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Prevention of COVID-19 with Oral Vitamin D Supplemental Therapy in Essential Healthcare Teams (PROTECT trial): protocol for a multicentre randomized placebo-controlled, triple-blind trial

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ABSTRACT

Introduction: In the COVID-19 pandemic, health care workers (HCWs) have been at high-risk of infection due to their exposure with COVID infections. HCWs are the backbone of our health care response to this pandemic, and every health care worker withdrawn or lost due to infection has an exponential impact on our capacity to deliver care. Primary prevention is a key approach to reduce infection. Vitamin D insufficiency is highly prevalent in Canadians and worldwide. Vitamin D supplementation has been shown to significantly decrease the risk of respiratory infections. Whether this risk reduction would apply to the COVID-19 infection remains to be determined. This study aims to determine the impact of high-dose vitamin D supplementation on incidence of laboratory-confirmed COVID19 infection in HCWs working in high COVID incidence areas.

Methods and analysis: This is a triple-blind, placebo-controlled, parallel-group multicentre trial of vitamin D supplementation in HCWs at high-risk of infection. Participants were randomly allocated in a 1:1 ratio in variable block size to: Intervention—1 oral loading dose of 100,000 IU vitamin D3 + 10000 IU weekly vitamin D3 or control—identical placebo loading dose + weekly placebo. The primary outcome is incidence of laboratory-confirmed COVID-19 infection, documented by RT-qPCR on salivary (or nasopharyngeal) specimens obtained for screening or diagnostic purposes, as well as self-obtained salivary specimens obtained at endpoint and COVID-19 seroconversion at endpoints. Secondary outcomes include disease severity; duration of COVID-19 related symptoms; COVID-19 seroconversion documented at endpoint; duration of work absenteeism; duration of unemployment support; and adverse health events.

Ethics and dissemination: This study was approved by the Research Ethics Board of the CHU Sainte-Justine and participating institutions. If proven effective in reducing the risk and morbidity

of COVID-19 infection, vitamin D supplementation could offer the cheapest, most easily implementable primary prevention strategy for HCWs.

Strengths and limitations of this study

- This multicentre randomised controlled trial will be the largest adult study testing the impact of high-dose vitamin D supplementation, compared to placebo, on the risk of infection and severity of COVID-19 in health care professionals.
- This trial was designed as a hybrid study enabling partially or totally remote screening, randomisation, follow-up, as well as outcome documentation by use of home capillary blood sampling, rapid capillary SARS-CoV2 serology, salivary self-sampling, videoconference, electronic reminders and questionnaires and communication by phone, text messaging or emails.
- This trial used a pragmatic subject selection and easily applicable intervention to maximise subsequent implementation in practice.
- The main outcome is clinically meaningful as it explores the primary prevention impact of vitamin D on the risk of laboratory-confirmed infection; it is likely to change practice if a 20% reduction is documented.
- Because of the variability in diet, vitamin D supplement use, sun exposure, and skin color, it is impossible to control all factors that may affect circulating 25-hydroxyvitamin D levels; however, it is expected that these factors will be balanced between groups due to randomization.
- A loading and regular doses have been shown to lead to rapid and sustained increase in serum level of 25-hydroxy-vitamin D and ensure adequate group separation. A rapid increase is particularly desired in the context of a rapidly expanding epidemic while weekly doses facilitate adherence in exhausted frontline health workers.

- Given the uncertainty in the progression of the infection rate, the use of a Bayesian adaptive design allows for adaptations (early stopping or prolongation of duration of follow-up) at the interim analysis according to the projection of infection rates.

Introduction

The Coronavirus 2 (SARS-CoV-2) disease (COVID-19) outbreak has rapidly expanded to a global pandemic. Healthcare workers (HCWs) play a crucial role in the fight against the COVID-19 pandemic. It was therefore a public health priority to develop strategies to decrease the risk and severity of COVID-19 in this vulnerable population. Indeed, there was rising concern as HCW are overrepresented in terms of infections (3.8% of infected individuals in Wuhan, China,[1] 10% in Italy and 12% in Spain, 10-20% in US)[2 3] and perhaps severity. Working in long-term care facilities (LTCF) and with aerosol generating medical procedures (e.g., hospitals) further increased the risk (Odds Ratio [OR]: 2.3).[4] The risk of reporting COVID-19 infection in front-line HCWs, defined as those in direct contact with patients, was 10-fold greater than the general population at the beginning of the pandemic (Hazard Ratio [HR]= 11.61). [5] Recent research also indicated that HCWs who were Blacks, Asians, or other minority ethnic populations, had a higher likelihood of contracting COVID-19.[5] Compared to those unexposed to COVID-19 patients; the risk was two to five-fold higher in HCWs exposed to COVID-19 suspected (HR= 2.39) or confirmed (HR= 4.83) patients, even with adequate personal protection equipment (PPE).[5] Although infections may be due to contact with infected patients, community, or family acquired disease, cases were rapidly emerging from cross-infection with asymptomatic infected HCW.

Vitamin D is an immunomodulatory micronutrient, and its levels in the body may vary due to diet and environmental conditions. Vitamin D insufficiency has been associated with increased risk of respiratory infections, and possibly COVID-19,[6] asthma exacerbations, and acute respiratory distress syndrome (ARDS) among others.[7-9] Optimal pro-immune and anti-inflammatory impacts likely occur at 25-hydroxyvitamin D (25OHD) levels above 75 nmol/L (30 ng/mL).[10,11] In a systematic review of 25 randomized controlled trials (RCT) of 11321 individuals, daily/weekly vitamin D supplementation decreased by 19% the rate of acute respiratory infections (two-step

analysis; OR 0.81, 95% CI 0.72 to 0.91),[12,13] with a stronger effect in subjects with baseline 25OHD <25 nmol/L. Whereas subgroup analyses suggested a protective effect primary in individuals receiving daily or weekly vitamin D supplement without an additional bolus, but not in those with bolus,[14] other important differences in population (e.g., malnutrition),[15,16] age (infant),[16] chronic disease (e.g. asthma, COPD)[17-21] and type of infection (e.g. bacterial)[15,16] could have contributed to the apparent lesser effect. Vitamin D supplementation was also found to be associated with a decreased load of rhinovirus (common cold), consistent with an increased antiviral immune response.[22] A systematic review and several studies reported an inverse association between serum vitamin D levels and COVID-19 severity, inpatient mortality, as well as serum levels of C-reactive protein (CRP) and lymphocyte percentage.[23,24] These findings suggest that vitamin D status is linked with the severity and mortality of the COVID-19 infection in the general population, particularly in severe COVID-19 cases.

The vitamin D receptor is expressed on innate and adaptive immune cells which also synthesize the active metabolite 1,25-hydroxyvitamin D_3 ($1,25(OH)_2D_3$); thus, vitamin D can strengthen innate and adaptive cellular immunity by increasing local production of antimicrobial peptides, decreasing secretion of pro-inflammatory cytokines, inhibiting dendritic cell activation, suppressing T helper cell type 1 response, and promoting T regulatory cells induction. These cellular effects are crucial for host responses against infection and can reduce the survival and replication of respiratory viruses.[13,24] $1,25(OH)_2D_3$ is also produced locally in bronchial epithelial cells and downregulates inflammatory cytokines (e.g. interleukin-8) and chemokines (e.g. leucocyte attracting CXCL10) expression from stimulated cells.[25]

The protocol of a placebo-controlled parallel-group triple-blind RCT to explore the impact of vitamin D_3 supplementation on reducing the risk of laboratory-confirmed COVID19 infection in HCWs is described herein, as per Standard Protocol Items: Recommendation for Intervention Trials guidelines **(Online supplemental file 1).** After funding, but prior to the start of recruitment, the protocol underwent four amendments (8 protocol versions) in view of the rapidly evolving science, multiple challenges faced with conducting a large scale COVID-19 trial of high-risk health-care workers during the pandemic, including difficulty in obtaining large-scale supplies, as well as favorable pilot results of two novel technologies **(Table 1).** These original and final (1.8, January 18, 2021) protocol versions are described below.

Research questions and study hypothesis

Objectives

The primary question was whether one oral dose of 100,000 IU vitamin D_3 (administered at baseline) plus weekly supplement of 10,000 IU vitamin D_3 can decrease the risk of laboratory-confirmed COVID-19 infection, versus placebo, in frontline HCWs in high COVID-19 incidence areas.

Additionally, the study aimed to examine if, compared with placebo, the vitamin D intervention reduces: (i) illness severity, (ii) symptom duration, (iii) work absenteeism, and (iv) unemployment among frontline health care workers (HCW) in high COVID-19 incidence areas. This study will also assess various exploratory outcomes.

Hypothesis

We hypothesised that compared to placebo, vitamin D supplementation would decrease the incidence of laboratory-confirmed symptomatic COVID-19 infection by 20% in frontline HCWs working in high COVID-19 incidence area.

Table 1. Study Amendments and Notifications

	Clinical trial Application (CTA)				Investigational testing Authorization (ITA)	
Version number	**Changes**	**Description**	**Submitted**	**Approved**	**Submitted**	**Approval**
Version 0.0 11-05-2020						
Version 1.0 23-08-2020	• Eligibility • Outcomes & Covariates	• Strengthening of exclusion of 'suspected or previously undocumented COVID-19 infection' by adding: (i) a questionnaire of symptoms elaborated by Menni et al. and (ii) a rapid (15-minute) serology test, not yet approved nor tested in Canada, to be pre-tested in a pilot study. • Addition of capillary blood self-collection with Tasso-SST device (to be pre-tested in a pilot study).	23-08-2020	16-09-2020	N/A	N/A
Amendment 1 **Version 1.1** 23-10-2020	• Eligibility • Exploratory Outcomes • Main outcome	• Clarification of the NADAL® COVID-19 IgG/IgM Rapid Test (Teracero Pharma Inc., Lachine Canada) on venous whole blood as the rapid serology test to exclude prior infection (following pilot comparative study) • Validation of Nadal serology test compared to IgG SARS-CoV2 serology as endpoint exploratory outcome • Salivary COVID-19 RT-PCR test method prioritized over nasopharyngeal samples for twice-monthly self-collection or accepted for clinical diagnostic by qPRC	23-10-2020 (CTA-A) †	02-11-2020	23-10-2020 (NADAL)	14-111-2020
Amendment 2 Versions 1.2-1.4 **Version 1.5** 27-11-2020	• Primary Outcome • Outcomes	• Removal of q2 weeks saliva sample for COVID-19 rt-PCR analysis due to supply problem with 50 mL Falcon centrifuge tube caused by a global plastics shortage combined with un-acceptable delay for public tender for a contract with courier service for q2weeks biological samples • Addition of questions and procedures to account for the possible effect of the Vitamin D supplementation on immune response to COVID-19 vaccine, including a saliva sampling for COVID-19 by RT-PCR and a blood test for serology (and vitamin D) prior to vaccination	27-11-2020 (CTA-A) †	30-11-2020	23-11-2020 (TASSO & NADAL)	2-12-2020

	Clinical trial Application (CTA)				Investigational testing Authorization (ITA)	
Version number	**Changes**	**Description**	**Submitted**	**Approved**	**Submitted**	**Approval**
	• Covariates & Outcome (Device)	• Specification of the TASSO SST on Demand (Tasso Inc, Seattle, USA) as choice for capillary blood self-sampling (following pilot comparative study)				
Amendment 3 versions 1.6 & **Version 1.7** 12-12-2020	• Eligibility • Exploratory Outcomes	• Exclusion of health care workers who have received the COVID-19 vaccine prior to enrolment • Addition of (i) effect of high-dose vitamin D on SARS-CoV-2 IgG titers before & after 2nd dose of COVID-19 vaccine and (ii) the long-term infection rate up to 12 months after end-of-study • Modifying exploratory outcome to allow exploration of modulating effect of vitamin D, not only on the risk of COVID-19 infection but also on response to vaccine	12-12-2020 (CTA-A) †	16-12-2020	12-12-2020 (TASSO & NADAL)	23-12-2020
Amendment 4 Version 1.8 18-01-2021	• Exploratory Outcomes • Eligibility	• Clarification that the serology to be done just prior the second dose of a COVID-19 vaccine may not always be at 3 or 4 weeks (as recommended by vaccine manufacturer) to reflect the recent governmental decision to delay the timing of the 2nd vaccine dose to 12 to 16 weeks • Slightly modifying wording to target healthcare workers at risk of contact with infected individuals that were not suspected of being infected (e.g., patients, colleagues, students, etc.)	18-01-2021 (CTA-N) ‡	N/A	18-01-2020 ¶	01-02-2021

† CTA-A, Clinical Trial Application -Amendment
‡ CTA-N, Clinical Trial Application -Notification
¶ As there is no 'notification' category for the Investigational Testing Authorization (ITA), each amendment or notification to the Clinical Trial Application (CTA) must be submitted as a new amendment for the devices to be reviewed by ITA.

Methods and analysis

Study design

This was a pragmatic 16-week, triple-blind, placebo-controlled, parallel-group, randomized trial comparing supplemental vitamin D3 and placebo in HCWs with the possibility of extending the study up to 24 weeks, depending on infection rate progression during an interim analysis **(Figure 1)**.

Subjects

HCWs (i.e., physicians, allied health care workers, orderlies, etc.) were eligible if they: (i) were aged ≥18 and <70 years old; (ii) were authorized to practice in Quebec; (iii) were working or scheduled to work over the next 16 weeks in a setting at high-risk of contact with COVID-19 infected individuals, particularly (but not only) those involved with aerosol generating medical procedures in hospitals and/or caring for patients in long-term care facilities; (iv) were working in high COVID incidence areas in the greater Montreal area and surroundings; (v) were covered by the provincial universal public health insurance (*Régie de l'assurance-maladie du Québec* [RAMQ] for medical services and hospitalisations; (vi) had a personal email or phone (to which to send reminders and questionnaire by email or text messages); (vii) had a fixed address (to which to send the material) in the greater Montreal or surrounding areas.

HCWs were excluded if they met any of the following criteria: vitamin D supplementation (cholecalciferol or calcitriol) intake >400 IU/day (or >12,000 IU/month) in past 3 months; intention to take >400 IU per day during the study period; suspected or previously documented COVID-19 infection; history of nephrolithiasis, hypercalcemia, hyperphosphatemia, hyperparathyroidism, granulomatosis disease (e.g., tuberculosis, sarcoidosis), renal failure, or

active cancer; current intake of medications that may cause hypercalcemia such as lithium, teriparatide, or digoxin; anticipated prolonged absence from work during the study period (i.e., pregnancy); anticipated difficult follow-up; enrollment in a concurrent interventional randomized trial; have already received the vaccine against COVID-19. Participation in this trial did not preclude subsequent enrollment in a COVID-19 therapeutic (but not preventive) trial, which would be documented.

Study intervention

Participants in the intervention group received 100,000 IU vitamin D_3 at randomization followed by a weekly dose of 10,000 IU vitamin D_3 for 16 weeks. Participants in the control groups received an identical placebo bolus followed by placebo weekly supplement for 16 weeks. Sufficient supply was provided for 24 weeks, in case of prolongation study based on the interim analysis. Participants in both groups were asked to take the study intervention with their most copious meal. Treatment of co-morbidities were permitted. Vitamin D intake up to 400 IU per day was allowed.

Randomization

Randomization was implemented using a computer-generated random list stratified by one of 11 workplaces (*Centre Hospitalier Universitaire* [CHU]) or health region (*Centre Intégré Universitaire de Santé et Services Sociaux* [CIUSSS] or *Centre Intégré de Santé et Services Sociaux* [CISSS]). HCWs were allocated (1:1) using permuted block randomization to enhance concealment. Group allocation codes for each stratum was held in a secure location with restricted access by the Central Pharmacy and Data Management.

Patient and public involvement

Participant burden of research measures was assessed using feedback from patients participating in one pilot round. Patients were not involved in study deign, recruitment of participants or conduct of the study. Results of this study will be disseminated through public fora.

Outcomes

Primary outcome

The original primary outcome, incidence of laboratory-confirmed COVID-19 infection, was originally based on (i) bi-monthly self-obtained mid-turbinate nasopharyngeal (NP) swabs, complemented by (ii) NP swabs obtained clinically for screening or diagnostic purposes throughout the study, both analysed by RT-qPRC approved by Health Canada. Faced with the unexpected cancellation of our large order of Falcon tubes to collect saliva sample for qPRC combined with the unacceptable additional delay for a public tender to securing a contract with a private courier service and in view of the uniform protocol for screening symptomatic or COVID-19 exposed health care workers throughout the Province of Quebec and the reliability of IgG serology, we decided to forgo the twice-monthly saliva sampling for qPRC analysis. The revised definition of the primary outcome became the incidence of laboratory-confirmed COVID-19 infection, documented by RT-qPCR based on salivary (or nasopharyngeal) specimens (i) obtained for screening or diagnostic purposes throughout the study and (ii) self-obtained salivary specimens obtained at endpoint as well as (ii) COVID-19 IgG seroconversion at endpoint (in COVID-unvaccinated individuals: ≥15 UA on the anti-S SARS-CoV-2 IgG Diasorin on Liaison XL platform; in COVID-vaccinated individuals : ≥1.40 index (S/C) on the anti-N SARS-CoV-2 IgG on ARCHITECT platform)

Secondary outcomes

(i) *Distribution of disease severity* on a 5-category ordinal scale [asymptomatic; mild (managed at home); moderate (hospitalisation without supplemental oxygen); severe (oxygen supplementation); critical (mechanical ventilation/death)], (self-reported, RAMQ); (ii) *Duration of COVID-19 positivity* between 1st COVID+ to first COVID- test) revised to *Duration of COVID-19 related symptoms* in individuals with laboratory confirmation of COVID infection, (self-reported on diary); (iii) COVID-19 IgG seroconversion documented at endpoint (see above); (iv) duration of work absenteeism (self-reported, medical records or human resources databases); (iv) duration of unemployment support (human resource databases); (v) Adverse health events (self-reported). Several *exploratory outcomes* pertained to the: incidence of post-acute and chronic symptoms; long-term (1-year) morbidity and work absence related to COVID-19; change in gene expression of ACE2 and TMPRSS2 in saliva cells; change in inflammatory markers (i.e., CRP), immune response post vaccination; other viral infections; and genetic markers (including changes in gene expression).

Study Procedures

To facilitate the recruitment of participants, this study is conceived as hybrid trial enabling partially or totally remote trial participation including screening, randomization, follow-up, and end-of-study visit.

Pre-Screening

Advertisements were placed in health institutions, newspapers, social media and online, where participants were invited to complete an online pre-screening form, read and download the consent forms; and if eligible and interested, to book a virtual screening appointment (via a secured videoconferencing platform) with research team who would confirm eligibility, explain the study, obtain informed consent, and schedule a virtual or in-person randomization visit.

Screening

At the virtual screening visit by videoconferencing, research coordinators completed with the individual a more extensive eligibility questionnaire, which included additional questions about: anticipated work exposure over the next 16 weeks to COVID-infected or suspected individuals and to high-risk medical procedures; work place (*Centre Hospitalier Universitaire* [CHU]) or Centre Hospitalier Universitaire Sainte-Justine) or health region (CIUSSS or CISSS), serving as randomization stratum; prior laboratory-confirmed or physician-suspected COVID-19 infection; assessment of the likelihood of prior/current, yet undocumented, COVID-19 infection using the 5-item questionnaire developed by Menni et al[26] (score >0.50 interpreted as high likelihood of prior infection); and finally, the comfort level with the study design and procedures, including saliva and capillary blood sampling self-collection demonstrated by instructional videos. Eligible and consenting individuals electronically signed an online consent form (with the signed PDF consent form automatically emailed to participants). Then, two additional questionnaires were completed on line with the research coordinators namely: (i) the baseline questionnaire collecting information about household, ethnicity, part- vs. full-time work, personal health, skin color (measured with the Fitzpatrick scale),[27] concomitant medications or supplements, and (ii) the nominative CRF collecting demographic information essential to opening a medical and pharmaceutical research

record (i.e., public health insurance number, allergies) and maintaining contact with the research team throughout the study (preferred means to receive electronic reminders/questionnaires and to be notified of positive test results; address to receive study material or for biological sample pick-up; and next-of-kin contact in case of inability to respond to questionnaire due to illness), and to document work absence (employee number).

Finally, at the screening visit, the participant was asked to choose an appointment for a *virtual* (via a secured videoconferencing platform) or *in-person* randomization visit at one of several locations. To help select their preferred visit format, videos of key procedures (such as home blood collection) were shown. Only in participants with a significant likelihood of a current or past undocumented (Menni score > 0.5) was an *in-person* randomization visit mandatory to receive the rapid COVID-19 serology test, prior to randomisation.

Preparation and shipment of Study drug by Research Pharmacy

The list of new participants approved by one of the PIs was sent daily by email to the CHUM research team to be open a medical chart and send an electronically signed prescription for the Study medication, to the Research Pharmacy for preparation of study drug.

Prior to randomization, a list of all consenting and eligible participants was automatically sent every night to the one of the co-principal investigators (FMD or CT) who screening and baseline questionnaires to approve or refuse study entry and electronically signed their decision. The list daily list of new PI-approved participants was sent electronically daily to the CHUM research team. Medical and pharmaceutical records were opened and an electronically signed prescription

for the Study medication sent to the Research Pharmacy for preparation of study drug for a given target date.

To enable remote randomization, the randomization took place about one week prior to the randomization visit to allow enough time for the preparation and shipment of patient-specific study supplement to the research team and, in turn, the shipment of the Study supplement and all materials required for the randomization visit by the research team to the participant.

Randomization visit

Seventy-two and 24 hours prior to, and at, the randomisation visit, participants were screened by questionnaire for recent travel, symptoms suggestive of SARS-Cov2 infection, or exposure to COVID-19 infected individuals. Those who responded positively were asked to get tested, notify their institutional health service and await end of quarantine and/or confirmed negative test to reschedule the randomisation visit.

Randomization visit (week 0) was performed *in person* (60 minutes) or *remotely* (90 minutes), depending on the availability and preference of participants as well as their likelihood of a past COVID-19 infection.

In-person visits were conducted—by appointment only—in designated rooms with restricted access. The research coordinators wore personal protection equipment (PPE), and all procedures, from participant arrival to departure, were approved by the institutional Infection Control and Safety committee. The *in-person* visit entailed (i) ascertainment of the signed consent form, (ii) capillary blood sample collection to perform NADAL® COVID-19 IgG/IgM Rapid Test (Teracero

Pharma Inc., Lachine Canada), (iii) venous blood sample collection for baseline serum 25(OH)D and SARS-CoV-2 IgG serology analyses and if genetic consent, DNA; (iv) viewing of the saliva collection video and instruction pamphlet, (v) collection of the first specimen under supervision, (vi) a final verification of the eligibility and exclusion criteria; (vii) randomization; (viii) oral administration of 100,000 IU vitamin D_3 or an identical placebo, and (ix) distribution of the study material including study supplement, saliva sampling kit for end-of-study, biohazard and sampling bag, and, if a remote visit was anticipated at week 16, capillary blood collection kits (TASSO OnDemand SST device). Any patient with a positive NADAL COVID-19 IgM/IgG Rapid Test serology test were excluded prior to randomisation.

The *remote* randomization visit, conducted by video-conference, was similar to the *in-person* randomization visit with the following additions: (a) viewing of the capillary blood collection video and instructional pamphlet; (b) remote capillary sampling under guidance using the TASSO-SST device (TASSO Inc., Seattle, USA); (c) viewing of the saliva collection video and instructional pamphlet (OG-600 Oragene DNA Collection Kit, DNA Genotek Inc., Ottawa, Canada); (d) remote DNA salivary sampling under guidance; (e) preparation of biological samples for shipment with phase change and insulated envelopes under guidance and (f) organising collection of biological specimens by approved courier service to respective laboratories. Note that a Nadal serology test was not conducted remotely.

Follow-up

Participants received *weekly electronic (SMS or email) reminders* to take their weekly Study Supplement (10,000 IU vitamin D or an identical placebo) and to start completing an *online daily*

Diary if they tested positive to SARS-CoV2 or they developed symptoms suggestive of COVID-19 infection.

Every two weeks, participants received a link to complete a *brief online questionnaire* asking to report: their adherence to weekly Study Supplement intake; health status including recent COVID-19 related exposure, symptoms, or testing; adverse health events or new co-morbidities; change in concomitant medications or supplement intake; work status (active duty, quarantined, holiday, sick) and work setting (ED, ICU, etc.); as well as expected/recent COVID-19 vaccination (date and vaccine name) if any; the latter question served to enable timely shipment of materials for additional sampling prior to vaccination, as COVID-19 vaccination was permitted during the study. In participants who planned to get vaccinated during the study, three additional blood, and one additional saliva, samplings, either *on-site* or *remotely*, were planned including: saliva (for COVID-19 qPCR analysis) and blood (for SARS-CoV-2 anti-S IgG serology) sampled prior to vaccination, a blood sample (for SARS-CoV-2 anti-S and anti-N IgG serology) collected just prior to the second vaccine dose, and a blood sample (for SARS-CoV-2 anti-S and anti-N IgG serology) collected one month of after second vaccine dose and endpoint. Regardless of their vaccination status, participants were asked to continue taking the weekly Study Supplement and complete the bi-monthly questionnaire until the end of the study. If questionnaires were not completed within 2 days of the target date, the research coordinator reached out the participant to complete the information.

End-of-Study visit

An end-of-study visit was conducted either in-person (60 minutes) or remotely (90 minutes), depending on the availability and preference of participants and likelihood of a current COVID-

19 infection. The *in-person end-of-study visit* entailed the collection of a (i) venous blood sample for serum 25(OH)D and SARS-CoV-2 anti-S IgG serological results and in vaccinated participants a SARS-CoV-2 anti-N IgG serology, (ii) capillary blood sample to perform the NADAL® COVID-19 IgG/IgM Rapid Test, (iii) a saliva sample for SARS-CoV-2 qPCR analysis as well as guessing of allocation and return of the study supplement bottle to assess adherence and any unused material.

The *remote end-of-study visit* conducted via videoconference entailed the same procedures as the in-person end-of-study visit with one exception: the self-collection of a capillary (instead of venous) blood using TASSO-SST devices (for the serum 25(OH)D and SARS-CoV-2 anti-S with/without anti-N serology). Individuals were guided into self-performing the pinprick capillary method to perform the NADAL® COVID-19 IgG/IgM Rapid Test and return of biological samples and materials by pre-paid approved courier.

Covariates

Several covariates that may act as confounders or interaction variables in the magnitude of effect associated with the intervention were documented, namely: baseline serum 25OHD level; smoking; concomitant supplements or drug(s) that alter calcium or vitamin D absorption or metabolism such as diuretics and anti-epileptics (reported at baseline and every 2 weeks); skin color (documented at baseline); obesity (documented by height & weight [BMI] at baseline); other comorbidities (i.e., diabetes, hypertension, etc.) that may affect the severity of COVID-19 infection and receipt of a COVID-19 vaccine (documented by verbal report bi-monthly). All

external (governmental and institutional) databases were to be obtained 3 months before, and up to 16 months following, randomization (as well as 12 months after then study endpoint).

During an event

During COVID-19 related symptoms or documented SARS-CoV infection, participants were instructed to complete a daily symptom diary from date of onset of symptoms or positive test, until two days with no symptoms or 14 days if asymptomatic,

Risk management

Clinical and biochemical adverse health events (AHEs) were monitored throughout the study and reported for all patients at the end of the study. No specific laboratory safety monitoring was planned given the established safety of the loading and weekly doses.[28] Adverse Health Events (AHE) were recorded via electronic questionnaires throughout the study. Participant who reported symptoms suggestive of vitamin D intoxication had a venous blood sampling (total and ionised calcium, phosphorus, alkaline phosphatase, albumin, and creatinine). Any abnormal laboratory values was interpreted as 'clinically significant' or 'not clinically significant' by the Site endocrinologist blinded to study allocation. Further investigation or action for individual participants (including interruption, cessation, or unblinding of the study drug via pharmacy or by analysis of serum 25OHD) was be determined by the Site endocrinologist, if indicated to ensure participant safety. The AHE's occurrence was reviewed periodically by the Data and Safety Monitoring Board (DSMB). Code breaking was allowed only if deemed essential for participant management. If relevant, summary reports aggregating (or not if requested) both groups were to be provided to the DSMB.

