High Dose Vitamin-D-substitution in patients with COVID-19: study protocol for a randomized, double blind, placebo controlled, multi-centre study- VitCov Trial

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Study protocol

Keywords: Coronavirus, SARS-CoV-2, Covid-19, Vitamin D, Vitamin D deficiency

Posted Date: June 25th, 2021

DOI: <https://doi.org/10.21203/rs.3.rs-633931/v1>

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Version of Record: A version of this preprint was published at Trials on February 4th, 2022. See the published version at <https://doi.org/10.1186/s13063-022-06016-2>.

Abstract

Background

The Coronavirus disease 19 (Covid-19) pandemic has caused more than a million deaths and new treatments are urgently needed. Factors associated with a worse Covid-19 prognosis include old age (> 65 years), ethnicity, male sex, obesity and people with comorbidities such as hypertension, diabetes, cardiovascular disease, and respiratory diseases. Further, vitamin D deficiency has been reported to be a predictor of poor prognosis in patients with acute respiratory failure due to Covid-19. Vitamin D deficiency is a modifiable risk factor, which - according to a recent clinical case series – has the prospect of reducing hospital stay, intensive care and fatal outcomes. Vitamin D has potent immunomodulatory property and its supplementation might improve important outcomes in critically ill and vitamin D deficient Covid-19 patients. Despite the evidence that supports an association between vitamin D deficiency and Covid-19 severity, there is uncertainty about the direct link. The aim of the trial is therefore to assess if high dose vitamin D supplementation has a therapeutic effect in vitamin D deficient patients with Covid-19.

Methods

Randomized, placebo-controlled double blind, multi-centre study trial to compare a high single dose of vitamin D (140'000 IU) followed by treatment as usual (TAU) (VitD + TAU) with treatment as usual only (placebo + TAU) in patients with Covid-19 and vitamin D deficiency.

Discussion

Vitamin D substitution in patients with Covid-19 and vitamin D deficiency should be investigated for efficacy and safety. The objective of the study is to test the hypothesis that patients with vitamin D deficiency suffering from Covid-19 treated under standardized conditions in hospital will recover faster when additionally treated with high dose vitamin D supplementation. Latest studies suggest, that vitamin D supplementation in patients with Covid-19 is highly recommended to positively influence the course of the disease. With this randomised controlled trial, a contribution to new treatment guidelines shall be made.

Trial registration: clinicaltrials.gov: NCT04525820 and SNCTP: 2020 – 01401

Introduction

Background and rationale {6a}

The world is currently experiencing a coronavirus (CoV) pandemic. A new CoV infection epidemic began in Wuhan, Hubei, China, in late 2019, originally called 2019- nCoV (1) and renamed Covid-19 by the World Health Organization on February 11, 2020. Previous CoV epidemics included severe acute respiratory syndrome (SARS)-CoV, which started in China in 2003 (2) and Middle East respiratory syndrome (MERS)-

CoV in the Middle East, which started in 2012 (3). The mortality rates were > 10% for SARS and > 35% for MERS (4). So far, this pandemic is responsible for more than a million deaths. The direct cause of death is generally due to ensuing severe atypical pneumonia and followed by acute respiratory distress syndrome (ARDS) (4,5). Pneumonia is a widespread complication in people with influenza type A although the mortality rate is lower (1%-3% for the influenza A H5N1 pandemic of 1918–1919 in the United States) (6).

Risk factors for a poor outcome of SARS-CoV-2 infection have so far been found to include, older age (> 65 years) and co-morbidities such as chronic respiratory conditions, hypertension or diabetes, current smoking status, male sex and Black or Asian ethnic origin (7–10).

There are observational studies reporting independent associations between low serum concentrations of 25-hydroxyvitamin D (the major circulating vitamin D metabolite) and susceptibility to acute respiratory tract infection (11,12). 25-hydroxyvitamin D supports induction of antimicrobial peptides in response to both viral and bacterial stimuli (7–9,13) suggesting a potential mechanism by which vitamin D inducible protection against respiratory pathogens might be mediated. A systematic review and meta-analysis of individual patient data showed in 25 eligible randomized controlled trials (total 11 321 participants, aged 0 to 95 years), that vitamin D supplementation showed a relative risk reduction of 12% for acute respiratory tract infection among all participants. The protective effects were stronger in those with baseline 25-hydroxyvitamin D levels < 25 nmol/L (14). Furthermore, the same research group have recently observed dysregulated vitamin D metabolism in patients with chronic obstructive lung disease (COPD) (15). Previously, another group had observed that vitamin D deficiency is prevalent among patients with ARDS and likely contributes to its pathogenesis through alveolar capillary damage (16).

