Vitamin D to Improve Outcomes by Leveraging Early Treatment

ACRONYM: ***VIOLET***

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_____ ___06/25/18___

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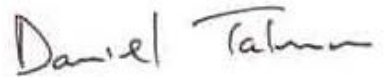

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Contents

1. Introduction 3
2. Trial Summary 3
2.1 Title 3
2.2 Objective 3
2.3 Hypothesis 3
2.4 Study Design 3
2.5 Study Flow 4
2.5.1 Inclusion Criteria 4
2.5.2 Exclusion Criteria 5
2.6 Randomization and Study Initiation Time Window 5
2.7 Primary endpoint 5
2.8 Secondary endpoint 5
2.9 Sample Size/Interim Monitoring 6
2.10Endpoints 7
2.10.1 Primary Endpoint 7
2.10.2 Secondary Endpoints 7
2.11 Subgroups 10
3. Main Manuscript and Statistical Methods 10
3.1 Statistical Methods for Main Manuscript 11
3.2 Primary Endpoint 12
3.3 Secondary Endpoints 12
3.3.1Clinical Endpoints 12
3.3.2 Physiological Endpoints 12
3.3.3 Safety Endpoints 13
3.3.4 Baseline Characteristics 13
3.3.5 Figures 13
4. Supplementary Materials Outline and Statistical Methods 13
4.1Statistical Methods for Supplementary Materials 15
4.2 Subgroup Analyses of Primary Endpoint 15
4.3 Adverse Events 15
References 16

1. Introduction

The following document is the Statistical Analysis Plan for the VIOLET Study. It describes the study and the statistical methods used to analyze the primary and secondary outcomes and the supplemental material for the primary manuscript.

2. Trial Summary

2.1 Title

Vitamin D to Improve Outcomes by Leveraging Early Treatment (VIOLET)

2.2 Objective

To assess the efficacy and safety of early administration of vitamin D_3 (cholecalciferol) in reducing mortality and morbidity for vitamin D deficient patients at high risk for ARDS and mortality.

2.3 Hypothesis

Early administration of vitamin D3 (cholecalciferol) will improve all-cause, all-location mortality to day 90 in vitamin D deficient patients at high risk for ARDS and mortality