Data management and monitoring

The principle investigator (FMD) and statistical group (SG, RP) oversaw randomization, data management, progress monitoring, and all analyses, including those for Data Monitoring Safety Board (DMSB). The DSMB membership included: Lehana Thabane, biostatistican (Chair), Gary Kobinger, infectious disease specialist, Kevin Thorpe, biostatistician, and Edgar Delvin, biochemist & expert in Vitamin D. DACIMA was used for online data entry and management.

A combination of *remote* monitoring activities and *in-person* routine monitoring visits were conducted by an independent Study Monitor with the first randomised participants at each site and on a bi-monthly basis, to ensure that each Site adhered to the study protocol, Good Clinical Practice guidelines and data collection completeness.

Sample size calculation

Given uncertainties in infection progression, a Bayesian adaptive design was used where the posterior probability of effectiveness, i.e., P(OR<1|data) was the basis of inference and decision making.[29] Assuming an expected OR of 0.80 and 1:1 treatment allocation, a total net sample size of 2100 was required to identify a 20% reduction in the risk of COVID-19 in the vitamin D vs. control group, with 80% power with the design described above. Considering a drop-out rate of 15%, 2414 participants were targeted. An interim analysis was planned when 75% of participants reached week 12, at which time the following assessments were made: the *progression over time*

in the incidence of infection (slope of the curve of infection) was updated and if the probability of effectiveness exceeded 0.95 [p(OR<1)>0.95], the trial should be terminated for efficacy at the interim point (12 weeks); otherwise, the study would continue to 16 weeks. Simulation results showed that, with the net sample size of 2100 (assuming an expected OR of 0.80 and 1:1 treatment allocation), there was about a 55% chance that the trial would be terminated for efficacy at the interim analysis. The overall infection rate was monitored on a monthly basis: note that the study could have been extended to 24 weeks based on the progress of the infection rate, if required.

Statistical analysis

Primary outcome

An intention-to-treat (ITT) analysis was to be carried out with all randomized participants. For the primary outcome, the posterior distribution of the odds ratio of COVID-19 infection (OR) was the basis of inference in interim and final analyses. The posterior distribution of the OR was to be estimated by drawing samples from the posterior risks under each arm, which could be obtained analytically in a Beta-binomial model. Flat prior distributions were assumed for the risks (Beta(1,1)). Posterior 95% credible intervals were to be reported as interval estimates for the OR. Crude analyses as well as analyses adjusted for important covariates (i.e., potential confounders, effect modification, and baseline group imbalances) where to be conducted. Subgroup analyses would be conducted on baseline 25OHD, age, sex, BMI, occupational risk, and COVID-19 vaccination. A stratified analysis on geographical infection rate would be explored; sensitivity analysis censoring to date of COVID-19 vaccination, would be conducted if applicable.

Secondary outcomes

Distribution of disease severity defined as a 5-level ordinal outcome would be examined with a Bayesian analysis using a proportional odds (PO) model; the posterior probability of OR would be obtained by Markov chain Monte Carlo sampling implemented in Stan.[29] Duration of symptoms, duration of workday absences and of unemployment would be examined by a zero-inflated Poisson distribution.

Ethics and dissemination

This study has been reviewed and approved by the research ethics board (REB) of the CHU Sainte-Justine, serving as the local REB of all participating institutions. A non-objection letter (NOL) from Health Canada has been obtained to use high-dose Vitamin D loading dose as well as Tasso OnDemand device for home blood sampling and the NADAL COVID-19 IgM/IgG Rapid serology test. Written informed consent for study participation, for biobanking specimens for ancillary studies, and for subsequent publication of results was obtained from all participants, with the knowledge that participation is voluntary and can be withdrawn at any time with no effect on their current/future medical care. As part of the informed consent, enrolees had the option to participate in the HostSeq COVID-19 Canadian biobank conducted under the supervision of CGen, a national Canadian platform for sequencing and genome analysis **(Supplementary file 2)**. In Canada, health care is provided to those who suffer harm from trial participation.

All protocol amendments were submitted to Health Canada, investigators and REB; if these changes implied a revision of consent forms, ongoing trial participants were informed of new modifications to provide informed consent. All information obtained during the study were and will be kept confidential as per the law. Data was collected directly by electronic data capture on Dacima Clinical Suite (DACIMA Software Inc., Montreal, Canada). Data safety and

confidentiality was upheld at all data collection stages by assigning a unique subject ID to each participant, with data and samples kept under lock and key, electronic password protection and access restricted to study personnel. Samples collected during the study were labelled with the unique research code, prior to transfer and storage at the CHUSJ biobank, with access restricted to authorised personnel.

This trial uses pragmatic patient (irrespective of baseline 25OHD level) and intervention to maximise subsequent implementation into practice. If effective in reducing infection and morbidity, this approach would be readily implementable and could markedly influence practice during the COVID-19 pandemic. No participant identifiers will be used in the dissemination of this research. Health care professionals serving as partners informed the study design and pre-test all questionnaires and will contribute to a disseminating plan. Results will be disseminated to the medical community and public health departments via national/international conferences and publications in peer-reviewed journals as well as to the public and study participants via the Direction Collaboration-Partenariat Patient of the University of Montreal and the Canadian Respiratory Research Network (CRRN) patient platform who would contribute to a disseminating plan to reach as many individuals as possible.

Trial Status

The study was conducted as per version 1.8 (January 18, 2021). The recruitment started on February 9, 2021. Upon the DSMB recommendation, recruitment was stopped prematurely on March 18, 2021 after 34 participants enrolled due to the inability to target sample size of 2415 participants. The DSMB advised that the continuation of the trial, as originally designed, would not be able to answer the research question and recommended that recruitment be stopped for futility. Recruitment

difficulties were attributed in part to the high use of vitamin D and high concurrent vaccination rate among our target population, healthcare workers, the first targeted to be vaccinated from January 2021 onwards. Based on the recommendations of the study's endocrinologist, a premature cessation of follow-up after a minimum of 4 weeks from randomization to monitor the safety of intervention in all participants. The timeframe was deemed sufficient to ensure participant safety while learning for the study, that is, transforming the PROTECT study into a pilot study to document the impact of the Study intervention on the rise in Vitamin D serum level, participants' adherence the Study intervention and procedures in the context of a hybrid study, etc. The last end-of-visit was conducted on May 4 202.

Potential redirection of the study were discussed. The first option was to change the main outcome for an immunogenicity study in the general adult population. However, after strong consideration of the amont of changes to be made to the protocol and related documents (standards of procedures, case report forms, participant' instructions and notification, etc.), the expected delay in obtaining approval by all regulatory and ethical authorities, the impossible logistic of recruiting participants after the same duration of exposure to the Study intervention prior to their vaccination, combined with the government of Quebec announcement that all willing adults would be vaccinated by June 24, 2021, the PI judged that it would unfeasible to perform a scientific solid and feasible trial on immunogenicity if one could not control the timing of immunization, combined with the expected very short recruitment timeframe.

A second option that received very strong consideration was to replicate the PROTECT trial in children aged 9 years and over. Again, after considering changes to be made to the protocol and

related documents, the expected delay for obtaining approval by all regulatory and ethical authorities including school boards, combined with the Pfizer-BioNtech announcement that their vaccine was not only 100% successful for preventing COVID-19 infection in adolescents aged 12 to 15 years but that they forecast vaccinating teenagers in time for September 2021 school entry, the PI judge it was unrealistic to aim for the large recruitment target within such a short timeframe.

The protocol was submitted after the last patient end-of-study visit, due to the incredible amount of work done to conducted to set-up and initiate this large hybrid trial, including two pilot studies testing two experimental devices to enable partially or totally remote participation, in the context of the pandemic which imposed large protocol and space restrictions for recruiting on-site potentially COVID-19 infected health care workers, several protocol amendments to facilitate and adjust the trial in the context of emerging science and anticipated vaccination campaign and their impact of all electronic documents, manual of procedures, and regulatory approvals, coupled with the premature end-of-follow-up in enrolled participants. The publication of this protocol is meant to share our experience, enables protocol uptake in the context of another epidemic/pandemic, and serves as reference for the publication of pilot studies and lessons learned from this experience.

Authors' contribution

FMD designed the study protocol, secured funding, and oversaw the overall conduct of the project. CLT contributed to the protocol and amendments, directed the study implementation at the CHUM, and coordinated the prescription of study drug, pharmacy dispensation, as well as salivary sample reception and interpretation. SG conceived the statistical approach and sample size calculation and along with RWP, oversaw randomization and statistical analysis. CL, JHW and CQ contributed to the study design and amendments. BH wrote the first manuscript draft. LGSM oversaw the safety assessment. DC is responsible for the work absenteeism analysis. All co-authors approved the manuscript. Authorship eligibility on resulting manuscripts will follow standard guidelines.[30]

Competing interests

The authors have no competing interests.

Funding

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Data access.

The datasets used and analyzed during the current study will be made available by the corresponding author on reasonable request.

Provenance and peer review

This study was not commissioned. It was peer-reviewed for funding and ethical approval.

Acknowledgements

We acknowledge the precious collaboration of Danny Germain from Quebec Riva Laboratories who agree to provide free of charge Study Preparations (vitamin D and matching placebo), available in bottles of 60 tablets, allowing for study prolongation. We sincerely thank Benoit Hebert of Teracero Pharma Inc, for providing free-of-charge the NADAL COVID-19 IgM/IgG Rapid serology test kits. We are indebted to Martin Sauvageau for implementing and coordinating the RT-qPRC analysis of saliva samples at the Montreal Clinical Research Institute, Christian Renaud for coordinating the COVID-19 serology analysis, and Claude Bourassa for coordinating all other blood analyses at the Sainte-Justine University Health Centre. We acknowledge the precious collaboration of Raymond Loyer from EFS Solution Santé who adapted their appointment software for our needs as well as John Padoba, Rabie Razgallah, and Mustapha Gharb who programmed and revised the eCRF to our needs. We sincerely thank Anna Smyrnova for coordinating the development of the eCRF and data management. We are indebted to Catherine Lamontague from Orokom Communication Marketing who developed the communication strategy and tools and oversaw the publicity campaign with Marie-Line Bénard-Cyr of the CHUSJ who also developed the PROTECT website and Laureanne Marceau of the CHUM. We sincerely thank the members of the Data Monitoring Safety Board namely Lehana Thabane (Chair), Gary Kobinger, Kevin Thorpe and Edgar Delvin.

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Randomized Controlled Trial

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Figure Legend

Figure 1- Study outline

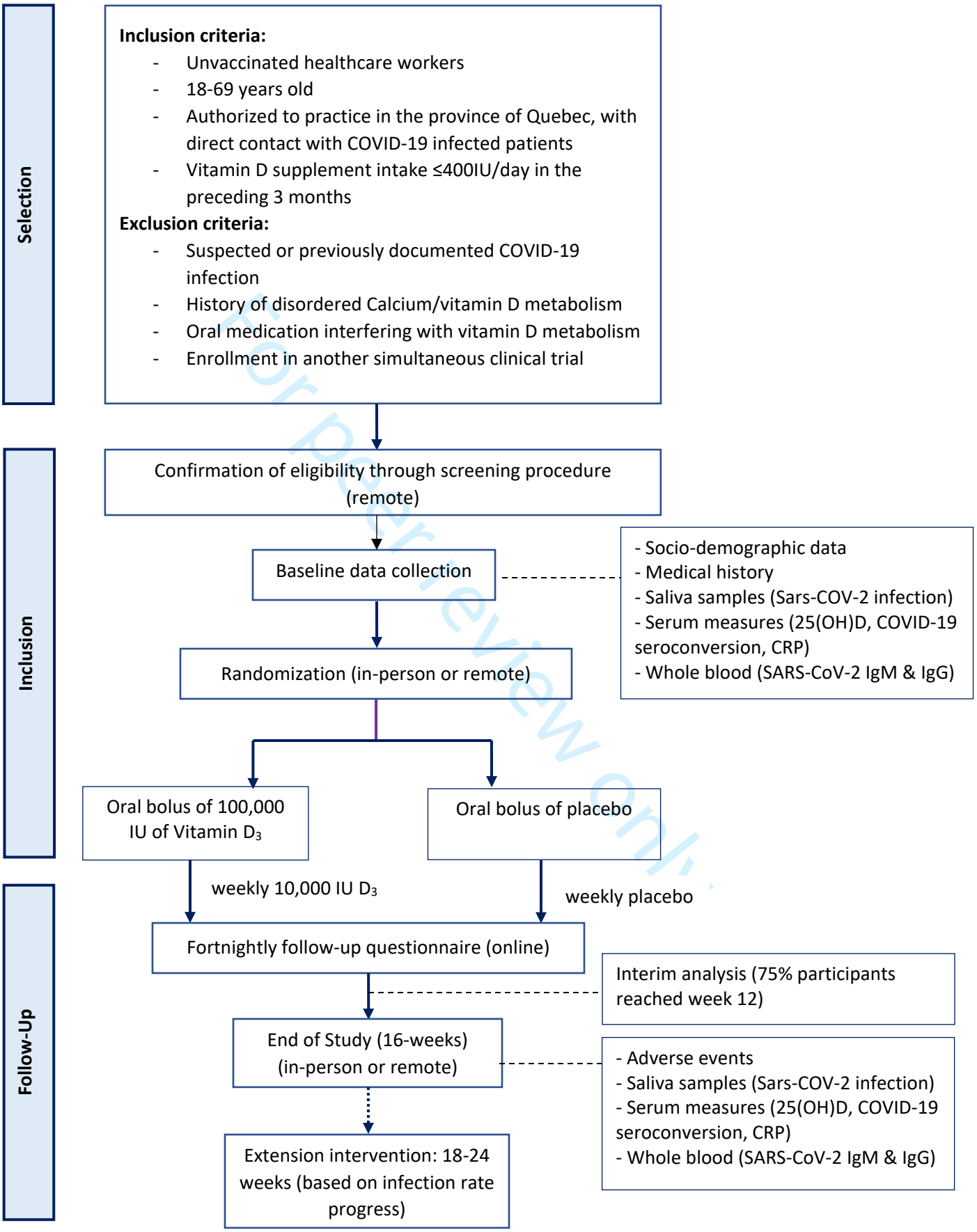

Figure 1

SPIRIT 2013 Checklist: Recommended items to address in a clinical trial protocol and related documents*

Section/item	ItemNo	Description	Addressed on page number
Administrative information			
Title	1	Descriptive title identifying the study design, population, interventions, and, if applicable, trial acronym	1
Trial registration	2a	Trial identifier and registry name. If not yet registered, name of intended registry	3
	2b	All items from the World Health Organization Trial Registration Data Set	**N/A**
Protocol version	3	Date and version identifier	8 (line 138)
Funding	4	Sources and types of financial, material, and other support	1, 25, supplemental funding file
Roles and responsibilities	5a	Names, affiliations, and roles of protocol contributors	1, 26-27
	5b	Name and contact information for the trial sponsor	1, 26
	5c	Role of study sponsor and funders, if any, in study design; collection, management, analysis, and interpretation of data; writing of the report; and the decision to submit the report for publication, including whether they will have ultimate authority over any of these activities	26

	5d	Composition, roles, and responsibilities of the coordinating centre, steering committee, endpoint adjudication committee, data management team, and other individuals or groups overseeing the trial, if applicable (see Item 21a for data monitoring committee)	19, 20, 26,27
Introduction			
Background and rationale	6a	Description of research question and justification for undertaking the trial, including summary of relevant studies (published and unpublished) examining benefits and harms for each intervention	6, 7
	6b	Explanation for choice of comparators	6, 7
Objectives	7	Specific objectives or hypotheses	8, 9
Trial design	8	Description of trial design including type of trial (e.g., parallel group, crossover, factorial, single group), allocation ratio, and framework (e.g., superiority, equivalence, noninferiority, exploratory)	9, 10
Methods: Participants, interventions, and outcomes			
Study setting	9	Description of study settings (e.g., community clinic, academic hospital) and list of countries where data will be collected. Reference to where list of study sites can be obtained	13

Eligibility criteria	10	Inclusion and exclusion criteria for participants. If applicable, eligibility criteria for study centres and individuals who will perform the interventions (eg, surgeons, psychotherapists)	9, 10
Interventions	11a	Interventions for each group with sufficient detail to allow replication, including how and when they will be administered	10, 11
	11b	Criteria for discontinuing or modifying allocated interventions for a given trial participant (eg, drug dose change in response to harms, participant request, or improving/worsening disease)	10
	11c	Strategies to improve adherence to intervention protocols, and any procedures for monitoring adherence (eg, drug tablet return, laboratory tests)	17, 18
	11d	Relevant concomitant care and interventions that are permitted or prohibited during the trial	10
Outcomes	12	Primary, secondary, and other outcomes, including the specific measurement variable (eg, systolic blood pressure), analysis metric (eg, change from baseline, final value, time to event), method of aggregation (eg, median, proportion), and time point for each outcome. Explanation of the clinical relevance of chosen efficacy and harm outcomes is strongly recommended	11, 12

Participant timeline	13	Time schedule of enrolment, interventions (including any run-ins and washouts), assessments, and visits for participants. A schematic diagram is highly recommended (see Figure)	15, 16, **17, 18, Figure** 1
Sample size	14	Estimated number of participants needed to achieve study objectives and how it was determined, including clinical and statistical assumptions supporting any sample size calculations	20-21
Recruitment	15	Strategies for achieving adequate participant enrolment to reach target sample size	12,13
Methods: Assignment of interventions (for controlled trials)			
Allocation:			
Sequence generation	16a	Method of generating the allocation sequence (eg, computer-generated random numbers), and list of any factors for stratification. To reduce predictability of a random sequence, details of any planned restriction (eg, blocking) should be provided in a separate document that is unavailable to those who enrol participants or assign interventions	11
Allocation concealment mechanism	16b	Mechanism of implementing the allocation sequence (eg, central telephone; sequentially numbered, opaque, sealed envelopes), describing any steps to conceal the sequence until interventions are assigned	11

Implementation	16c	Who will generate the allocation sequence, who will enrol participants, and who will assign participants to interventions	11
Blinding (masking)	17a	Who will be blinded after assignment to interventions (eg, trial participants, care providers, outcome assessors, data analysts), and how	5, 19
	17b	If blinded, circumstances under which unblinding is permissible, and procedure for revealing a participant's allocated intervention during the trial	19
Methods: Data collection, management, and analysis			
Data collection methods	18a	Plans for assessment and collection of outcome, baseline, and other trial data, including any related processes to promote data quality (eg, duplicate measurements, training of assessors) and a description of study instruments (eg, questionnaires, laboratory tests) along with their reliability and validity, if known. Reference to where data collection forms can be found, if not in the protocol	12 to 19
	18b	Plans to promote participant retention and complete follow-up, including list of any outcome data to be collected for participants who discontinue or deviate from intervention protocols	12 to 19

Data management	19	Plans for data entry, coding, security, and storage, including any related processes to promote data quality (eg, double data entry; range checks for data values). Reference to where details of data management procedures can be found, if not in the protocol	20
Statistical methods	20a	Statistical methods for analysing primary and secondary outcomes. Reference to where other details of the statistical analysis plan can be found, if not in the protocol	21, 22
	20b	Methods for any additional analyses (eg, subgroup and adjusted analyses)	22
	20c	Definition of analysis population relating to protocol non-adherence (eg, as randomised analysis), and any statistical methods to handle missing data (eg, multiple imputation)	21-22
Methods: Monitoring			
Data monitoring	21a	Composition of data monitoring committee (DMC); summary of its role and reporting structure; statement of whether it is independent from the sponsor and competing interests; and reference to where further details about its charter can be found, if not in the protocol. Alternatively, an explanation of why a DMC is not needed	20,

	21b	Description of any interim analyses and stopping guidelines, including who will have access to these interim results and make the final decision to terminate the trial	20
Harms	22	Plans for collecting, assessing, reporting, and managing solicited and spontaneously reported adverse events and other unintended effects of trial interventions or trial conduct	17, 19, 20, 23
Auditing	23	Frequency and procedures for auditing trial conduct, if any, and whether the process will be independent from investigators and the sponsor	20
Ethics and dissemination			
Research ethics approval	24	Plans for seeking research ethics committee/institutional review board (REC/IRB) approval	25
Protocol amendments	25	Plans for communicating important protocol modifications (eg, changes to eligibility criteria, outcomes, analyses) to relevant parties (eg, investigators, REC/IRBs, trial participants, trial registries, journals, regulators)	8
Consent or assent	26a	Who will obtain informed consent or assent from potential trial participants or authorised surrogates, and how (see Item 32)	13
	26b	Additional consent provisions for collection and use of participant data and biological specimens in ancillary studies, if applicable	N/A

A	27	How personal information about potential and enrolled participants will be collected, shared, and maintained in order to protect confidentiality before, during, and after the trial	14-15, 25
Declaration of interests	28	Financial and other competing interests for principal investigators for the overall trial and each study site	27
Access to data	29	Statement of who will have access to the final trial dataset, and disclosure of contractual agreements that limit such access for investigators	27
Ancillary and post-trial care	30	Provisions, if any, for ancillary and post-trial care, and for compensation to those who suffer harm from trial participation	25
Dissemination policy	31a	Plans for investigators and sponsor to communicate trial results to participants, healthcare professionals, the public, and other relevant groups (eg, via publication, reporting in results databases, or other data sharing arrangements), including any publication restrictions	23
	31b	Authorship eligibility guidelines and any intended use of professional writers	25
	31c	Plans, if any, for granting public access to the full protocol, participant-level dataset, and statistical code	27

Appendices

Informed consent materials	32	Model consent form and other related documentation given to participants and authorised surrogates	Supplementary file 2
Biological specimens	33	Plans for collection, laboratory evaluation, and storage of biological specimens for genetic or molecular analysis in the current trial and for future use in ancillary studies, if applicable	Supplementary file 2

*It is strongly recommended that this checklist be read in conjunction with the SPIRIT 2013 Explanation & Elaboration for important clarification on the items. Amendments to the protocol should be tracked and dated. The SPIRIT checklist is copyrighted by the SPIRIT Group under the Creative Commons "Attribution-NonCommercial-NoDerivs 3.0 Unported" license.

INFORMATION AND CONSENT FORM

Project Title: PRevention of COVID-19 with Oral Vitamin D supplemental Therapy in Essential healthCare Teams (PROTECT)

Protocol Number: PROTECT 2020

- **Principal investigators at CHUSJ:** Dr. Francine M. Ducharme, MD, FRCPC, Paediatrician, Centre hospitalier universitaire Sainte-Justine (CHUSJ)
- **Principal investigator at the CHUM:** Dr. Cecile Tremblay, MD, FRCPC, Microbiologist/Infectiologist, Centre hospitalier universitaire de Montréal (CHUM)

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- Shirin Golchi, biostatistician, McGill university
- Cristina Longo, epidemiologist, University of Montreal
- Robert Platt, biostatistician, McGill University
- Caroline Quach, MD, Paediatrician microbiologist, CHUSJ
- Christian Renaud, pediatric microbiologist, CHUSJ
- John White, biochemist, McGill University

Co-investigators at the CHUM:

- Dr. Louis-Georges Sainte-Marie, MD, endocrinologist, CHUM
- Dr. Emil Toma, MD, Dsc, FRCPC,CHUM

Industrial Collaborators: Laboratoires Riva, Blainville, Quebec

Funding source: Canadian Institutes of Health Research (CIHR), in the context of the COVID-19 Rapid Research Funding Opportunity

Multicenter identifier: MP-21-2021-3044
CHUM project number : 20.319

WHY ARE YOU BEING INVITED TO TAKE PART IN THIS STUDY?

Today, we are inviting you to participate in this research study because you are a healthcare worker who is working in a high COVID-19 incidence area and in a setting with a high risk of contact with COVID-19 infected cases. Please read this information to help you decide if you want to participate in this research project. It is important that you understand this information. We encourage you to ask questions. Please take all the time you need to make your decision. You may also want to discuss this study with your family doctor, a family member or a close friend.

WHY IS THIS STUDY BEING DONE?

During the current COVID-19 pandemic, many healthcare workers are working in an environment which increases their probability of contracting this viral infection. Healthcare workers are more frequently infected than the rest of the population. Infected healthcare workers can infect their family, their patients, and their contacts. In addition to being withdrawn from work, they could have transmitted the disease to other colleagues, which further impedes our ability to deliver care to the population.

Vitamin D supplementation can decrease the risk of having the common cold, but it is not known if it could have an effect on the COVID-19 infection. Vitamin D is produced in our bodies from exposure to the sun and can be obtained from supplements and certain foods. However, many Canadians do not have an adequate intake of vitamin D throughout the year.

However, studies testing supplementation with other seemingly harmless vitamins, such as beta carotene and vitamin E, have shown unexpected important adverse reactions. Therefore, it is necessary to properly assess the benefits and the potential unexpected adverse reactions in the context of a clinical study.

This study will investigate whether a high-dose vitamin D supplementation could reduce the risk and severity of COVID-19 infection and work absence in healthcare workers.

HOW MANY PEOPLE WILL TAKE PART IN THIS STUDY?

We will be recruiting 2414 graduate healthcare workers, men and women, aged 18 to 69 years old, actively working and scheduled to continue working in a setting at high-risk of contact with people infected with COVID-19 and are working in high COVID incidence areas.

WHAT DOES THE STUDY INVOLVE?

If you agree to participate in this study, you will be assigned by chance to one of two groups. One group will receive one dose of 100,000 IU of vitamin D by mouth at the first visit and then take at home 1 pill of 10,000 IU of vitamin D once a week. The other group will receive a placebo dose at the first visit and then a placebo pill once a week. The vitamin D supplement is the active substance, meaning it could have an effect in the body. A placebo is an inactive substance, meaning it has no effect in the body. A placebo is used in clinical studies, such as this one, to ensure that observed changes are due to the active treatment and not to chance. You will have an equal chance of being assigned to each

group. The placebo and the vitamin D supplement look and taste exactly the same, so no one will know which treatment you are given, including the people involved in the study. In this informed consent form, we will use "Study supplement" to refer either to the vitamin D or the placebo.

This study should last 16 weeks and involves two visits. But prior to the first visit, we will conduct a screening/enrolment visit remotely by videoconferencing (or phone). If eligible and consenting, there will be a randomisation visit and an end-of-study visit, the latter two could be conducted in-person or remotely by videoconferencing, at your preference. Because of the pandemic, we wish to reduce the need and/or length of any in-person visit by doing as much as possible remotely.

Note that the study could finish earlier or be prolonged to 24 weeks, depending on the evolution of the pandemic. We ask that you don't change your usual diet or intake of vitamin supplements (if any) during the study.

Screening/Enrolment (pre-visit: about 45-60 minutes)

- We will review the eligibility questionnaire you have completed online, complete it with additional questions, explain the study in detail, and answer your questions.
- If eligible and consenting, you will be asked to sign the consent form, complete a few short study questionnaires and provide your contact information.
- We will ask you questions about your demographics (household, ethnicity), work-related activities and personal health (weight, height, skin color, smoking, medication, vitamins, supplements, health problems).
- To enable the creation of a medical and research pharmacy records, obtain information on COVID test, and ensure optimal contact with you throughout the study, we will ask personal information namely your RAMQ number, names (yours, your parents, your spouse), any drug allergy, your employee number (or practice number for physicians), your postal and email addresses, phone numbers and that of a next of kin and your preferred means to reach you.
- We could show you videos of key procedures (e.g. home blood collection) to help you chose your preferred type of randomisation visit.
- We will schedule the randomisation visit at a mutually convenient time and place given your choice of in-person or remote visit by videoconferencing. However, in case of a suspected prior COVID-19 infection, we would prefer you do an in-person visit to perform a rapid screening test for COVID-19 antibodies.

