 relative to placebo. A two-sided alpha of 0.05 was used for the calculation.~~ ~~Approximately 390 evaluable subjects with confirmed influenza A infection will be enrolled in this study.~~	New paragraph due to decreased sample size

Location in Protocol	Changes	Rationale
Section 10.2 *Randomization and Blinding*	An IWRS will be used to allocate the randomized treatments to subjects with stratification by presence or absence of positive pressure ventilation (PPV) at baseline. Pharmacists must obtain the status of the subject relative to PPV at baseline via query, and this data must be input into the IWRS prior to randomization. The randomized treatment assignment will be transferred electronically for integration with the clinical study data at the appropriate time. PPV includes any respiratory assistance using a mechanical ventilation device and can be either invasive or noninvasive ventilation. Invasive PPV includes intubation with endotracheal tube with mechanical ventilation (most common) or tracheostomy with mechanical ventilation. Noninvasive PPV includes use of facemask, nasal plugs, or nasal mask with mechanical ventilation. The devices are named according to the type of mechanical ventilation given continuous positive airway pressure (CPAP) or bi-level positive airway pressure (BiPAP). Randomization will be used to avoid bias in the assignment of subjects to treatment, to increase the likelihood that known and unknown subject attributes (eg, demographic and baseline characteristics) are evenly balanced across treatment arms. The maintenance of the study blind is critical for an unbiased assessment of the safety and efficacy of the study drug. All study staff that evaluate subjects and render decisions regarding subject care will remain blinded to the study treatment each subject receives. The designated study site pharmacist or designee will remain unblinded. Also, unblinded clinical research associates will handle study drug accountability. The study will be conducted in a double-blind manner. The CRO, site study personnel, and Visterra personnel will not be aware of which treatment (VIS410 or placebo) the subjects have been given. Subjects will not be aware of which treatment they have been administered. Subjects will be randomized 1:1:1 to receive VIS410 2000 mg, VIS410 4000 mg, or placebo over a 2-hour infusion with pretreatment regimen of PO or IV diphenhydramine 50 mg plus either ibuprofen 400 mg PO or acetylsalicylic acid 320-325 mg PO approximately 60 minutes before IV infusion of VIS410/placebo. Allocation of each subject to a given treatment sequence will be described in a randomization schedule prepared by Visterra or designee. The randomization will be balanced using randomly permuted blocks across the treatment groups. Based on this randomization code, the site's unblinded pharmacist or trained designee will prepare and dispense VIS410/placebo. ~~If the interim analysis determines that one of the VIS410 treatment arms can be terminated early for futility, then a second randomization schedule with a unique set of randomization numbers will be created for all subsequent subject enrollment and randomization. As with the initial~~	Explanation Due to decrease in sample size an interim analysis is not going to be performed.

Location in Protocol	Changes	Rationale
	~~randomization procedure, randomization will be stratified by presence or absence of PPV at baseline to ensure an even distribution across treatment arms.~~ The randomization schedule will not be available to the subjects, Investigators, blinded monitors, or employees of the clinical center involved in the management of the study before unblinding of the data, unless in case of emergency. The Sponsor's clinical team will also be blinded during the study, as they will not have direct access to the randomization schedule. ~~The Sponsor medical representative(s) may be partially unblinded during the interim analyses. A separate interim analysis plan will provide details on access to unblinded data and the level of unblinding.~~ The CRO personnel performing data management and statistical activities will receive a copy of the randomization schedule during database lock. Other team members will not have access to any data that could lead to unblinding. Unblinding of the individual subject's treatment (via IWRS) by the Investigator should be limited to medical emergencies or urgent clinical situations in which knowledge of the subject's study treatment is necessary for clinical management. In such cases, the Investigator is encouraged to contact the Medical Monitor to discuss and agree to the need for unblinding to occur. In situations in which the Investigator has tried, but is unable to reach the Medical Monitor, they should use their best judgment, based on the nature and urgency of the clinical situation, and may proceed with unblinding without having successfully reached and discussed the situation with the Medical Monitor. Once a subject's treatment assignment has been unblinded, the Medical Monitor should be notified within 24 hours of unblinding of the treatment, without revealing the study treatment. Information relating to unblinding (eg, reason and date) shall be clearly recorded in the subject's study file, as part of relevant standard operating procedures. In addition, the Investigator should consider whether the clinical event prompting unblinding should be considered an SAE, according to the regulatory definitions or criteria for SAEs, and if so, submit an SAE report to Visterra or designee. If the code is broken by the Investigator or by someone of his/her staff, the subject must be withdrawn from the study and must be followed as appropriate. Visterra or designee will also unblind any SAE reports that are serious, unexpected, and considered to be related to investigational product, in accordance with safety reporting guidance and regulations. If the code is broken by the Sponsor for safety reporting purposes, the subject may remain in the study. To maintain study blinding, the infusion bag will be covered by an opaque sleeve in the pharmacy.	
Section 10.5 *Handling of Missing Data*	~~No imputations~~ Procedures for handling missing data will be ~~performed~~ defined in the Statistical Analysis Plan, and may include last observation carried forward or an interpolation method.	Clarification

Location in Protocol	Changes	Rationale
Section 10.6 *Interim Analysis*	~~An interim analysis of efficacy may be performed by an unblinded third party after 50% of subjects have been enrolled (n = 195) and have an assessment of the primary endpoint (time to normalization of respiratory function). This interim analysis may be conducted to assess if one of the active VIS410 treatment arms will need to be terminated for futility. Using the methods of Lan and DeMets an alpha spending function will be created and if the test statistic for one of the active VIS410 treatment arms is less than the lower critical point then the VIS410 treatment arm may be terminated for futility. The interim analysis may also test the primary efficacy objective to assess if the primary efficacy endpoint has been met early.~~ ~~The effect size for this study was estimated without the benefit of any prior randomized, controlled trial. If the VIS410 effect size proves to be greater than anticipated, then it is possible that the primary study endpoint will be met by the time the interim efficacy analysis is performed. In the event the interim analysis reveals that the primary efficacy endpoint has been satisfied, this Phase 2 study could be terminated for efficacy.~~ ~~The DSMB and the unblinded statistician will review the interim analysis results to provide their recommendation on discontinuation of one of the VIS410 treatment arms, if needed, based on pre-specified criteria as outlined in the SAP. The interim analysis may also review the following key secondary endpoints, when available, to assist in the decision of discontinuing a VIS410 treatment arm:~~ ~~• Time to clinical resolution of vital signs~~ ~~• Total number of hours on oxygen support or PPV~~ ~~• Viral load from nasopharyngeal swabs~~ ~~A sample size re-estimation (SSRE) will also be performed at this point in order to determine if the current sample size is sufficient based on the observed effect size in the study. The unblinded statisticians will work directly with the Sponsor on the SSRE. The details of SSRE and individuals with access to the information will be outlined in a separate plan. The MITT population will be used for the interim analysis.~~	Due to decrease in sample size an interim analysis is not going to be performed.
Section 10.7 *Efficacy Analyses*	Section number changed to 10.6 **Efficacy Analyses** Efficacy analyses will be performed using the MITT population. Efficacy analysis will also be performed in the PP population to demonstrate consistency with the primary analysis population. **Day 7 Ordinal Scale Status** The ordinal scale outcomes will be measured daily from Day 1 (baseline) through Day 14 using the seven-level hierarchical scale with the classifications presented in Table 8~~Table 2~~. The clinical outcomes therein are ordered from the worst clinical outcome to the best clinical outcome in descending order. For each day, subjects will be classified by the worst clinical outcome for which they qualify.	Renumbered due to deletion of previous section 10.6 Moved this section to the top of the efficacy analyses as a component of the ordinal scale is now the primary objective/primary endpoint