2.4 Study Design

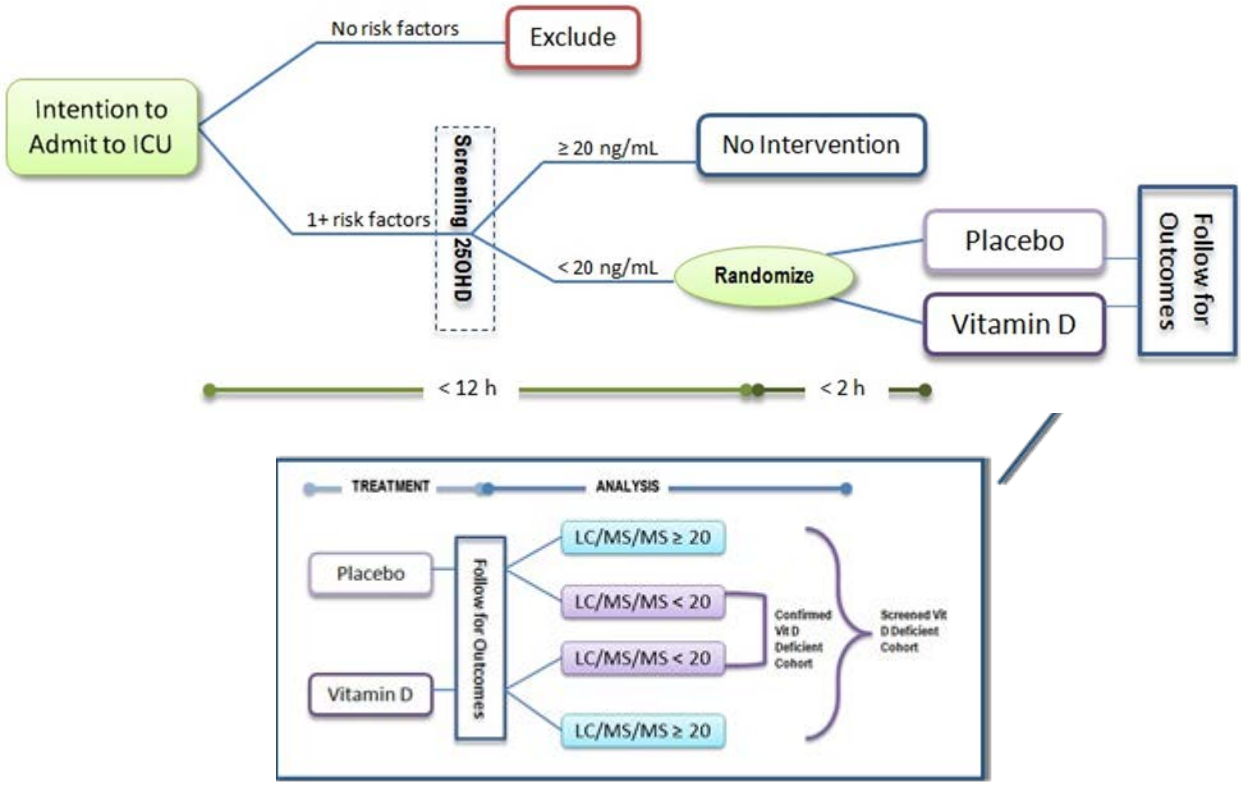
VIOLET is a randomized, double-blinded, placebo-controlled, phase III trial (up to maximum n=3000) of early vitamin D_3 in vitamin D deficient patients at high risk for ARDS and mortality. We will screen all patients for whom there is an intention to admit to ICU for study eligibility and will approach patients meeting inclusion/exclusion criteria for study enrollment. Screening will require screening for vitamin D deficiency using an FDA approved testing method for 25OHD, either by the hospital's clinical laboratory or using the FastPack IP device (Qualigen Inc., Carlsbad, CA). We will obtain written informed consent for the protocol prior to the vitamin D screening test.

We will randomize enrolled participants who are vitamin D deficient (initial screening 25OHD levels <20 ng/mL) to receive either 540,000 IU vitamin D_3 (cholecalciferol) or placebo as a single, liquid enteral dose, administered either orally or via naso/orogastric tube. Randomization must occur within 12 hours of ICU admission decision and study drug administered within 2 hours of randomization. We will assess trial endpoints for randomized participants by chart review and will contact participants or proxies at day 90.
At baseline for all randomized participants, we will measure serum calcium (in real time, if not clinically available) and plasma 25OHD levels (batched and measured initially in monthly increments using the gold-standard liquid chromatography-tandem mass spectrometry [LC/MS/MS] method). We will collect additional plasma and whole blood for DNA banking. For the first 300 randomized participants, we will measure day 3 serum calcium (in real time, if not clinically available) and baseline plasma IL-6. We will also collect plasma at day 3 in the first 300 randomized participants to measure 25OHD by LC/MS/MS and IL-6 (both batched) and for cryopreservation in the biorepository.

The primary efficacy analysis cohort (confirmed vitamin D deficient cohort) will be defined as randomized participants having LC/MS/MS levels less than 20 ng/mL. The primary efficacy analysis will only involve these participants. The FastPack device had some discordance with

LC/MS/MS at a 20 ng/mL cutoff, and the previous study only showed a treatment effect in patients with low vitamin D levels. [1]Since LC/MS/MS values will take at least 24 hours to process, we need to treat participants on the basis of the screening test result (FastPack or clinical laboratory test) available within three hours of patient consent. A secondary analysis cohort (screened vitamin D deficient cohort), which will also be used for the primary safety analysis, will include all randomized participants who receive vitamin D or placebo.