Randomisation visit (First visit: Week 0)

During the visit, which will last approximately an hour,

- If not already done, we will ask you to sign the consent forms, complete missing study questionnaires and your contact information.
- We will take a venous blood sample of about 15 mL (3 teaspoons) to measure the level of vitamin D, look for COVID-19 antibodies and to do an optional genetic analysis to examine a possible genetic predisposition to respond to vitamin D and to severity of COVID-19 symptoms.

- We would like to obtain a small drop of blood either from the venous puncture or via a finger-prick to look for COVID-19 antibodies in your blood using NADAL® COVID-19 IgG/IgM Rapid Test: if positive, you would not be eligible for this study. This test is not yet licensed for use in Canada and its use in this study is investigational. It has been selected for use prior to enrolment because it provides antibody results in 15 minutes. The results of this investigational test will be shared with you, acknowledging the risk of false positive or negative results. They will also be subsequently compared to the approved (Liaison IgG COVID-19) serology test.
- We will show you how a video on the TASSO home blood sampling kit at home for the last visit; if interested, we will show you how to use it, identify your sample, package it, and send it back to us (see below under First Remote visit). This device is not yet licensed for use in Canada and its use in this study is investigational. However, we have successfully pre-tested it and have validated the concordance between test results obtained with the TASSO and venous sampling.
- We will show you how to take a saliva sample by spitting into a tube. You will receive a pamphlet with instructions and could watch a video. You should not brush your teeth, eat, drink, smoke or chew gum for 30 minutes before spitting a small volume of saliva (2 mL). If you prefer to do an oro-nasopharyngeal sample, you would need to insert a swab (a small tube with a cotton tip) into the back of your mouth, then in one of your nostrils gently rotating the swab for about 5 seconds. We will ask you to take the saliva (or oro-nasopharyngeal) sample under our guidance. We will then show you how to identify it with our prepared labels, record the date and time, package it, and sent it back for analysis for COVID-19, and possibly other viruses and cells.
- You will be asked to take ten (10) pills of the Study supplement at this first visit only in front of the research personnel (in person or by videoconference). You will take home the bottle of Study supplement and be asked to take one (1) pill once a week until the end of the study.
- We will send you by text message or email as per your preference, a first reminder with a link to a questionnaire to confirm that you have received it and are able to complete and submit the brief questionnaire. The same approach will be used every week.
- We will give you all the other study materials including the saliva collection tube pre-printed labels, biohazard bags, insulated envelopes or boxes for shipment, prepaid courier waybills, and if you are interested in a **Remote end-of-study visit**, the TASSO home blood collection kit. It is possible that we ask you to use the NADAL® COVID-19 IgG/IgM Rapid Test at the last visit for validation purposes.

For participants choosing to have a **Remote First Visit**, in whom there is a suspicion that you may have had a prior undiagnosed COVID-19 infection in the past, we would prefer that you come for an in-person visit. Alternatively, we may send you first a finger-prick test kit to look for COVID-19 antibodies in your blood; if so, we would ask that you use it on your fingertip in front of us by videoconference. If positive, you would not be eligible for this study. If negative, the Study supplement and required materials would then be sent to the participant's home prior to the randomisation visit. We will ask you to take in front of us by videoconference, the Study supplement, the saliva sample, as well as the blood test and 2nd optional saliva sample (for genetic analysis).

- We will show you how to take a small sample (<1 mL) of capillary blood, using a blood collection kit specifically conceived for home collection, called TASSO-SST OnDemand. This device is not yet licensed for use in Canada and its use in this study is investigational. However, we have successfully pre-tested it and have validated the concordance between test results obtained with the TASSO and venous sampling. We will ask you to watch a short video and read the brochure explaining the procedure, then ask you to use it under our guidance. Briefly, you will need to warm the skin of your upper arm by rubbing it for about 45 seconds, disinfecting it, applying the little device on your arm, pressing on a button that will puncture a very small hole in the skin, then leave the device in place for about 5 minutes while blood flows slowly in a small tube. As only a small sample of blood can be obtained, it is very likely that we ask you to repeat this with a second kit. We will show you how to remove the small tube, close it with a small cap, identify the sample with our prepared labels, record the sampling date and time, package it, and prepare it to be sent for analysis for vitamin D and COVID-19 antibodies. We will ask your feedback on this type of blood collection method.
- If you wish to participate in the optional genetic analysis, we will ask you to collect 2 mL of saliva in another small tube (as the blood sample made by TASSO is not enough for this analysis), identify and date the sample with our prepared labels, and send it to us.

Between visits

- For the following weeks 0 to 16 (or later, should the study be extended up to 24 weeks) participants will take every week one (1) Study pill.
- You will receive a message via email or text message, according to your indicated preference, to remind you
  - Once a week to take the Study supplement
  - Once every two weeks to take the Study supplement, and fill out the brief electronic questionnaire (duration of 3-5 minutes) regarding your health and work status in the previous 2 weeks.
  - At any point in time if you have symptoms, to complete the daily symptoms diary (duration of 1-2 minutes) until 48 hours after the resolution of symptoms. If you have symptoms that should prompt testing for COVID-19, as listed on the cv19quebec.ca website, we will ask you to contact your health office for this purpose.
- If there is no response from you within a few days of sending the electronic questionnaire, we will contact you; if there is still no response from you within 7 days of us sending the electronic questionnaire, we will contact your next-of-kin indicated by you.

If you are infected during the study

If we obtain a positive COVID result from one of your saliva (or oro-nasopharyngeal) samples, you will be notified by a Study team member, by the preferred communication means you have indicated (phone, text message or email). As all positive results will be reported to the public health authorities you will be contacted as soon as possible by them for an assessment and instructions.

If you receive a positive COVID result from a test done outside this study (i.e., for clinical reasons), we ask that you inform us immediately and to indicate it in the follow-up questionnaire.

At the reception of a positive COVID test result,

- We will ask you to complete the daily diary of symptoms (duration of 1-2 minutes)
  - Until 48 hours after resolution of symptoms
  - Or if you remain asymptomatic, for a minimum of 14 days;
- If symptoms reappear, we will ask you to restart documenting them in the daily diary of symptoms
  - Until 48 hours after resolution of symptoms;
- If your symptoms continue beyond 14 days, we will ask you to complete the weekly diary of symptoms (duration of 1-2 minutes), once a week, until resolution of symptoms
- We will ask you to continue taking your weekly supplement and completing the follow-up questionnaire once every two weeks.

In case of an imminent vaccination against COVID-19

- If you expect to receive a vaccine against COVID-19 in the next few weeks, we will ask you to notify us immediately or via the questionnaire every two weeks.
  - We will rapidly organise a visit in person or remotely, before the scheduled date of the vaccination, to obtain a saliva sample to test for COVID-19 infection and a blood sample either in your vein (9 mL) or with the TASSO device at home to look for COVID-19 antibodies and level of vitamin D prior to the vaccination.
  - Just before, and about 1 month after, your second vaccine dose against COVID-19, we will ask you for another blood sample either in your vein (4.5 mL) or with the TASSO device at home to look for COVID-19 antibodies.
- We will ask you to continue
  - Once a week to take the Study supplement
  - Once every two weeks to fill out the brief electronic questionnaire (duration of 3-5 minutes) regarding your health and work status in the previous 2 weeks.
  - At any point in time if you have symptoms, to complete the daily symptoms diary (duration of 1-2 minutes) until 48 hours after the resolution of symptoms. If you have symptoms that should prompt testing for COVID-19, as listed on the cv19quebec.ca website, we will ask you to contact your health office for this purpose, whether you have been vaccinated or not.

End-of-study – Week 16 (or later if the study is prolonged)

At the end of the study, you will be invited to a last in-person or remote visit. This research visit will take approximately 30 minutes and involve the following:

- You will be asked to return the Study supplement bottle containing the unused pills.

- A venous (or capillary blood if done remotely) sample and, if you have not tested positive at COVID-19 before, a saliva (or oro-nasopharyngeal) sample will be collected.
- A rapid COVID-19 antibody test (NADAL®) may also be done on a blood drop (finger-prick or from the venous puncture) for validation purposes. If done remotely, this test may be done on blood sampled by finger-prick under our guidance by videoconference.
- You will complete the last few short study questionnaires and any missing information in the previous ones, if applicable.
- If conducted remotely, the samples, Study supplement bottle and unused material should also be shipped to the Coordinating Center.

Collecting information on COVID-19 tests made for clinical reasons

The results for a COVID-19 test performed for clinical reasons outside this study will be documented by you in the follow-up questionnaire (*faster means to inform us*) as well as in your institution's (Pandemic) or provincial database of COVID-19 cases, namely Trajectoire Santé publique (TSP) including all individuals who tested positive and all healthcare workers who tested positive under the supervision of the Ministère de la santé et des services sociaux (MSSS). If you are unable to answer the follow-up questionnaires, the information documented in these databases would ensure that we have complete information on the primary outcome of the study and thus allow us to determine accurately the impact of the intervention on the risk of infection with COVID-19.

Collecting information on healthcare services

The date, diagnosis, type of professional and of health care services which you have received during medical visits and hospitalisations will be obtained from the administrative databases of the Régie de l'assurance maladie du Québec (RAMQ) and Quebec hospital discharges (MED-ECHO). This will allow us to accurately determine the impact of the intervention on the severity of COVID-19 infection and other concomitant illnesses.

Collecting information of work absence

The number of days of work absence, overall, by type (i.e., holiday, illness, etc.), and specifically due to COVID-19, including absences due to an infection acquired at work or outside of work, preventive withdrawal due to pregnancy or other health conditions, awaiting test results/investigation, or other reason for quarantine will be collected from you via the follow-up questionnaire (*faster and most detailed means*), as well as from your institution's *Direction of Health Resources* or, if you are an attending physician, from the *Direction of professional services*. If you are unable to answer the follow-up questionnaires, the information documented in these databases would ensure that we accurately ascertain the impact of the intervention on work absences.

BIOBANK

For the purposes of this study, we will keep the biological samples collected (blood, saliva and/or oro-nasopharyngeal) in a biobank as well as the clinical and administrative data collected during the course of this study in order to complete the study's objectives, and to

conduct research on vitamin D, COVID-19 and its treatments and other related diseases. We would like to quantify specific cellular receptors which allow entry of COVID-19 into cells (for example, the angiotensin converting enzyme-ACE2) and inflammatory markers (such as the C-reactive protein). The collected samples will be kept in a biobank in the Research Center of CHU Sainte-Justine under the supervision of Dr Francine M. Ducharme. The samples will be kept as long as the research team can guarantee their proper management. Confidentiality of the identity of the samples will be guaranteed by assigning them a specific code. Your sample will not be identified by your name and cannot be used to identify you directly. After 5 years, the code key will be destroyed, and the samples will become completely anonymous. Your samples could possibly be shared with other researchers in other institutions. However, the access to data will only be allowed for approved projects by an independent research ethics board.

GENETIC ANALYSIS (optional)

Each person has their own set of unique genes or "genome". Genetic research aims to determine if there are genetic predispositions which make you more susceptible to a COVID-19 infection, to respond to vitamin D, to modulate disease severity and the interaction of these factors.

If you accept to participate in the genetic analysis, these analyses will be done on a small part (4 mL) of the venous blood sample provided during the first visit. If you decide to participate remotely, we will ask you to provide a saliva sample in a small tube.

We would like to sequence your entire genome and conduct gene expression analyses. We would also like to share your genetic data as well as other collected clinical data during the PROTECT study with the Canadian database Hostseq COVID-19 for use for COVID-19 related research and other aspects of human health. This biobank will serve as a centralized resource in Canada for COVID-19 research and other health-related studies. The data in the HostSeq database are under the supervision of CGen, a national Canadian platform financed by the federal government for sequencing and genome analysis. The principal investigators of the PROTECT study as well as the administrators of the Hostseq biobank COVID-19 will share your genetic and clinical information with other Canadian and international researchers whom are approved by CGen (the sponsor). The data could also be used for commercial use. However, your data will not be shared with until after an examination by a data access committee. This committee will verify that the use of the proposed research is in line with the objectives of the database HostSeq and that the research team which requests access has already been granted the required approval in accordance in terms of research ethics requirements. Approved researchers will sign agreements. These agreements will control how the data will be used. Individual results of any research conducted using your samples or any individual incidental findings will not be shared with you, as the research conducted on your data will have no individual diagnostic or therapeutic significance to you.

WHAT ARE THE BENEFITS AND RISKS OF THIS STUDY?

Benefits:

You may not benefit directly from the study intervention if it is not efficacious or if you have been assigned in the placebo group. However, the screening may identify earlier an active or past COVID-19 infection that was not apparent. Vour participation will help advance our knowledge on vitamin D and on the prevention of COVID-19 infection in healthcare workers and other individuals at risk of infection.

Each positive COVID-19 result from the saliva (or oro-nasopharyngeal) or blood sample will be shared with you according to your preferred way of communication: telephone, text message or email. All positive saliva (or oro-nasopharyngeal) results will also be shared by the Microbiology Laboratory of CHUM with the Public health authorities and will be added into your file at the CHUM and Dossier Santé Québec. No other research result will be provided to you. Research findings resulting from your participation to this study could potentially contribute to creating commercial products from which you would not be able to claim any financial benefit.

Risks:

• Related to study medication:

The vitamin D dose used in this study has been shown to be safe in adults. This dose is approved by Health Canada for the purpose of this study only, but not for clinical use yet. It is unlikely that you will have any side effects because of the amount of vitamin D used in this study as when combining the first and weekly doses, the total remains below the maximum amount allowed.

However, we will ask you to notify us immediately if you have any of the following, as they could be signs of an acute excess intake of vitamin D: mainly, a marked increase in thirst or an increase in the volume and frequency of urination (with or without fatigue, loss of appetite, nausea or vomiting, headaches, drowsiness, cardiac arrhythmias, constipation, muscle or bone or chest pain, mouth dryness or a metallic taste).

Later signs and symptoms that may indicate a chronic excess intake of vitamin D are: a marked increase in thirst, an increase in the volume and frequency of urination including during the night, loss of appetite, weight loss, red eye or conjunctivitis, inflammation of the pancreas, light sensitivity, runny nose, itching, fever, reduced libido, kidney stones, increased concentration of some analytes in the blood (BUN, AST, ALT, cholesterol), or in urine (albumin), ectopic calcification, hypertension, cardiac arrhythmias and rarely, a psychosis.

It's possible that other currently unknown risks are associated with Vitamin D intake.

One of the reasons we collect a blood sample is to measure the concentration of vitamin D in the blood at the start and end of the study. this will allow us to see if the vitamin D blood level is linked to the number and severity of COVID-19 confirmed cases.

• Related to study procedures:

The salivary collection sample is painless. If done, an oro-nasopharyngeal swab may cause slight discomfort during collection that will subside after its removal. The side effects of having blood collected by venous puncture or TASSO can include bleeding, bruising, discomfort and pain at the sample site. It is possible that the NADAL COVID-19 IgG/IgM Test may give false positive or false negative results. In case of divergence of results, we will communicate to you the results of the approved IgG test when available.

• Related to confidentiality:

There is always a small risk that your data could one day be re-identified. The genetic information is unique to each person, just as your fingerprint. This means that theoretically, you could be identified using your genetic code; however, this is not easy to do. Considering the advances in technology, there could be new ways to link you to data that we have not foreseen today, despite the strict confidentiality measures in place. Possible re-identification or unintentional disclosure of your genetic and clinical research data could lead to a loss in confidentiality and a possible future discrimination against yourself or your biological parents. But all security measures will be put in place to protect your privacy.

WHAT ARE THE COSTS OF TAKING PART IN THIS STUDY?

The Study supplements are provided free of charge by the manufacturer, Laboratoire RIVA.

WHAT ARE THE OTHER FINANCIAL ASPECTS?

For each completed visit (0 and 16 weeks), you will receive a $25 check by mail to compensate for your time. The check may arrive at your home between 4 and 8 weeks after the visit.

HOW IS PRIVACY INSURED?

During your participation in this research study, the investigators responsible for this study as well as the members of their research team will collect, in a research file, the required personal information to answer the scientific objectives of this research project.

These information could include your demographic data (name, sex, date of birth, ethnic origin, weight and height), your past and present health status, your health-related habits, medication you take, your work absences, and the results of all tests, exams, and procedures which you will participate in. Your personal file will include your address, email, telephone numbers, RAMQ number, and employee or practice number be kept in a separate file with restricted access; this information is required to create a medical and pharmacy file at the CHUM and for communication purposes during the study.

The coded blood, saliva (and/or oro-nasopharyngeal) samples will be sent to the biobank located at the Research Center of CHU Sainte-Justine under the supervision of Dr Francine M. Ducharme. The coded results of completed analyses will be kept on a protected server with restricted access at DACIMA company during the study, and thereby transferred to a secure server with restricted access in the Research Center of CHU Sainte-Justine under the supervision of Dr Francine M. Ducharme. During the study, the personal information

used to arrange virtual and in-person study visit appointments will be kept on a protected server with restricted access at the company providing the appointment-making software. Following the conclusion of the study, this information of yours will be transferred to a secure server with restricted access in the Research Center of CHU Sainte-Justine under the supervision of Dr Francine M. Ducharme. The database of HostSeq will be kept on secure cloud servers (online) that are based in Canada and will be indefinitely kept or until they are not useful for research.

To ensure your privacy, a copy of the consent form as well as the results to the diagnostic tests required for conducting the research project, will be copied in the research and medical file of the CHUM. Therefore, each person or company which you authorize to consult your medical file, will have access to this information.

The research data will be kept for at least 25 years by the principle investigator. The data collected could be published or discussed during scientific meetings, but it would not be possible to identify you.

All collected information will remain confidential within the limits provided by law. You will only be identified by a code number. The key to the code linking your name to your research file will be kept by the investigator responsible for this research project.

To ensure your safety, a copy of the consent form as well as the results of the diagnostic tests required for research purposes will be placed in the research file and the medical file of the CHUM. Consequently, any person or company to whom you give access to your medical file will have access to this information.

Research data will be kept for at least 25 years by the investigator responsible for this research project. Research data may be published or be the subject of scientific discussion, but it will not be possible to identify you.

For the purposes of surveillance, control, safety and marketing of the Study drug, your research as well as your medical files could be consulted by a person mandated by a regulatory organization, in Canada or elsewhere, such as Health Canada, as well as sponsor representatives of the company manufacturing the vitamin D pills for this project (Laboratoire RIVA), the institution or research ethics committee. These people and organizations adhere to a strict confidentiality agreement.

You have the right to consult your research file to verify the collected data and to correct them, if needed. Moreover, access to certain information before the end of the study could mean your removal from this study in order to maintain the study's integrity.

IS YOUR PARTICIPATION VOLUNTARY?

Yes. Taking part in this study is voluntary. You may choose not to be in this study. You can decide to stop being in the study at any time, without needing to provide any reason, but simply informing the research team.

Your decision to refuse participation or to stop participating in the study at a later time, will have no effect on the quality of care or services to which you are entitled or on your relationship with the people that provide them.

The principal investigators of this study, the research ethics board, the funding agency or the sponsor could decide to end your participation in the study without your consent. This could happen if there are new information or findings that indicate your participation is no longer in the best of your interests, or if you have not been following the study instructions as explained, or if there are other administrative-related reasons to stop the project.

If you stop participating in the study or if you have been removed from it, the collected information and material already received will be kept (as well as the data pertaining to healthcare services and work absences will continue to be collected) and analysed to ensure the validity of this project, unless you specifically ask for them to be destroyed. If this is the case, these data and/or material will be removed from the biobank provided that the code key (linking between nominal data and the study code) is still available, that is, up to 5 years after the end of the study.

If you decide to drop out of the HostSeq database, your data will no longer be shared, and no new data will be collected. The data already in the HostSeq database will be destroyed once informed about this decision. However, it could be impossible to remove the results once they have been compiled with the results of other participants or if they have been published. Moreover, if certain data have been shared with other researchers, it could be possible not to be able to remove this part of the data. In such a case of unsuccessful withdrawal from the study, your identity will always be protected.

All new information acquired during the course of the study which could have an impact on your decision to continue participation will be shared with you rapidly, which is the reason why we would like to keep your personal information and have your approval to communicate with you after the end of the study (optional).

WHOM DO I CALL IF I HAVE QUESTIONS OR PROBLEMS?

If you have any questions about the research project or if you have any problems that you believe are related to your participation in the project, you can call the researchers responsible for the project:

- Dr. Francine M. Ducharme at 514 345 4931, extension 4398
- Dr. Cecile Tremblay at 514 890-8000, extension 14645

If you would like information about your rights related to your participation in the research, you may contact the Ombudsman - complaints and quality services of the CHU Sainte-Justine at 514 345-4749, of the CHUM at 514 890-8484 or your CISSS/CIUSSS:

- CIUSSS de l'Est-de-l'Île-de-Montréal : 514 252-3510
- CIUSSS de l'Ouest-de-l'Île-de-Montréal : 514-989-1885, extension 1010
- CIUSSS du Centre-Sud-de-l'Île-de-Montréal : 514 593-3600
- CISSS de la Montérégie-Est : 450-468-8447
- CISSS de la Montérégie-Centre : 450-466-5434

RESEARCH ETHICS COMMITTEE

The Research Ethics Board of CHU Sainte-Justine has approved this study and will continue to monitor it for all participating institutions of the Quebec Health and Social Services network.

LIABILITY

This research is not funded by a private industry. In case of side effects resulting from the study medication or from procedures required for this research project, you will receive all necessary medical care covered by the Quebec's provincial health insurance plan (RAMQ) or by your private drug insurance plan. You will be responsible for paying the portion of any costs not covered.

CONSENT FORM

Research project title: PRevention of COVID-19 with Oral Vitamin D supplemental Therapy in Essential healthCare Teams (PROTECT)

The nature and procedures of this research project were explained to me. I have read the information and consent forms and I kept a copy, or a copy has been provided to me. I was able to ask my questions and they were answered to my satisfaction. After consideration, I agree to participate in this research project.

I authorize the research team to consult the collected data about me in the COVID infection database (Pandemic) of my institution and/or the provincial TSP database, the medical and hospitalisation services database (RAMQ and MED-ECHO), and the workplace absenteeism database (Human Resources Directorate or Professional Services Directorate) to obtain information that is pertinent to this project.

By agreeing to participate in this study, you are not waiving any of my rights under the law. You are not releasing the investigators from their legal and professional liability.

Name of participant (Print)	Signature	Date

1. I consent to the analysis of gene expression and the sequencing of the whole genome of my coded biological material (blood, saliva, and/or oro-nasopharyngeal). The whole genome sequence could be hosted in the Canadian HostSeq COVID-19 biobank and linked to a database containing the viral genome. This would serve to explore any genetic predisposition to COVID-19, the severity of the disease and response to vaccine.

❒ Yes _____ (Initials) ❒ No_____ (Initials)

2. I consent to prolonging the access to my coded data on healthcare use, COVID-19 infections and work absenteeism for 12 months following the study end date, to explore the long-term impact of COVID-19 infection and vaccination.

❒ Yes _____ (Initials) ❒ No_____ (Initials)

3. I consent to being contacted to update my personal information, obtain additional information about my health or to be invited to participate in new research.

❒ Yes _____ (Initials) ❒ No_____ (Initials)

4. In case I receive a vaccine against COVID-19 during the study, I agree to do the blood samples before the first and second vaccine dose as well as 1 month after the 2nd vaccine dose, even if these samples were to be done after the end-of-study's visit planned at week 16 (or 24).

❒ Yes _____ (Initials) ❒ No_____ (Initials)

Participant's signature: _____

I have explained the research study and the terms of this information and consent form to the research participant, and I answered all his/her questions. I explained that participation in a research project is free and voluntary and could be stopped at any time they choose.

Name of person obtaining consent (Print)	Signature	Date

(FOR THE CHUM PARTICIPANTS ONLY)

COMMITMENT OF THE PRINCIPAL INVESTIGATOR AT THE CHUM

I certify that this information and consent form was explained to the research participant, and that the questions the participant had were answered.

I undertake, together with the research team, to respect what was agreed upon in the information and consent form, and to give a signed and dated copy of this form to the research participant.┼

Name (Print)	Signature of the principal investigator at the CHUM	Date

BMJ Open

Prevention of COVID-19 with oral vitamin D supplemental therapy in essential healthcare teams (PROTECT trial): protocol for a multicentre, randomized, placebo-controlled, triple-blind trial

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Keywords:	COVID-19, PREVENTIVE MEDICINE, Clinical trials < THERAPEUTICS

Prevention of COVID-19 with oral vitamin D supplemental therapy in essential healthcare teams (PROTECT trial): protocol for a multicentre, randomized, placebo-controlled, triple-blind trial

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ABSTRACT

Introduction: In the COVID-19 pandemic, health care workers (HCWs) have been at high-risk of infection due to their exposure with COVID infections. HCWs are the backbone of our health care response to this pandemic, and every health care worker withdrawn or lost due to infection has an exponential impact on our capacity to deliver care. Primary prevention is a key approach to reduce infection. Vitamin D insufficiency is highly prevalent in Canadians and worldwide. Vitamin D supplementation has been shown to significantly decrease the risk of respiratory infections. Whether this risk reduction would apply to the COVID-19 infection remains to be determined. This study aims to determine the impact of high-dose vitamin D supplementation on incidence of laboratory-confirmed COVID19 infection rate and severity in HCWs working in high COVID incidence areas.

Methods and analysis: This is a triple-blind, placebo-controlled, parallel-group multicentre trial of vitamin D supplementation in HCWs at high-risk of infection. Participants were randomly allocated in a 1:1 ratio in variable block size to: Intervention—1 oral loading dose of 100,000 IU vitamin D3 + 10000 IU weekly vitamin D3 or control—identical placebo loading dose + weekly placebo. The primary outcome is incidence of laboratory-confirmed COVID-19 infection, documented by RT-qPCR on salivary (or nasopharyngeal) specimens obtained for screening or diagnostic purposes, as well as self-obtained salivary specimens obtained at endpoint and COVID-19 seroconversion at endpoints. Secondary outcomes include disease severity; duration of COVID-19 related symptoms; COVID-19 seroconversion documented at endpoint; duration of work absenteeism; duration of unemployment support; and adverse health events.

Ethics and dissemination: This study was approved by the Research Ethics Board of the CHU Sainte-Justine and participating institutions. If proven effective in reducing the risk and morbidity

of COVID-19 infection, vitamin D supplementation could offer the cheapest, most easily implementable primary prevention strategy for HCWs. [NCT04483635]

Strengths and limitations of this study

- This trial was designed as a hybrid study enabling partially or totally remote screening, randomisation, follow-up, as well as outcome documentation by use of home capillary blood and saliva sampling, visits conducted by videoconference, monitoring by electronic reminders and questionnaires, and communication by phone, text messaging or emails.
- This trial used a pragmatic subject selection and easily applicable intervention to maximise subsequent implementation in practice.
- The main outcome was clinically meaningful as it explored the primary prevention impact of vitamin D on the risk of laboratory-confirmed infection and would likely change practice if a 20% reduction was documented.
- A single loading dose followed by regular doses have been shown to lead to rapid and sustained increase in serum level of 25-hydroxy-vitamin D and ensure adequate group separation, both properties desired in the context of a rapidly expanding epidemic while facilitating adherence in exhausted frontline health workers.
- Given the uncertainty in the progression of the infection rate, the use of a Bayesian adaptive design allows for adaptations (early stopping or prolongation of duration of follow-up) at the interim analysis according to the projection of infection rates.