Location in Protocol	Changes	Rationale
	The number and percentage of subjects in each treatment group with each classification will be summarized for each day from Day 1 through Day 14, inclusive. **Table 1. Hierarchical Seven-Level Ordinal Scale for Clinical Outcomes** **Clinical Parameter** Death ICU stay with mechanical ventilation ICU stay without mechanical ventilation Non-ICU hospitalization with supplemental oxygen Non-ICU hospitalization without supplemental oxygen Discharge with partial resumption of normal activities Discharge with full resumption of normal activities **Note:** Clinical outcomes listed from worst clinical outcome to best clinical outcome in descending order. **Time to Cessation of O_2 Support** Time to cessation of O_2 support resulting in a stable SpO_2will be analyzed using a Cox model. Time from onset of symptoms to VIS410 treatment and the number of doses of oseltamivir (Tamiflu) prior to VIS410 treatment for the current case of influenza will be included as a covariate in the analysis. A *P*-value (using the Wald statistic) for each VIS410 dose vs placebo will be presented. Secondarily, subgroup analyses will be performed to describe the time to cessation of O_2 for subgroups, such as use of positive pressure ventilation, use of endotracheal intubation, time from onset of symptom to VIS410 therapy, number of oseltamivir doses prior to VIS410, elderly, and underlying lung disease. Clinical and virologic endpoints will also be evaluated by influenza A subtypes. **Time to Clinical Response** Time to clinical response is defined as resolution of 4 of 5 vital signs that will be determined upon physical examination. Clinical response is defined as: • Afebrile with core temperature ≤ 37.8°C, without use of antipyretics (oral ≤ 37.2°C) • Respiratory rate ≤ 24 beats per minute • Oxygen saturation ≥ 95% on room air without support or a return to pre-infection status, if pre-infection status was < 95% • Pulse rate ≤ 100/min • SBP ≥ 90 mm/Hg, without vasopressor use The probability of time to clinical response (defined as resolution of vital signs) will be calculated via Kaplan-Meier. A ~~P value~~ significance test (using the log-rank test) for each VIS410 dose vs placebo will be presented. The number and percentage of subjects in each treatment group with clinical response will be summarized. Results will be tabulated and presented graphically as well.	

	Time to Cessation of Ventilator Support The probability of time to cessation of ventilator support will be calculated via Kaplan-Meier. A P-value (using the log-rank test) for each VIS410 dose vs placebo will be presented. Results will be tabulated and presented graphically as well. **Healthcare Resource Utilization** Descriptive statistics will be used to compare the total number of days in the hospital and/or ICU from admission to discharge, number of subjects requiring ICU admission post-randomization, overall number of days in the ICU, number of hours on ventilation, rehospitalization due to influenza A relapse/reinfection, the total number of days of oseltamivir therapy, and the total number of days to resumption of usual activities by treatment group. Time (number of days) to resumption of usual activities will be determined from the VAS (scale ranged from 0 to 10, where 0 indicates subject is unable to perform any of his/her usual activities prior to influenza onset, and 10 indicates subject is able to fully perform all usual activities). This evaluation will be performed using the MITT population. **Signs and Symptoms of Influenza** Descriptive statistics will be used to compare the duration of symptoms of influenza-like illness in the subset of subjects able to complete the FluPRO Questionnaire at baseline and post-dose by treatment group. The number and percentage of subjects who were not able to complete the assessment at each visit will be summarized. Frequency tabulation of the occurrence and severity of each subject-reported symptom of influenza-like illness (via FluPRO Questionnaire) will be summarized by assessment time point and by treatment group. Time to resolution of symptoms will be evaluated by Kaplan Meier analysis. Analyses of the signs and symptoms of influenza will be conducted only on the subset of the MITT population who had baseline FluPRO Questionnaire assessments. Additional populations may be analyzed as described in the SAP. ~~**Clinical Outcome by Seven-Level Ordinal Scale**~~ ~~The ordinal scale outcomes will be measured daily from Day 1 (baseline) through Day 14 using the seven-level hierarchical scale with the classifications presented in **Table 2**. The clinical outcomes therein are ordered from the worst clinical outcome to the best clinical outcome in descending order. For each day, subjects will be classified by the worst clinical outcome for which they qualify.~~ ~~The number and percentage of subjects in each treatment group with each classification will be summarized for each day from Day 1 through Day 14, inclusive.~~ ~~**Table 2. Hierarchical Seven-Level Ordinal Scale for Clinical Outcomes**~~ ~~**Clinical Parameter**~~	Moved this section to the top of the efficacy analyses as a component of the ordinal scale is now the primary objective/primary endpoint.

Location in Protocol	Changes	Rationale
	~~Death~~ ~~ICU stay with mechanical ventilation~~ ~~ICU stay without mechanical ventilation~~ ~~Non-ICU hospitalization with supplemental oxygen~~ ~~Non-ICU hospitalization without supplemental oxygen~~ ~~Discharge with partial resumption of normal activities~~ ~~Discharge with full resumption of normal activities~~ ~~**Note:** Clinical outcomes listed from worst clinical outcome to best clinical outcome in descending order.~~	
Section 10.8.3 *Complications of Influenza*	Now Section 10.7.3 Complications of influenza are defined as pneumonia, myocarditis, worsening of chronic bronchitis, sinusitis, otitis or other chronic pulmonary diseases, encephalopathy, and death. Complications of influenza and use of antibiotic therapy for bacterial pneumonia will be reported by variable, treatment group, and time point. Diagnosis of bacterial pneumonia will be based on local clinical practice, usually an assessment of clinical features and any radiographic evidence of bacterial pneumonia (eg, infiltrate) and/or microbiologic evidence of bacterial pneumonia. All-cause and attributable mortality will be evaluated on Days 14 and 28.	Clarification
Sections 10.8 - 10.12	Renumbered sections due to deletion of original section 10.6; now 10.7 - 10.11	Correction
Section 12.1.2 *Subject Informed Consent*	Each subject or a legally ~~authorized~~ acceptable representative must give written consent according to local requirements after the nature of the study has been fully explained. The consent form must be signed before performance of any study-related activity. The consent form that is used must be approved by both the Sponsor and by the reviewing IEC/IRB. The informed consent should be in accordance with principles that originated in the Declaration of Helsinki, current ICH and GCP guidelines, applicable regulatory requirements, and Sponsor policy. Before enrolling potential subjects in the study, the Investigator or an authorized member of the investigational staff must explain to the subject the aims, methods, reasonably anticipated benefits, and potential hazards of the study, and any discomfort that participation in the study may entail. Subjects will be told that the Investigator will maintain a subject identification register for the purposes of long-term follow up if needed and that their records may be accessed by health authorities and authorized Sponsor staff without violating the confidentiality of the subject, to the extent permitted by the applicable law(s) or regulations. By signing the Informed Consent Form (ICF) the subject is authorizing such access and agrees to allow his/her study physician to recontact the subject for the purpose of obtaining consent for additional safety evaluations, if needed. The language used in the oral and written information about the study, including the ICF, should be nontechnical	Change requested by an Ethics Committee