2.5 Study Flow



2.5.1 Inclusion Criteria

1. Age ≥ 18 years
2. Intention to admit to ICU from emergency department, hospital ward, operating room, or outside facility
3. One or more of the following acute risk factors for ARDS and mortality contributing directly to the need for ICU admission:

<u>Pulmonary</u>

a) Pneumonia
b) Aspiration
c) Smoke Inhalation
d) Lung contusion
e) Mechanical ventilation for acute hypoxemic or hypercarbic respiratory failure

Extra-Pulmonary

f) Shock
g) Sepsis
h) Pancreatitis

4. Vitamin D deficiency (screening 25OHD level <20 ng/mL)

2.5.2 Exclusion Criteria

1. Inability to obtain informed consent
2. Unable to randomize within 12 hours of ICU admission decision
3. Unable to take study medication by mouth or enteral tube
4. Baseline serum calcium >10.2 mg/dL (2.54 mmol/L) or ionized calcium >5.2 mg/dL (1.30 mmol/L)
5. Known kidney stone in past year or history of multiple (>1) prior kidney stone episodes
6. Decision to withhold or withdraw life-sustaining treatment (patients are still eligible if they are committed to full support except cardiopulmonary resuscitation if a cardiac arrest occurs)
7. Expect <48 hour survival
8. If no other risk factors present, a) mechanical ventilation primarily for airway protection, pain/agitation control, or procedure; or b) elective surgical patients with routine postoperative mechanical ventilation; or c) anticipated mechanical ventilation duration <24 hours; or d) chronic/home mechanical ventilation for chronic lung or neuromuscular disease (non-invasive ventilation used solely for sleep-disordered breathing is not an exclusion).
9. Prisoner
10. Pregnancy
11. Greater than 72 hours since hospital presentation (or > 3 calendar days since hospital presentation if transferred with no time stamp).

2.6 Randomization and Study Initiation Time Window

All participants must be enrolled and randomized within 12 hours of meeting inclusion criteria. Study medication must be administered within 2 hours of randomization.

2.7 Primary endpoint

The primary endpoint is all-cause, all-location mortality to day 90. The primary analysis will be based on randomized participants with baseline 25OHD levels <20 ng/mL, as measured by LC/MS/MS (***confirmed vitamin D deficient cohort***). As below, additional secondary analyses will be based on all randomized participants (***screened vitamin D deficient cohort***).

2.8 Secondary endpoint

Clinical Endpoints

- Hospital length of stay among survivors to day 90
- Healthcare facility length of stay among survivors to day 90
- Alive and home (prior level of care) at day 90
- Ventilator-free days to day 28

- Time to mortality to day 90

Physiological Endpoints

- Development and severity of ARDS to day 7
- Change in organ failure severity to day 7
- 25OHD levels to day 3
- IL-6 levels to day 3

Safety Endpoints

- Calcium levels to day 14
- Kidney stones to day 90
- Fall-related fractures to day 90

The primary analyses will be in the confirmed vitamin D deficient cohort. These analyses will include an assessment of the secondary endpoints defined above. In addition, we will estimate the treatment effects on the primary and secondary endpoints in the screened vitamin D deficient cohort. If the analysis in the confirmed vitamin D deficient cohort is significant then estimates in the screened vitamin D deficient cohort will measure the treatment effect in patients who would be given the treatment if early LC/MS/MS is not available (or until next generation screening tests more closely approximate LC/MS/MS results). We will also estimate the treatment effects on the endpoints above as a continuous function of baseline 25OHD level by LC/MS/MS. This function will provide an estimate of the extent to which efficacy (and safety) depends on vitamin D level.

2.9 Sample Size/Interim Monitoring

1. We base our assumptions on finding a 5% or greater absolute difference in mortality. With a 20% mortality rate in the control arm and a 15% mortality rate in the intervention arm, the maximum required total sample size is 3000 randomized participants with 87.4% power based on the assumption that 80% of the randomized participants will have baseline 25OHD levels < 20 ng/mL, as measured by LC/MS/MS, and one sided $\alpha = 0.025$.
2. The principal analysis will be based on randomized participants with baseline 25OHD levels <20 ng/mL, as measured by LC/MS/MS (confirmed vitamin D deficient cohort), following an intention-to-treat principle (analysis according to randomization assignment). We anticipate that approximately 80% of randomized participants will have baseline 25OHD levels <20 ng/mL by LC/MS/MS. The maximum size of the screened vitamin D deficient cohort will be 3000 patients and the futility stopping boundaries will consider the possibility that the sample size will not reach 2400 (80% of 3,000) in the confirmed vitamin D deficient cohort if too small a fraction of randomized patients would have baseline 25OHD levels <20 ng/mL by LC/MS/MS.
3. An independent Data and Safety Monitoring Board (DSMB) will determine if the study should stop for superiority or futility of the intervention, or safety concerns. There will be three interim analyses throughout the trial (at subject enrollments 750, 1500, and 2250 randomized participants).
4. For the first 300 randomized participants, we will analyze calcium levels at baseline and day 3 and 25OHD levels at day 3 using LC/MS/MS. This analysis will evaluate the