Introduction

The Coronavirus 2 (SARS-CoV-2) disease (COVID-19) outbreak has rapidly expanded to a global pandemic. Healthcare workers (HCWs) play a crucial role in the fight against the COVID-19 pandemic. It was therefore a public health priority to develop strategies to decrease the risk and severity of COVID-19 in this vulnerable population. Indeed, there was rising concern as HCW are overrepresented in terms of infections (3.8% of infected individuals in Wuhan, China,[1] 10% in Italy and 12% in Spain, 10-20% in US)[2 3] and perhaps severity. Working in long-term care facilities (LTCF) and with aerosol generating medical procedures (e.g., hospitals) further increased the risk (Odds Ratio [OR]: 2.3).[4] The risk of reporting COVID-19 infection in front-line HCWs, defined as those in direct contact with patients, was 10-fold greater than the general population at the beginning of the pandemic (Hazard Ratio [HR]= 11.61). [5] Recent research also indicated that HCWs who were Blacks, Asians, or other minority ethnic populations, had a higher likelihood of contracting COVID-19.[5] Compared to those unexposed to COVID-19 patients; the risk was two to five-fold higher in HCWs exposed to COVID-19 suspected (HR= 2.39) or confirmed (HR= 4.83) patients, even with adequate personal protection equipment (PPE).[5] Although infections may be due to contact with infected patients, community, or family acquired disease, cases were rapidly emerging from cross-infection with asymptomatic infected HCW.

Vitamin D is an immunomodulatory micronutrient, and its levels in the body may vary due to diet and environmental conditions. Vitamin D insufficiency has been associated with increased risk of respiratory infections, and possibly COVID-19,[6] asthma exacerbations, and acute respiratory distress syndrome (ARDS) among others.[7-9] Optimal pro-immune and anti-inflammatory impacts likely occur at 25-hydroxyvitamin D (25OHD) levels above 75 nmol/L (30 ng/mL).[10,11] In a systematic review of 25 randomized controlled trials (RCT) of 11321 individuals, daily/weekly vitamin D supplementation decreased by 19% the rate of acute respiratory infections (two-step

analysis; OR 0.81, 95% CI 0.72 to 0.91),[12,13] with a stronger effect in subjects with baseline 25OHD <25 nmol/L. Whereas subgroup analyses suggested a protective effect primary in individuals receiving daily or weekly vitamin D supplement without an additional bolus, but not in those with bolus,[14] other important differences in population (e.g., malnutrition),[15,16] age (infant),[16] chronic disease (e.g. asthma, COPD)[17-21] and type of infection (e.g. bacterial)[15,16] could have contributed to the apparent lesser effect. Of interest, Vitamin D supplementation significantly reduced the rate of severe exacerbations (i.e., requiring rescue systemic corticosteroids), a condition association with airway inflammation, with no impact of bolus use or not. [14] Vitamin D supplementation was also found to be associated with a decreased load of rhinovirus (common cold), consistent with an increased antiviral immune response.[22] A systematic review and several studies reported an inverse association between serum vitamin D levels and COVID-19 severity, inpatient mortality, as well as serum levels of C-reactive protein (CRP) and lymphocyte percentage.[23,24] These findings suggest that vitamin D status was linked with the severity and mortality of the COVID-19 infection in the general population, particularly in severe COVID-19 cases. Whether Vitamin D could prevent or lessen infection and/or the inflammatory response associated with the COVID-19 remained to be explored.[25] At the time of the funding in June 2020 and study initiation (February 2021), no other primary prevention trials were published. Since then, one positive and two negative trials testing different vitamin D intervention were published. [26-28]

The vitamin D receptor is expressed on innate and adaptive immune cells which also synthesize the active metabolite 1,25-hydroxyvitamin D_3 ($1,25(OH)_2D_3$); thus, vitamin D can strengthen innate and adaptive cellular immunity by increasing local production of antimicrobial peptides, decreasing secretion of pro-inflammatory cytokines, inhibiting dendritic cell activation,

suppressing T helper cell type 1 response, and promoting T regulatory cells induction. These cellular effects are crucial for host responses against infection and can reduce the survival and replication of respiratory viruses.[13,24] $1,25(OH)_2D_3$ is also produced locally in bronchial epithelial cells and downregulates inflammatory cytokines (e.g. interleukin-8) and chemokines (e.g. leucocyte attracting CXCL10) expression from stimulated cells.[29]

The protocol of a placebo-controlled parallel-group triple-blind RCT to explore the impact of vitamin D_3 supplementation on reducing the risk and severity of laboratory-confirmed COVID19 infection in HCWs is described herein, as per Standard Protocol Items: Recommendation for Intervention Trials guidelines **(Online supplemental file 1).** After funding, but prior to the start of recruitment, the protocol underwent four amendments (8 protocol versions) in view of the rapidly evolving science, multiple challenges faced with conducting a large scale COVID-19 trial of high-risk health-care workers during the pandemic, including difficulty in obtaining large-scale supplies, as well as favorable pilot results of two novel technologies **(Table 1).** These original and final (1.8, January 18, 2021) protocol versions are described below. The trial was initiated but interrupted prematurely due to recruitment difficulty.

Research questions and study hypothesis

Objectives

The primary question was whether one oral dose of 100,000 IU vitamin D_3 (administered at baseline) plus weekly supplement of 10,000 IU vitamin D_3 can decrease the risk of laboratory-confirmed COVID-19 infection, versus placebo, in frontline HCWs in high COVID-19 incidence areas.

Additionally, the study aimed to examine if, compared with placebo, the vitamin D intervention reduces: (i) illness severity, (ii) symptom duration, (iii) work absenteeism, and (iv) unemployment among frontline health care workers (HCW) in high COVID-19 incidence areas. This study will also assess various exploratory outcomes.

Hypothesis

We hypothesised that compared to placebo, vitamin D supplementation would decrease the incidence of laboratory-confirmed symptomatic COVID-19 infection by 20% in frontline HCWs working in high COVID-19 incidence area.

Table 1. Study Amendments and Notifications

	Clinical trial Application (CTA)				Investigational Testing Authorization (ITA)	
Version number	**Changes**	**Description**	**Submitted**	**Approved**	**Submitted**	**Approval**
Version 0.0 11-05-2020						
Version 1.0 23-08-2020	• Eligibility • Outcomes & Covariates	• Strengthening of exclusion of 'suspected or previously undocumented COVID-19 infection' by adding: (i) a questionnaire of symptoms elaborated by Menni et al. and (ii) a rapid (15-minute) serology test, not yet approved nor tested in Canada, to be pre-tested in a pilot study. • Addition of capillary blood self-collection with Tasso-SST device (to be pre-tested in a pilot study).	23-08-2020	16-09-2020	N/A	N/A
Amendment 1 **Version 1.1** 23-10-2020	• Eligibility • Exploratory Outcomes • Main outcome	• Clarification of the NADAL® COVID-19 IgG/IgM Rapid Test (Teracero Pharma Inc., Lachine Canada) on venous whole blood as the rapid serology test to exclude prior infection (following pilot comparative study) • Validation of Nadal serology test compared to IgG SARS-CoV2 serology as endpoint exploratory outcome • Salivary COVID-19 RT-PCR test method prioritized over nasopharyngeal samples for twice-monthly self-collection or accepted for clinical diagnostic by qPRC	23-10-2020 (CTA-A) †	02-11-2020	23-10-2020 (NADAL)	14-111-2020
Amendment 2 Versions 1.2-1.4 **Version 1.5** 27-11-2020	• Primary Outcome • Outcomes	• Removal of q2 weeks saliva sample for COVID-19 rt-PCR analysis due to supply problem with 50 mL Falcon centrifuge tube caused by a global plastics shortage combined with un-acceptable delay for public tender for a contract with courier service for q2weeks biological samples • Addition of questions and procedures to account for the possible effect of the Vitamin D supplementation on immune response to COVID-19 vaccine, including a saliva sampling for COVID-19 by RT-PCR and a blood test for serology (and vitamin D) prior to vaccination	27-11-2020 (CTA-A) †	30-11-2020	23-11-2020 (TASSO & NADAL)	2-12-2020

	Clinical trial Application (CTA)				Investigational Testing Authorization (ITA)	
Version number	**Changes**	**Description**	**Submitted**	**Approved**	**Submitted**	**Approval**
	• Covariates & Outcome (Device)	• Specification of the TASSO SST on Demand (Tasso Inc, Seattle, USA) as choice for capillary blood self-sampling (following pilot comparative study)				
Amendment 3 versions 1.6 & **Version 1.7** 12-12-2020	• Eligibility • Exploratory Outcomes	• Exclusion of health care workers who have received the COVID-19 vaccine prior to enrolment • Addition of (i) effect of high-dose vitamin D on SARS-CoV-2 IgG titers before & after 2nd dose of COVID-19 vaccine and (ii) the long-term infection rate up to 12 months after end-of-study • Modifying exploratory outcome to allow exploration of modulating effect of vitamin D, not only on the risk of COVID-19 infection but also on response to vaccine	12-12-2020 (CTA-A) †	16-12-2020	12-12-2020 (TASSO & NADAL)	23-12-2020
Amendment 4 Version 1.8 18-01-2021	• Exploratory Outcomes • Eligibility	• Clarification that the serology to be done just prior the second dose of a COVID-19 vaccine may not always be at 3 or 4 weeks (as recommended by vaccine manufacturer) to reflect the recent governmental decision to delay the timing of the 2nd vaccine dose to 12 to 16 weeks • Slightly modifying wording to target healthcare workers at risk of contact with infected individuals that were not suspected of being infected (e.g., patients, colleagues, students, etc.)	18-01-2021 (CTA-N) ‡	N/A	18-01-2020 ¶	01-02-2021

† CTA-A, Clinical Trial Application -Amendment
‡ CTA-N, Clinical Trial Application -Notification
¶ As there is no 'notification' category for the Investigational Testing Authorization (ITA), each amendment or notification to the Clinical Trial Application (CTA) must be submitted as a new amendment for the devices to be reviewed by ITA.

Methods and analysis

Study design

This was a pragmatic 16-week, triple-blind, placebo-controlled, parallel-group, randomized trial comparing supplemental vitamin D3 and placebo in HCWs with the possibility of extending the study up to 24 weeks, depending on infection rate progression during an interim analysis **(Figure 1)**.

Subjects

HCWs (i.e., physicians, allied health care workers, orderlies, etc.) were eligible if they: (i) were aged ≥18 and <70 years old; (ii) were authorized to practice in Quebec; (iii) were working or scheduled to work over the next 16 weeks in a setting at high-risk of contact with COVID-19 infected individuals, particularly (but not only) those involved with aerosol generating medical procedures in hospitals and/or caring for patients in long-term care facilities; (iv) were working in high COVID incidence areas in the greater Montreal area and surroundings; (v) were covered by the provincial universal public health insurance (*Régie de l'assurance-maladie du Québec* [RAMQ] for medical services and hospitalisations; (vi) had a personal email or phone (to which to send reminders and questionnaire by email or text messages); (vii) had a fixed address (to which to send the material) in the greater Montreal or surrounding areas.

HCWs were excluded if they met any of the following criteria: vitamin D supplementation (cholecalciferol or calcitriol) intake >400 IU/day (or >12,000 IU/month) in past 3 months; intention to take >400 IU per day during the study period; suspected or previously documented COVID-19 infection; history of nephrolithiasis, hypercalcemia, hyperphosphatemia, hyperparathyroidism, granulomatosis disease (e.g., tuberculosis, sarcoidosis), renal failure, or

active cancer; current intake of medications that may cause hypercalcemia such as lithium, teriparatide, or digoxin; anticipated prolonged absence from work during the study period (i.e., pregnancy); anticipated difficult follow-up; enrollment in a concurrent interventional randomized trial; have already received the vaccine against COVID-19. Participation in this trial did not preclude subsequent enrollment in a COVID-19 therapeutic (but not preventive) trial, which would be documented.

Study intervention

Participants in the intervention group received 100,000 IU vitamin D_3 (cholecalciferol) at randomization followed by a weekly dose of 10,000 IU vitamin D_3 for 16 weeks. Participants in the control groups received an identical placebo bolus followed by placebo weekly supplement for 16 weeks. Sufficient supply was provided for 24 weeks, in case of prolongation study based on the interim analysis. Participants in both groups were asked to take the study intervention with their most copious meal. Treatment of co-morbidities were permitted. Vitamin D intake up to 400 IU per day was allowed.

Randomization

Randomization was implemented using a computer-generated random list stratified by one of 11 workplaces (*Centre Hospitalier Universitaire* [CHU]) or health region (*Centre Intégré Universitaire de Santé et Services Sociaux* [CIUSSS] or *Centre Intégré de Santé et Services Sociaux* [CISSS]). HCWs were allocated (1:1) using permuted block randomization to enhance concealment. Group allocation codes for each stratum was held in a secure location with restricted access by the Central Pharmacy and Data Management.

Patient and public involvement

Participant burden of research measures was assessed using feedback from patients participating in one pilot round. Patients were not involved in study deign, recruitment of participants or conduct of the study. Results of this study will be disseminated through public fora.

Outcomes

Primary outcome

The original primary outcome, incidence of laboratory-confirmed COVID-19 infection, was originally based on (i) bi-monthly self-obtained mid-turbinate nasopharyngeal (NP) swabs, complemented by (ii) NP swabs obtained clinically for screening or diagnostic purposes throughout the study, both analysed by RT-qPRC approved by Health Canada. Faced with the unexpected cancellation of our large order of Falcon tubes to collect saliva sample for qPRC combined with the unacceptable additional delay for a public tender to securing a contract with a private courier service and in view of the uniform protocol for screening symptomatic or COVID-19 exposed health care workers throughout the Province of Quebec and the reliability of IgG serology, we decided to forgo the twice-monthly saliva sampling for qPRC analysis. The revised definition of the primary outcome became the incidence of laboratory-confirmed COVID-19 infection, documented by RT-qPCR based on salivary (or nasopharyngeal) specimens (i) obtained for screening or diagnostic purposes throughout the study and (ii) self-obtained salivary specimens obtained at endpoint as well as (ii) COVID-19 IgG seroconversion at endpoint (in COVID-unvaccinated individuals: ≥15 UA on the anti-S SARS-CoV-2 IgG Diasorin on Liaison XL

platform; in COVID-vaccinated individuals : ≥1.40 index (S/C) on the anti-N SARS-CoV-2 IgG on ARCHITECT platform)

Secondary outcomes

(i) *Distribution of disease severity* on a 5-category ordinal scale [asymptomatic; mild (managed at home); moderate (hospitalisation without supplemental oxygen); severe (oxygen supplementation); critical (mechanical ventilation/death)], (self-reported, RAMQ); (ii) *Duration of COVID-19 positivity* between 1st COVID+ to first COVID- test) revised to *Duration of COVID-19 related symptoms* in individuals with laboratory confirmation of COVID infection, (self-reported on diary); (iii) COVID-19 IgG seroconversion documented at endpoint (see above); (iv) duration of work absenteeism (self-reported, medical records or human resources databases); (iv) duration of unemployment support (human resource databases); (v) Adverse health events (self-reported). Several *exploratory outcomes* pertained to the: incidence of post-acute and chronic symptoms; long-term (1-year) morbidity and work absence related to COVID-19; change in gene expression of ACE2 and TMPRSS2 in saliva cells; change in inflammatory markers (i.e., CRP), immune response post vaccination; other viral infections; and genetic markers (including changes in gene expression).

Study Procedures

To facilitate the recruitment of participants, this study is conceived as hybrid trial enabling partially or totally remote trial participation including screening, randomization, follow-up, and end-of-study visit.

Pre-Screening

Advertisements were placed in health institutions, newspapers, social media and online, where participants were invited to complete an online pre-screening form, read and download the consent forms; and if eligible and interested, to book a virtual screening appointment (via a secured videoconferencing platform) with research team who would confirm eligibility, explain the study, obtain informed consent, and schedule a virtual or in-person randomization visit.

Screening

At the virtual screening visit by videoconferencing, research coordinators completed with the individual a more extensive eligibility questionnaire, which included additional questions about: anticipated work exposure over the next 16 weeks to COVID-infected or suspected individuals and to high-risk medical procedures; work place (*Centre Hospitalier Universitaire* [CHU]) or Centre Hospitalier Universitaire Sainte-Justine) or health region (CIUSSS or CISSS), serving as randomization stratum; prior laboratory-confirmed or physician-suspected COVID-19 infection; assessment of the likelihood of prior/current, yet undocumented, COVID-19 infection using the 5-item questionnaire developed by Menni et al[30] (score >0.50 interpreted as high likelihood of prior infection); and finally, the comfort level with the study design and procedures, including saliva and capillary blood sampling self-collection demonstrated by instructional videos. Eligible and consenting individuals electronically signed an online consent form (with the signed PDF consent form automatically emailed to participants). Then, two additional questionnaires were completed on line with the research coordinators namely: (i) the baseline questionnaire collecting information about household, ethnicity, part- vs. full-time work, personal health, skin color (measured with the Fitzpatrick scale),[31] concomitant medications or supplements, and (ii) the nominative CRF

collecting demographic information essential to opening a medical and pharmaceutical research record (i.e., public health insurance number, allergies) and maintaining contact with the research team throughout the study (preferred means to receive electronic reminders/questionnaires and to be notified of positive test results; address to receive study material or for biological sample pick-up; and next-of-kin contact in case of inability to respond to questionnaire due to illness), and to document work absence (employee number).

Finally, at the screening visit, the participant was asked to choose an appointment for a *virtual* (via a secured videoconferencing platform) or *in-person* randomization visit at one of several locations. To help select their preferred visit format, videos of key procedures (such as home blood collection) were shown. Only in participants with a significant likelihood of a current or past undocumented (Menni score > 0.5) was an *in-person* randomization visit mandatory to receive the rapid COVID-19 serology test, prior to randomisation.

Preparation and shipment of Study drug by Research Pharmacy

The list of new participants approved by one of the PIs was sent daily by email to the CHUM research team to be open a medical chart and send an electronically signed prescription for the Study medication, to the Research Pharmacy for preparation of study drug.

Prior to randomization, a list of all consenting and eligible participants was automatically sent every night to the one of the co-principal investigators (FMD or CT) who screening and baseline questionnaires to approve or refuse study entry and electronically signed their decision. The list daily list of new PI-approved participants was sent electronically daily to the CHUM research

team. Medical and pharmaceutical records were opened and an electronically signed prescription for the Study medication sent to the Research Pharmacy for preparation of study drug for a given target date.

To enable remote randomization, the randomization took place about one week prior to the randomization visit to allow enough time for the preparation and shipment of patient-specific study supplement to the research team and, in turn, the shipment of the Study supplement and all materials required for the randomization visit by the research team to the participant.

Randomization visit

Seventy-two and 24 hours prior to, and at, the randomisation visit, participants were screened by questionnaire for recent travel, symptoms suggestive of SARS-Cov2 infection, or exposure to COVID-19 infected individuals. Those who responded positively were asked to get tested, notify their institutional health service and await end of quarantine and/or confirmed negative test to reschedule the randomisation visit.

Randomization visit (week 0) was performed *in person* (60 minutes) or *remotely* (90 minutes), depending on the availability and preference of participants as well as their likelihood of a past COVID-19 infection.

In-person visits were conducted—by appointment only—in designated rooms with restricted access. The research coordinators wore personal protection equipment (PPE), and all procedures, from participant arrival to departure, were approved by the institutional Infection Control and Safety committee. The *in-person* visit entailed (i) ascertainment of the signed consent form, (ii)

capillary blood sample collection to perform NADAL® COVID-19 IgG/IgM Rapid Test (Teracero Pharma Inc., Lachine Canada), (iii) venous blood sample collection for baseline serum 25(OH)D and SARS-CoV-2 IgG serology analyses and if genetic consent, DNA; (iv) viewing of the saliva collection video and instruction pamphlet, (v) collection of the first specimen under supervision, (vi) a final verification of the eligibility and exclusion criteria; (vii) randomization; (viii) oral administration of 100,000 IU vitamin D_3 or an identical placebo, and (ix) distribution of the study material including study supplement, saliva sampling kit for end-of-study, biohazard and sampling bag, and, if a remote visit was anticipated at week 16, capillary blood collection kits (TASSO OnDemand SST device). Any patient with a positive NADAL COVID-19 IgM/IgG Rapid Test serology test were excluded prior to randomisation.

The *remote* randomization visit, conducted by video-conference, was similar to the *in-person* randomization visit with the following additions: (a) viewing of the capillary blood collection video and instructional pamphlet; (b) remote capillary sampling under guidance using the TASSO-SST device (TASSO Inc., Seattle, USA); (c) viewing of the saliva collection video and instructional pamphlet (OG-600 Oragene DNA Collection Kit, DNA Genotek Inc., Ottawa, Canada); (d) remote DNA salivary sampling under guidance; (e) preparation of biological samples for shipment with phase change and insulated envelopes under guidance and (f) organising collection of biological specimens by approved courier service to respective laboratories. Note that a Nadal serology test was not conducted remotely.

Follow-up

Participants received *weekly electronic (SMS or email) reminders* to take their weekly Study Supplement (10,000 IU vitamin D or an identical placebo) and to start completing an *online daily Diary* if they tested positive to SARS-CoV2 or they developed symptoms suggestive of COVID-19 infection.

Every two weeks, participants received a link to complete a *brief online questionnaire* asking to report: their adherence to weekly Study Supplement intake; health status including recent COVID-19 related exposure, symptoms, or testing; adverse health events or new co-morbidities; change in concomitant medications or supplement intake; work status (active duty, quarantined, holiday, sick) and work setting (ED, ICU, etc.); as well as expected/recent COVID-19 vaccination (date and vaccine name) if any; the latter question served to enable timely shipment of materials for additional sampling prior to vaccination, as COVID-19 vaccination was permitted during the study. In participants who planned to get vaccinated during the study, three additional blood, and one additional saliva, samplings, either *on-site* or *remotely*, were planned including: saliva (for COVID-19 qPCR analysis) and blood (for SARS-CoV-2 anti-S IgG serology) sampled prior to vaccination, a blood sample (for SARS-CoV-2 anti-S and anti-N IgG serology) collected just prior to the second vaccine dose, and a blood sample (for SARS-CoV-2 anti-S and anti-N IgG serology) collected one month of after second vaccine dose and endpoint. Regardless of their vaccination status, participants were asked to continue taking the weekly Study Supplement and complete the bi-monthly questionnaire until the end of the study. If questionnaires were not completed within 2 days of the target date, the research coordinator reached out the participant to complete the information.

End-of-Study visit

An end-of-study visit was conducted either in-person (60 minutes) or remotely (90 minutes), depending on the availability and preference of participants and likelihood of a current COVID-19 infection. The *in-person end-of-study visit* entailed the collection of a (i) venous blood sample for serum 25(OH)D and SARS-CoV-2 anti-S IgG serological results and in vaccinated participants a SARS-CoV-2 anti-N IgG serology, (ii) capillary blood sample to perform the NADAL® COVID-19 IgG/IgM Rapid Test, (iii) a saliva sample for SARS-CoV-2 qPCR analysis as well as guessing of allocation and return of the study supplement bottle to assess adherence and any unused material.

The *remote end-of-study visit* conducted via videoconference entailed the same procedures as the in-person end-of-study visit with one exception: the self-collection of a capillary (instead of venous) blood using TASSO-SST devices (for the serum 25(OH)D and SARS-CoV-2 anti-S with/without anti-N serology). Individuals were guided into self-performing the pinprick capillary method to perform the NADAL® COVID-19 IgG/IgM Rapid Test and return of biological samples and materials by pre-paid approved courier.

Covariates

Several covariates that may act as confounders or interaction variables in the magnitude of effect associated with the intervention were documented, namely: baseline serum 25OHD level; smoking; concomitant supplements or drug(s) that alter calcium or vitamin D absorption or metabolism such as diuretics and anti-epileptics (reported at baseline and every 2 weeks); skin color (documented at baseline); obesity (documented by height & weight [BMI] at baseline); other comorbidities (i.e., diabetes, hypertension, etc.) that may affect the severity of COVID-19

infection and receipt of a COVID-19 vaccine (documented by verbal report bi-monthly). All external (governmental and institutional) databases were to be obtained 3 months before, and up to 16 months following, randomization (as well as 12 months after then study endpoint).

During an event

During COVID-19 related symptoms or documented SARS-CoV infection, participants were instructed to complete a daily symptom diary from date of onset of symptoms or positive test, until two days with no symptoms or 14 days if asymptomatic,

Risk management

Clinical and biochemical adverse health events (AHEs) were monitored throughout the study and reported for all patients at the end of the study. No specific laboratory safety monitoring was planned given the established safety of the loading dose of 100,000 IU and weekly dose of 10,000 IU.[32,33] Adverse Health Events (AHE) were recorded via electronic questionnaires throughout the study. Participant who reported symptoms suggestive of vitamin D intoxication had a venous blood sampling (total and ionised calcium, phosphorus, alkaline phosphatase, albumin, and creatinine). Any abnormal laboratory values was interpreted as 'clinically significant' or 'not clinically significant' by the Site endocrinologist blinded to study allocation. Further investigation or action for individual participants (including interruption, cessation, or unblinding of the study drug via pharmacy or by analysis of serum 25OHD) was be determined by the Site endocrinologist, if indicated to ensure participant safety. The AHE's occurrence was reviewed periodically by the Data and Safety Monitoring Board (DSMB). Code breaking was allowed only if deemed essential

for participant management. If relevant, summary reports aggregating (or not if requested) both groups were to be provided to the DSMB.

Data management and monitoring

The principle investigator (FMD) and statistical group (SG, RP) oversaw randomization, data management, progress monitoring, and all analyses, including those for Data Monitoring Safety Board (DMSB). The DSMB membership included: Lehana Thabane, biostatistician (Chair), Gary Kobinger, infectious disease specialist, Kevin Thorpe, biostatistician, and Edgar Delvin, biochemist & expert in Vitamin D. DACIMA was used for online data entry and management.

A combination of *remote* monitoring activities and *in-person* routine monitoring visits were conducted by an independent Study Monitor with the first randomised participants at each site and on a bi-monthly basis, to ensure that each Site adhered to the study protocol, Good Clinical Practice guidelines and data collection completeness.

Sample size calculation

Given uncertainties in infection progression, a Bayesian adaptive design was used where the posterior probability of effectiveness, i.e., P(OR<1|data) was the basis of inference and decision making.[34] Assuming an expected OR of 0.80 and 1:1 treatment allocation, a total net sample size of 2100 was required to identify a 20% reduction in the risk of COVID-19 in the vitamin D vs. control group, with 80% power with the design described above. Considering a drop-out rate of 15%, 2414 participants were targeted. An interim analysis was planned when 75% of participants reached week 12, at which time the following assessments were made: the *progression over time*

in the incidence of infection (slope of the curve of infection) was updated and if the probability of effectiveness exceeded 0.95 [p(OR<1)>0.95], the trial should be terminated for efficacy at the interim point (12 weeks); otherwise, the study would continue to 16 weeks. Simulation results showed that, with the net sample size of 2100 (assuming an expected OR of 0.80 and 1:1 treatment allocation), there was about a 55% chance that the trial would be terminated for efficacy at the interim analysis.[35] The overall infection rate was monitored on a monthly basis: note that the study could have been extended to 24 weeks based on the progress of the infection rate, if required.

Statistical analysis

Primary outcome

An intention-to-treat (ITT) analysis was to be carried out with all randomized participants. For the primary outcome, the posterior distribution of the odds ratio of COVID-19 infection (OR) was the basis of inference in interim and final analyses. The posterior distribution of the OR was to be estimated by drawing samples from the posterior risks under each arm, which could be obtained analytically in a Beta-binomial model. Flat prior distributions were assumed for the risks (Beta(1,1)). Posterior 95% credible intervals were to be reported as interval estimates for the OR. Crude analyses as well as analyses adjusted for important covariates (i.e., potential confounders, effect modification, and baseline group imbalances) where to be conducted. Subgroup analyses would be conducted on baseline 25OHD, age, sex, BMI, occupational risk, and COVID-19 vaccination. A stratified analysis on geographical infection rate would be explored; sensitivity analysis censoring to date of COVID-19 vaccination, would be conducted if applicable.