In the one retrospective study it was reported that vitamin D is a predictor of poor prognosis in patients with acute respiratory failure due to Covid-19 (17). So far, vitamin D can be seen as a cheap and easily modifiable factor in the treatment of Covid-19 (18,19). Several retrospective studies have assessed vitamin D levels in patients with Covid-19, so far only one interventional trial is published (20) to evaluate the recommendations and observations from these retrospective studies (13). Leading experts in the field have therefore called for well-powered randomised controlled trials of vitamin D supplementation for the treatment of Covid-19 to test for causality(21).

Vitamin D and respiratory tract infection

Martineau and colleagues found in their meta-analysis of vitamin D supplementation in the prevention of acute respiratory tract infections, that the overall number needed to treat (NNT) was 33 and that in patients with profound vitamin D deficiency at baseline (25-OH-vitamin D serum level < 25 nmol/L) the NNT was 4 (14). The use of vitamin D as a prophylactic agent for influenza has shown promise in preventing illness and reducing secondary asthma in children (22). In this study, there was a benefit for influenza A but not influenza B or the common cold. In a small study of 30 patients, high-dose cholecalciferol supplementation rapidly and safely improves 25-hydroxyvitamin D and bioavailable 25-hydroxyvitamin D levels in patients with severe sepsis or septic shock (23). Changes in bioavailable 25-

hydroxyvitamin D are associated with concomitant increases in circulating LL-37 levels (23). The mechanism shown by Beard and colleagues (24) relies on CYP27B1- dependent cathelicidin expression in macrophages and keratinocytes which is significantly impaired in the absence of 25-hydroxyvitamin D, VDR, or CYP27B1. Cathelicidin production correlates positively with the serum level of 1,25-dihydroxyvitamin D. As shown by Lang and Samaras (25), 30 ng/mL 1,25-dihydroxyvitamin D is necessary for the optimal induction of cathelicidin mRNA. Khare and colleagues (26) noted that treatment of human lung A549 epithelial cells with 100 or 30 nM of 1,25- dihydroxyvitamin D prior to or post-H1N1 (influenza A virus) exposure significantly decreased the levels of infection-induced TNF α , IFN β , and IFN-stimulated gene-15 (ISG15) and downregulated IL-8 and IL-6 RNA levels. According to Helming (27) 1,25-dihydroxyvitamin D also potentially diminishes the proinflammatory cytokine production by the modulation of macrophages, which prevents them from secreting too many cyto- and chemokines. On the basis of the obtained data, the authors suggested that 1,25-dihydroxyvitamin D participates in a negative feedback loop in which IFN γ - activated macrophages induce the release of 1,25-dihydroxyvitamin D.

Vitamin D and chronic respiratory disease

Vitamin D deficiency is common in patients with asthma and COPD and according to new evidence, vitamin D metabolism might be dysregulated in these conditions (15). Inadequate vitamin D status is associated with susceptibility to upper respiratory infections in patients with COPD (28). In the ViDiCo-trial (29), vitamin D supplementation protected against moderate or severe exacerbation, but not upper respiratory tract infection, in patients with COPD with baseline 25-hydroxyvitamin D levels of less than 50 nmol/L.

Vitamin D, Critical illness and ARDS

A randomized placebo-controlled trial of high-dose vitamin D (540 000 IU followed by monthly maintenance doses of 90 000 IU for 5 months) in 492 critically ill adult patients with vitamin D deficiency (25-hydroxyvitamin D $\leq$ 50 nmol/L) found a significant in-hospital mortality reduction among patients with severe vitamin D deficiency (< 30 nmol/L) compared to placebo (hazard ratio 0.56, 95% confidence interval 0.35–0.9) (30). In a study looking specifically at ARDS, Dancer and colleagues found that esophagectomy patients with 25-hydroxyvitamin D < 50 nmol/L (< 20 ng/mL) ($n = 25$) had a 37.5% risk of postoperative lung injury compared to a 15% risk in patients with higher 25-hydroxyvitamin D concentrations ($n = 32$) (16). Eight patients received pre-operative high dose vitamin D supplementation and had higher ratios of postoperative oxygen saturations to fraction of inspired oxygen. In a further study of vitamin D status in patients with ARDS, Park and colleagues retrospectively examined data from 108 patients with ARDS for whom a vitamin D status was available at the time of diagnosis (31). Over 95% of these patients had vitamin D deficiency. When examined according to quartile of serum 25-hydroxyvitamin D, a consistent inverse relationship between serum 25-hydroxyvitamin D and length of hospital and ICU stay among survivors ($n = 36$) was observed (31). A large, multi-centre, multi-ethnic study (vitamin D to improve outcomes by leveraging early treatment (VIOLET) trial) investigated ultra-high-dose vitamin D supplementation (single dose 540 000 IU, without maintenance doses) on decision

to admit to ITU for any reason (primarily pneumonia and sepsis) did not show any reduction in 90-day mortality compared to placebo (32). In this study, only 12% had or developed ARDS and 25% of patients in the treatment group did not show a treatment response of a plasma 25-hydroxyvitamin D concentration of at least 75 nmol/L (30 ng/mL), which has led some colleagues to interpret the results with caution (32).