effectiveness and safety of the vitamin D intervention in correcting vitamin D deficiency (clinical outcomes will not be evaluated at this time). Based on pre-specified rules and in consultation with the DSMB (who will have access to additional adverse event data), we may require changes to the vitamin D dose or extend the duration of 25OHD/calcium monitoring.

2.10Endpoints

2.10.1 Primary Endpoint

The primary endpoint is all-cause, all-location mortality to day 90. We will ascertain vital status at day 90 by chart review (for participants that remain in the hospital at day 90 or died in hospital prior to day 90). For participants that were discharged alive from the hospital prior to day 90, we will call the patient or proxy (e.g., research contact list and next-of-kin) on/after day 90 to determine vital status. For those that we are unable to verify vital status at day 90, we will use evidence from the medical record (e.g., healthcare visits after day 90), review of obituaries, or phone calls to healthcare facilities to determine that participants are alive. Finally, for participants that have missing 90-day mortality, we will use the Centers for Disease Control and Prevention's National Death Index (NDI) to determine vital status, using each patient's social security number (SSN) for an exact NDI match. At the time of final analysis, we will use best available mortality data and if vital status remains unknown (anticipated for <5%), will impute 90-day mortality based on last known location.

2.10.2 Secondary Endpoints

<u>Clinical Endpoints</u>

1. **Study hospital length of stay among survivors to day 90:** Study hospital length of stay is defined as the number of days from enrollment (typically the day of hospital admission) to the day of study hospital discharge up to day 90. This endpoint will be analyzed only in survivors because hospital length of stay in those who die in the hospital is non-informative for this endpoint. In addition, we will use survivor average causal effect (SACE) methods to estimate hospital length of stay among participants that would have survived in both treatment and control groups.[50] Accordingly, these methods adjust for the impact of differential survival between treatment groups.

2. **Healthcare facility length of stay among survivors to day 90:** Healthcare facility length of stay is the time spent in another hospital or healthcare facility (e.g. long-term acute care [LTAC] hospitals or acute rehabilitation/skilled nursing facility), for the subgroup of participants that were discharged to another healthcare facility after the initial hospitalization. This measure is defined as the number of days from initial hospital discharge to the first facility discharge to home (pre-hospitalization level of care) up to day 90. Healthcare facility LOS is zero for patients discharged to home (pre-hospitalization level of care) from the study hospital. This endpoint will be analyzed only in survivors using SACE methods because healthcare facility length of stay in those who die during the follow-up period is non-informative for this endpoint.

3. **Alive and home at day 90:** This endpoint is the proportion of participants who have survived and are present at home, defined as pre-hospitalization level of care, at day 90.

4. **Ventilator free days (VFD) to day 28:** VFD depends on both duration of ventilation and mortality through study day 28. In participants who survive 28 days, VFD is defined as 28 minus duration of ventilation. Duration of ventilation is counted from the first study day of assisted breathing through the last day of assisted breathing provided the last day is prior to day 28. Otherwise, it is counted from the first study day of assisted breathing through day 28. For participants discharged with assisted ventilation (e.g., to LTAC facility) prior to day 28, a phone call will be required to assess ventilator status at day 28. Participants discharged prior to day 28 (but not to home) on unassisted breathing will be assumed to remain on unassisted breathing through day 28. Isolated periods of ventilation briefer than 24 hours for surgical procedures and ventilation solely for sleep disordered breathing do not count towards duration of ventilation. In participants who never require assisted breathing, duration of ventilation is zero. Participants who do not survive 28 days will be assigned zero VFD. VFD is undefined in participants with chronic/home mechanical ventilation (except solely for sleep disordered breathing) and they will be excluded from this analysis.