Location in Protocol	Changes	Rationale
	and practical and should be understandable to the subject or the subject's legal representative. The subject will be given sufficient time to read the ICF and the opportunity to ask questions. After this explanation and before entry into the study, consent should be appropriately recorded by means of the subject's personally dated signature. After having obtained the consent, a copy of the ICF must be given to the subject. If the subject is unable to read or write, an impartial witness should be present for the entire informed consent process (which includes reading and explaining all written information) and should personally date and sign the ICF after the oral consent of the subject is obtained, if permitted by local law.	
Section 14.2 *Laboratory Assessments*	**Other Assessments** Urine pregnancy test Erythrocyte Sedimentation Rate (ESR) C-Reactive Protein (CRP)	Added to complete the assays being performed

[i] Treanor JJ, Hayden FG, Vrooman PS, et al. Efficacy and safety of the oral neuraminidase inhibitor oseltamivir in treating acute influenza: a randomized controlled trial. US Oral Neuraminidase Study Group. *JAMA* 2000;283(8):1016–24.
[ii] Schanzer DL, Langley JM, Tam TW. Co-morbidities associated with influenza attributed mortality, 1994-2000, Canada. *Vaccine* 2008;26(36):4697–703.
[iii] CDC. Estimates of deaths associated with seasonal influenza US, 1976-2007. MMWR. 2012;59(33):1057–62.
[iv] WHO. Seasonal Influenza Fact sheet N°211. 2014. http://www.who.int/mediacentre/factsheets/fs211/en. Accessed 10 Mar 2014.
[v] World Health Organization (WHO). Pandemic (H1N1) 2009-update 70. http://www.who.int/csr/don/2009_10_16/en/index.html. Accessed 25 October 2009.

Visterra

PROTOCOL AMENDMENT – SUMMARY OF CHANGES

Phase 2b, Multicenter, Randomized, Double-blind, Controlled Study to Evaluate the Efficacy and Safety of Intravenous VIS410 in Addition to Oseltamivir (Tamiflu®) Compared With Oseltamivir Alone in Hospitalized Adults With Influenza A Infection Requiring Oxygen Support

Product	VIS410
Protocol Number	VIS410-203
EudraCT Number	2016-004009-15
Clinical Phase	2b
Clinical Indication	Influenza A infection

Sponsor	Visterra, Inc. One Kendall Square, Suite B3301 Cambridge, MA 02139 United States of America
Sponsor Representative	[redacted]

ORIGINAL PROTOCOL DATE: **25 October 2016**

AMENDMENT 1 DATE (SOUTH AFRICA): **3 July 2017**

**SUMMARY OF CHANGES TO PROTOCOL VIS410-203
FROM VERSION 1.1 TO VERSION 2.1 (SOUTH AFRICA)**

Substantive changes to the protocol for Amendment 1 and their location within the protocol are noted below. Additions are marked as red underlined text and deletions are marked as red strikethrough text. Administrative, stylistic and formatting changes that do not alter the conduct of the study are not summarized in this document.

<table>
<tr><th>Location in Protocol</th><th>Changes</th><th>Rationale</th></tr>
<tr><td>Cover Page;
Signature of Sponsor Representative</td><td>Issue Date (Version): ~~25 October 2016~~ (Version ~~1~~.1)
3 July 2017 (Version 2.1 – South Africa)

Sponsor Representative [redacted]</td><td>Updated date and version number; added South Africa to identify country level protocol.

Sponsor representative updated.</td></tr>
<tr><td>Protocol History</td><td>~~Protocol History~~
~~Visterra, Inc. VIS410-203~~<table><tr><th>~~Document~~</th><th>~~Issue Date~~</th><th>~~Amendment Type~~</th><th>~~Comments~~</th></tr><tr><td>~~Clinical Study Protocol~~</td><td>~~25 October 2016 (Version 1.1)~~</td><td>~~-~~</td><td>~~This document~~</td></tr></table></td><td>Deleted because changes will be summarized in a separate Summary of Changes document (this file).</td></tr>
<tr><td>Synopsis and Section 2.2 Secondary Objectives</td><td>• Evaluate the effect of VIS410 + oseltamivir vs oseltamivir alone on the following endpoints:
▪ Viral ~~titer~~ load in upper respiratory samples
▪ Time to clinical response
▪ Time to cessation of ventilator support
▪ Time to resumption of normal activities
▪ All-cause and attributable 14- and 28-day mortality
▪ Healthcare resource utilization
▪ Time to alleviation of clinical symptoms of influenza in subset of subjects able to complete the FluPRO Questionnaire at baseline and post-dose
▪ Proportion of subjects with new documented bacterial pneumonia/superinfection
▪ Proportion of subjects with influenza-related complications</td><td>Viral load is a more accurate term to represent the test being conducted.</td></tr>
<tr><td>Synopsis;
Section 2.3 Exploratory Objectives;
Section 3.3 Exploratory Endpoints;
Section 6.3.12.3 Serum Cytokines</td><td>• Evaluate the effect of VIS410 + oseltamivir vs oseltamivir alone on clinical outcomes as measured by a seven-level ordinal scale
• Evaluate the pharmacokinetics of VIS410 from nasopharyngeal secretions and tracheal aspirate (ventilated subjects only)
• Assess the effects of VIS410 on viral ~~titer~~ load in tracheal aspirate (ventilated subjects only)
• Assess correlations between virology, safety, VIS410 dose, pharmacokinetics, viral shedding, ~~immunogenicity~~ immunology, signs and symptoms of influenza, and other endpoints
• Assess the anti-influenza immune response</td><td>Viral load and immunology are more accurate terms.