Potential risks associated with trial participation

Every treatment with drugs has a risk of side effects, but due to the short treatment period of only one single high dose and the careful surveillance of the included patients through qualified personnel, we anticipate the risk of side effects in this study to be very small. The reported undesired effects of vitamin D are hypercalcemia, a greater urge to urinate, polydipsia, renal stones, loss of appetite and therefore weight loss. However, these effects can be expected after prolonged exposure to vitamin D over several months.

Secondary side effects can rarely occur because of blood sampling, such as haematoma or infections of the sampling point. Furthermore, the study only involves routine blood sampling that will be performed by highly qualified personnel who work according to high standards of hygiene, so patients will not be exposed to more or riskier interventions than necessary. With the outlined safety measures, we will reduce the risk of side effects in this vulnerable group of patients.

Objectives {7}

We hypothesised that time to recovery is shorter in the single high dose vitamin D group relative to the standard treatment group. The overall objective is to determine whether patients with vitamin D deficiency and a Covid-19 infection recover faster when they receive a single high dose of vitamin D in addition to standard care compared to placebo and standard care.

The primary objective of this study is to investigate if high dose vitamin D reduces the length of the hospital stay in patients with Covid-19 and vitamin D deficiency. Further we have defined the following parameters as secondary outcomes: necessity of an ICU stay, overall mortality, percentage of patients with 25-hydroxyvitamin D > 50 nmol/L (> 20 ng/mL) at day 7, change in serum calcium, phosphorus, 25-hydroxyvitamin D, 1.25-dihydroxyvitamin D, parathyroid hormone (PTH) and the development of sepsis.

Methods: Participants, Interventions And Outcomes

Trial design {8}

We decided to conduct the study as a prospective, randomized, placebo controlled double blind trial. This study compares a single high dose of vitamin D in addition to treatment as usual (VitD + TAU) to placebo and treatment as usual only (Placebo + TAU). Ethically it is not justifiable not to treat a known vitamin D deficiency, therefore the consequence is that we compare an intervention (single high dose of vitamin D) with treatment as usual (daily small dose of vitamin D).

Study setting {9}

The study will be conducted in four hospitals in Switzerland – planned are two in north-western Switzerland, one in the canton of Tessin and one in eastern Switzerland. If necessary, other hospitals will be asked to participate.

Eligibility criteria {10}

Participants fulfilling all of the following inclusion criteria are eligible to participate in the study after giving written, informed consent:

Hospitalized patients aged ≥ 18 years with an ongoing Covid-19 infection and laboratory confirmed vitamin D deficiency defined as a serum 25-hydroxyvitamin D concentration ≤ 50 nmol/l (≤ 20 ng/ml).

The presence of any one of the following exclusion criteria will lead to exclusion of the participant:

Known hypersensitivity to one of the Vitamin D products used in this study or to one of the adjuvants in the drug's composition, active malignancy, hypercalcemia, granulomatous disease such as sarcoidosis, history of renal stones within the past year, pregnancy/breastfeeding, previous enrolment into the current study, or another interventional study.

All hospitalized Patients suffering from Covid-19, a vitamin D deficiency and do fulfil the above-mentioned eligibility criteria and none of the exclusion criteria are able to participate in this trial.

Who will take informed consent? {26a}

After a patient is identified as potential participant, all inclusion criteria are fulfilled, and none of the exclusion criteria is present, the patient will be asked by their treating physician if he/she wants to participate in this study.

Additional consent provisions for collection and use of participant data and biological specimens {26b}

It is not planned to collect any more data from participants and/ or biological material, no additional consent is needed.

Interventions

Explanation for the choice of comparators {6b}

This study will be conducted as a randomized, placebo controlled, double blind trial comparing high dose vitamin D in addition to TAU with placebo + TAU. The recommended supplementation of vitamin D in adults is 800 IU vitamin D3/day (33,34).

As there is no ethical justification for not supplementing vitamin D in patients with laboratory confirmed vitamin D deficiency, we decided against a “placebo only” design. Instead, we will compare single high

dose vitamin D + TAU versus placebo + TAU.

Intervention description {11a}

Intervention Group

Participants will receive a single oral dose of 140'000 IU of vitamin D3 as an oily solution and then continue with the standard oral dose of 800 IU vitamin D3 per day until discharge. The medication will be administered by instructed personnel - preferably in the morning along with the patients` other prescribed medication. No information was found about significant differences resulting from the timing of administration.