Secondary outcomes

Distribution of disease severity defined as a 5-level ordinal outcome would be examined with a Bayesian analysis using a proportional odds (PO) model; the posterior probability of OR would be obtained by Markov chain Monte Carlo sampling implemented in Stan.[34] Duration of symptoms, duration of workday absences and of unemployment would be examined by a zero-inflated Poisson distribution.

Ethics and dissemination

This study has been reviewed and approved by the research ethics board (REB) of the CHU Sainte-Justine, serving as the local REB of all participating institutions. A non-objection letter (NOL) from Health Canada has been obtained to use high-dose Vitamin D loading dose as well as Tasso OnDemand device for home blood sampling and the NADAL COVID-19 IgM/IgG Rapid serology test. Written informed consent for study participation, for biobanking specimens for ancillary studies, and for subsequent publication of results was obtained from all participants, with the knowledge that participation is voluntary and can be withdrawn at any time with no effect on their current/future medical care. As part of the informed consent, enrolees had the option to participate in the HostSeq COVID-19 Canadian biobank conducted under the supervision of CGen, a national Canadian platform for sequencing and genome analysis **(Supplementary file 2)**. In Canada, health care is provided to those who suffer harm from trial participation.

All protocol amendments were submitted to Health Canada, investigators and REB; if these changes implied a revision of consent forms, ongoing trial participants were informed of new modifications to provide informed consent. All information obtained during the study were and will be kept confidential as per the law. Data was collected directly by electronic data capture on Dacima Clinical Suite (DACIMA Software Inc., Montreal, Canada). Data safety and

confidentiality was upheld at all data collection stages by assigning a unique subject ID to each participant, with data and samples kept under lock and key, electronic password protection and access restricted to study personnel. Samples collected during the study were labelled with the unique research code, prior to transfer and storage at the CHUSJ biobank, with access restricted to authorised personnel.

This trial uses pragmatic patient (irrespective of baseline 25OHD level) and intervention to maximise subsequent implementation into practice. If effective in reducing infection and morbidity, this approach would be readily implementable and could markedly influence practice during the COVID-19 pandemic. No participant identifiers will be used in the dissemination of this research. Health care professionals serving as partners informed the study design and pre-test all questionnaires and will contribute to a disseminating plan. Results will be disseminated to the medical community and public health departments via national/international conferences and publications in peer-reviewed journals as well as to the public and study participants via the Direction Collaboration-Partenariat Patient of the University of Montreal and the Canadian Respiratory Research Network (CRRN) patient platform who would contribute to a disseminating plan to reach as many individuals as possible.

Trial status

The study was conducted as per version 1.8 (January 18, 2021). The recruitment started on February 9, 2021. Upon the DSMB recommendation, recruitment was stopped prematurely on March 18, 2021 after 34 participants enrolled due to the inability to recruit approximatively 200 participants/week required to meet the target sample size of 2415 participants. The DSMB advised that the continuation of the trial, as originally designed, would not be able to answer the research question

and recommended that recruitment be stopped for futility. Recruitment difficulties were attributed in part to the high use of vitamin D and high concurrent vaccination rate among our target population, healthcare workers, the first targeted to be vaccinated from January 2021 onwards. Based on the recommendations of the study's endocrinologist, a premature cessation of follow-up after a minimum of 4 weeks from randomization to monitor the safety of intervention in all participants. The timeframe was deemed sufficient to ensure participant safety while learning for the study, that is, transforming the PROTECT study into a pilot study to document the impact of the study intervention on the rise in Vitamin D serum level, participants' adherence the study intervention and procedures in the context of a hybrid study, etc. The last end-of-visit was conducted on May 4, 2022.

Potential redirections of the study were discussed. The first option was to change the main outcome for an immunogenicity study in the general adult population. However, after strong consideration of the amount of changes to be made to the protocol and related documents (standards of procedures, case report forms, participant' instructions and notification, etc.), the expected delay in obtaining approval by all regulatory and ethical authorities, the impossible logistic of recruiting participants after the same duration of exposure to the study intervention prior to their vaccination, combined with the government of Quebec announcement that all willing adults would be vaccinated by June 24, 2021, the research team judged that it would unfeasible to perform a scientific solid and feasible trial on immunogenicity if one could not control the timing of immunization, combined with the expected very short recruitment timeframe.

A second option that received very strong consideration was to replicate the PROTECT trial in children aged 9 years and over. Again, after considering changes to be made to the protocol and related documents, the expected delay for obtaining approval by all regulatory and ethical authorities including school boards, combined with the Pfizer-BioNtech announcement that their vaccine was not only 100% successful for preventing COVID-19 infection in adolescents aged 12 to 15 years but that they forecast vaccinating teenagers in time for September 2021 school entry, the PI judge it was unrealistic to aim for the large recruitment target within such a short timeframe.

The protocol was submitted after the last patient end-of-study visit, due to the incredible amount of work done to conducted to set-up and initiate this large hybrid trial; the latter included two pilot studies testing two experimental devices to enable partially or totally remote participation, in the context of the pandemic which imposed large protocol and space restrictions for recruiting on-site potentially COVID-19 infected health care workers, several protocol amendments to facilitate and adjust the trial in the context of emerging science and anticipated vaccination campaign and their impact of all electronic documents, manual of procedures, and regulatory approvals, coupled with the premature end-of-follow-up in enrolled participants.

With the gained experience and knowledge, it is crucial that a future trial must begin fast prior to widespread vaccination and in populations where infection rate is high.[28] Permitting study entry to individuals with prior infection and prior vaccination (given common reinfections and temporary vaccine protection) [36] could have been considered, but it would have significantly reduced the event rate, required prolongation beyond 24 weeks (and additional funding), and compromised study power as was noted in other primary prevention trials.[26,27] Restricting

eligibility to patients with vitamin D deficiency (<25 nmol/l) would have severely interfere with recruitment ability in population-based or health care workers studies. [26-28] Use of calciferol may be associated with more potency and rapid rise in serum 25OHD than expected for cholecalciferol[37] although the choice is debated[38] and rapid access to study drug and matching placebo remain a crucial challenge at the onset of a pandemic. Revisiting the intervention dose and frequency of administration in light of the latest literature on SARS-CoV2 and related virus should be considered, although current evidence suggest that, with similar doses, high-incidence population may be more important than dosing in primary prevention [26,28] and high doses are effective in tertiary prevention. [35] A pragmatic design with fewer outcomes and monitoring via administrative databases appears theoretically more efficient, but requires rapid access to data when interim analyses are planned to monitor event rate, a serious challenge. Pursuing a hybrid approach to facilitate enrollment in the context of a pandemic is feasible, although electronic self-screening and outcome monitoring required a lot of programming that may delay implementation.

The publication of this protocol is meant to share our experience, including conducting a hybrid (virtual and/or in-person) trial and lessons learned, enable protocol uptake and its improvement in the context of another epidemic/pandemic, and serve as reference for the publication of our pilot studies that enabled this trial, and lessons learned from this experience.

Contributors

FMD designed the study protocol, secured funding, and oversaw the overall conduct of the project. CLT contributed to the protocol and amendments, directed the study implementation at the CHUM, and coordinated the prescription of study drug, pharmacy dispensation, as well as salivary sample reception and interpretation. SG conceived the statistical approach and sample size calculation and along with RWP, oversaw randomization and statistical analysis. CL, JHW and CQ contributed to the study design and amendments. BH wrote the first manuscript draft. LGSM oversaw the safety assessment. DC is responsible for the work absenteeism analysis. All co-authors approved the manuscript. Authorship eligibility on resulting manuscripts will follow standard guidelines.

Competing interests

The authors declare that they have no competing interests.

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Data availability statement

The datasets used and analyzed during the current study will be made available by the corresponding author on reasonable request.

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Figure legend

Figure 1. Study outline

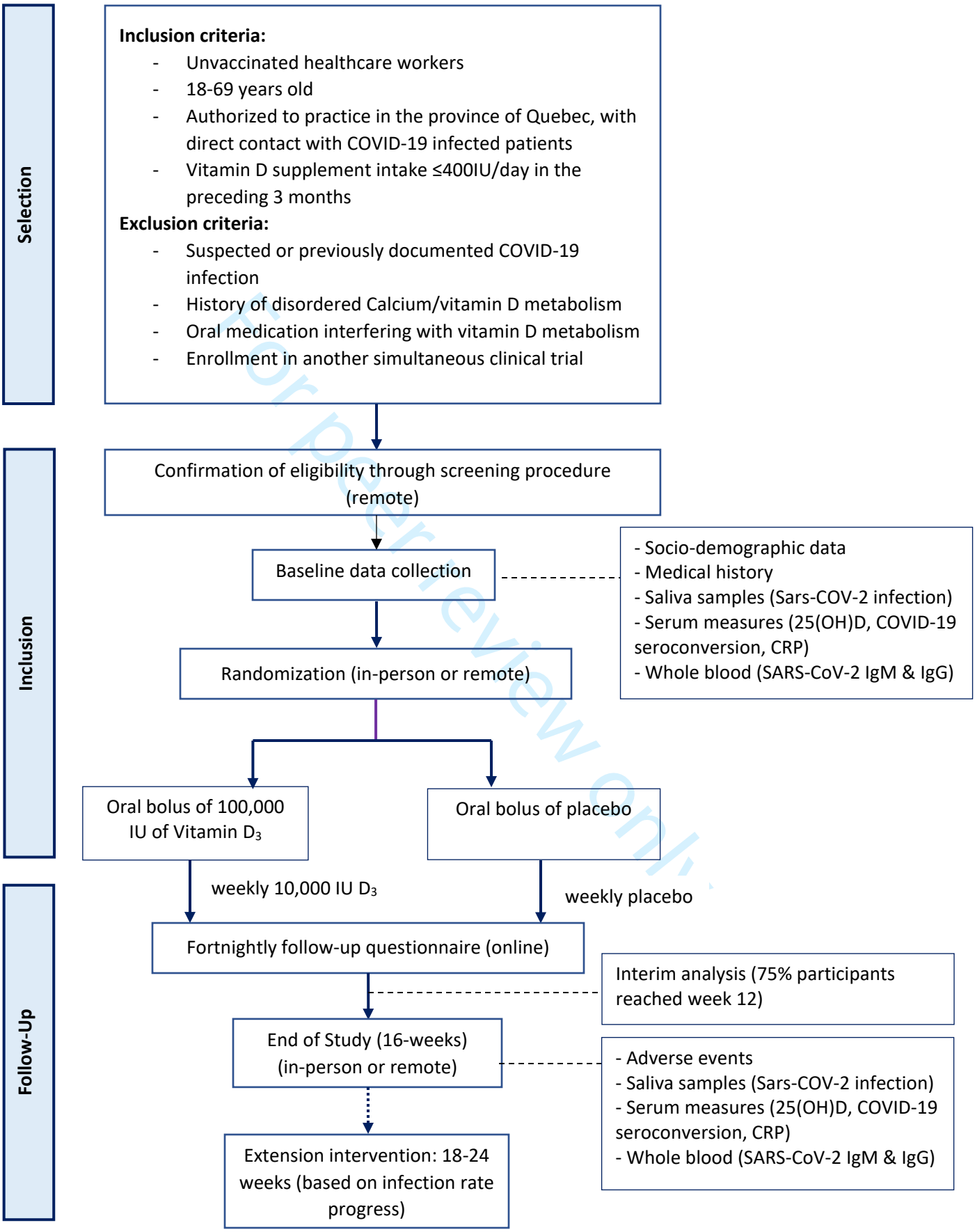

Figure 1

SPIRIT 2013 Checklist: Recommended items to address in a clinical trial protocol and related documents*

Section/item	ItemNo	Description	Addressed on page number
Administrative information			
Title	1	Descriptive title identifying the study design, population, interventions, and, if applicable, trial acronym	1
Trial registration	2a	Trial identifier and registry name. If not yet registered, name of intended registry	3
	2b	All items from the World Health Organization Trial Registration Data Set	**N/A**
Protocol version	3	Date and version identifier	8 (line 138)
Funding	4	Sources and types of financial, material, and other support	1, 25, supplemental funding file
Roles and responsibilities	5a	Names, affiliations, and roles of protocol contributors	1, 26-27
	5b	Name and contact information for the trial sponsor	1, 26
	5c	Role of study sponsor and funders, if any, in study design; collection, management, analysis, and interpretation of data; writing of the report; and the decision to submit the report for publication, including whether they will have ultimate authority over any of these activities	26

	5d	Composition, roles, and responsibilities of the coordinating centre, steering committee, endpoint adjudication committee, data management team, and other individuals or groups overseeing the trial, if applicable (see Item 21a for data monitoring committee)	19, 20, 26,27
Introduction			
Background and rationale	6a	Description of research question and justification for undertaking the trial, including summary of relevant studies (published and unpublished) examining benefits and harms for each intervention	6, 7
	6b	Explanation for choice of comparators	6, 7
Objectives	7	Specific objectives or hypotheses	8, 9
Trial design	8	Description of trial design including type of trial (e.g., parallel group, crossover, factorial, single group), allocation ratio, and framework (e.g., superiority, equivalence, noninferiority, exploratory)	9, 10
Methods: Participants, interventions, and outcomes			
Study setting	9	Description of study settings (e.g., community clinic, academic hospital) and list of countries where data will be collected. Reference to where list of study sites can be obtained	13

Eligibility criteria	10	Inclusion and exclusion criteria for participants. If applicable, eligibility criteria for study centres and individuals who will perform the interventions (eg, surgeons, psychotherapists)	9, 10
Interventions	11a	Interventions for each group with sufficient detail to allow replication, including how and when they will be administered	10, 11
	11b	Criteria for discontinuing or modifying allocated interventions for a given trial participant (eg, drug dose change in response to harms, participant request, or improving/worsening disease)	10
	11c	Strategies to improve adherence to intervention protocols, and any procedures for monitoring adherence (eg, drug tablet return, laboratory tests)	17, 18
	11d	Relevant concomitant care and interventions that are permitted or prohibited during the trial	10
Outcomes	12	Primary, secondary, and other outcomes, including the specific measurement variable (eg, systolic blood pressure), analysis metric (eg, change from baseline, final value, time to event), method of aggregation (eg, median, proportion), and time point for each outcome. Explanation of the clinical relevance of chosen efficacy and harm outcomes is strongly recommended	11, 12

Participant timeline	13	Time schedule of enrolment, interventions (including any run-ins and washouts), assessments, and visits for participants. A schematic diagram is highly recommended (see Figure)	15, 16, **17, 18, Figure** 1
Sample size	14	Estimated number of participants needed to achieve study objectives and how it was determined, including clinical and statistical assumptions supporting any sample size calculations	20-21
Recruitment	15	Strategies for achieving adequate participant enrolment to reach target sample size	12,13
Methods: Assignment of interventions (for controlled trials)			
Allocation:			
Sequence generation	16a	Method of generating the allocation sequence (eg, computer-generated random numbers), and list of any factors for stratification. To reduce predictability of a random sequence, details of any planned restriction (eg, blocking) should be provided in a separate document that is unavailable to those who enrol participants or assign interventions	11
Allocation concealment mechanism	16b	Mechanism of implementing the allocation sequence (eg, central telephone; sequentially numbered, opaque, sealed envelopes), describing any steps to conceal the sequence until interventions are assigned	11

Implementation	16c	Who will generate the allocation sequence, who will enrol participants, and who will assign participants to interventions	11
Blinding (masking)	17a	Who will be blinded after assignment to interventions (eg, trial participants, care providers, outcome assessors, data analysts), and how	5, 19
	17b	If blinded, circumstances under which unblinding is permissible, and procedure for revealing a participant's allocated intervention during the trial	19
Methods: Data collection, management, and analysis			
Data collection methods	18a	Plans for assessment and collection of outcome, baseline, and other trial data, including any related processes to promote data quality (eg, duplicate measurements, training of assessors) and a description of study instruments (eg, questionnaires, laboratory tests) along with their reliability and validity, if known. Reference to where data collection forms can be found, if not in the protocol	12 to 19
	18b	Plans to promote participant retention and complete follow-up, including list of any outcome data to be collected for participants who discontinue or deviate from intervention protocols	12 to 19

Data management	19	Plans for data entry, coding, security, and storage, including any related processes to promote data quality (eg, double data entry; range checks for data values). Reference to where details of data management procedures can be found, if not in the protocol	20
Statistical methods	20a	Statistical methods for analysing primary and secondary outcomes. Reference to where other details of the statistical analysis plan can be found, if not in the protocol	21, 22
	20b	Methods for any additional analyses (eg, subgroup and adjusted analyses)	22
	20c	Definition of analysis population relating to protocol non-adherence (eg, as randomised analysis), and any statistical methods to handle missing data (eg, multiple imputation)	21-22
Methods: Monitoring			
Data monitoring	21a	Composition of data monitoring committee (DMC); summary of its role and reporting structure; statement of whether it is independent from the sponsor and competing interests; and reference to where further details about its charter can be found, if not in the protocol. Alternatively, an explanation of why a DMC is not needed	20,

	21b	Description of any interim analyses and stopping guidelines, including who will have access to these interim results and make the final decision to terminate the trial	20
Harms	22	Plans for collecting, assessing, reporting, and managing solicited and spontaneously reported adverse events and other unintended effects of trial interventions or trial conduct	17, 19, 20, 23
Auditing	23	Frequency and procedures for auditing trial conduct, if any, and whether the process will be independent from investigators and the sponsor	20
Ethics and dissemination			
Research ethics approval	24	Plans for seeking research ethics committee/institutional review board (REC/IRB) approval	25
Protocol amendments	25	Plans for communicating important protocol modifications (eg, changes to eligibility criteria, outcomes, analyses) to relevant parties (eg, investigators, REC/IRBs, trial participants, trial registries, journals, regulators)	8
Consent or assent	26a	Who will obtain informed consent or assent from potential trial participants or authorised surrogates, and how (see Item 32)	13
	26b	Additional consent provisions for collection and use of participant data and biological specimens in ancillary studies, if applicable	N/A

A	27	How personal information about potential and enrolled participants will be collected, shared, and maintained in order to protect confidentiality before, during, and after the trial	14-15, 25
Declaration of interests	28	Financial and other competing interests for principal investigators for the overall trial and each study site	27
Access to data	29	Statement of who will have access to the final trial dataset, and disclosure of contractual agreements that limit such access for investigators	27
Ancillary and post-trial care	30	Provisions, if any, for ancillary and post-trial care, and for compensation to those who suffer harm from trial participation	25
Dissemination policy	31a	Plans for investigators and sponsor to communicate trial results to participants, healthcare professionals, the public, and other relevant groups (eg, via publication, reporting in results databases, or other data sharing arrangements), including any publication restrictions	23
	31b	Authorship eligibility guidelines and any intended use of professional writers	25
	31c	Plans, if any, for granting public access to the full protocol, participant-level dataset, and statistical code	27

Appendices

Informed consent materials	32	Model consent form and other related documentation given to participants and authorised surrogates	Supplementary file 2
Biological specimens	33	Plans for collection, laboratory evaluation, and storage of biological specimens for genetic or molecular analysis in the current trial and for future use in ancillary studies, if applicable	Supplementary file 2

*It is strongly recommended that this checklist be read in conjunction with the SPIRIT 2013 Explanation & Elaboration for important clarification on the items. Amendments to the protocol should be tracked and dated. The SPIRIT checklist is copyrighted by the SPIRIT Group under the Creative Commons "Attribution-NonCommercial-NoDerivs 3.0 Unported" license.

CHU Sainte-Justine
Le centre hospitalier universitaire mère-enfant
Université de Montréal

CHUM

Centre intégré universitaire de santé et de services sociaux de l'Ouest-de-l'Île-de-Montréal Québec
Centre intégré de santé et de services sociaux de la Montérégie-Centre Québec
Centre intégré de santé et de services sociaux de la Montérégie-Est Québec
Centre intégré universitaire de santé et de services sociaux de l'Est-de-l'Île-de-Montréal Québec
Centre intégré universitaire de santé et de services sociaux du Centre-Sud-de-l'Île-de-Montréal Québec

INFORMATION AND CONSENT FORM

Project Title: PRevention of COVID-19 with Oral Vitamin D supplemental Therapy in Essential healthCare Teams (PROTECT)

Protocol Number: PROTECT 2020

- **Principal investigators at CHUSJ:** Dr. Francine M. Ducharme, MD, FRCPC, Paediatrician, Centre hospitalier universitaire Sainte-Justine (CHUSJ)
- **Principal investigator at the CHUM:** Dr. Cecile Tremblay, MD, FRCPC, Microbiologist/Infectiologist, Centre hospitalier universitaire de Montréal (CHUM)

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- Shirin Golchi, biostatistician, McGill university
- Cristina Longo, epidemiologist, University of Montreal
- Robert Platt, biostatistician, McGill University
- Caroline Quach, MD, Paediatrician microbiologist, CHUSJ
- Christian Renaud, pediatric microbiologist, CHUSJ
- John White, biochemist, McGill University

Co-investigators at the CHUM:

- Dr. Louis-Georges Sainte-Marie, MD, endocrinologist, CHUM
- Dr. Emil Toma, MD, Dsc, FRCPC,CHUM

Industrial Collaborators: Laboratoires Riva, Blainville, Quebec

Funding source: Canadian Institutes of Health Research (CIHR), in the context of the COVID-19 Rapid Research Funding Opportunity

Multicenter identifier: MP-21-2021-3044
CHUM project number : 20.319

WHY ARE YOU BEING INVITED TO TAKE PART IN THIS STUDY?

Today, we are inviting you to participate in this research study because you are a healthcare worker who is working in a high COVID-19 incidence area and in a setting with a high risk of contact with COVID-19 infected cases. Please read this information to help you decide if you want to participate in this research project. It is important that you understand this information. We encourage you to ask questions. Please take all the time you need to make your decision. You may also want to discuss this study with your family doctor, a family member or a close friend.

WHY IS THIS STUDY BEING DONE?

During the current COVID-19 pandemic, many healthcare workers are working in an environment which increases their probability of contracting this viral infection. Healthcare workers are more frequently infected than the rest of the population. Infected healthcare workers can infect their family, their patients, and their contacts. In addition to being withdrawn from work, they could have transmitted the disease to other colleagues, which further impedes our ability to deliver care to the population.

Vitamin D supplementation can decrease the risk of having the common cold, but it is not known if it could have an effect on the COVID-19 infection. Vitamin D is produced in our bodies from exposure to the sun and can be obtained from supplements and certain foods. However, many Canadians do not have an adequate intake of vitamin D throughout the year.

However, studies testing supplementation with other seemingly harmless vitamins, such as beta carotene and vitamin E, have shown unexpected important adverse reactions. Therefore, it is necessary to properly assess the benefits and the potential unexpected adverse reactions in the context of a clinical study.

This study will investigate whether a high-dose vitamin D supplementation could reduce the risk and severity of COVID-19 infection and work absence in healthcare workers.

HOW MANY PEOPLE WILL TAKE PART IN THIS STUDY?

We will be recruiting 2414 graduate healthcare workers, men and women, aged 18 to 69 years old, actively working and scheduled to continue working in a setting at high-risk of contact with people infected with COVID-19 and are working in high COVID incidence areas.

WHAT DOES THE STUDY INVOLVE?

If you agree to participate in this study, you will be assigned by chance to one of two groups. One group will receive one dose of 100,000 IU of vitamin D by mouth at the first visit and then take at home 1 pill of 10,000 IU of vitamin D once a week. The other group will receive a placebo dose at the first visit and then a placebo pill once a week. The vitamin D supplement is the active substance, meaning it could have an effect in the body. A placebo is an inactive substance, meaning it has no effect in the body. A placebo is used in clinical studies, such as this one, to ensure that observed changes are due to the active treatment and not to chance. You will have an equal chance of being assigned to each

group. The placebo and the vitamin D supplement look and taste exactly the same, so no one will know which treatment you are given, including the people involved in the study. In this informed consent form, we will use "Study supplement" to refer either to the vitamin D or the placebo.

This study should last 16 weeks and involves two visits. But prior to the first visit, we will conduct a screening/enrolment visit remotely by videoconferencing (or phone). If eligible and consenting, there will be a randomisation visit and an end-of-study visit, the latter two could be conducted in-person or remotely by videoconferencing, at your preference. Because of the pandemic, we wish to reduce the need and/or length of any in-person visit by doing as much as possible remotely.

Note that the study could finish earlier or be prolonged to 24 weeks, depending on the evolution of the pandemic. We ask that you don't change your usual diet or intake of vitamin supplements (if any) during the study.

Screening/Enrolment (pre-visit: about 45-60 minutes)

- We will review the eligibility questionnaire you have completed online, complete it with additional questions, explain the study in detail, and answer your questions.
- If eligible and consenting, you will be asked to sign the consent form, complete a few short study questionnaires and provide your contact information.
- We will ask you questions about your demographics (household, ethnicity), work-related activities and personal health (weight, height, skin color, smoking, medication, vitamins, supplements, health problems).
- To enable the creation of a medical and research pharmacy records, obtain information on COVID test, and ensure optimal contact with you throughout the study, we will ask personal information namely your RAMQ number, names (yours, your parents, your spouse), any drug allergy, your employee number (or practice number for physicians), your postal and email addresses, phone numbers and that of a next of kin and your preferred means to reach you.
- We could show you videos of key procedures (e.g. home blood collection) to help you chose your preferred type of randomisation visit.
- We will schedule the randomisation visit at a mutually convenient time and place given your choice of in-person or remote visit by videoconferencing. However, in case of a suspected prior COVID-19 infection, we would prefer you do an in-person visit to perform a rapid screening test for COVID-19 antibodies.

Randomisation visit (First visit: Week 0)

During the visit, which will last approximately an hour,

- If not already done, we will ask you to sign the consent forms, complete missing study questionnaires and your contact information.
- We will take a venous blood sample of about 15 mL (3 teaspoons) to measure the level of vitamin D, look for COVID-19 antibodies and to do an optional genetic analysis to examine a possible genetic predisposition to respond to vitamin D and to severity of COVID-19 symptoms.

- We would like to obtain a small drop of blood either from the venous puncture or via a finger-prick to look for COVID-19 antibodies in your blood using NADAL® COVID-19 IgG/IgM Rapid Test: if positive, you would not be eligible for this study. This test is not yet licensed for use in Canada and its use in this study is investigational. It has been selected for use prior to enrolment because it provides antibody results in 15 minutes. The results of this investigational test will be shared with you, acknowledging the risk of false positive or negative results. They will also be subsequently compared to the approved (Liaison IgG COVID-19) serology test.
- We will show you how a video on the TASSO home blood sampling kit at home for the last visit; if interested, we will show you how to use it, identify your sample, package it, and send it back to us (see below under First Remote visit). This device is not yet licensed for use in Canada and its use in this study is investigational. However, we have successfully pre-tested it and have validated the concordance between test results obtained with the TASSO and venous sampling.
- We will show you how to take a saliva sample by spitting into a tube. You will receive a pamphlet with instructions and could watch a video. You should not brush your teeth, eat, drink, smoke or chew gum for 30 minutes before spitting a small volume of saliva (2 mL). If you prefer to do an oro-nasopharyngeal sample, you would need to insert a swab (a small tube with a cotton tip) into the back of your mouth, then in one of your nostrils gently rotating the swab for about 5 seconds. We will ask you to take the saliva (or oro-nasopharyngeal) sample under our guidance. We will then show you how to identify it with our prepared labels, record the date and time, package it, and sent it back for analysis for COVID-19, and possibly other viruses and cells.
- You will be asked to take ten (10) pills of the Study supplement at this first visit only in front of the research personnel (in person or by videoconference). You will take home the bottle of Study supplement and be asked to take one (1) pill once a week until the end of the study.
- We will send you by text message or email as per your preference, a first reminder with a link to a questionnaire to confirm that you have received it and are able to complete and submit the brief questionnaire. The same approach will be used every week.
- We will give you all the other study materials including the saliva collection tube pre-printed labels, biohazard bags, insulated envelopes or boxes for shipment, prepaid courier waybills, and if you are interested in a **Remote end-of-study visit**, the TASSO home blood collection kit. It is possible that we ask you to use the NADAL® COVID-19 IgG/IgM Rapid Test at the last visit for validation purposes.