Exploratory cytokine objective removed from protocol.</td></tr>
</table>

Location in Protocol	Changes	Rationale
	• ~~Evaluate serum cytokine profiles~~	

Location in Protocol	Changes	Rationale
Synopsis; Section 4.0 *Study Design;* Section 4.3 *Data Safety Monitoring Board;* Section 6.1.3 *Study Procedures;* Section 7.5 *Pretreatment;* Section 8.3.3 *Post-Treatment Procedures (Day 3 to 56);* Section 11.4 *Stopping Rules or Discontinuation Criteria*	This is a Phase 2b multicenter, randomized, double-blind, controlled study comparing the efficacy and safety of 2 dose levels of VIS410 (2000 and 4000 mg) in combination with oseltamivir vs oseltamivir alone in hospitalized ~~severely ill~~ subjects with influenza A infection requiring oxygen. Subjects will be randomly assigned at a ratio of 1:1:1 using a permuted block randomization method to receive VIS410 at a dose of 2000 mg (low dose) or 4000 mg (high dose) or placebo (0.9% sodium chloride), administered as a single IV infusion over 2 hours. Randomization will be stratified by presence or absence of positive pressure ventilation (PPV) at baseline to ensure an even distribution across treatment arms (see Section 10.2). All subjects will receive oseltamivir 75 mg twice daily (BID) for 5 days (total of 10 doses) as part of standard of care (SOC). Treatment with oseltamivir may be extended for up to 5 additional days (up to 20 doses of total therapy) if clinical symptoms warrant further treatment as assessed by the Investigator. Note: based on SOC, subjects may have started oseltamivir therapy prior to randomization. All subjects will also receive a pretreatment regimen of diphenhydramine 50 mg PO or IV plus either ibuprofen 400 mg PO or acetylsalicylic acid 320-325 mg PO 60 minutes before VIS410/placebo infusion. Approximately 390 evaluable subjects (130/arm) with confirmed influenza A infection will be treated. Subjects admitted to the hospital within 5 days of onset of initial symptoms who require supplemental oxygen ~~of at least 40% (4 L/min) and/or hypoxemia defined as SpO_2 of less than 90%~~ will undergo a rapid influenza test (supplied by the Sponsor) or a PCR test, fluorescent immunoassay (FIA) test, or enzyme-linked immunosorbent assay (ELISA) test to confirm influenza A infection. Subjects diagnosed with influenza A will undergo the screening procedures to confirm eligibility. Study assessments are outlined in Table 1, Schedule of Assessments. Eligible subjects will be randomized to receive either oseltamivir + VIS410 2000 mg, oseltamivir + VIS410 4000 mg, or oseltamivir + placebo (0.9% sodium chloride) at a ratio of 1:1:1 using a permuted block randomization method summarized in Table 6. **(Randomization table, unchanged)** Oseltamivir (Tamiflu®) will be provided by the Sponsor. Treatment with oseltamivir may be extended for up to 5 additional days (up to 20 doses of total therapy) if clinical symptoms ~~are not resolved by Day 5 or~~ warrant further treatment as assessed by the Investigator. Note: based on SOC, subjects may have started oseltamivir therapy prior to randomization. Study assessments are outlined in Table 1. Subjects discharged from ~~will be monitored daily while in~~ the hospital prior ~~up~~ to Day 14 will attend ~~(± 3 days) with additional visits on Day 28 (± 3 days) and~~ the applicable outpatient ~~last~~ follow-up visit(s) (eg, Day 3, Day 5, ~~on~~ Day~~56 (± 7 days). Study assessments are outlined in~~, Day 14) per Table 1 ~~(Schedule of Assessments)~~.	Based on change to Inclusion Criteria regarding oxygen requirement, it is more accurate to define subjects as hospitalized, not severely ill. Clarified that subjects will receive either ibuprofen or acetylsalicylic acid, not both. Dose for acetylsalicylic acid changed to comply with standard dose in various countries. Inclusion Criteria regarding oxygen requirement has been modified to allow participation of less severe infections, hence subjects no longer require a specific concentration or amount of oxygen support to be eligible for the study. Clarified that subjects may have started oseltamivir treatment prior to enrollment in the study; total minimum and maximum doses the same for all subjects regardless of when a subject began oseltamivir. Clarified that subjects discharged from the hospital will attend follow-up visits; the number of outpatient follow-up visits are dependent on when the subject is discharged from the hospital. Defined GI TEAEs that require intervention. Updated statistical language related to the error rate and the interim analysis. The DSMB will review safety data after

Location in Protocol	Changes	Rationale
	An independent data safety monitoring board (DSMB) will be established to review all available safety data after 30 subjects as well as when ~~and~~ approximately 120 subjects have completed study Day 14 ~~7~~. The assessment of safety will be determined from vital sign measurements, physical examinations, hematology, chemistry and urinalysis laboratory testing, 12-lead electrocardiograms (ECGs), use of concomitant medications, and review of adverse events (AEs). Based on the safety data provided, the DSMB will determine if it is acceptable to continue the study or reduce the dose as appropriate. Dosing will temporarily pause while the DSMB meets if there are 4 treatment-related serious adverse events (SAEs) or 4 severe gastrointestinal (GI) treatment–emergent adverse events (TEAEs) that require intervention, which is defined as requiring IV fluid and medication to decrease the frequency of diarrhea (ie, loperamide). Additional DSMB reviews can occur throughout the trial as deemed necessary by the DSMB or Sponsor to ensure subject safety and well-being. Study enrollment and dosing will continue while the DSMB evaluates data. In addition, following 50% enrollment (195 subjects), an interim analysis may ~~will~~ be conducted, by an unblinded third party, to assess if one of the VIS410 treatment arms can be terminated early for futility. A prespecified sample size reanalysis may also be conducted based on the observed effect size. The interim analysis may ~~will~~ also test the primary efficacy objective to assess if the primary efficacy endpoint has been met. An alpha spending function will be designed in order to determine the amount of alpha that will be spent at the interim analysis. The effect size for this study was estimated without the benefit of any prior randomized controlled trial. If the VIS410 effect size proves to be greater than anticipated, then it is possible that the primary study endpoint will be met by the time the interim efficacy analysis is performed. In the event the interim analysis reveals that the primary efficacy endpoint has been satisfied, this Phase 2 study could be terminated for efficacy.	approximately 30 subjects and 120 subjects complete Day 14 to allow more safety data to be reviewed.
Synopsis and Section 5.2.1 *Inclusion Criteria*	**Only revised list items shown:** 4. Requirement for oxygen support including any positive pressure ventilation (PPV). ~~4. Need for supplemental oxygen of at least 40% (4 L/min) and/or hypoxemia defined as SpO2 of less than 90%~~	Inclusion Criteria regarding oxygen requirement has been modified to allow participation of less severe infections, hence subjects no longer require a specific concentration or amount of oxygen support to be eligible for the study.
Synopsis and Section 5.2.2 *Exclusion Criteria*	**Only revised list items shown:** 2. Subjects who have received VIS410 in the past ~~History of receiving monoclonal antibody products (including VIS410) within 3 months prior to VIS410/placebo dosing or planned administration during the study period~~. 3. Subjects who have a h~~i~~story of receiving monoclonal antibody products within 3 months prior to VIS410/placebo dosing or planned administration of	Clarified that subjects are not eligible for the study if they have received VIS410 in the past. Clarified exclusion criteria of subjects that have Influenza B or another viral