Control Group

Participants will receive a single dose of placebo orally and then continue with the standard dose of 800 IU vitamin D3 per day. The procedure of administration is the same as in the intervention group.

Criteria for discontinuing or modifying allocated interventions {11b}

Treating doctors can change the dose of vitamin D or stop the treatment if clinically indicated. In such cases, a note to file has to be made and reported to the lead centre immediately. The patient will be considered as a drop out. Throughout the whole trial, treatment can be re-evaluated and treating physicians can independently decide about additional treatment options. The patient has to be informed about all treatment options and has to be treated according to the most recent local or national Covid-19 guidelines.

Strategies to improve adherence to interventions {11c}

Patient adherence is not assessed, since the participants are all hospitalized, and qualified personnel will administer medication. The nurses and doctors will ensure that the patient receives the study medication correctly. If there is any complication with the administration (e.g. vomiting after taking the study medication) it will be reported as a note to file and directly to the lead centre. The used medication bottles will be collected and if needed can be used for additional evaluation of adherence after the patient has been discharged.

Relevant concomitant care permitted or prohibited during the trial {11d}

All treatments or medication considered necessary by treating doctors are permitted and their use will be recorded in the Case Report Form. There is no restriction in using other treatments or interventions during this trial.

Provisions for post-trial care {30}

Patients are carefully followed up until discharge from hospital. If there is any after-care necessary due to the Covid-19 it will be organized by the treating physicians and is not in connection with this trial. This trial has an insurance, which covers any harm that might happen to participants during this trial.

Outcomes {12}

As primary outcome, we fixed the length of the hospital stay. The measurement will therefore be the overall duration of the hospitalisation from admission until the discharge from hospital care.

As mentioned above, to measure the secondary objectives we will assess if there are any differences between the TAU + VitD and the TAU + Placebo group in terms of:

ICU stay	Yes/No
	If yes, length of the ICU stays (admission to discharge)
	Requirement for mechanical ventilation
Overall mortality	
Vitamin D serum concentration	% of patients with 25-(OH)D > 50 nmol/L at day 7
	25-(OH)D
Laboratory parameters	calcium
	Phosphorus
	parathyroid hormone (PTH)
Sepsis	% of patients who develop a sepsis
Complications	complications during the hospitalization due to Covid-19
Vital signs	Blood pressure (BP)
	Heart rate (HR)
	respiratory rate (RR)
	Blood oxygen saturation
	Need of oxygen and how much
Glasgow Coma Scale	

Participant timeline {13}

STUDY PERIOD								
	Enrolment	Randomization	Follow- Up Period					Discharge
TIMEPOINT**	-t ₁	0	t ₁	t ₂	t _{...}	t ₇	t _x	t _x
ENROLMENT:								
Eligibility screen	X							
Informed consent	X							
Allocation		X						
INTERVENTIONS:								
High Dose Vitamin D			X					
Standard Treatment for both groups								
Placebo			X					
ASSESSMENTS:								
Baseline Day 1		X						
Daily Visits until Discharge or fatality			X	X	X	X	etc.	X
Laboratory every 7 days and at Discharge			X			X	etc.	X

Sample size {14}

In order to calculate sample size, we looked at an interventional study of vitamin D, which examined length of hospital stay as an outcome and what is known about median length of hospital stay in patients hospitalised with Covid19. Two years ago a randomized, double-blind, placebo-controlled trial examined the effects of adjunctive vitamin D in adults hospitalized with community-acquired pneumonia (CAP) (35). The primary outcome was the complete resolution of chest radiograph infiltrate at 6 weeks post-study treatment. Secondary outcomes included length of hospital stay, intensive care admission and return to normal activity. Adjunctive vitamin D did not have any effect on the primary outcome (OR 0.78, 95% CI 0.31 to 1.86, p = 0.548). However, there was evidence it increased the complete resolution of pneumonia in participants with baseline vitamin D levels < 25 nmol/L (OR 17.0, 95% CI 1.40–549.45, P = 0.043). This effect was not observed when the data was analysed by baseline 25-OHD levels < 50 nmol/L [1]. Although not significant, the secondary outcomes indicated that the treatment group had lower probability of ICU admission OR = 0.95 (95% CI: 0.89, 1.01) and shorter length of hospital stay in the intervention group (mean = 3.8, SD = ± 3.1, n = 60) compared with the placebo group (mean = 4.8, SD = ± 6.5, n = 57), which indicates a small effect size (Cohen's d) of 0.1980. The study indicated a 20% reduction in length of hospital stay.