For participants choosing to have a **Remote First Visit**, in whom there is a suspicion that you may have had a prior undiagnosed COVID-19 infection in the past, we would prefer that you come for an in-person visit. Alternatively, we may send you first a finger-prick test kit to look for COVID-19 antibodies in your blood; if so, we would ask that you use it on your fingertip in front of us by videoconference. If positive, you would not be eligible for this study. If negative, the Study supplement and required materials would then be sent to the participant's home prior to the randomisation visit. We will ask you to take in front of us by videoconference, the Study supplement, the saliva sample, as well as the blood test and 2nd optional saliva sample (for genetic analysis).

- We will show you how to take a small sample (<1 mL) of capillary blood, using a blood collection kit specifically conceived for home collection, called TASSO-SST OnDemand. This device is not yet licensed for use in Canada and its use in this study is investigational. However, we have successfully pre-tested it and have validated the concordance between test results obtained with the TASSO and venous sampling. We will ask you to watch a short video and read the brochure explaining the procedure, then ask you to use it under our guidance. Briefly, you will need to warm the skin of your upper arm by rubbing it for about 45 seconds, disinfecting it, applying the little device on your arm, pressing on a button that will puncture a very small hole in the skin, then leave the device in place for about 5 minutes while blood flows slowly in a small tube. As only a small sample of blood can be obtained, it is very likely that we ask you to repeat this with a second kit. We will show you how to remove the small tube, close it with a small cap, identify the sample with our prepared labels, record the sampling date and time, package it, and prepare it to be sent for analysis for vitamin D and COVID-19 antibodies. We will ask your feedback on this type of blood collection method.
- If you wish to participate in the optional genetic analysis, we will ask you to collect 2 mL of saliva in another small tube (as the blood sample made by TASSO is not enough for this analysis), identify and date the sample with our prepared labels, and send it to us.

Between visits

- For the following weeks 0 to 16 (or later, should the study be extended up to 24 weeks) participants will take every week one (1) Study pill.
- You will receive a message via email or text message, according to your indicated preference, to remind you
  - Once a week to take the Study supplement
  - Once every two weeks to take the Study supplement, and fill out the brief electronic questionnaire (duration of 3-5 minutes) regarding your health and work status in the previous 2 weeks.
  - At any point in time if you have symptoms, to complete the daily symptoms diary (duration of 1-2 minutes) until 48 hours after the resolution of symptoms. If you have symptoms that should prompt testing for COVID-19, as listed on the cv19quebec.ca website, we will ask you to contact your health office for this purpose.
- If there is no response from you within a few days of sending the electronic questionnaire, we will contact you; if there is still no response from you within 7 days of us sending the electronic questionnaire, we will contact your next-of-kin indicated by you.

If you are infected during the study

If we obtain a positive COVID result from one of your saliva (or oro-nasopharyngeal) samples, you will be notified by a Study team member, by the preferred communication means you have indicated (phone, text message or email). As all positive results will be reported to the public health authorities you will be contacted as soon as possible by them for an assessment and instructions.

If you receive a positive COVID result from a test done outside this study (i.e., for clinical reasons), we ask that you inform us immediately and to indicate it in the follow-up questionnaire.

At the reception of a positive COVID test result,

- We will ask you to complete the daily diary of symptoms (duration of 1-2 minutes)
  - Until 48 hours after resolution of symptoms
  - Or if you remain asymptomatic, for a minimum of 14 days;
- If symptoms reappear, we will ask you to restart documenting them in the daily diary of symptoms
  - Until 48 hours after resolution of symptoms;
- If your symptoms continue beyond 14 days, we will ask you to complete the weekly diary of symptoms (duration of 1-2 minutes), once a week, until resolution of symptoms
- We will ask you to continue taking your weekly supplement and completing the follow-up questionnaire once every two weeks.

In case of an imminent vaccination against COVID-19

- If you expect to receive a vaccine against COVID-19 in the next few weeks, we will ask you to notify us immediately or via the questionnaire every two weeks.
  - We will rapidly organise a visit in person or remotely, before the scheduled date of the vaccination, to obtain a saliva sample to test for COVID-19 infection and a blood sample either in your vein (9 mL) or with the TASSO device at home to look for COVID-19 antibodies and level of vitamin D prior to the vaccination.
  - Just before, and about 1 month after, your second vaccine dose against COVID-19, we will ask you for another blood sample either in your vein (4.5 mL) or with the TASSO device at home to look for COVID-19 antibodies.
- We will ask you to continue
  - Once a week to take the Study supplement
  - Once every two weeks to fill out the brief electronic questionnaire (duration of 3-5 minutes) regarding your health and work status in the previous 2 weeks.
  - At any point in time if you have symptoms, to complete the daily symptoms diary (duration of 1-2 minutes) until 48 hours after the resolution of symptoms. If you have symptoms that should prompt testing for COVID-19, as listed on the cv19quebec.ca website, we will ask you to contact your health office for this purpose, whether you have been vaccinated or not.

End-of-study – Week 16 (or later if the study is prolonged)

At the end of the study, you will be invited to a last in-person or remote visit. This research visit will take approximately 30 minutes and involve the following:

- You will be asked to return the Study supplement bottle containing the unused pills.

- A venous (or capillary blood if done remotely) sample and, if you have not tested positive at COVID-19 before, a saliva (or oro-nasopharyngeal) sample will be collected.
- A rapid COVID-19 antibody test (NADAL®) may also be done on a blood drop (finger-prick or from the venous puncture) for validation purposes. If done remotely, this test may be done on blood sampled by finger-prick under our guidance by videoconference.
- You will complete the last few short study questionnaires and any missing information in the previous ones, if applicable.
- If conducted remotely, the samples, Study supplement bottle and unused material should also be shipped to the Coordinating Center.

Collecting information on COVID-19 tests made for clinical reasons

The results for a COVID-19 test performed for clinical reasons outside this study will be documented by you in the follow-up questionnaire (*faster means to inform us*) as well as in your institution's (Pandemic) or provincial database of COVID-19 cases, namely Trajectoire Santé publique (TSP) including all individuals who tested positive and all healthcare workers who tested positive under the supervision of the Ministère de la santé et des services sociaux (MSSS). If you are unable to answer the follow-up questionnaires, the information documented in these databases would ensure that we have complete information on the primary outcome of the study and thus allow us to determine accurately the impact of the intervention on the risk of infection with COVID-19.

Collecting information on healthcare services

The date, diagnosis, type of professional and of health care services which you have received during medical visits and hospitalisations will be obtained from the administrative databases of the Régie de l'assurance maladie du Québec (RAMQ) and Quebec hospital discharges (MED-ECHO). This will allow us to accurately determine the impact of the intervention on the severity of COVID-19 infection and other concomitant illnesses.

Collecting information of work absence

The number of days of work absence, overall, by type (i.e., holiday, illness, etc.), and specifically due to COVID-19, including absences due to an infection acquired at work or outside of work, preventive withdrawal due to pregnancy or other health conditions, awaiting test results/investigation, or other reason for quarantine will be collected from you via the follow-up questionnaire (*faster and most detailed means*), as well as from your institution's *Direction of Health Resources* or, if you are an attending physician, from the *Direction of professional services*. If you are unable to answer the follow-up questionnaires, the information documented in these databases would ensure that we accurately ascertain the impact of the intervention on work absences.

BIOBANK

For the purposes of this study, we will keep the biological samples collected (blood, saliva and/or oro-nasopharyngeal) in a biobank as well as the clinical and administrative data collected during the course of this study in order to complete the study's objectives, and to

conduct research on vitamin D, COVID-19 and its treatments and other related diseases. We would like to quantify specific cellular receptors which allow entry of COVID-19 into cells (for example, the angiotensin converting enzyme-ACE2) and inflammatory markers (such as the C-reactive protein). The collected samples will be kept in a biobank in the Research Center of CHU Sainte-Justine under the supervision of Dr Francine M. Ducharme. The samples will be kept as long as the research team can guarantee their proper management. Confidentiality of the identity of the samples will be guaranteed by assigning them a specific code. Your sample will not be identified by your name and cannot be used to identify you directly. After 5 years, the code key will be destroyed, and the samples will become completely anonymous. Your samples could possibly be shared with other researchers in other institutions. However, the access to data will only be allowed for approved projects by an independent research ethics board.

GENETIC ANALYSIS (optional)

Each person has their own set of unique genes or "genome". Genetic research aims to determine if there are genetic predispositions which make you more susceptible to a COVID-19 infection, to respond to vitamin D, to modulate disease severity and the interaction of these factors.

If you accept to participate in the genetic analysis, these analyses will be done on a small part (4 mL) of the venous blood sample provided during the first visit. If you decide to participate remotely, we will ask you to provide a saliva sample in a small tube.

We would like to sequence your entire genome and conduct gene expression analyses. We would also like to share your genetic data as well as other collected clinical data during the PROTECT study with the Canadian database Hostseq COVID-19 for use for COVID-19 related research and other aspects of human health. This biobank will serve as a centralized resource in Canada for COVID-19 research and other health-related studies. The data in the HostSeq database are under the supervision of CGen, a national Canadian platform financed by the federal government for sequencing and genome analysis. The principal investigators of the PROTECT study as well as the administrators of the Hostseq biobank COVID-19 will share your genetic and clinical information with other Canadian and international researchers whom are approved by CGen (the sponsor). The data could also be used for commercial use. However, your data will not be shared with until after an examination by a data access committee. This committee will verify that the use of the proposed research is in line with the objectives of the database HostSeq and that the research team which requests access has already been granted the required approval in accordance in terms of research ethics requirements. Approved researchers will sign agreements. These agreements will control how the data will be used. Individual results of any research conducted using your samples or any individual incidental findings will not be shared with you, as the research conducted on your data will have no individual diagnostic or therapeutic significance to you.

WHAT ARE THE BENEFITS AND RISKS OF THIS STUDY?

Benefits:

You may not benefit directly from the study intervention if it is not efficacious or if you have been assigned in the placebo group. However, the screening may identify earlier an active or past COVID-19 infection that was not apparent. Vour participation will help advance our knowledge on vitamin D and on the prevention of COVID-19 infection in healthcare workers and other individuals at risk of infection.

Each positive COVID-19 result from the saliva (or oro-nasopharyngeal) or blood sample will be shared with you according to your preferred way of communication: telephone, text message or email. All positive saliva (or oro-nasopharyngeal) results will also be shared by the Microbiology Laboratory of CHUM with the Public health authorities and will be added into your file at the CHUM and Dossier Santé Québec. No other research result will be provided to you. Research findings resulting from your participation to this study could potentially contribute to creating commercial products from which you would not be able to claim any financial benefit.

Risks:

• Related to study medication:

The vitamin D dose used in this study has been shown to be safe in adults. This dose is approved by Health Canada for the purpose of this study only, but not for clinical use yet. It is unlikely that you will have any side effects because of the amount of vitamin D used in this study as when combining the first and weekly doses, the total remains below the maximum amount allowed.

However, we will ask you to notify us immediately if you have any of the following, as they could be signs of an acute excess intake of vitamin D: mainly, a marked increase in thirst or an increase in the volume and frequency of urination (with or without fatigue, loss of appetite, nausea or vomiting, headaches, drowsiness, cardiac arrhythmias, constipation, muscle or bone or chest pain, mouth dryness or a metallic taste).

Later signs and symptoms that may indicate a chronic excess intake of vitamin D are: a marked increase in thirst, an increase in the volume and frequency of urination including during the night, loss of appetite, weight loss, red eye or conjunctivitis, inflammation of the pancreas, light sensitivity, runny nose, itching, fever, reduced libido, kidney stones, increased concentration of some analytes in the blood (BUN, AST, ALT, cholesterol), or in urine (albumin), ectopic calcification, hypertension, cardiac arrhythmias and rarely, a psychosis.

It's possible that other currently unknown risks are associated with Vitamin D intake.

One of the reasons we collect a blood sample is to measure the concentration of vitamin D in the blood at the start and end of the study. this will allow us to see if the vitamin D blood level is linked to the number and severity of COVID-19 confirmed cases.

• Related to study procedures:

The salivary collection sample is painless. If done, an oro-nasopharyngeal swab may cause slight discomfort during collection that will subside after its removal. The side effects of having blood collected by venous puncture or TASSO can include bleeding, bruising, discomfort and pain at the sample site. It is possible that the NADAL COVID-19 IgG/IgM Test may give false positive or false negative results. In case of divergence of results, we will communicate to you the results of the approved IgG test when available.

• Related to confidentiality:

There is always a small risk that your data could one day be re-identified. The genetic information is unique to each person, just as your fingerprint. This means that theoretically, you could be identified using your genetic code; however, this is not easy to do. Considering the advances in technology, there could be new ways to link you to data that we have not foreseen today, despite the strict confidentiality measures in place. Possible re-identification or unintentional disclosure of your genetic and clinical research data could lead to a loss in confidentiality and a possible future discrimination against yourself or your biological parents. But all security measures will be put in place to protect your privacy.

WHAT ARE THE COSTS OF TAKING PART IN THIS STUDY?

The Study supplements are provided free of charge by the manufacturer, Laboratoire RIVA.

WHAT ARE THE OTHER FINANCIAL ASPECTS?

For each completed visit (0 and 16 weeks), you will receive a $25 check by mail to compensate for your time. The check may arrive at your home between 4 and 8 weeks after the visit.

HOW IS PRIVACY INSURED?

During your participation in this research study, the investigators responsible for this study as well as the members of their research team will collect, in a research file, the required personal information to answer the scientific objectives of this research project.

These information could include your demographic data (name, sex, date of birth, ethnic origin, weight and height), your past and present health status, your health-related habits, medication you take, your work absences, and the results of all tests, exams, and procedures which you will participate in. Your personal file will include your address, email, telephone numbers, RAMQ number, and employee or practice number be kept in a separate file with restricted access; this information is required to create a medical and pharmacy file at the CHUM and for communication purposes during the study.

The coded blood, saliva (and/or oro-nasopharyngeal) samples will be sent to the biobank located at the Research Center of CHU Sainte-Justine under the supervision of Dr Francine M. Ducharme. The coded results of completed analyses will be kept on a protected server with restricted access at DACIMA company during the study, and thereby transferred to a secure server with restricted access in the Research Center of CHU Sainte-Justine under the supervision of Dr Francine M. Ducharme. During the study, the personal information

used to arrange virtual and in-person study visit appointments will be kept on a protected server with restricted access at the company providing the appointment-making software. Following the conclusion of the study, this information of yours will be transferred to a secure server with restricted access in the Research Center of CHU Sainte-Justine under the supervision of Dr Francine M. Ducharme. The database of HostSeq will be kept on secure cloud servers (online) that are based in Canada and will be indefinitely kept or until they are not useful for research.

To ensure your privacy, a copy of the consent form as well as the results to the diagnostic tests required for conducting the research project, will be copied in the research and medical file of the CHUM. Therefore, each person or company which you authorize to consult your medical file, will have access to this information.

The research data will be kept for at least 25 years by the principle investigator. The data collected could be published or discussed during scientific meetings, but it would not be possible to identify you.

All collected information will remain confidential within the limits provided by law. You will only be identified by a code number. The key to the code linking your name to your research file will be kept by the investigator responsible for this research project.

To ensure your safety, a copy of the consent form as well as the results of the diagnostic tests required for research purposes will be placed in the research file and the medical file of the CHUM. Consequently, any person or company to whom you give access to your medical file will have access to this information.

Research data will be kept for at least 25 years by the investigator responsible for this research project. Research data may be published or be the subject of scientific discussion, but it will not be possible to identify you.

For the purposes of surveillance, control, safety and marketing of the Study drug, your research as well as your medical files could be consulted by a person mandated by a regulatory organization, in Canada or elsewhere, such as Health Canada, as well as sponsor representatives of the company manufacturing the vitamin D pills for this project (Laboratoire RIVA), the institution or research ethics committee. These people and organizations adhere to a strict confidentiality agreement.

You have the right to consult your research file to verify the collected data and to correct them, if needed. Moreover, access to certain information before the end of the study could mean your removal from this study in order to maintain the study's integrity.

IS YOUR PARTICIPATION VOLUNTARY?

Yes. Taking part in this study is voluntary. You may choose not to be in this study. You can decide to stop being in the study at any time, without needing to provide any reason, but simply informing the research team.

Your decision to refuse participation or to stop participating in the study at a later time, will have no effect on the quality of care or services to which you are entitled or on your relationship with the people that provide them.

The principal investigators of this study, the research ethics board, the funding agency or the sponsor could decide to end your participation in the study without your consent. This could happen if there are new information or findings that indicate your participation is no longer in the best of your interests, or if you have not been following the study instructions as explained, or if there are other administrative-related reasons to stop the project.

If you stop participating in the study or if you have been removed from it, the collected information and material already received will be kept (as well as the data pertaining to healthcare services and work absences will continue to be collected) and analysed to ensure the validity of this project, unless you specifically ask for them to be destroyed. If this is the case, these data and/or material will be removed from the biobank provided that the code key (linking between nominal data and the study code) is still available, that is, up to 5 years after the end of the study.

If you decide to drop out of the HostSeq database, your data will no longer be shared, and no new data will be collected. The data already in the HostSeq database will be destroyed once informed about this decision. However, it could be impossible to remove the results once they have been compiled with the results of other participants or if they have been published. Moreover, if certain data have been shared with other researchers, it could be possible not to be able to remove this part of the data. In such a case of unsuccessful withdrawal from the study, your identity will always be protected.

All new information acquired during the course of the study which could have an impact on your decision to continue participation will be shared with you rapidly, which is the reason why we would like to keep your personal information and have your approval to communicate with you after the end of the study (optional).

WHOM DO I CALL IF I HAVE QUESTIONS OR PROBLEMS?

If you have any questions about the research project or if you have any problems that you believe are related to your participation in the project, you can call the researchers responsible for the project:

- Dr. Francine M. Ducharme at 514 345 4931, extension 4398
- Dr. Cecile Tremblay at 514 890-8000, extension 14645

If you would like information about your rights related to your participation in the research, you may contact the Ombudsman - complaints and quality services of the CHU Sainte-Justine at 514 345-4749, of the CHUM at 514 890-8484 or your CISSS/CIUSSS:

- CIUSSS de l'Est-de-l'Île-de-Montréal : 514 252-3510
- CIUSSS de l'Ouest-de-l'Île-de-Montréal : 514-989-1885, extension 1010
- CIUSSS du Centre-Sud-de-l'Île-de-Montréal : 514 593-3600
- CISSS de la Montérégie-Est : 450-468-8447
- CISSS de la Montérégie-Centre : 450-466-5434

RESEARCH ETHICS COMMITTEE

The Research Ethics Board of CHU Sainte-Justine has approved this study and will continue to monitor it for all participating institutions of the Quebec Health and Social Services network.

LIABILITY

This research is not funded by a private industry. In case of side effects resulting from the study medication or from procedures required for this research project, you will receive all necessary medical care covered by the Quebec's provincial health insurance plan (RAMQ) or by your private drug insurance plan. You will be responsible for paying the portion of any costs not covered.

CONSENT FORM

Research project title: PRevention of COVID-19 with Oral Vitamin D supplemental Therapy in Essential healthCare Teams (PROTECT)

The nature and procedures of this research project were explained to me. I have read the information and consent forms and I kept a copy, or a copy has been provided to me. I was able to ask my questions and they were answered to my satisfaction. After consideration, I agree to participate in this research project.

I authorize the research team to consult the collected data about me in the COVID infection database (Pandemic) of my institution and/or the provincial TSP database, the medical and hospitalisation services database (RAMQ and MED-ECHO), and the workplace absenteeism database (Human Resources Directorate or Professional Services Directorate) to obtain information that is pertinent to this project.

By agreeing to participate in this study, you are not waiving any of my rights under the law. You are not releasing the investigators from their legal and professional liability.

Name of participant (Print)	Signature	Date

1. I consent to the analysis of gene expression and the sequencing of the whole genome of my coded biological material (blood, saliva, and/or oro-nasopharyngeal). The whole genome sequence could be hosted in the Canadian HostSeq COVID-19 biobank and linked to a database containing the viral genome. This would serve to explore any genetic predisposition to COVID-19, the severity of the disease and response to vaccine.

❒ Yes _____ (Initials) ❒ No_____ (Initials)

2. I consent to prolonging the access to my coded data on healthcare use, COVID-19 infections and work absenteeism for 12 months following the study end date, to explore the long-term impact of COVID-19 infection and vaccination.

❒ Yes _____ (Initials) ❒ No_____ (Initials)

3. I consent to being contacted to update my personal information, obtain additional information about my health or to be invited to participate in new research.

❒ Yes _____ (Initials) ❒ No_____ (Initials)

4. In case I receive a vaccine against COVID-19 during the study, I agree to do the blood samples before the first and second vaccine dose as well as 1 month after the 2nd vaccine dose, even if these samples were to be done after the end-of-study's visit planned at week 16 (or 24).

❒ Yes _____ (Initials) ❒ No_____ (Initials)

Participant's signature: _____

I have explained the research study and the terms of this information and consent form to the research participant, and I answered all his/her questions. I explained that participation in a research project is free and voluntary and could be stopped at any time they choose.

Name of person obtaining consent (Print)	Signature	Date

(FOR THE CHUM PARTICIPANTS ONLY)

COMMITMENT OF THE PRINCIPAL INVESTIGATOR AT THE CHUM

I certify that this information and consent form was explained to the research participant, and that the questions the participant had were answered.

I undertake, together with the research team, to respect what was agreed upon in the information and consent form, and to give a signed and dated copy of this form to the research participant.┼

Name (Print)	Signature of the principal investigator at the CHUM	Date

BMJ Open

Prevention of COVID-19 with oral vitamin D supplemental therapy in essential healthcare teams (PROTECT): protocol for a multicentre, triple-blind, randomized, placebo-controlled trial

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Primary Subject Heading:	Nutrition and metabolism
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Keywords:	COVID-19, PREVENTIVE MEDICINE, Clinical trials < THERAPEUTICS

Prevention of COVID-19 with oral vitamin D supplemental therapy in essential healthcare teams (PROTECT): protocol for a multicentre, triple-blind, randomized, placebo-controlled trial

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ABSTRACT

Introduction: In the COVID-19 pandemic, health care workers (HCWs) were at high-risk of infection due to their exposure to COVID infections. HCWs were the backbone of our health care response to this pandemic; every health care worker withdrawn or lost due to infection had a substantial impact on our capacity to deliver care. Primary prevention was a key approach to reduce infection. Vitamin D insufficiency is highly prevalent in Canadians and worldwide. Vitamin D supplementation has been shown to significantly decrease the risk of respiratory infections. Whether this risk reduction would apply to COVID-19 infections remained to be determined. This study aimed to determine the impact of high-dose vitamin D supplementation on incidence of laboratory-confirmed COVID19 infection rate and severity in HCWs working in high COVID incidence areas.

Methods and analysis: PROTECT was a triple-blind, placebo-controlled, parallel-group multicentre trial of vitamin D supplementation in HCWs. Participants were randomly allocated in a 1:1 ratio in variable block size to intervention (one oral loading dose of 100,000 IU vitamin D3 + 10000 IU weekly vitamin D3) or control (identical placebo loading dose + weekly placebo). The primary outcome was the incidence of laboratory-confirmed COVID-19 infection, documented by RT-qPCR on salivary (or nasopharyngeal) specimens obtained for screening or diagnostic purposes, as well as self-obtained salivary specimens and COVID-19 seroconversion at endpoint. Secondary outcomes included disease severity; duration of COVID-19 related symptoms; COVID-19 seroconversion documented at endpoint; duration of work absenteeism; duration of unemployment support; and adverse health events. The trial was terminated prematurely, due to recruitment difficulty.

Ethics and dissemination: This study was approved by the Research Ethics Board of the CHU Sainte-Justine as the central committee for participating institutions ((MP-21-2021-3044). Participants provided written informed consent. Results are being disseminated to the medical community via national/international conferences and publications in peer-reviewed journals.

Trial registration: NCT04483635.

Strengths and limitations of this study

- This trial was designed as a hybrid study enabling partially or totally remote screening, randomisation, follow-up, as well as outcome documentation by use of home capillary blood and saliva sampling, visits conducted by videoconference, monitoring by electronic reminders and questionnaires, and communication by phone, text messaging or emails.
- The trial used a pragmatic subject selection and easily applicable intervention to maximise subsequent implementation in practice and selected a primary outcome, the risk of laboratory-confirmed infection, that would likely change practice.
- A single loading dose followed by regular doses have been shown to lead to rapid and sustained increase in serum level of 25-hydroxy-vitamin D and ensure adequate group separation, both properties desired in the context of a rapidly expanding epidemic while facilitating adherence in exhausted frontline health workers.
- Given the uncertainty in the progression of the infection rate, the use of a Bayesian adaptive design allowed for adaptations (early stopping or prolongation of duration of follow-up) at the interim analysis according to the projection of infection rates.
- Although the trial aimed for high-intensity recruitment, the delay in setting up a remote study in the context of the pandemic, combined with high use of vitamin D and successful vaccination program in health care workers, resulted in severe recruitment difficulty and early stopping of the trial for futility.

Introduction

The Coronavirus 2 (SARS-CoV-2) disease (COVID-19) outbreak has rapidly expanded to a global pandemic. Healthcare workers (HCWs) played a crucial role in the fight against the COVID-19 pandemic. It was therefore a public health priority to develop strategies to decrease the risk and severity of COVID-19 in this vulnerable population. Indeed, there was rising concern as HCW were overrepresented in terms of infections (3.8% of infected individuals in Wuhan, China,[1] 10% in Italy and 12% in Spain, 10-20% in US)[2] [3] and perhaps severity. Working in long-term care facilities (LTCF) and with aerosol generating medical procedures (e.g., hospitals) further increased the risk (Odds Ratio [OR]: 2.3).[4] The risk of reporting COVID-19 infection in front-line HCWs, defined as those in direct contact with patients, was 10-fold greater than the general population at the beginning of the pandemic (Hazard Ratio [HR]= 11.61). [5] Recent research also indicated that HCWs who were Blacks, Asians, or other minority ethnic populations, had a higher likelihood of contracting COVID-19.[5] Compared to those unexposed to COVID-19 patients, the risk was two to five-fold higher in HCWs exposed to suspected (HR= 2.39) or confirmed (HR= 4.83) COVID-19 cases, even with adequate personal protection equipment (PPE).[5] Although infections may have been due to contact with infected patients, community-, or family-acquired disease, cases were rapidly emerging from cross-infection with asymptomatic infected HCW.