Location in Protocol	Changes	Rationale
	another monoclonal antibody during the study period. 5. Subjects with known co-infection with influenza B or other viral respiratory infections (eg, respiratory syncytial virus [RSV], parainfluenza viruses, respiratory adenoviruses).~~infection~~ 6. Subjects with lung transplant or history of severe chronic lung disease, including cystic fibrosis or any condition requiring ~~> 2 L/minute of~~ home oxygen therapy.~~(ie, severe chronic obstructive pulmonary disease [COPD], pulmonary fibrosis)~~ 7. Subjects on extracorporeal membrane oxygenation ~~(ECMO)~~ at time of randomization. ~~8. Hospitalization for > 48 hours prior to randomization~~ 8. Subjects with end-stage renal disease (ESRD) who are not undergoing hemodialysis.	respiratory infection. Clarified exclusion criteria of cystic fibroses subjects; due to changes in patient population , subjects that require oxygen therapy at home are not eligible for the study. Subjects hospitalized before enrollment in the study are eligible as long as the onset of influenza symptoms are no more than 5 days before treatment with VIS410/ placebo (Inclusion Criteria 3). For consistency with oseltamivir prescribing information, subjects with ESRD who are not undergoing hemodialysis are excluded from the study.
Synopsis; Section 7.1 *Test Product, Dose, Mode of Administration;* Section 7.2 *Reference Product, Dose, Mode of Administration*	**¶ 2 only:** VIS410 (2000 mg or 4000 mg) will be administered IV over 2 hours as a single, 200-mL infusion, followed by a 25-mL saline flush to ensure all product is administered, in addition to oseltamivir (Tamiflu) 75 mg BID for total of 5 days (10 doses). Treatment with oseltamivir may be extended for up to 5 additional days (up to 20 doses of total therapy) if clinical symptoms ~~are not resolved by Day 5 and/or~~ warrant further treatment as assessed by the Investigator. Note: based on SOC, subjects may have started oseltamivir therapy prior to randomization. No dose adjustment is necessary for VIS410 based on renal or hepatic impairment. For patients with renal insufficiency, r~~R~~efer to prescribing information for administration of oseltamivir ~~in patients with renal insufficiency~~.	Clarified that subjects are required to be treated with oseltamivir for a minimum of 5 days but that treatment may be extended up to 10 days. Clarified that subjects may have started oseltamivir treatment prior to enrollment in the study; the total minimum and maximum doses are the same for all subjects regardless of when a subject began oseltamivir.
Synopsis and Section 7.3 *Instructions for Preparation, Use, and Administration*	The infusion bag will be covered with an opaque sleeve in the pharmacy to maintain the study blind. The VIS410/placebo will be administered IV at a rate of 100 mL/h, over 2 hours. After 200 mL of the diluted dose has been administered, the infusion will be stopped, followed by a 25-mL saline flush. The infusion time may be extended up to 4 hours at the Investigator's discretion based on local infusion site–related symptoms.~~ VIS410 or placebo will be administered within 48 hours of being prepared by the study pharmacist.~~ When 200 mL has been administered, the line will be flushed with 25 mL of normal saline. The infusion time may be longer at the Investigator's discretion based only on local infusion site–related symptoms up to a maximum of 4 hours.	The window for administration of VIS410/placebo after preparation by the pharmacist has been deleted and will be noted in the Pharmacy Manual.

Location in Protocol	Changes	Rationale
	~~VIS410 or placebo will be administered within 48 hours of being prepared by the study pharmacist. After preparation, the VIS410/placebo infusion bag should be stored at room temperature.~~	
Synopsis and Section 3.1.1 *Primary Efficacy Endpoint*	Time to cessation of O_2 support resulting in ~~a~~ stable SpO_2 by pulse oximetry. Stable SpO_2 is defined as two consecutive SpO_2 values of > 92% on room air that are~~> 95% for~~ at least ~~8~~6 hours apart. ~~on room air, return to baseline respiratory status, or hospital discharge with no need for additional O_2 support, whichever occurs first.~~	Time to cessation of O_2 support redefined based on update to Inclusion Criteria regarding oxygen requirement.
Synopsis and Section 3.2 *Secondary Endpoints*	**Only revised bullet with its sub-bullets shown:** • Time to clinical response defined as resolution of at least 4 of 5 vital signs ▪ Afebrile with core temperature ≤ 37.8°C, without use of antipyretics (oral ≤ 37.2°C) ▪ Oxygen saturation ≥~~>~~ 95% on room air without support or a return to pre-infection status, if pre-infection status was < 95% ▪ Pulse rate ≤ 100/min ▪ Systolic blood pressure ~~(SBP)~~ ≥ 90 mm/Hg, without vasopressor use ▪ Respiratory rate ≤~~<~~ 24 beats per minute	Specified that temperature endpoint specific to core temperature; also defined equivalent oral temperature. Updated definition of time to clinical response for oxygen saturation and respiratory rate.
Section 6.1.1 *Prescreening Procedures*	Subjects may be asked to sign a Prescreening Consent prior to undergoing a single nasopharyngeal swab (one nostril) for rapid influenza A testing. Subjects will be given a full explanation of the nature of the study and provide full written informed consent before any study-specific assessments or procedures are performed. Note: subjects that tested positive for influenza A within 48 hours prior to screening do not need to be retested. ~~This screening test is not necessary if the subject has a prior influenza A positive test by RAT or with another commercially available test including PCR, FIA, or ELISA within the prior 48 hours of screening.~~	Clarified that a pre-screening consent to perform rapid influenza testing is optional as this may depend on country and site requirements.
Section 6.2 *Pregnancy Safeguards*	Pregnancy will be determined by evaluation of β-human chorionic gonadotropin in serum or urine for all women of childbearing potential. Subjects who are pregnant or nursing will be excluded from the study. During the course of the study drug administration period within the study, any nursing mother(s) or subject(s) with suspected or confirmed pregnancy will be discontinued from study drug therapy but will be encouraged to undergo follow-up for safety monitoring for themselves (ie, the pregnant female) and the baby. All women of childbearing potential and all male subjects must practice effective contraception from screening until 60 days post-VIS410/placebo infusion. For the purposes of this study, women who do not satisfy at least one of the following criteria listed below (ie, the criteria for defining non-childbearing potential) are considered to be physiologically capable of becoming pregnant and are, therefore, defined as women of childbearing potential. The criteria for defining women as being of non-childbearing potential are:	Clarified that any pregnancy should be reported up until 60 days post VIS410/placebo dosing. Clarified the contraception requirements for women of childbearing potential and all male subjects.