5. **Time to mortality to day 90:** Time to mortality will use best available data on vital status and date of death and censor based on last known alive date for participants with missing vital status.

<u>Physiological Endpoints</u>

6. **Development and severity of ARDS to day 7:** We will determine the presence and severity of ARDS for each day of mechanical ventilation to day 7 using the following approach:
   a. For each ventilator day: if ABG available between 2:00 AM and 8:00 AM, measure P/F (PaO2, FiO2 and PEEP) for all ABGs during this time window daily to day 7. Or, for ventilator days that no ABG available between 2:00 AM and 8:00 AM, determine lowest imputed P/F from measured S/F (SpO2, FiO2, and PEEP). See Appendix B for P/F imputation table.
   b. For participants with P/F <300 or imputed P/F <300, FiO2 ≥40%, and PEEP ≥5 cm H_2O, determine if hypoxemia is valid, acute, and not fully explained by CHF or fluid overload
   c. If yes to item (b), local investigators will:
      i. Review the first CXR (or CT) performed on each ventilated day with valid P/F or imputed P/F <300 (to day 7). If no chest imaging studies are present that day, site investigators may review imaging one day before or after to determine if ARDS imaging criteria met.
      ii. Assess if the images are consistent with ARDS (bilateral opacities not fully explained by effusions, lobar/lung collapse, or nodules). If equivocal, the reviewing investigator will adjudicate with additional investigators.

7. **Change in organ failure severity to day 7:** We will measure acute organ failure severity daily to day 7 for three key organ systems: acute hypoxemic respiratory failure, acute kidney injury, and cardiovascular failure (i.e., shock). We will measure acute hypoxemic respiratory failure using data from the ARDS assessment, except this outcome does not require chest imaging consistent with ARDS. We will measure acute kidney injury by highest daily creatinine values or new use of dialysis/ renal replacement therapy (chronic dialysis

participants are excluded). We will measure cardiovascular failure by highest daily dose of vasopressors (epinephrine, norepinephrine, dopamine, phenylephrine, or vasopressin).

8. **Plasma 25OHD levels:** In addition to the screening vitamin D test (for trial entry), we will measure gold-standard plasma 25OHD levels at baseline for all randomized participants and at day 3 in the first 300 randomized participants to confirm biomarker response to the intervention. The gold-standard test will be done in batch using LC/MS/MS methods. Individual results will not be available to investigators and coordinators to avoid unblinding. Based on pre-specified rules and in consultation with the DSMB (who will have access to additional adverse event data), we may require changes to the vitamin D dose or extend the duration of 25OHD/calcium monitoring.

9. **Plasma IL-6 levels:** We will measure plasma IL-6 levels at baseline and day 3 in the first 300 randomized participants in batch concurrent with 25OHD testing. IL-6 is a pro-inflammatory cytokine associated with increased incidence and severity of ARDS severity and worse clinical outcomes. Vitamin D may improve outcomes in critically ill patients by reducing levels of IL-6 (either causal association or as a result of improved recovery)

Safety Endpoints

10. **Calcium levels to day 14:** Because the half-life of 25OHD is approximately 2 weeks, we will record clinically available serum or ionized calcium levels through day 14 for all randomized participants. In addition, we will measure baseline and day 3 in the first 300 randomized participants in real time, either as part of routine clinical care or as a research procedure if not available. Day 3 is the when 25OHD levels peak in ICU patients given 540,000 IU of vitamin D.[1,49] While no prior study of acute vitamin D repletion, using similar high doses as our trial, has observed clinically important hypercalcemia, we will confirm these findings in the VIOLET trial. Because little change in calcium levels is anticipated, access to individual-level calcium results will not unblind study staff. During the trial, the DSMB may seek additional subject measurements for safety assessment, though this is not anticipated.

11. **Kidney stones to day 90:** While an association between hypervitaminosis D and kidney stones has been suggested, the relationship with high dose vitamin D supplementation and kidney stones remains controversial. We will assess for incident kidney stones by chart review at the end of the hospitalization and by self-report at the 90 day phone call for those discharged from the hospital prior to day 90.