Vitamin D is an immunomodulatory micronutrient, and its levels in the body may vary due to diet and environmental conditions. Vitamin D insufficiency had been associated with increased risk of respiratory infections, and possibly COVID-19,[6] asthma exacerbations, and acute respiratory distress syndrome (ARDS) among others.[7-9] Optimal pro-immune and anti-inflammatory impacts likely occur at 25-hydroxyvitamin D (25OHD) levels above 75 nmol/L (30 ng/mL).[10,11] In a systematic review of 25 randomized controlled trials (RCT) of 11321 individuals, daily/weekly vitamin D supplementation decreased by 19% the rate of acute

respiratory infections (two-step analysis; OR 0.81, 95% CI 0.72 to 0.91),[12,13] with a stronger effect in subjects with baseline 25OHD <25 nmol/L. Whereas subgroup analyses suggested a protective effect, primary in individuals receiving daily or weekly vitamin D supplement, and not in those with bolus,[14] other important differences in population (e.g., malnutrition),[15,16] age (infant),[16] chronic disease (e.g. asthma, COPD)[17-21] and type of infection (e.g. bacterial)[15,16] could have contributed to the apparent lesser effect. Of interest, Vitamin D supplementation significantly reduced the rate of severe exacerbations (i.e., requiring rescue systemic corticosteroids), a condition association with airway inflammation, with no impact according to bolus use or not. [14] Vitamin D supplementation was also found to be associated with a decreased load of rhinovirus (common cold), consistent with an increased antiviral immune response.[22] A systematic review and several studies reported an inverse association between serum vitamin D levels and COVID-19 severity, in-patient mortality, as well as serum levels of C-reactive protein (CRP) and lymphocyte percentage.[23,24] These findings suggested that vitamin D status was linked with the severity and mortality of the COVID-19 infection in the general population, particularly in severe COVID-19 cases. Whether Vitamin D could have prevented or lessened infection and/or the inflammatory response associated with the COVID-19 remained to be explored.[25] At the time of funding (June 2020) and study initiation (February 2021), no other primary prevention trials were published. Since then, one positive and two negative trials testing different vitamin D intervention as primary prevention were published. [26-28]

The vitamin D receptor is expressed on innate and adaptive immune cells which also synthesize the active metabolite 1,25-hydroxyvitamin D_3 ($1,25(OH)_2D_3$); thus, vitamin D could strengthen innate and adaptive cellular immunity by increasing local production of antimicrobial peptides,

decreasing secretion of pro-inflammatory cytokines, inhibiting dendritic cell activation, suppressing T helper cell type 1 response, and promoting T regulatory cells induction. These cellular effects are crucial for host responses against infection and can reduce the survival and replication of respiratory viruses.[13,24] $1,25(OH)_2D_3$ is also produced locally in bronchial epithelial cells and downregulates inflammatory cytokines (e.g. interleukin-8) and chemokines (e.g. leucocyte attracting CXCL10) expression from stimulated cells.[29]

The protocol of a placebo-controlled parallel-group triple-blind RCT to explore the impact of vitamin D_3 supplementation on reducing the risk and severity of laboratory-confirmed COVID19 infection in HCWs is described herein, as per Standard Protocol Items: Recommendation for Intervention Trials guidelines (**Supplementary file 1**). After funding, but prior to the start of recruitment, the protocol underwent four amendments (8 protocol versions) in view of the rapidly evolving science, multiple challenges faced with conducting a large scale COVID-19 trial of high-risk health-care workers during the pandemic, including difficulty in obtaining large-scale supplies, as well as favorable pilot results of two novel technologies (**Table 1**). These original and final (1.8, January 18, 2021) protocol versions are described below. The trial was initiated but stopped prematurely due to recruitment difficulty.

Objectives

The primary research question was whether one oral dose of 100,000 IU vitamin D_3 (administered at baseline) plus weekly supplement of 10,000 IU vitamin D_3 could decrease the risk of laboratory-confirmed COVID-19 infection, versus placebo, in frontline HCWs in high COVID-19 incidence areas.

Additionally, the study aimed to examine if, compared with placebo, the vitamin D intervention reduced: (i) illness severity, (ii) symptom duration, (iii) work absenteeism, and (iv) unemployment among frontline health care workers (HCW) in high COVID-19 incidence areas. This study was to also assess various exploratory outcomes.

Hypothesis

We hypothesised that compared to placebo, vitamin D supplementation would decrease the incidence of laboratory-confirmed symptomatic COVID-19 infection by 20% in frontline HCWs working in high COVID-19 incidence area.

Table 1. Study amendments and notifications

	Clinical trial Application (CTA)				Investigational Testing Authorization (ITA)	
Version number	**Changes**	**Description**	**Submitted**	**Approved**	**Submitted**	**Approval**
Version 0.0 11-05-2020						
Version 1.0 23-08-2020	• Eligibility • Outcomes & Covariates	• Strengthening of exclusion of 'suspected or previously undocumented COVID-19 infection' by adding: (i) a questionnaire of symptoms elaborated by Menni et al. and (ii) a rapid (15-minute) serology test, not yet approved nor tested in Canada, to be pre-tested in a pilot study. • Addition of capillary blood self-collection with Tasso-SST device (to be pre-tested in a pilot study).	23-08-2020	16-09-2020	N/A	N/A
Amendment 1 **Version 1.1** 23-10-2020	• Eligibility • Exploratory Outcomes • Main outcome	• Clarification of the NADAL® COVID-19 IgG/IgM Rapid Test (Teracero Pharma Inc., Lachine Canada) on venous whole blood as the rapid serology test to exclude prior infection (following pilot comparative study) • Validation of Nadal serology test compared to IgG SARS-CoV2 serology as endpoint exploratory outcome • Salivary COVID-19 RT-PCR test method prioritized over nasopharyngeal samples for twice-monthly self-collection or accepted for clinical diagnostic by qPRC	23-10-2020 (CTA-A) †	02-11-2020	23-10-2020 (NADAL)	14-111-2020
Amendment 2 Versions 1.2-1.4 **Version 1.5** 27-11-2020	• Primary Outcome • Outcomes	• Removal of q2 weeks saliva sample for COVID-19 rt-PCR analysis due to supply problem with 50 mL Falcon centrifuge tube caused by a global plastics shortage combined with un-acceptable delay for public tender for a contract with courier service for q2weeks biological samples • Addition of questions and procedures to account for the possible effect of the Vitamin D supplementation on immune response to COVID-19 vaccine, including a saliva sampling for COVID-19 by RT-PCR and a blood test for serology (and vitamin D) prior to vaccination	27-11-2020 (CTA-A) †	30-11-2020	23-11-2020 (TASSO & NADAL)	2-12-2020

	Clinical trial Application (CTA)				Investigational Testing Authorization (ITA)	
Version number	**Changes**	**Description**	**Submitted**	**Approved**	**Submitted**	**Approval**
	• Covariates & Outcome (Device)	• Specification of the TASSO SST on Demand (Tasso Inc, Seattle, USA) as choice for capillary blood self-sampling (following pilot comparative study)				
Amendment 3 versions 1.6 & **Version 1.7** 12-12-2020	• Eligibility • Exploratory Outcomes	• Exclusion of health care workers who have received the COVID-19 vaccine prior to enrolment • Addition of (i) effect of high-dose vitamin D on SARS-CoV-2 IgG titers before & after 2nd dose of COVID-19 vaccine and (ii) the long-term infection rate up to 12 months after end-of-study • Modifying exploratory outcome to allow exploration of modulating effect of vitamin D, not only on the risk of COVID-19 infection but also on response to vaccine	12-12-2020 (CTA-A) †	16-12-2020	12-12-2020 (TASSO & NADAL)	23-12-2020
Amendment 4 Version 1.8 18-01-2021	• Exploratory Outcomes • Eligibility	• Clarification that the serology to be done just prior the second dose of a COVID-19 vaccine may not always be at 3 or 4 weeks (as recommended by vaccine manufacturer) to reflect the recent governmental decision to delay the timing of the 2nd vaccine dose to 12 to 16 weeks • Slightly modifying wording to target healthcare workers at risk of contact with infected individuals that were not suspected of being infected (e.g., patients, colleagues, students, etc.)	18-01-2021 (CTA-N) ‡	N/A	18-01-2020 ¶	01-02-2021

† CTA-A, Clinical Trial Application -Amendment
‡ CTA-N, Clinical Trial Application -Notification
¶ As there is no 'notification' category for the Investigational Testing Authorization (ITA), each amendment or notification to the Clinical Trial Application (CTA) must be submitted as a new amendment for the devices to be reviewed by ITA.

Methods and analysis

Study design

PROTECT was a pragmatic 16-week, triple-blind, placebo-controlled, parallel-group, randomized trial comparing supplemental vitamin D3 and placebo in HCWs with the possibility of extending the study follow-up up to 24 weeks, depending on infection rate progression during an interim analysis (**Figure 1**).

Participants

HCWs (i.e., physicians, allied health care workers, orderlies, etc.) were eligible if they: (i) were aged ≥18 and <70 years old; (ii) were authorized to practice in Quebec; (iii) were working or scheduled to work over the next 16 weeks in a setting at high-risk of contact with COVID-19 infected individuals, particularly (but not only) those involved with aerosol generating medical procedures in hospitals and/or caring for patients in long-term care facilities; (iv) were working in high COVID incidence areas in the greater Montreal area and surroundings; (v) were covered by the provincial universal public health insurance (*Régie de l'assurance-maladie du Québec* [RAMQ] for medical services and hospitalisations; (vi) had a personal email or phone (to which to send reminders and questionnaire by email or text messages); (vii) had a fixed address (to which to send the material) in the greater Montreal or surrounding areas.

HCWs were excluded if they met any of the following criteria: vitamin D supplementation (cholecalciferol or calcitriol) intake >400 IU/day (or >12,000 IU/month) in past 3 months; intention to take >400 IU per day during the study period; suspected or previously documented COVID-19 infection; history of nephrolithiasis, hypercalcemia, hyperphosphatemia, hyperparathyroidism, granulomatosis disease (e.g., tuberculosis, sarcoidosis), renal failure, or

active cancer; current intake of medications that may cause hypercalcemia such as lithium, teriparatide, or digoxin; anticipated prolonged absence from work during the study period (i.e., pregnancy); anticipated difficult follow-up; enrolment in a concurrent interventional randomized trial; have already received the vaccine against COVID-19. Participation in this trial did not preclude subsequent enrolment in a COVID-19 therapeutic (but not preventive) trial, which would be documented.

Study intervention

Participants in the intervention group received 100,000 IU vitamin D_3 (cholecalciferol) at randomization followed by a weekly dose of 10,000 IU vitamin D_3 for 16 weeks. Participants in the control groups received an identical placebo bolus followed by placebo weekly supplement for 16 weeks. Sufficient supply was provided for 24 weeks, in case of prolongation study based on the interim analysis. Participants in both groups were asked to take the study intervention with their most copious meal. Treatment of co-morbidities were permitted. Vitamin D intake up to 400 IU per day was allowed.

Randomization

Randomization was implemented using a computer-generated random list stratified by one of 11 workplaces (*Centre Hospitalier Universitaire* [CHU]) or health region (*Centre Intégré Universitaire de Santé et Services Sociaux* [CIUSSS] or *Centre Intégré de Santé et Services Sociaux* [CISSS]). HCWs were allocated (1:1) using permuted block randomization to enhance concealment. Group allocation codes for each stratum was held in a secure location with restricted access by the Central Pharmacy and Data Management.

Patient and public involvement

Participant burden of research measures was assessed using feedback from patients participating in one pilot round. Patients were not involved in study design, recruitment of participants or conduct of the study.

Outcomes

Primary outcome

The original primary outcome, incidence of laboratory-confirmed COVID-19 infection, was originally based on (i) bi-monthly self-obtained mid-turbinate nasopharyngeal (NP) swabs, complemented by (ii) NP swabs obtained clinically for screening or diagnostic purposes throughout the study, both analysed by RT-qPRC approved by Health Canada. Faced with the unexpected cancellation of our large order of Falcon tubes to collect saliva sample for qPRC, combined with the unacceptable additional delay for a public tender to securing a contract with a private courier service, and in view of the uniform protocol for screening symptomatic or COVID-19 exposed health care workers throughout the Province of Quebec and the reliability of IgG serology, we decided to forgo the twice-monthly saliva sampling for qPRC analysis. The revised definition of the primary outcome became the incidence of laboratory-confirmed COVID-19 infection, documented by RT-qPCR based on salivary (or nasopharyngeal) specimens (i) obtained for screening or diagnostic purposes throughout the study and (ii) self-obtained salivary specimens obtained at endpoint as well as (iii) COVID-19 IgG seroconversion at endpoint (in COVID-unvaccinated individuals: ≥15 UA on the anti-S SARS-CoV-2 IgG Diasorin on Liaison XL

platform; in COVID-vaccinated individuals : ≥1.40 index (S/C) on the anti-N SARS-CoV-2 IgG on ARCHITECT platform)

Secondary outcomes

(i) *Distribution of disease severity* on a 5-category ordinal scale [asymptomatic; mild (managed at home); moderate (hospitalisation without supplemental oxygen); severe (oxygen supplementation); critical (mechanical ventilation/death)], (self-reported, RAMQ); (ii) *Duration of COVID-19 positivity* between 1st COVID+ to first COVID- test) revised to *Duration of COVID-19 related symptoms* in individuals with laboratory confirmation of COVID infection, (self-reported on diary); (iii) COVID-19 IgG seroconversion documented at endpoint (see above); (iv) duration of work absenteeism (self-reported, medical records or human resources databases); (iv) duration of unemployment support (human resource databases); (v) Adverse health events (self-reported). Several *exploratory outcomes* pertained to the: incidence of post-acute and chronic symptoms; long-term (1-year) morbidity and work absence related to COVID-19; change in gene expression of ACE2 and TMPRSS2 in saliva cells; change in inflammatory markers (i.e., CRP), immune response post vaccination; other viral infections; and genetic markers (including changes in gene expression).

Study procedures

To facilitate the recruitment of participants, this study was conceived as hybrid trial enabling partially or totally remote trial participation including screening, randomization, follow-up, and end-of-study visit.

Pre-screening

Advertisements were placed in health institutions, newspapers, social media and online, where participants were invited to complete an online pre-screening form, read and download the consent forms; and if eligible and interested, to book a virtual screening appointment (via a secured videoconferencing platform) with research team who would confirm eligibility, explain the study, obtain informed consent, and schedule a virtual or in-person randomization visit.

Screening

At the virtual screening visit by videoconferencing, research coordinators completed with the individual a more extensive eligibility questionnaire, which included additional questions about: anticipated work exposure over the next 16 weeks to COVID-infected or suspected individuals and to high-risk medical procedures; work place (*Centre Hospitalier Universitaire* [CHU]) or Centre Hospitalier Universitaire Sainte-Justine) or health region (CIUSSS or CISSS), serving as randomization stratum; prior laboratory-confirmed or physician-suspected COVID-19 infection; assessment of the likelihood of prior/current, yet undocumented, COVID-19 infection using the 5-item questionnaire developed by Menni et al[30] (score >0.50 interpreted as high likelihood of prior infection); and finally, the comfort level with the study design and procedures, including saliva and capillary blood sampling self-collection demonstrated by instructional videos. Eligible and consenting individuals electronically signed an online consent form (with the signed PDF consent form automatically emailed to participants). Then, two additional questionnaires were completed on line with the research coordinators namely: (i) the baseline questionnaire collecting information about household, ethnicity, part- vs. full-time work, personal health, skin color (measured with the Fitzpatrick scale),[31] concomitant medications or supplements, and (ii) the

nominative CRF collecting demographic information essential to opening a medical and pharmaceutical research record (i.e., public health insurance number, allergies) and maintaining contact with the research team throughout the study (preferred means to receive electronic reminders/questionnaires and to be notified of positive test results; address to receive study material or for biological sample pick-up; and next-of-kin contact in case of inability to respond to questionnaire due to illness), and to document work absence (employee number).

Finally, at the screening visit, the participant was asked to choose an appointment for a *virtual* (via a secured videoconferencing platform) or *in-person* randomization visit at one of several locations. To help select their preferred visit format, videos of key procedures (such as home blood collection) were shown. Only in participants with a significant likelihood of a current or past undocumented (Menni score > 0.5) was an *in-person* randomization visit mandatory to receive the rapid COVID-19 serology test, prior to randomisation.

Preparation and shipment of study drug by research pharmacy

The list of new participants approved by one of the PIs was sent daily by email to the CHUM research team to be open a medical chart and send an electronically signed prescription for the Study medication, to the Research Pharmacy for preparation of study drug.

Prior to randomization, a list of all consenting and eligible participants was automatically sent every night to the one of the co-principal investigators (FMD or CT) who reviewed screening and baseline questionnaires to approve or refuse study entry and electronically signed their decision. The daily list of new approved participants was sent electronically daily to the CHUM research

team. Medical and pharmaceutical records were opened and an electronically signed prescription for the Study medication sent to the Research Pharmacy for preparation of study drug for a given target date.

To enable remote randomization, the randomization took place about one week prior to the randomization visit to allow enough time for the preparation and shipment of patient-specific study supplement to the research team and, in turn, the shipment of the Study supplement and all materials required for the randomization visit by the research team to the participant.

Randomization visit

Seventy-two and 24 hours prior to, and at, the randomisation visit, participants were screened by questionnaire for recent travel, symptoms suggestive of SARS-Cov2 infection, or exposure to COVID-19 infected individuals. Those who responded positively were asked to get tested, notify their institutional health service, and await end of quarantine and/or confirmed negative test to reschedule the randomisation visit.

Randomization visit (week 0) was performed *in person* (60 minutes) or *remotely* (90 minutes), depending on the availability and preference of participants as well as their likelihood of a past COVID-19 infection.

In-person visits were conducted—by appointment only—in designated rooms with restricted access. The research coordinators wore personal protection equipment (PPE); all procedures, from participant arrival to departure, were approved by the institutional Infection Control and Safety committee. The *in-person* visit entailed (i) ascertainment of the signed consent form, (ii) capillary

blood sample collection to perform NADAL® COVID-19 IgG/IgM Rapid Test (Teracero Pharma Inc., Lachine Canada), (iii) venous blood sample collection for baseline serum 25(OH)D and SARS-CoV-2 IgG serology analyses and if genetic consent, DNA; (iv) viewing of the saliva collection video and instruction pamphlet, (v) collection of the first specimen under supervision, (vi) a final verification of the eligibility and exclusion criteria; (vii) randomization; (viii) oral administration of 100,000 IU vitamin D_3 or an identical placebo, and (ix) distribution of the study material including study supplement, saliva sampling kit for end-of-study, biohazard and sampling bag, and, if a remote visit was anticipated at endpoint, capillary blood collection kits (TASSO OnDemand SST device). Any patient with a positive NADAL COVID-19 IgM/IgG Rapid Test serology test were excluded prior to randomisation.

The *remote* randomization visit, conducted by video-conference, was similar to the *in-person* randomization visit with the following additions: (a) viewing of the capillary blood collection video and instructional pamphlet; (b) remote capillary sampling under guidance using the TASSO-SST device (TASSO Inc., Seattle, USA); (c) viewing of the saliva collection video and instructional pamphlet (OG-600 Oragene DNA Collection Kit, DNA Genotek Inc., Ottawa, Canada); (d) remote DNA salivary sampling under guidance; (e) preparation of biological samples for shipment with phase change and insulated envelopes under guidance and (f) organising collection of biological specimens by approved courier service to respective laboratories. Note that a Nadal serology test was not conducted remotely.

Follow-up

Participants received *weekly electronic (SMS or email) reminders* to take their weekly Study Supplement (10,000 IU vitamin D or an identical placebo) and to start completing an *online daily diary* if they tested positive to SARS-CoV2 or they developed symptoms suggestive of COVID-19 infection.

Every two weeks, participants received a link to complete a *brief online questionnaire* asking to report: their adherence to weekly Study Supplement intake; health status including recent COVID-19 related exposure, symptoms, or testing; adverse health events or new co-morbidities; change in concomitant medications or supplement intake; work status (active duty, quarantined, holiday, sick) and work setting (ED, ICU, etc.); as well as expected/recent COVID-19 vaccination (date and vaccine name) if any; the latter question served to enable timely shipment of materials for additional sampling prior to vaccination, as COVID-19 vaccination was permitted during the study. In participants who planned to get vaccinated during the study, three additional blood, and one additional saliva, samplings, either *on-site* or *remotely*, were planned including: saliva (for COVID-19 qPCR analysis) and blood (for SARS-CoV-2 anti-S IgG serology) sampled prior to vaccination, a blood sample (for SARS-CoV-2 anti-S and anti-N IgG serology) collected just prior to the second vaccine dose, and a blood sample (for SARS-CoV-2 anti-S and anti-N IgG serology) collected one month after second vaccine dose and endpoint. Regardless of their vaccination status, participants were asked to continue taking the weekly Study Supplement and complete the bi-monthly questionnaire until the end of the study. If questionnaires were not completed within 2 days of the target date, the research coordinator reached out the participant to complete the information.

End-of-study visit

An end-of-study visit was conducted either in-person (60 minutes) or remotely (90 minutes), depending on the availability and preference of participants and likelihood of a current COVID-19 infection. The *in-person end-of-study visit* entailed the collection of a (i) venous blood sample for serum 25(OH)D and SARS-CoV-2 anti-S IgG serological results and in vaccinated participants a SARS-CoV-2 anti-N IgG serology, (ii) capillary blood sample to perform the NADAL® COVID-19 IgG/IgM Rapid Test, (iii) a saliva sample for SARS-CoV-2 qPCR analysis as well as guessing of allocation and return of the study supplement bottle to assess adherence and any unused material.

The *remote end-of-study visit* conducted via videoconference entailed the same procedures as the in-person end-of-study visit with one exception: the self-collection of a capillary (instead of venous) blood using TASSO-SST devices (for the serum 25(OH)D and SARS-CoV-2 anti-S with/without anti-N serology). Individuals were guided into self-performing the pinprick capillary method to perform the NADAL® COVID-19 IgG/IgM Rapid Test and return of biological samples and materials by pre-paid approved courier.

Covariates

Several covariates that could act as confounders or interaction variables in the magnitude of effect associated with the intervention were documented, namely: baseline serum 25OHD level; smoking; concomitant supplements or drug(s) that can alter calcium or vitamin D absorption or metabolism such as diuretics and anti-epileptics (reported at baseline and every 2 weeks); skin color (documented at baseline); obesity (documented by height & weight [BMI] at baseline); other comorbidities (i.e., diabetes, hypertension, etc.) that may affect the severity of COVID-19

infection and receipt of a COVID-19 vaccine (documented by verbal report bi-monthly). All external (governmental and institutional) databases were to be obtained 3 months before, and up to 16 months following, randomization (as well as 12 months after then study endpoint).

During an event

During COVID-19 related symptoms or documented SARS-CoV infection, participants were instructed to complete a daily symptom diary from date of onset of symptoms or positive test, until two days with no symptoms or 14 days if asymptomatic,

Risk management

Clinical and biochemical adverse health events (AHEs) were monitored throughout the study and reported for all patients at the end of the study. No specific laboratory safety monitoring was planned given the established safety of the loading dose of 100,000 IU and weekly dose of 10,000 IU.[32,33] Adverse Health Events (AHE) were recorded via electronic questionnaires throughout the study. Participant who reported symptoms suggestive of vitamin D intoxication had a venous blood sampling (total and ionised calcium, phosphorus, alkaline phosphatase, albumin, and creatinine). Any abnormal laboratory value was interpreted as 'clinically significant' or 'not clinically significant' by the Site endocrinologist blinded to study allocation. Further investigation or action for individual participants (including interruption, cessation, or unblinding of the study drug via pharmacy or by analysis of serum 25OHD) was determined by the Site endocrinologist, if indicated to ensure participant safety. The AHE's occurrence was reviewed periodically by the Data and Safety Monitoring Board (DSMB). Code breaking was allowed only if deemed essential

for participant management. If relevant, summary reports aggregating (or not if requested) both groups were to be provided to the DSMB.

Data management and monitoring

The principal investigator (FMD) and statistical group (SG, RP) oversaw randomization, data management, progress monitoring, and all analyses, including those for Data Monitoring Safety Board (DMSB). The DSMB membership included: Lehana Thabane, biostatistican (Chair), Gary Kobinger, infectious disease specialist, Kevin Thorpe, biostatistician, and Edgar Delvin, biochemist & expert in Vitamin D. DACIMA was used for online data entry and management.

A combination of *remote* monitoring activities and *in-person* routine monitoring visits were conducted by an independent Study Monitor with the first randomised participants at each site and on a bi-monthly basis, to ensure that each Site adhered to the study protocol, Good Clinical Practice guidelines, and data collection completeness.

Sample size calculation

Given uncertainties in infection progression, a Bayesian adaptive design was used where the posterior probability of effectiveness, i.e., P(OR<1|data) was the basis of inference and decision making.[34] Assuming an expected OR of 0.80 and 1:1 treatment allocation, a total net sample size of 2100 was required to identify a 20% reduction in the risk of COVID-19 in the vitamin D vs. control group, with 80% power with the design described above. Considering a drop-out rate of 15%, 2414 participants were targeted. An interim analysis was planned when 75% of participants would have reached week 12, at which time the following assessments were to be

made: the *progression over time in the incidence of infection* (slope of the curve of infection) was to be updated and if the probability of effectiveness exceeded 0.95 [p(OR<1)>0.95], the trial would have been terminated for efficacy at the interim point (12 weeks); otherwise, the study would have continued to 16-week follow-up. Simulation results showed that, with the net sample size of 2100 (assuming an expected OR of 0.80 and 1:1 treatment allocation), there was about a 55% chance that the trial would be terminated for efficacy at the interim analysis.[35] The overall infection rate was monitored on a monthly basis: note that the study could have been extended to 24 weeks based on the progress of the infection rate, if required.

Statistical analysis

Primary outcome

An intention-to-treat (ITT) analysis was to be carried out with all randomized participants. For the primary outcome, the posterior distribution of the odds ratio of COVID-19 infection (OR) was the basis of inference in interim and final analyses. The posterior distribution of the OR was to be estimated by drawing samples from the posterior risks under each arm, which could be obtained analytically in a Beta-binomial model. Flat prior distributions were assumed for the risks (Beta(1,1)). Posterior 95% credible intervals were to be reported as interval estimates for the OR. Crude analyses as well as analyses adjusted for important covariates (i.e., potential confounders, effect modification, and baseline group imbalances) where to be conducted. Subgroup analyses would be conducted on baseline 25OHD, age, sex, BMI, occupational risk, and COVID-19 vaccination. A stratified analysis on geographical infection rate would be explored; sensitivity analysis censoring to date of COVID-19 vaccination, would be conducted if applicable.

Secondary outcomes

Distribution of disease severity defined as a 5-level ordinal outcome would have been examined with a Bayesian analysis using a proportional odds (PO) model; the posterior probability of OR would have been obtained by Markov chain Monte Carlo sampling implemented in Stan.[34] Duration of symptoms, duration of workday absences and of unemployment would have been examined by a zero-inflated Poisson distribution.

Ethics and dissemination

This study was reviewed and approved by the research ethics board (REB) of the CHU Sainte-Justine, serving as the central REB of all participating institutions (MP-21-2021-3044). A non-objection letter (NOL) from Health Canada had been obtained to use high-dose Vitamin D loading dose as well as the Tasso OnDemand device for home blood sampling and the NADAL COVID-19 IgM/IgG Rapid serology test. Written informed consent for study participation, for biobanking specimens for ancillary studies, and for subsequent publication of results was obtained from all participants, with the knowledge that participation was voluntary and could be withdrawn at any time with no effect on their current/future medical care. As part of the informed consent, enrolees had the option to participate in the HostSeq COVID-19 Canadian biobank conducted under the supervision of CGen, a national Canadian platform for sequencing and genome analysis (**Supplementary file 2**). In Canada, health care is provided to those who suffer harm from trial participation.

All protocol amendments were submitted to Health Canada, investigators, and REB; if these changes implied a revision of consent forms, ongoing trial participants were informed of new modifications to provide informed consent. All information obtained during the study were and

would continue to be kept confidential as per the law. Data was collected directly by electronic data capture on Dacima Clinical Suite (DACIMA Software Inc., Montreal, Canada). Data safety and confidentiality was upheld at all data collection stages by assigning a unique subject ID to each participant, with data and samples kept under lock and key, electronic password protection and access restricted to study personnel. Samples collected during the study were labelled with the unique research code, prior to transfer and storage at the CHUSJ biobank, with access restricted to authorised personnel.