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	• Post-menopausal: ≥ 12 months of natural (spontaneous) amenorrhea, or ~~• Six weeks after surgical bilateral oophorectomy with or without hysterectomy, or~~ • Follicle stimulating hormone > 40 mIU/mL as documented in their medical history, or • Surgical bilateral oophorectomy with or without hysterectomy, or • Hysterectomy, or • Bilateral tubal ligation Women of childbearing potential and all male subjects participating in heterosexual relations must be willing to practice effective contraception from screening until 60 days post-VIS410/placebo infusion. For the purposes of the study, highly effective contraception is defined as: • Male vasectomy with negative semen analysis documentation at least 6 months prior to dosing. OR • Combination of an established form of hormonal contraception (oral, injected, or implanted) or an intrauterine device or intrauterine system Plus one of the following: ~~And~~ • A physical barrier method of contraception with use of a spermicide, such as condom or occlusive cap (diaphragm or cervical/vault caps) with spermicidal foam/gel/film/cream suppository. The use of barrier contraceptives should always be supplemented with the use of a spermicide, unless not available in a country. OR • Male vasectomy with negative semen analysis documentation less than ~~The use of contraception does not apply if the male partner has been vasectomized at least~~ 6 months prior to dosing. OR • Complete abstinence can be considered an acceptable method of contraception at the discretion of the Investigator. Periodic abstinences (eg, calendar, ovulation, symptothermal, or post-ovulation methods) and withdrawal are NOT considered acceptable methods of contraception. The combination of 2 barrier methods, periodic abstinence (eg, calendar, ovulation, symptothermal, or post-ovulation methods), and withdrawal are not considered acceptable methods of contraception. Pregnancy reporting is described in Section 9.7.3.	
Section 6.3.1 *SpO_2 Oxygen Support*	Baseline SpO_2 on room air to be documented, if available. Once randomized, SpO_2 will be measured using pulse oximetry 3 times per day (approximately every ~~preferably~~ 8 hours ~~apart~~) at approximately the same time each day until stable according to Table 1. Stable ~~the schedule of assessment.~~ SpO_2 is defined as two consecutive SpO_2 values of > 92% ~~will be measured using pulse oximetry until > 95%~~	Clarified time points for obtaining SpO_2 measurements and definition of stable SpO_2.

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	~~for at least 6 hours~~ on room air that are at least 8 hours apart.	
Synopsis; Section 6.3.2; Section 6.3.11; Section 10.10	**Section subheading titles only** 6.3.2 *Viral Load ~~Titer~~* 6.3.11 Pharmacokinetics and Viral Load ~~Titer~~ in Tracheal Aspirate 10.10 *Viral Load ~~Titer~~*	Section heading only revised.
Section 6.3.4 *Resumption of Normal Activities*	Subjects will complete a visual analog scale (VAS) daily from Day 1 (baseline) until either the subject reports that all usual activities (prior to influenza onset) can be performed or Day 56, whichever comes first. The VAS scale ranges ~~scale ranged~~ from 0 to 10, where 0 indicates subject is unable to perform usual activities at all, and 10 indicates subject is able to perform all usual~~) according to the time points defined in~~ Error! Reference source not found.activities fully)	Added language to require a subject to complete VAS from baseline until the subject reports they are able to perform all usual activities they were performing prior to influenza onset.
Section 6.3.8 *Viral Resistance*	Samples for virologic and PK analysis ~~A nasopharyngeal swab~~ will be collected ~~from each nostril~~ as described in Table 1 and the laboratory manual. These samples ~~swabs~~ will be sent to a central virology laboratory for processing into appropriate aliquots for virology and PK analysis, including resistance testing. Further procedures for sample collection, processing, shipment, and storage will be described in the laboratory manual. Note**:** Nasopharyngeal and tracheal samples taken from all subjects may be infectious and will be classified as "diagnostic specimens" for dispatch purposes.	Updated language to more accurately describe virologic analysis which includes PK analysis and resistance testing.
Section 6.3.12 *Immunology ~~Immunogenicity~~*	**¶ 1 only:** The immunology ~~immunogenicity~~ parameters under evaluation are the detection of ADA titers and anti-influenza A antibody titers by ~~hemagglutination inhibition assay (~~HAI~~), and serum cytokine concentrations~~.	Updated language to more accurately describe immunology evaluation. Exploratory cytokine objective removed from protocol.
Section 6.5.3 *Complications of Influenza;* Section 10.8.3 *Complications of Influenza*	Complications of influenza are defined as pneumonia, myocarditis, worsening of chronic bronchitis, sinusitis, otitis, other chronic pulmonary diseases, encephalopathy, and death. Complications of influenza and use of antibiotic therapy for bacterial pneumonia will be reported by variable, treatment group, and time point. Diagnosis of bacterial pneumonia will be based on local clinical practice, usually an assessment of clinical features and any radiographic evidence of bacterial pneumonia (eg, infiltrate) and/or microbiologic evidence of bacterial pneumonia. All-cause and attributable mortality will be evaluated on Day 14 and Day 28.	Clarified that diagnosis of bacterial pneumonia will be based on local clinical practice.
Section 6.5.4 *Clinical Laboratory Tests*	Blood samples will be collected by venipuncture or via indwelling cannula at the time points indicated in Table 1. ~~Standard laboratory tests will be performed by a central or local laboratory.~~ Appendix 14.2 lists the biochemistry, hematology, and coagulation~~, and serology~~ tests that will be performed by the central laboratory on the safety blood samples. Note: subjects to be enrolled based on the Investigator's discretion including any local laboratory	Updated language to state that the required baseline laboratory tests will be sent to the central laboratory for consistency of data for trial analysis. However, for enrollment, Investigators should use local laboratory

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	results per SOC at institution. Creatine kinase-MB, creatinine kinase, and troponin are to be measured by the central laboratory in the event of a subject having chest pain. Subjects with chest pain should be managed per SOC. In the event of a subject exhibiting the signs and symptoms of an anaphylactic reaction, when possible at the time of the event, a 5-ml serum sample should be collected for further assessment (ie, tryptase, chymase) to be measured by the central laboratory. A midstream urine sample will be collected for the central laboratory for urinalysis by dipstick, flow cytometry, and microscopic examination. Appendix 14.2 lists the urinalysis parameters that will be assessed. A serum ~~Serum or urine~~ pregnancy tests will be performed by ~~at~~ the central laboratory for all time points indicated in Table 1. In addition, a negative Pregnancy test (serum or urine) result within 2 days prior to dosing ~~results~~ must be available from the local laboratory prior to randomization. ~~A negative result is required for randomization.~~ For oseltamivir dosing, creatinine clearance (Cockcroft-Gault Equation) must be calculated using the serum creatinine value from the local laboratory prior to randomization. The Investigator must review the laboratory report, document this review, and record any change occurring during the study he/she considers to be clinically relevant in the EDC system. Laboratory values outside the normal range will be flagged, and their clinical relevance will be assessed by the Investigator.	testing for pregnancy and serum creatinine. Subject's overall health for participation in the trial should be based on institutional practices and local laboratory results. Clarified that laboratory tests required in the case of a subject experiencing chest pain will be sent to the central laboratory for analysis while these subjects should be managed based on institutional practices and Investigator discretion. Clarified that all pregnancy testing will be performed at the central laboratory but that a negative pregnancy test performed by the local laboratory is required within 2 days of dosing. Specified that creatinine clearance should be calculated by using the Cockcroft-Gault equation.
Section 6.5.5 *Vital Signs and Body Temperature*	Vital sign parameters will be assessed after 5 minutes in supine position at the time points indicated in Table 1. The vital sign parameters that will be assessed are supine SBP, DBP, heart rate, and respiratory rate. Any change from baseline in vital sign values occurring during the study that is considered to be clinically relevant by the Investigator should be recorded as an AE ~~adverse event~~. Oral body temperature will be recorded at the end of infusion then BID while in the hospital and then throughout the study according to the study procedures outlined in Table 1**Error! Reference source not found.**. In cases where obtaining oral temperature is not possible, core temperature (eg, tympanic, axillary) will be obtained. While the patient is hospitalized, the maximum temperature should be recorded for each 12-hour interval (from 12 AM to 12 PM and from 12 PM to 12 AM). Fever is defined as a core body temperature ≥ 38°C.	Provided examples of core temperature methods. Clarified that fever is defined based on core temperature.
Section 6.5.8 *Radiological Assessment*	A baseline chest x-ray or CT scan will be performed for the assessment of pneumonia. However, a chest x-ray or CT scan performed as part of routine SOC ~~standard of care~~ within 72 ~~48~~ hours before randomization will be acceptable. The results of any such studies will be recorded in the EDC.	The window has been expanded to allow use of local tests for diagnosis of pneumonia and minimize unnecessary tests for the purpose of the study. The