12. **Fall-related fractures to day 90:** The association between acute vitamin D supplementation and falls/fractures remains unclear. Most data suggest that high dose vitamin D in healthy outpatients may improve muscle function, balance, and bone mineral density, and thus decrease fall-related fractures, but other data suggest that high dose vitamin D supplementation may actually increase the incidence of falls/fractures. Because of this uncertainty and limited data in hospitalized patients, we will assess for incident fall-related fractures by chart review at the end of the hospitalization and by self-report at the 90 day phone call for those discharged from the hospital prior to day 90.

2.11 Subgroups

The primary analysis will be based on randomized participants with baseline 25OHD levels <20 ng/mL by LC/MS/MS (confirmed vitamin D deficient cohort). Secondary analysis will be based on the screened vitamin D deficient cohort (all randomized patients). *A priori* subgroups for analysis will include age, sex, race/ethnicity, residence (independent vs. long-term care facility), BMI, pre-hospitalization vitamin D supplementation, baseline 25OHD level (by the LC/MS/MS method), baseline renal function, ARDS risk factor (e.g., infectious vs non-infectious), LIPS score, pre-randomization mechanical ventilation status, pre-randomization presence of ARDS, and source of ICU admission.

3. Main Manuscript and Statistical Methods

The primary analysis will be based on randomized participants with baseline 25OHD levels <20 ng/mL, as measured by LC/MS/MS (***confirmed vitamin D deficient cohort***). Additional analyses will be based on all randomized participants (***screened vitamin D deficient cohort***).

The main manuscript will include the following tables and figures.

Figure. CONSORT Diagram

Table. Baseline Characteristics (confirmed deficient cohort)

- Demographics
- Co-Morbidities—Charlson, BMI, eGFR, pre-hospital level of care
- VIOLET Risk Factors
- Location—intent to admit, ICU destination
- Illness Severity—SOFA, LIPS, mechanical ventilation, ARDS, vasopressor use, vital signs, WBC, lactate
- Baseline vitamin D (home meds, vitD level by LCMS, calcium level)

Figure. Change in LCMS from Day 0 to Day 3 for (screened deficient cohort)
Summary data in text of results or Supplementary Material

Table. Primary and Main Secondary Endpoints (confirmed deficient cohort)

Primary

- 90 day all-cause, all-location mortality

Secondary Clinical

- Hospital mortality to day 90
- Alive and home (prior level of care) at day 90
- All-cause, all-location 28 day mortality
- Hospital LOS
- Healthcare facility LOS
    - Include proportion that went to healthcare facility (vs home) separately
- VFD to day 28

    - Include proportion ventilated to help with interpretation

Secondary Physiological

- Respiratory failure
    - New mechanical ventilation ever to day 7 (Y/N)
    - Lowest P/F in the first 7 days if on mechanical ventilation
- ARDS
    - New ARDS ever to day 7 (Y/N)
    - ARDS severity by Berlin definition at time of new ARDS diagnosis[1]
    - Lowest P/F in the first 7 days if ARDS
- AKI to day 7
    - New RRT ever to day 7 (Y/N)
    - Worst AKI severity in first 7 days (no, mild, moderate, or severe AKI by KDIGO criteria[2]
    - Highest creatinine in first 7 days
- Cardiovascular failure to day 7
    - New vasopressor use ever to day 7 (Y/N)
    - Highest CV SOFA score in first 7 days

- IL-6 to day 3

Safety

- Calcium to day 14
    - Daily comparison to day 14
    - Hypercalcemia ever during the first 14 days (Y/N)
- Kidney stones to day 90
- Falls to day 90
- Fall-related fractures to day 90
- EQ-5D-5L at day 90 (adjusted for baseline)

Figure. Time to Mortality to Day 90 (confirmed deficient cohort)

Figure. 90-day Mortality by Baseline Vitamin D Level for (screened deficient cohort)

3.1 Statistical Methods for Main Manuscript

All analyses are intention to treat, that is, all randomized participants within each cohort (screened deficient, confirmed deficient) will be included in the analyses specified for each cohort. The confirmed deficient cohort is the primary efficacy cohort per the protocol.