Considering the above results in the light of Covid-19, we have to expect longer hospital stays in Covid-19 patients. In smaller observational studies describing the clinical course of hospitalized Covid-19 patients in China, median hospital stay of 12 days (IQR 9–15) (n = 137) (1) or of 10 days (IQR, 7.0–14.0) among those discharged alive (n = 47) (36). Guan et al. presented the clinical characteristics of 1099 patients with laboratory-confirmed Covid-19 and the median length of hospital stay was reported to be 12 (IQR = 10–14) (37), which we will take as a basis for our sample size calculation.

Sample size calculation was performed using the function “power.t.test” from the R package “stats” with the following parameters: alpha = 0.05, power = 0.80, sigma = 2.96, delta (difference in primary outcome between intervention and control group) = 2, alternative hypothesis = two-sided, requires a n of 35 patients per group, therefore a total of 70. We estimate a 5 to 10% dropout, and that would mean 77 patients, because we have a block size randomization of four we decided to include 80 patients. Sample size calculation was performed using R (38).

Recruitment {15}

After a patient is identified as a potential participant, all inclusion criteria are fulfilled, and none of the exclusion criteria is present, the patient will be asked by their treating physician if he/she wants to participate in this study.

Only patients on general wards will be asked. Patients who are already being treated on the intensive care unit or who are taking part in another interventional trial will not be considered as potential participants. Neither patients nor doctors will receive a financial reimbursement. Expenses for the study medication and diagnostics performed only during this study will be covered by the study budget. The recruitment will take place in the described way in all four centres (cantonal hospital of Baselland in Liestal and Bruderholz, the cantonal hospital of Aarau, the cantonal hospital of St. Gallen and the regional hospital Lugano. Collaboration with other hospitals in Switzerland will be considered if recruitment is slow.

Assignment of interventions: allocation

Sequence generation {16a}

Participants will be randomly assigned to either the intervention or the control group. A randomization list with study group allocation will be generated using R (38). The randomization master list will be handed over to the hospital pharmacy of the university hospital in Basel for the labelling and packaging of the study medication. In case of an adverse event (AE) or serious adverse event (SAE) the hospital, pharmacy is able to decode the randomization immediately. Because standard care may change during the study period as new data on the management of Covid-19 becomes available from other centres, the randomization will be carried out in a block size of four. This will reduce the risk of a timing effect between study participants randomized to receive standard care or standard care plus single high-dose vitamin D.

Concealment mechanism {16b}

The hospital pharmacy of the university hospital of Basel will pack and label the study medication and the placebo after receiving the randomization master list. They will provide the study centres with medication packages for each patient that includes either the high dose vitamin D and the standard medication or the placebo and the standard medication. Participating centres will receive one or two blocks at the time, which they have to use fully before receiving the next blocks.

Implementation {16c}

The treating physician will make the enrolment. After a patient has given his /her consent, the physician will take one of the prepared medication packages with the medication. As described above the assignment to either group will be made randomly.

Assignment of interventions: Blinding

Who will be blinded {17a}

As the medication are provided by the hospital pharmacy in packaging labelled as study medication, neither the treating physician nor the patient or other care providers know if the patient is in the intervention or placebo group.

Procedure for unblinding if needed {17b}

When a SAE/AE occurs or in the event of a medical emergency, the hospital pharmacy is able to perform a patient-specific unblinding at any time during the study. In such a case, treating doctors can call the pharmacist who has access to the concealed list.

Data collection and management

Plans for assessment and collection of outcomes {18a}

All outcomes are assessed by treating physicians and/or nurses until discharge or fatality. The CRF is provided either electronically (web-based) or in paper form in which case it will be sent by mail or fax to the coordinating study centre, where data will be entered into the web-based database. Data collection ends at the day of discharge.

The primary outcome is the length of hospital stay. The exact length will be calculated automatically by the electronic database into which only the date of the admission and the date of discharge have to be entered. With this method copying or calculation errors can be minimized.

Secondary outcomes are the following:

ICU stay	Yes/No
	If yes, length of the ICU stays (admission to discharge)
	Requirement for mechanical ventilation
Overall mortality	
Vitamin D serum concentration	% of patients with 25-(OH)D > 50 nmol/L at day 7
	25-(OH)D
Laboratory parameters	calcium
	phosphorus
	parathyroid hormone (PTH)
Sepsis	% of patients who develop a sepsis
Complications	complications during the hospitalization due to Covid-19
Vital signs	BP
	HR
	RR
	Blood oxygen saturation
	Need of oxygen and how much
Glasgow Coma Scale	
Demographic Data	Age
	Sex
Medical history	co-morbidities
	smoking status
Medication	current medication
	changes in medication
Influenza Vaccination Status	

The treating physicians will provide the research team with a copy of all laboratory results (anonymized, with participant identification number) as source data. This procedure was chosen to avoid transcription errors in the paper based CRF and/or the electronic CRF.