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		72-hour window will allow detection of pneumonia at baseline, if present.
Section 7.4 *The Use of Oseltamivir as Standard of Care*	All subjects will receive oseltamivir 75 mg BID for 5 days as part of ~~standard of care (~~SOC~~).~~ Treatment with oseltamivir may be extended for up to 5 additional days (up to 20 doses of total therapy) if clinical symptoms ~~are not resolved by Day 5 or~~ warrant further treatment as assessed by the Investigator. Oseltamivir (Tamiflu) will be provided by the Sponsor. Note: based on SOC, subjects may have started oseltamivir therapy prior to randomization. For patients unable to swallow, oseltamivir capsules can be opened and mixed with liquid for ease of administration. Dose adjustment is recommended for patients with a serum creatinine clearance of 60 mL/min or less[9] (see Table 7).	Clarified that subjects may have started oseltamivir treatment prior to enrollment in the study. Added guidelines on administration of oseltamivir in subjects unable to swallow in compliance with Tamiflu product information.
Section 7.7 *Treatment Compliance*	**¶ 2 only:** The number of oseltamivir doses administered while the subject is in the hospital will be documented by the Investigator or his/her designee. In case of discharge while on oseltamivir therapy, subjects will document the number of doses taken at home and any missed doses; subjects self-administering oseltamivir at home will bring the pack(s) ~~pill bottle~~ to each study visit so study staff may count and record the number of pills taken. Any unused capsules must be returned to the study site.	Clarified that oseltamivir will be provided in packs, not pill bottles.
Section 8.2.1 *Prohibited Prior Medications*	**¶ 1 only:** Per Exclusion Criterion 3 (Section 5.2.2) subjects are not allowed to receive monoclonal antibody products ~~(including VIS410)~~ within 3 months prior to VIS410/placebo dosing.	Removed incorrect reference to VIS410.
Section 8.2.2 *Prohibited Concomitant Medications*	**¶ 1 only:** Per Exclusion Criterion 2 (Section 5.2.2), subjects will not be allowed to receive additional monoclonal antibody products during the study period. Following randomization, use of other antiviral therapy for treatment of influenza A infection will not be permitted. Excluded antiviral medications include but are not limited to rimantadine, amantadine, peramivir, zanamivir, and laninamivir.	Clarified language since VIS410 is a monoclonal antibody.
Section 8.4 *Subject Diary*	A subject diary will be provided upon discharge from the hospital and will record the following: • Daily oseltamivir dosing to be completed for as long as the subject continues to take oseltamivir • Daily VAS for assessing resumption of usual activities to be completed up until either the subject reports that all pre-influenza usual activities can be performed or Day 56, whichever comes first • Daily Influenza Patient Reported Outcomes (FluPRO) Questionnaire to be completed until Day 14, if applicable	Added specific list of what will be included in the subject diary.
Section 9.1.3 *Adverse Event of Special Interest*	For VIS410, the AESIs include the following: • Abdominal cramping • Diarrhea/Loose stool • Nausea	Updated AESI list to include single episodes of loose stool.

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	• ~~and~~ vomiting • Pruritus • Rash • Hypotension • Throat tightening • Trouble breathing or wheezing	
Section 9.4 *Action Taken Regarding Investigational Product*	The action taken toward the study drug must be described as one of the following: • Permanently discontinued • Stopped temporarily • Modified infusion rate • No action Taken • Not applicable	Updated to match what is captured in the database if an AE occurs and include options in case AE occurs after end of infusion or did not result in a change with regards to VIS410 infusion.
Section 9.6 *Recording Adverse Events*	**¶ 1 only:** All (S)AEs occurring during the clinical investigation must be documented in the EDC system from the time of dosing with any study treatment through Day 56.	Clarified timeframe when AEs are captured.
Section 9.7.1 *Reporting Serious Adverse Events*	All SAEs, irrespective of the circumstances or suspected cause, must be reported on a Serious Adverse Event Form by the Investigator to the Sponsor or designee within 24 hours of their knowledge of the event, preferably by email. ~~fax. Other means of transmission can be decided when fax is not possible (email).~~ **Contact details for reporting SAEs:** [redacted]	Updated to specify that SAEs should be sent via email as preferred method and included additional contact numbers for reporting.
Section 9.7.3 *Reporting a Pregnancy*	Subjects should not become pregnant during the study. Pregnancy is not an AE; however, the Investigator must report any pregnancy which occurs in a female subject or the female partner of a male subject up to 60 ~~56~~ days after last dose of VIS410/placebo by emailing ~~faxing~~ the pregnancy notification form to the Sponsor or designee within 24 hours of the study site staff becoming aware of the pregnancy. The Investigator will also follow up with the subject to determine the outcome of the pregnancy and any fetal or neonatal sequelae will be obtained and documented. The Investigator or study site staff must report the outcome of the pregnancy to the Sponsor or designee. **Contact details for reporting Pregnancy:** Pharm-Olam Pharmacovigilance Email: [redacted]	Changed pregnancy reporting to 60 days for consistency with prior studies. Updated to specify that pregnancy should be reported on the pregnancy notification form and sent via email.