All significance testing will be two-tailed at alpha = 0.05 with no adjustment made for multiple comparisons[3]

With the exception of the primary outcome, analysis is based on all available data with no imputation of missing data.

The causal clinical effect of treatment on outcomes defined only in patients who are on study is complicated by potential case mix imbalance between the treatment groups due to differential recovery and mortality. However, the proposed statistical methods for these outcomes maintain the correct comparison-wise Type I error rate under the global null of no treatment effect whatsoever on any patient.

3.2 Primary Endpoint

The primary outcome of all-cause, all-location mortality to day 90 will be compared between treatment groups on the risk difference scale using a generalized linear model with a binomial distribution function and identity link function. This is equivalent to a Z-test comparing the difference between two proportions but extends naturally to assessment of sub-group by treatment interactions on the risk difference scale.

For participants that have missing 90-day mortality, we will use the Centers for Disease Control and Prevention's National Death Index (NDI) to determine vital status, using each patient's social security number (SSN) for an exact NDI match or other identifying information if SSN unavailable. If none of the patients with missing 90-day mortality are found in the NDI to have died within 90 days we will assume they are all alive at 90 days. If any are known to be deceased, we will impute 90-day mortality for the remaining patients with unknown status using a multi-state model to estimate the transition probabilities across recorded states (in hospital, post hospital facility, home, death). In addition, we will include a sensitivity analysis based on the assumption that the probability of death increases with elapsed time since last follow up.

3.3 Secondary Endpoints

3.3.1Clinical Endpoints

Hospital mortality to day 90, and alive and home (prior level of care) at day 90, and 28 day mortality will be compared between treatment groups using Pearson's chi-square test.

Hospital length of stay and healthcare facility length of stay will be analyzed only in survivors to day 90. We will use survivor average causal effect (SACE) methods to estimate each outcome among participants that would have survived in both treatment and control groups[4]. The SACE methodology includes a model for predicting survival using baseline covariates. The covariates used will be sex, age, Charlson, SOFA, baseline 25OHD as measured by LCMS, and race/ethnicity (non-Hispanic white versus other).

Ventilator-free days to day 28 will be compared between treatment groups using a t-test.

3.3.2 Physiological Endpoints

Binary endpoints will be compared between treatment groups using Pearson's chi-square test.

Ordinal endpoints will be will be scored as 1,2,3, … and compared between treatment groups using the CMH mean score test.

Continuous endpoints will be compared between treatment groups using a t-test with two exceptions. The highest creatinine and CV SOFA score will be analyzed controlling for baseline value using repeated measures ANOVA with a treatment by time interaction and shared intercept at time 0 (baseline). This model is equivalent to ANCOVA controlling for baseline but is able to make use of all available data.

3.3.3 Safety Endpoints

Calcium levels to day 14 will be compared daily between treatment groups using a t-test. Binary occurrence of hypercalcemia (as defined by the protocol) to day 14 will be compared between treatment groups using Pearson's chi-square test.

Kidney stones to day 90 will be compared between treatment groups using Pearson's chi-square test.

Falls and fall-related fractures to day 90 will be compared between treatment groups using Pearson's chi-square test.

EQ-5D-5L at day 90 (adjusted for baseline) will be analyzed only in survivors to day 90. We will use survivor average causal effect (SACE) methods to estimate the outcome among participants that would have survived in both treatment and control groups as described above.

3.3.4 Baseline Characteristics

Summary statistics will be tabulated to show the distribution in each treatment group and in aggregate. Categorical variables will be summarized as frequency and percent and continuous outcomes as mean and standard deviation or median and interquartile range as appropriate. No p-values testing for a baseline difference between treatment groups will be presented

3.3.5 Figures

A log-rank test will be used to compare the time to 90 day mortality in the two treatment groups.

A quadratic smoothing spline in each treatment group will be used to display the relationship between the primary mortality outcome and baseline 25OHD level as measured by LCMS. Pointwise bootstrap confidence intervals for the difference between the two curves will be calculated at LCMS values of 5, 10, 15, 20, 25, and 30.