Plans to promote participant retention and complete follow-up {18b}

- The study team and the ward staff are available for any questions or concerns a patient may have regarding this trial. As this trial only lasts for the duration of hospitalisation, we do not anticipate problems with participant retention.

Data management {19}

Data acquisition will be performed by participating centres, entering data directly through a web-based electronic Case Report Form (eCRF) or by sending a paper version of the CRF to the research team at the main study centre (Kantonsspital Baselland, Liestal), where data entry will be performed by one of the team members. For data verification purposes, data from paper CRFs entered at the study centre will be double-checked. Data about adverse events will be reported by participating centres to the research team at the lead centre. The lead centre will inform the relevant authorities (ethics committee and the Swiss agency for therapeutic products).

Data entered into the electronic CRF or paper version of the CRF will be treated as source data. Information about medical treatment and medical history of the participant is part of the CRF. Documents relating to the study (written informed consent) will be sent or faxed to the lead study centre by participating physicians and treated as source data as well. Source data corresponding to the CRF will be found either in the electronic database when entered by the participating centres or in the paper version stored at the lead study centre (paper version of CRF, written informed consent).

To minimize copying errors, a copy of the lab results will be treated as source data as well.

All study data will be archived at the lead study centre in Liestal for a minimum of ten years after study termination or premature termination of the clinical trial.

The data will be collected in a web-based electronic data capture (EDC) system, named secuTrial®. The EDC system runs on a server maintained by the IT-department of the university hospital Basel. The electronic CRF (eCRF) will be configured by the data management at the clinical trial unit (CTU) at the university hospital Basel.

The EDC system is accessible via a standard browser on a www -connected device. Password protection ensures that only authorized persons can enter the system to view, add or edit data according to their permissions. User administration and user training is performed by the CTU according to predefined processes. Back up of secuTrial® study data is performed according to the processes of the IT-department of the university hospital Basel.

The EDC will be locked after all data was monitored and all raised queries have been resolved. Data is exported and transferred to the investigator by the CTU according to internal defined processes. Data will be archived by the investigator.

Data is entered into the eCRF and can be validated for completeness and discrepancies automatically. An audit trail maintains a record of initial entries and changes (reasons for changes, time and date of

changes, user identification of entry and changes). The data entered into the eCRF will be reviewed by the responsible centres and an independent monitor will raise queries using the query management system implemented. The participating centre has to respond to the query and confirm or correct the corresponding data. Thereafter a monitor can close the query.

Confidentiality {27}

All collected data will be treated as confidential. All data will be anonymised by the participating centre and data will be stored and analysed in an anonymised way. Results of this study will be published in an anonymised fashion. Direct access to source documents will be permitted for purposes of monitoring, audits and inspections. The study protocol and dataset shall be accessible to any regulatory authority after publication for at least ten years.

- **Plans for collection, laboratory evaluation and storage of biological specimens for genetic or molecular analysis in this trial/future use {33}**

We have no plans for collection, laboratory evaluation and storage of any biological specimens for genetic or molecular analyses in this trial or for further use. This trial only uses blood samples, which are taken routinely during hospitalisation of the patients and no further samples of any other biological material is planned.

Statistical methods

Statistical methods for primary and secondary outcomes {20a}

Detailed methodology for summaries and statistical analyses of the data collected in this trial is documented in a statistical analysis plan. The statistical analysis plan is finalized before database closure and is under version control. We will summarize our results using descriptive statistics. R will be used for all statistical analysis (38). The full analysis set (FAS) consists of all patients who were randomized. The intention-to-treat (ITT) set includes all patients in the FAS for whom complete measurements are available. Each patient will be analysed according to the treatment he or she was randomly allocated. The per protocol (PP) set consists of all patients in the FAS without any major protocol deviation. Each participant will be analysed according to the treatment he or she actually received. Details will be given for patients who, for whatever reason, sign the consent form but have not been treated.

Primary Analysis

The primary analysis of comparing the mean length of hospital stay of both groups will be performed using a two-sided independent Student's t-test.

Secondary Analyses

For the secondary outcome analyses, mean values of ICU stay, the level of calcium, phosphorus, 25-hydroxyvitamin D, 1,25-dihydroxyvitamin D, PTH will be compared using two-sided independent Student's t-tests. In order to compare the proportion of patients who died, developed sepsis as well as the proportion of patients with 25-hydroxyvitamin D ≥ 30 ng/mL between the two groups, either a z-test (if data permits) or Fisher Exact probability test, will be used. The Fisher Exact probability test is an excellent non-parametric technique for comparing proportions, when the two independent samples are small in size.

Interim analyses {21b}

Due to the relatively small sample size and short study duration, we will not identify stopping criteria nor perform interim analyses.