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	~~Fax: +44 208 338 0380~~ Congenital abnormalities and birth defects in the offspring of male or female subjects should be reported as an SAE if conception occurred during the study treatment period or in the 60 ~~56~~-day period after the last dose of VIS410/placebo.	
Section 10.1 *Determination of Sample Size*	**¶ 2 only:** Using a log-rank test, a sample size of 130 influenza A–infected subjects per treatment group will provide 80% power to detect a 1.5-day difference (5 days for oseltamivir alone and 3.5 days for VIS410 plus oseltamivir) in the median time to cessation of O_2 support ~~resulting in a stable SpO_2 > 95% for at least 6 hours on room air, return to baseline respiratory status, or hospital discharge with no need for additional O_2 support, whichever occurs first~~ for VIS410 relative to placebo. A two-sided alpha of 0.05 was used for the calculation.	Section updated to maintain consistency with prior changes.
Section 10.6 *Interim Analysis*	An ~~The first~~ interim analysis of efficacy may ~~will~~ be performed by an unblinded third party after 50% of subjects have been enrolled (n = 195) and have an assessment of the primary endpoint (time to normalization of respiratory function). This interim analysis may ~~will~~ be conducted to assess if one of the active VIS410 treatment arms will need to be terminated for futility. Using the methods of Lan and DeMets an alpha spending function will be created and if the test statistic for one of the active VIS410 treatment arms is less than the lower critical point then the VIS410 treatment arm may be terminated for futility. The interim analysis may ~~The interim analysis will~~ also test the primary efficacy objective to assess if the primary efficacy endpoint has been met early. The effect size for this study was estimated without the benefit of any prior randomized, controlled trial. If the VIS410 effect size proves to be greater than anticipated, then it is possible that the primary study endpoint will be met by the time the interim efficacy analysis is performed. In the event the interim analysis reveals that the primary efficacy endpoint has been satisfied, this Phase 2 study could be terminated for efficacy. The DSMB and the unblinded statistician will review the interim analysis results to provide their recommendation on discontinuation of one of the VIS410 treatment arms, if needed, based on pre-specified criteria as outlined in the SAP. The interim analysis may ~~will~~ also review the following key secondary endpoints, when available, to assist in the decision of discontinuing a VIS410 treatment arm: • Time to clinical resolution of vital signs • Total number of hours on oxygen support or PPV • Viral load ~~titer~~ from nasopharyngeal swabs A sample size re-estimation (SSRE) will also be performed at this point in order to determine if the current sample size is sufficient based on the observed effect size in the study. The unblinded statisticians will work directly with the Sponsor on the SSRE. The details of SSRE and individuals with access to the information will be out	Clarified that an interim analysis may (or may not) be conducted due to rate of enrollment. Updated statistical methods and language to include more details related to the error rate and the interim analysis.

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Section 10.7.1 *Time to Cessation of O_2 Support*	**¶ 1 only:** Time to cessation of O_2 support resulting in a stable SpO_2 will ~~> 95% for at least 6 hours, return to baseline respiratory status, or hospital discharge with no need for O_2 support, whichever occurs first, will~~ be analyzed using a Cox model. Time from onset of symptoms to VIS410 treatment and the number of doses of oseltamivir (Tamiflu) prior to VIS410 treatment for the current case of influenza will be included as a covariate in the analysis. A *P*-value (using the Wald statistic) for each VIS410 dose vs placebo will be presented.	Section updated to maintain consistency with prior changes. Clarified the statistical method planned to determine P-value.
Section 10.7.2 *Time to Clinical Response;* Section 10.7.3 *Time to Cessation of Ventilator Support*	Time to clinical response is defined as resolution of 4 of 5 vital signs that will be determined upon physical examination. Clinical response is defined as: • Afebrile with core temperature ≤ 37.8°C, without use of antipyretics (oral ≤ 37.2°C) • Respiratory rate ≤~~<~~ 24 beats per minute • Oxygen saturation ≥~~>~~ 95% on room air without support or a return to pre-infection status, if pre-infection status was < 95% • Pulse rate ≤ 100/min • SBP ≥ 90 mm/Hg, without vasopressor use The probability of time to clinical response (defined as resolution of vital signs) will be calculated via Kaplan-Meier. A P-value (using the log-rank test) for each VIS410 dose vs placebo will be presented. The number and percentage of subjects in each treatment group with clinical response will be summarized. Results will be tabulated and presented graphically as well.	Section updated to maintain consistency with prior changes. Added details on statistical method planned to determine P-value.
Section 10.7.4 *Healthcare Resource Utilization*	**¶ 2 only:** Time (number of days) to resumption of usual activities will be determined from the VAS ~~visual analog scale~~ (scale ranged from 0 to 10, where 0 indicates subject is unable to perform any of his/her usual activities prior to influenza onset ~~at all~~, and 10 indicates subject is able to fully perform all usual ~~return to his/her normal~~ activities ~~fully~~).	Added language to clarify that a subject needs to complete VAS from baseline until the subject reports they are able to perform all usual activities they were performing prior to influenza onset.
Section 10.9.2 *Pharmacokinetics of Nasopharyngeal Secretions and Tracheal Aspirate*	Nasopharyngeal swabs will be obtained from both nostrils (1 swab per nostril). The first 50 randomized subjects will have nasopharyngeal swabs collected up to Day 56 (predose, end of infusion, Days 3, 5, 7, 14, 28 and 56); while in the remaining subjects, nasopharyngeal swabs will be obtained up to Day 14 only. If the subject remains in the hospital on Day 10, then additional nasopharyngeal swabs will be obtained on Day 10. The VIS410 concentrations in the nasopharyngeal secretions and tracheal aspirate will be listed by subject, and summary statistics by group will be reported as described for the serum concentrations. The computed PK parameters will be listed by subject for VIS410. Summary statistics and PK parameters will be presented, including means, geometric means, standard deviations, CV, medians, and ranges, as appropriate. Summary graphs, including mean concentration-time profiles by group, will also be presented. The nasopharyngeal concentration data may also be analyzed by population PK ~~pharmacokinetic~~ methods using nonlinear	Updated the planned PK analysis on nasopharyngeal swabs. to minimize the number of nasopharyngeal swabs in this study, the full nasal PK profile (Day 56) will be determined based on the first 50 subjects only. The remainder of the subjects will only have nasal PK along with nasal virology up to Day 14. PK analysis will be extrapolated beyond Day 14in these subjects based on population PK analysis.

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	mixed effects modeling as implemented in NONMEM or equivalent software. If necessary, the data may be pooled with data from previous studies. Additional analyses and summaries may be generated as appropriate. Results of population PK or PK/PD analyses may be reported outside the clinical study report.	
Section 10.11 *Immunological Analyses*	Immunological assessments will be summarized for the MITT population by parameter, treatment group, and time point using descriptive statistics: • Anti-influenza A antibodies by HAI in serum • ADA titers • ~~Concentrations of cytokines in serum~~	Updated language to more accurately describe all immunological analyses in one section.
Section 10.12 *Viral Resistance*	Viral sensitivity to VIS410 and oseltamivir will be assessed ~~over time~~ during the study. ~~This evaluation will be performed using the safety population~~	Clarified that viral resistance will be assessed. Analysis population will be defined in the SAP.
Appendix 14.1	Footnote to Questionnaire Copyright© 2014, 2015, 2016 Leidos Biomedical Research, Inc. All rights reserved. ~~Research Tool in Development: Do NOT copy or distribute~~	Added appropriate copyright information.
Schedule of Assessments	Visit window of ± 1 day added to Day 5 and Day 7	Visit window added to allow flexibility for subjects discharged from the hospital prior to Day 5.

Protocol History Visterra, Inc. – VIS410-203			
Document	**Issue Date**	**Amendment Type**	**Comments**
Clinical Study Protocol	21 October 2016 (Version 1)	-	This document
Revised Clinical Study Protocol	25 October 2016 (Version 1.1)	Country-specific: South Africa	Inclusion of HIV testing at baseline