4. Supplementary Materials Outline and Statistical Methods

The supplementary materials will include the following tables and figures.

Table. Baseline Characteristics for confirmed not deficient (LCMS≥20), confirmed deficient, and screened deficient (all randomized)

- Demographics
- Co-Morbidities—Charlson, BMI, eGFR, pre-hospital level of care
- VIOLET Risk Factors
- Location—intent to admit, ICU destination
- Illness Severity—SOFA, LIPS, mechanical ventilation, ARDS, vasopressor use, vital signs, WBC, lactate
- Baseline vitamin D (home meds, vitD level, calcium level)

Table. Study Initiation/Dosing for confirmed deficient and screened deficient (all randomized)

- Inclusion to Randomization (time)
- Randomization to Drug Administration (time)
- Study Drug Dosing
- Study Drug Tolerance

Figure. Subgroup Analysis of Primary Endpoint (confirmed deficient)

- Age: <60, ≥60
- Sex: M, F
- Race/ethnicity: NH white, NH black, Hispanic, other
- Residence: independent, long-term care facility
- BMI: <20, 20-24.9, 25-29.9, 30-34.9, ≥35
- Pre-hospitalization vitamin D supplementation: none, 1-800 IU/day, >800 IU/day
- Baseline 25OHD level (by the LCMS): <12, 12-19, 20-29, ≥30 AND <10, 10-19, 20-29, ≥30 AND <20, ≥20
- Baseline renal function: eGFR 15 (or on dialysis), 15-29, 30-59, 60-89, ≥90
- ARDS risk factor: sepsis, pneumonia, shock, mechanical ventilation, other AND infectious, non-infectious
- LIPS score: <4, ≥4
- Pre-randomization mechanical ventilation status: Y, N
- Pre-randomization presence of ARDS: Y, N
- Source of ICU admission: ED, other
- ICU destination: MICU, other
- Modified intention to treat (received full study drug dose without vomiting within 1 hour): yes, no

Figure. Subgroup Analysis of Primary Endpoint for screened deficient (all randomized)

- Age: <60, ≥60
- Sex: M, F
- Race/ethnicity: NH white, NH black, Hispanic, other
- Residence: independent, long-term care facility
- BMI: <20, 20-24.9, 25-29.9, 30-34.9, ≥35
- Pre-hospitalization vitamin D supplementation: none, 1-800 IU/day, >800 IU/day
- Baseline 25OHD level (by the LCMS): <12, 12-19, 20-29, ≥30 AND <10, 10-19, 20-29, ≥30 AND <20, ≥20
- Baseline renal function: eGFR 15 (or on dialysis), 15-29, 30-59, 60-89, ≥90
- ARDS risk factor: sepsis, pneumonia, shock, mechanical ventilation, other AND infectious, non-infectious
- LIPS score: <4, ≥4
- Pre-randomization mechanical ventilation status: Y, N
- Pre-randomization presence of ARDS: Y, N
- Source of ICU admission: ED, other
- ICU destination: MICU, other
- Modified intention to treat (received full study drug dose without vomiting within 1 hour): yes, no

Figure. Time to Mortality to Day 90 for screened deficient (all randomized)

- Baseline LCMS<12
- Baseline LCMS 12 to <20
- Baseline LCMS ≥20

Table. Primary and Main Secondary Endpoints for screened deficient (all randomized)

Table. Adverse Events and Safety Outcomes for confirmed deficient, confirmed not deficient (LCMS≥20), and screened deficient (all randomized)

4.1Statistical Methods for Supplementary Materials

Statistical methods for the supplement will be the same as those for similar outcomes in the main manuscript

4.2 Subgroup Analyses of Primary Endpoint

For the primary outcome, interactions between treatment groups and will be analyzed on the risk difference scale using a generalized linear model with a binomial distribution function and identity link function including a treatment effect, a sub-group effect, and a treatment by sub-group interaction.

4.3 Adverse Events

Adverse events will be analyzed using weighted Poisson regression with non-serious events weighted by one and serious events weighted by two. Events rather than patients will be the unit of analysis. Adverse events will be grouped and analyzed separately by MeDRA system organ classes

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