Methods for additional analyses (e.g. subgroup analyses) {20b}

To evaluate the safety of short-term vitamin D administration in the above-described population, linear/logistic mixed-effect models will be used to estimate treatment effects on safety parameters such as adverse events/laboratory parameters (e.g. hypercalcemia, hypercalciuria) and vital signs.

- Methods in analysis to handle protocol non-adherence and any statistical methods to handle missing data {20c}

Careful trial planning and execution will minimize the occurrence of missing data as far as possible. In cases where data is missing, treating physicians will be contacted with the aim to complete the data from patients' records. Multiple imputation might be used if none of the reasons cited in Jakobsen (39) practical guide is fulfilled.

Plans to give access to the full protocol, participant level-data and statistical code {31c}

We plan to publish this protocol, meaning it will be accessible to the public. As soon as data collection and analysis is completed, the complete statistical methods and results will also be published. Competent authorities (CA) will have full access to anonymised patient data and full statistical code upon request.

Oversight and monitoring

Composition of the coordinating centre and trial steering committee {5d}

Professor Leuppi at the medical university clinic of the cantonal hospital Baselland leads this study, which is the coordinating centre. The multidisciplinary research team includes medical doctors, a clinical pharmacologist, study nurses, a statistician and public health scientists. Furthermore, the study team is

supported by the clinical trial unit from the department of clinical research at the university hospital of Basel. A trial steering committee is not planned.

Composition of the data monitoring committee, its role and reporting structure {21a}

All study data and documents shall be accessible to monitors and questions will be answered during monitoring. The site initiation visits and the close out visit will be done by the sponsor. Scheduled monitoring time points are after inclusion of the first 10 patients and after the last visit of the last patient.

Adverse event reporting and harms {22}

Reporting of SAEs

All SAEs must be reported immediately and within a maximum of 24 hours to the Sponsor-Investigator of the study. The Sponsor-Investigator will re-evaluate the SAE and return the form to the site. SAEs resulting in death are reported to the Ethics Committee via BASEC within 7 days.

All involved Ethics Committees receive SAEs resulting in death in Switzerland via Sponsor-Investigator via BASEC within 7 days.

Reporting of suspected unexpected serious adverse reactions (SUSARs)

A SUSAR needs to be reported to the Ethics Committee (local event via local Investigator) via BASEC and to Swissmedic for category B and C studies (via Sponsor-Investigator) within 7 days, if the event is fatal, or within 15 days (all other events).

The Sponsor-Investigator must inform all Investigators participating in the clinical study of the occurrence of a SUSAR. All in the trial involved Ethics Committees will be informed about SUSARs in Switzerland via Sponsor-Investigator via BASEC according to the same timelines.

Reporting of Safety Signals

All suspected new risks and relevant new aspects of known adverse reactions that require safety-related measures, i.e. so-called safety signals, must be reported to the Sponsor-Investigator within 24 hours. The Sponsor-Investigator must report the safety signals within 7 days to the Ethics Committee (local event via local Investigator) via BASEC and to Swissmedic in case of a category B or C study.

The Sponsor-Investigator must immediately inform all participating Investigators about all safety signals. The other Ethics Committees involved in the trial will be informed about safety signals in Switzerland via the Sponsor-Investigator.

Reporting and Handling of Pregnancies

Pregnant participants must immediately be withdrawn from the clinical study. Any pregnancy occurring during the treatment phase of the study and within 30 days after discontinuation of study medication will be reported to the Sponsor-Investigator within 24 hours. Pregnancy is not a contraindication for Vitamin D. Nevertheless, pregnant women will be excluded from study participation.

Periodic reporting of safety

An annual safety report (ASR) is submitted once a year to the local Ethics Committee by the local investigator and to Swissmedic in case of a category B study, as this study is categorised, via Sponsor-Investigator.

Frequency and plans for auditing trial conduct {23}

Independently from investigators, regulatory authorities can audit this trial. Study documentation and data are accessible to auditors/inspectors and questions are answered during inspections. All involved parties must keep the participant data strictly confidential.

Plans for communicating important protocol amendments to relevant parties (e.g. trial participants, ethical committees) {25}

Substantial amendments are only implemented after approval of the local ethics committee (LEC) and Swissmedic respectively.

Under emergency circumstances, deviations from the protocol to protect the rights, safety and well-being of human subjects may proceed without prior approval of the sponsor and the LEC/CA. Such deviations shall be documented and reported to the sponsor and the LEC/CA as soon as possible.

All non-substantial amendments are communicated to the CA as soon as possible if applicable and to the CEC within the ASR.

Dissemination plans {31 a}

We aim to publish results of this study in a peer-reviewed journal. There is no intention to use professional writers. This trial is registered at www.clinicaltrials.gov and kofam.ch and therefore accessible to the public.

Discussion

The primary aim of this study is to determine, whether a high dose of vitamin D improves the course of Covid-19 or not. Two issues might affect the ability to recruit patients for this trial: first levels of vitamin D are the highest by the end of summer, (40) and this could lead to problems in. After the full ethics approval and the approval from Swissmedic, the start of the recruitment period is expected to be the beginning of fall 2020. Shorter days and less exposure to sunlight most certainly leads to a decreasing

serum concentration of vitamin D. We therefore anticipate, that we will not have recruiting problems due to high vitamin D levels.

Secondly, travel regulation and other policies can change day by day and is carefully monitored by the Swiss government, which leads to a more unpredictable situation for this and other studies investigating Covid-19.

While the number of patients hospitalised with Covid-19 declined between April and August, numbers have started to rise again. Latest studies have shown a link between international and domestic air travel and the number of Covid-19 cases, so unless air travel is restricted again, we can expect an increasing number of cases (41,42). The same can be applied for lockdown policies and other public health measures which lead to decreasing numbers of cases (43,44), and subsequent rebound after the measures were gradually released. Nevertheless, we aim to include the 80 patients as soon as possible and make a contribution to treatment guidelines for patients with Covid-19.

Trial Status

This study was submitted to the local ethics committee the first-time at the end of May 2020. After the feedback we revised the study protocol to change the design from an open label to randomized controlled double blind. This version (2, 05.08.2020) of the study protocol is approved by the regional ethics committee without further obligations.

We are awaiting the answer of Swissmedic so that we can start the trial. We plan to include the first patient at the latest at the end of October. recruitment should be completed within 10 months (mid 2021).

Abbreviations

AE	Adverse Event
ARDS	Acute respiratory distress syndrome
ASR	Annual Safety Reprot
BASEC	Business Administration System for Ethical Committees, (https://submissions.swissethics.ch/en/)
BP	Blood Pressure
CA	Competent Authority (e.g. Swissmedic)
CRF	Case Report Form
CoV	Coronavirus
eCRF	Electronic Case Report Form
CTU	Clinical Trial Unit
EKNZ	ethical committee of northwestern and central Switzerland
EKOS	ethical committee of eastern Switzerland
EOC	Ente Ospedaliero Cantonale (Regional Hospital Ticino)
FAS	Full analysis set
HR	Heart rate
KSA	Kantonsspital Aarau
KSBL	Kantonsspital Baselland
KSSG	Kantonsspital St. Gallen
LEC	Local Ethics Committee
MERS	Middle east respiratory syndrome
PI	Principal Investigator
PP	Per protocol
PTH	Parathyroid Hormone
RR	Respiratory rate
SAE	Serious Adverse Event
SARS	Severe acute respiratory syndrome
SUSAR	Suspected Unexpected Serious Adverse Reaction
TAU	Treatment as usual
TI	Ticino

Declarations

Acknowledgements

The authors of this manuscript thank hospital colleagues and colleagues from the Centre for Primary health care at the University of Basel for their support for this study.

Authors' contributions {31b}

JDL: Is the Sponsor-Investigator of this trial. SG is the trial statistician. FJ, CLG, KA,PH and PT are Co-Investigators at the lead centre. MB, PS,LE, LG are local Principal investigators. FJ wrote the first draft of this manuscript, which was revised critically by all authors. KA, FJ, GLC MB, PS, LG, LE, ALT, JM, PH, PT made contribution to the protocol with their expertise. FJ, KA, GLC and AM are responsible for the project administration.

Funding {4}

This study is financed by the research fund of Professor Leuppi. Additionally, it is foreseen to obtain more financial support from other foundations.

Availability of data and materials {29}

The primary owner of the data which are collected within this trial is Professor Leuppi. With all the local project leaders, an agreement is available which declares duties and responsibilities of all parties within this project.

Furthermore, the study protocol and dataset shall be accessible to any regulatory authority for at least ten years after publication.

Ethics approval and consent to participate {24}

Attached to this document you will find the statement of the Swiss ethics committee (BASEC No: 2020-01401). In this case several local ethical committees are involved.

EKNZ (ethical committee of north-western and central Switzerland) is the lead ethics committee in this trial, full ethical approval was granted on September 23rd 2020.

EKOS (ethical committee of eastern Switzerland), the obligation was fulfilled, we are awaiting the answer and the the full approval.

CEC TI (ethical committee of Ticino): We are in the process of translating the documents into Italian for submission.

A written informed consent will be obtained from all study participants.

Consent for publication {32}

No personal data and/or video of any patient is used in this manuscript therefore a consent for publication is not needed.

Competing interests {28}

None of the authors has any competing of interests.

Authors' information (optional)

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