This trial used pragmatic patient (irrespective of baseline 25OHD level) and intervention to attempt to maximise subsequent implementation into practice. If the intervention had been shown to be effective in reducing infection and morbidity, this approach would have been readily implementable and could have markedly influenced practice during the COVID-19 pandemic. No participant identifiers were used in the dissemination of this research. Health care professionals serving as partners were informed the study design and pre-tested all questionnaires.

Results are being disseminated to the medical community via national/international conferences and publications in peer-reviewed journals.

Trial status, challenges, and discussion

The study was conducted as per version 1.8 (January 18, 2021). The recruitment started on February 9, 2021. Upon the DSMB recommendation, recruitment was stopped prematurely on March 18, 2021, after 34 participants were enrolled, due to the inability to recruit approximatively 200 participants/week required to meet the target sample size of 2415 participants. The DSMB advised that the continuation of the trial, as originally designed, would not be able to answer the research question and recommended that recruitment be stopped for futility. Recruitment

difficulties were attributed in part to the high use of vitamin D and high concurrent vaccination rate among our target population, healthcare workers, the first targeted to be vaccinated from January 2021 onwards. Based on the recommendations of the study's endocrinologist, a premature end of follow-up after a minimum of 4 weeks from randomization was deemed sufficient to monitor the safety of the intervention in all participants. The timeframe was deemed sufficient to ensure participant safety while learning for the study, that is, transforming the PROTECT study into a pilot study to document the impact of the Study intervention on the rise in Vitamin D serum level, participants' adherence the Study intervention and procedures in the context of a hybrid study, etc. The last end-of-visit was conducted on May 4, 2022.

Potential redirections of the study were discussed. The first option was to change the main outcome for an immunogenicity study in the general adult population. However, after strong consideration of the amount of changes to be made to the protocol and related documents (standards of procedures, case report forms, participant' instructions and notification, etc.), the expected delay in obtaining approval by all regulatory and ethical authorities, the impossible logistic of recruiting participants after the same duration of exposure to the Study intervention prior to their vaccination, combined with the government of Quebec announcement that all willing adults would be vaccinated by June 24, 2021, the research team judged that it would unfeasible to perform a scientific solid and feasible trial on immunogenicity if one could not control the timing of immunization, combined with the expected very short recruitment timeframe.

A second option that received very strong consideration was to replicate the PROTECT trial in children aged 9 years and over. Again, after considering changes to be made to the protocol and

related documents, the expected delay for obtaining approval by all regulatory and ethical authorities including school boards, combined with the Pfizer-BioNtech announcement that their vaccine was not only 100% successful for preventing COVID-19 infection in adolescents aged 12 to 15 years, but that they forecast vaccinating teenagers in time for the September 2021 school entry, the PI judge that it was unrealistic to aim for the large recruitment target within such a short timeframe.

The protocol was submitted for publication after the last patient end-of-study visit, due to the incredible amount of work done to set-up and initiate this large hybrid trial. Of note, the latter included two pilot studies testing two experimental devices to enable partially or totally remote participation, in the context of the pandemic which also imposed large protocol and space restrictions for recruiting on-site potentially COVID-19 infected health care workers, several protocol amendments to facilitate and adjust the trial in the context of emerging science and anticipated vaccination campaign and their impact of all electronic documents, manual of procedures, and regulatory approvals, coupled with the premature end-of-follow-up in enrolled participants.

With the gained experience and knowledge, it is crucial that a future trial must begin fast prior to widespread vaccination and in populations where infection rate is high.[28] Permitting study entry to individuals with prior infection and prior vaccination (given common reinfections and temporary vaccine protection) [36] could have been considered, but it would have significantly reduced the event rate, required prolongation beyond 24 weeks (and additional funding), and compromised study power as was noted in other primary prevention trials.[26,27] Restricting

eligibility to patients with vitamin D deficiency (<25 nmol/l) would have severely interfered with recruitment ability in population-based or health care workers studies. [26-28] Use of calcifediol (25-hydroxy-vitamine D or (25OHD) may have been associated with more potency and rapid rise in serum 25OHD than expected for cholecalciferol[37] (Vitamin D3) although the choice is debated[38] and rapid access to study drug and matching placebo remain a crucial challenge at the onset of a pandemic. Revisiting the intervention dose and frequency of administration in light of the latest literature on SARS-CoV2 and related virus could be considered, although current evidence suggest that, with similar doses, high-incidence population may be more important than dosing in primary prevention [26,28] and high doses are effective in tertiary prevention.[35] Of interest, we have demonstrated that the intervention significantly rose 25OHD levels well above 75 nmol/L, that is, in the hypothesised range for optimal pro-immune and anti-inflammatory impact.[39] A pragmatic design with fewer outcomes and monitoring via administrative databases appeared theoretically more efficient, but required rapid access to data when interim analyses are planned to monitor event rate; any delayed in data access could raise serious challenges and hamper trial decisions. Pursuing a hybrid approach to facilitate enrolment in the context of a pandemic was feasible, although electronic self-screening and outcome monitoring required a lot of programming that have contributed to implementation delays.

The publication of this protocol is meant to share our experience, including conducting a hybrid (virtual and/or in-person) trial and lessons learned, to enable serve as template to accelerate protocol writing and its improvement in the context of another epidemic/pandemic, and to serve as reference for the publication of our pilot studies that enabled this trial, and lessons learned from this experience. As Vitamin D supplementation has shown a benefit as tertiary prevention in severe

564 COVID-19 cases, with insufficient data to conclude its impact as primary and secondary
565 prevention, testing this approach remains worthy to test. [40]
566

Contributors

FMD designed the study protocol, secured funding, and oversaw the overall conduct of the project. CLT contributed to the protocol and amendments, directed the study implementation at the CHUM, and coordinated the prescription of study drug, pharmacy dispensation, as well as salivary sample reception and interpretation. SG conceived the statistical approach and sample size calculation and along with RWP, oversaw randomization and statistical analysis. CL, JHW and CQ contributed to the study design and amendments. BH wrote the first manuscript draft. LGSM oversaw the safety assessment. DC is responsible for the work absenteeism analysis. All co-authors approved the manuscript. Authorship eligibility on resulting manuscripts will follow standard guidelines.

Competing interests

The authors have no competing interests.

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Data availability statement

After publication of the primary results, datasets used and analyzed during the current study will be made available by the corresponding author on reasonable request.

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Figure legend

Figure 1. Study outline

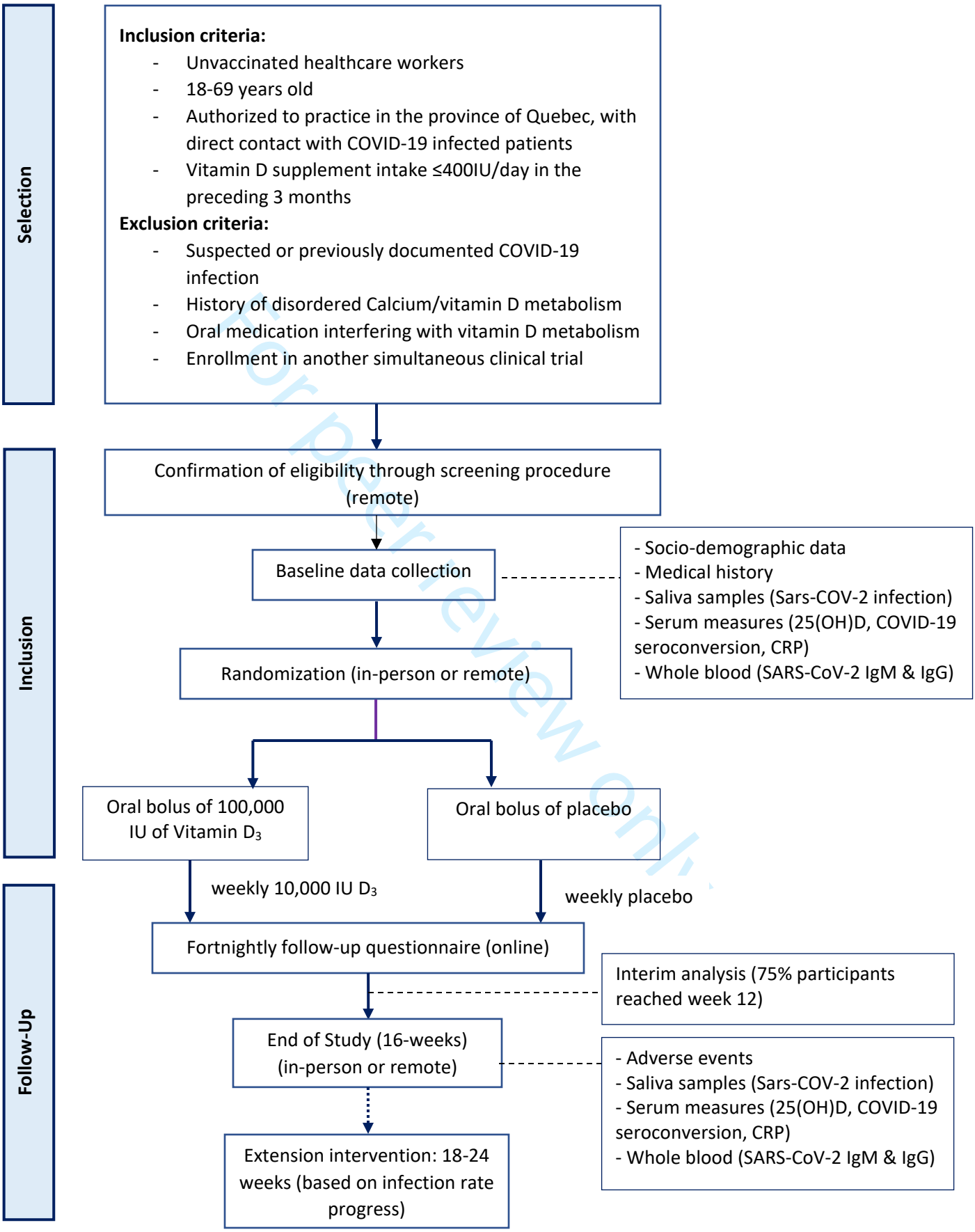

Figure 1

SPIRIT 2013 Checklist: Recommended items to address in a clinical trial protocol and related documents*

Section/item	ItemNo	Description	Addressed on page number
Administrative information			
Title	1	Descriptive title identifying the study design, population, interventions, and, if applicable, trial acronym	1
Trial registration	2a	Trial identifier and registry name. If not yet registered, name of intended registry	3
	2b	All items from the World Health Organization Trial Registration Data Set	**N/A**
Protocol version	3	Date and version identifier	8 (line 138)
Funding	4	Sources and types of financial, material, and other support	1, 25, supplemental funding file
Roles and responsibilities	5a	Names, affiliations, and roles of protocol contributors	1, 26-27
	5b	Name and contact information for the trial sponsor	1, 26
	5c	Role of study sponsor and funders, if any, in study design; collection, management, analysis, and interpretation of data; writing of the report; and the decision to submit the report for publication, including whether they will have ultimate authority over any of these activities	26

	5d	Composition, roles, and responsibilities of the coordinating centre, steering committee, endpoint adjudication committee, data management team, and other individuals or groups overseeing the trial, if applicable (see Item 21a for data monitoring committee)	19, 20, 26,27
Introduction			
Background and rationale	6a	Description of research question and justification for undertaking the trial, including summary of relevant studies (published and unpublished) examining benefits and harms for each intervention	6, 7
	6b	Explanation for choice of comparators	6, 7
Objectives	7	Specific objectives or hypotheses	8, 9
Trial design	8	Description of trial design including type of trial (e.g., parallel group, crossover, factorial, single group), allocation ratio, and framework (e.g., superiority, equivalence, noninferiority, exploratory)	9, 10
Methods: Participants, interventions, and outcomes			
Study setting	9	Description of study settings (e.g., community clinic, academic hospital) and list of countries where data will be collected. Reference to where list of study sites can be obtained	13

Eligibility criteria	10	Inclusion and exclusion criteria for participants. If applicable, eligibility criteria for study centres and individuals who will perform the interventions (eg, surgeons, psychotherapists)	9, 10
Interventions	11a	Interventions for each group with sufficient detail to allow replication, including how and when they will be administered	10, 11
	11b	Criteria for discontinuing or modifying allocated interventions for a given trial participant (eg, drug dose change in response to harms, participant request, or improving/worsening disease)	10
	11c	Strategies to improve adherence to intervention protocols, and any procedures for monitoring adherence (eg, drug tablet return, laboratory tests)	17, 18
	11d	Relevant concomitant care and interventions that are permitted or prohibited during the trial	10
Outcomes	12	Primary, secondary, and other outcomes, including the specific measurement variable (eg, systolic blood pressure), analysis metric (eg, change from baseline, final value, time to event), method of aggregation (eg, median, proportion), and time point for each outcome. Explanation of the clinical relevance of chosen efficacy and harm outcomes is strongly recommended	11, 12

Participant timeline	13	Time schedule of enrolment, interventions (including any run-ins and washouts), assessments, and visits for participants. A schematic diagram is highly recommended (see Figure)	15, 16, **17, 18, Figure** 1
Sample size	14	Estimated number of participants needed to achieve study objectives and how it was determined, including clinical and statistical assumptions supporting any sample size calculations	20-21
Recruitment	15	Strategies for achieving adequate participant enrolment to reach target sample size	12,13
Methods: Assignment of interventions (for controlled trials)			
Allocation:			
Sequence generation	16a	Method of generating the allocation sequence (eg, computer-generated random numbers), and list of any factors for stratification. To reduce predictability of a random sequence, details of any planned restriction (eg, blocking) should be provided in a separate document that is unavailable to those who enrol participants or assign interventions	11
Allocation concealment mechanism	16b	Mechanism of implementing the allocation sequence (eg, central telephone; sequentially numbered, opaque, sealed envelopes), describing any steps to conceal the sequence until interventions are assigned	11

Implementation	16c	Who will generate the allocation sequence, who will enrol participants, and who will assign participants to interventions	11
Blinding (masking)	17a	Who will be blinded after assignment to interventions (eg, trial participants, care providers, outcome assessors, data analysts), and how	5, 19
	17b	If blinded, circumstances under which unblinding is permissible, and procedure for revealing a participant's allocated intervention during the trial	19
Methods: Data collection, management, and analysis			
Data collection methods	18a	Plans for assessment and collection of outcome, baseline, and other trial data, including any related processes to promote data quality (eg, duplicate measurements, training of assessors) and a description of study instruments (eg, questionnaires, laboratory tests) along with their reliability and validity, if known. Reference to where data collection forms can be found, if not in the protocol	12 to 19
	18b	Plans to promote participant retention and complete follow-up, including list of any outcome data to be collected for participants who discontinue or deviate from intervention protocols	12 to 19

Data management	19	Plans for data entry, coding, security, and storage, including any related processes to promote data quality (eg, double data entry; range checks for data values). Reference to where details of data management procedures can be found, if not in the protocol	20
Statistical methods	20a	Statistical methods for analysing primary and secondary outcomes. Reference to where other details of the statistical analysis plan can be found, if not in the protocol	21, 22
	20b	Methods for any additional analyses (eg, subgroup and adjusted analyses)	22
	20c	Definition of analysis population relating to protocol non-adherence (eg, as randomised analysis), and any statistical methods to handle missing data (eg, multiple imputation)	21-22
Methods: Monitoring			
Data monitoring	21a	Composition of data monitoring committee (DMC); summary of its role and reporting structure; statement of whether it is independent from the sponsor and competing interests; and reference to where further details about its charter can be found, if not in the protocol. Alternatively, an explanation of why a DMC is not needed	20,

	21b	Description of any interim analyses and stopping guidelines, including who will have access to these interim results and make the final decision to terminate the trial	20
Harms	22	Plans for collecting, assessing, reporting, and managing solicited and spontaneously reported adverse events and other unintended effects of trial interventions or trial conduct	17, 19, 20, 23
Auditing	23	Frequency and procedures for auditing trial conduct, if any, and whether the process will be independent from investigators and the sponsor	20
Ethics and dissemination			
Research ethics approval	24	Plans for seeking research ethics committee/institutional review board (REC/IRB) approval	25
Protocol amendments	25	Plans for communicating important protocol modifications (eg, changes to eligibility criteria, outcomes, analyses) to relevant parties (eg, investigators, REC/IRBs, trial participants, trial registries, journals, regulators)	8
Consent or assent	26a	Who will obtain informed consent or assent from potential trial participants or authorised surrogates, and how (see Item 32)	13
	26b	Additional consent provisions for collection and use of participant data and biological specimens in ancillary studies, if applicable	N/A

A	27	How personal information about potential and enrolled participants will be collected, shared, and maintained in order to protect confidentiality before, during, and after the trial	14-15, 25
Declaration of interests	28	Financial and other competing interests for principal investigators for the overall trial and each study site	27
Access to data	29	Statement of who will have access to the final trial dataset, and disclosure of contractual agreements that limit such access for investigators	27
Ancillary and post-trial care	30	Provisions, if any, for ancillary and post-trial care, and for compensation to those who suffer harm from trial participation	25
Dissemination policy	31a	Plans for investigators and sponsor to communicate trial results to participants, healthcare professionals, the public, and other relevant groups (eg, via publication, reporting in results databases, or other data sharing arrangements), including any publication restrictions	23
	31b	Authorship eligibility guidelines and any intended use of professional writers	25
	31c	Plans, if any, for granting public access to the full protocol, participant-level dataset, and statistical code	27

Appendices

Informed consent materials	32	Model consent form and other related documentation given to participants and authorised surrogates	Supplementary file 2
Biological specimens	33	Plans for collection, laboratory evaluation, and storage of biological specimens for genetic or molecular analysis in the current trial and for future use in ancillary studies, if applicable	Supplementary file 2

*It is strongly recommended that this checklist be read in conjunction with the SPIRIT 2013 Explanation & Elaboration for important clarification on the items. Amendments to the protocol should be tracked and dated. The SPIRIT checklist is copyrighted by the SPIRIT Group under the Creative Commons "Attribution-NonCommercial-NoDerivs 3.0 Unported" license.

CHU Sainte-Justine
Le centre hospitalier universitaire mère-enfant

Université de Montréal

APPROUVÉ PAR LE COMITÉ D'ÉTHIQUE
27 JANV. 2021
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CHU SAINTE-JUSTINE

CHUM

Centre intégré universitaire de santé et de services sociaux de l'Ouest-de-l'Île-de-Montréal Québec

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Centre intégré universitaire de santé et de services sociaux du Centre-Sud-de-l'Île-de-Montréal Québec

INFORMATION AND CONSENT FORM

Project Title: PRevention of COVID-19 with Oral Vitamin D supplemental Therapy in Essential healthCare Teams (PROTECT)

Protocol Number: PROTECT 2020

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- **Principal investigator at the CHUM:** Dr. Cecile Tremblay, MD, FRCPC, Microbiologist/Infectiologist, Centre hospitalier universitaire de Montréal (CHUM)

Co-investigators:

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- Shirin Golchi, biostatistician, McGill university
- Cristina Longo, epidemiologist, University of Montreal
- Robert Platt, biostatistician, McGill University
- Caroline Quach, MD, Paediatrician microbiologist, CHUSJ
- Christian Renaud, pediatric microbiologist, CHUSJ
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CHUM project number : 20.319

WHY ARE YOU BEING INVITED TO TAKE PART IN THIS STUDY?

Today, we are inviting you to participate in this research study because you are a healthcare worker who is working in a high COVID-19 incidence area and in a setting with a high risk of contact with COVID-19 infected cases. Please read this information to help you decide if you want to participate in this research project. It is important that you understand this information. We encourage you to ask questions. Please take all the time you need to make your decision. You may also want to discuss this study with your family doctor, a family member or a close friend.

WHY IS THIS STUDY BEING DONE?

During the current COVID-19 pandemic, many healthcare workers are working in an environment which increases their probability of contracting this viral infection. Healthcare workers are more frequently infected than the rest of the population. Infected healthcare workers can infect their family, their patients, and their contacts. In addition to being withdrawn from work, they could have transmitted the disease to other colleagues, which further impedes our ability to deliver care to the population.

Vitamin D supplementation can decrease the risk of having the common cold, but it is not known if it could have an effect on the COVID-19 infection. Vitamin D is produced in our bodies from exposure to the sun and can be obtained from supplements and certain foods. However, many Canadians do not have an adequate intake of vitamin D throughout the year.

However, studies testing supplementation with other seemingly harmless vitamins, such as beta carotene and vitamin E, have shown unexpected important adverse reactions. Therefore, it is necessary to properly assess the benefits and the potential unexpected adverse reactions in the context of a clinical study.

This study will investigate whether a high-dose vitamin D supplementation could reduce the risk and severity of COVID-19 infection and work absence in healthcare workers.

HOW MANY PEOPLE WILL TAKE PART IN THIS STUDY?

We will be recruiting 2414 graduate healthcare workers, men and women, aged 18 to 69 years old, actively working and scheduled to continue working in a setting at high-risk of contact with people infected with COVID-19 and are working in high COVID incidence areas.

WHAT DOES THE STUDY INVOLVE?

If you agree to participate in this study, you will be assigned by chance to one of two groups. One group will receive one dose of 100,000 IU of vitamin D by mouth at the first visit and then take at home 1 pill of 10,000 IU of vitamin D once a week. The other group will receive a placebo dose at the first visit and then a placebo pill once a week. The vitamin D supplement is the active substance, meaning it could have an effect in the body. A placebo is an inactive substance, meaning it has no effect in the body. A placebo is used in clinical studies, such as this one, to ensure that observed changes are due to the active treatment and not to chance. You will have an equal chance of being assigned to each

group. The placebo and the vitamin D supplement look and taste exactly the same, so no one will know which treatment you are given, including the people involved in the study. In this informed consent form, we will use "Study supplement" to refer either to the vitamin D or the placebo.

This study should last 16 weeks and involves two visits. But prior to the first visit, we will conduct a screening/enrolment visit remotely by videoconferencing (or phone). If eligible and consenting, there will be a randomisation visit and an end-of-study visit, the latter two could be conducted in-person or remotely by videoconferencing, at your preference. Because of the pandemic, we wish to reduce the need and/or length of any in-person visit by doing as much as possible remotely.

Note that the study could finish earlier or be prolonged to 24 weeks, depending on the evolution of the pandemic. We ask that you don't change your usual diet or intake of vitamin supplements (if any) during the study.

Screening/Enrolment (pre-visit: about 45-60 minutes)

- We will review the eligibility questionnaire you have completed online, complete it with additional questions, explain the study in detail, and answer your questions.
- If eligible and consenting, you will be asked to sign the consent form, complete a few short study questionnaires and provide your contact information.
- We will ask you questions about your demographics (household, ethnicity), work-related activities and personal health (weight, height, skin color, smoking, medication, vitamins, supplements, health problems).
- To enable the creation of a medical and research pharmacy records, obtain information on COVID test, and ensure optimal contact with you throughout the study, we will ask personal information namely your RAMQ number, names (yours, your parents, your spouse), any drug allergy, your employee number (or practice number for physicians), your postal and email addresses, phone numbers and that of a next of kin and your preferred means to reach you.
- We could show you videos of key procedures (e.g. home blood collection) to help you chose your preferred type of randomisation visit.
- We will schedule the randomisation visit at a mutually convenient time and place given your choice of in-person or remote visit by videoconferencing. However, in case of a suspected prior COVID-19 infection, we would prefer you do an in-person visit to perform a rapid screening test for COVID-19 antibodies.

Randomisation visit (First visit: Week 0)

During the visit, which will last approximately an hour,

- If not already done, we will ask you to sign the consent forms, complete missing study questionnaires and your contact information.
- We will take a venous blood sample of about 15 mL (3 teaspoons) to measure the level of vitamin D, look for COVID-19 antibodies and to do an optional genetic analysis to examine a possible genetic predisposition to respond to vitamin D and to severity of COVID-19 symptoms.

- We would like to obtain a small drop of blood either from the venous puncture or via a finger-prick to look for COVID-19 antibodies in your blood using NADAL® COVID-19 IgG/IgM Rapid Test: if positive, you would not be eligible for this study. This test is not yet licensed for use in Canada and its use in this study is investigational. It has been selected for use prior to enrolment because it provides antibody results in 15 minutes. The results of this investigational test will be shared with you, acknowledging the risk of false positive or negative results. They will also be subsequently compared to the approved (Liaison IgG COVID-19) serology test.
- We will show you how a video on the TASSO home blood sampling kit at home for the last visit; if interested, we will show you how to use it, identify your sample, package it, and send it back to us (see below under First Remote visit). This device is not yet licensed for use in Canada and its use in this study is investigational. However, we have successfully pre-tested it and have validated the concordance between test results obtained with the TASSO and venous sampling.
- We will show you how to take a saliva sample by spitting into a tube. You will receive a pamphlet with instructions and could watch a video. You should not brush your teeth, eat, drink, smoke or chew gum for 30 minutes before spitting a small volume of saliva (2 mL). If you prefer to do an oro-nasopharyngeal sample, you would need to insert a swab (a small tube with a cotton tip) into the back of your mouth, then in one of your nostrils gently rotating the swab for about 5 seconds. We will ask you to take the saliva (or oro-nasopharyngeal) sample under our guidance. We will then show you how to identify it with our prepared labels, record the date and time, package it, and sent it back for analysis for COVID-19, and possibly other viruses and cells.
- You will be asked to take ten (10) pills of the Study supplement at this first visit only in front of the research personnel (in person or by videoconference). You will take home the bottle of Study supplement and be asked to take one (1) pill once a week until the end of the study.
- We will send you by text message or email as per your preference, a first reminder with a link to a questionnaire to confirm that you have received it and are able to complete and submit the brief questionnaire. The same approach will be used every week.
- We will give you all the other study materials including the saliva collection tube pre-printed labels, biohazard bags, insulated envelopes or boxes for shipment, prepaid courier waybills, and if you are interested in a **Remote end-of-study visit**, the TASSO home blood collection kit. It is possible that we ask you to use the NADAL® COVID-19 IgG/IgM Rapid Test at the last visit for validation purposes.

For participants choosing to have a **Remote First Visit**, in whom there is a suspicion that you may have had a prior undiagnosed COVID-19 infection in the past, we would prefer that you come for an in-person visit. Alternatively, we may send you first a finger-prick test kit to look for COVID-19 antibodies in your blood; if so, we would ask that you use it on your fingertip in front of us by videoconference. If positive, you would not be eligible for this study. If negative, the Study supplement and required materials would then be sent to the participant's home prior to the randomisation visit. We will ask you to take in front of us by videoconference, the Study supplement, the saliva sample, as well as the blood test and 2nd optional saliva sample (for genetic analysis).

- We will show you how to take a small sample (<1 mL) of capillary blood, using a blood collection kit specifically conceived for home collection, called TASSO-SST OnDemand. This device is not yet licensed for use in Canada and its use in this study is investigational. However, we have successfully pre-tested it and have validated the concordance between test results obtained with the TASSO and venous sampling. We will ask you to watch a short video and read the brochure explaining the procedure, then ask you to use it under our guidance. Briefly, you will need to warm the skin of your upper arm by rubbing it for about 45 seconds, disinfecting it, applying the little device on your arm, pressing on a button that will puncture a very small hole in the skin, then leave the device in place for about 5 minutes while blood flows slowly in a small tube. As only a small sample of blood can be obtained, it is very likely that we ask you to repeat this with a second kit. We will show you how to remove the small tube, close it with a small cap, identify the sample with our prepared labels, record the sampling date and time, package it, and prepare it to be sent for analysis for vitamin D and COVID-19 antibodies. We will ask your feedback on this type of blood collection method.
- If you wish to participate in the optional genetic analysis, we will ask you to collect 2 mL of saliva in another small tube (as the blood sample made by TASSO is not enough for this analysis), identify and date the sample with our prepared labels, and send it to us.

Between visits

- For the following weeks 0 to 16 (or later, should the study be extended up to 24 weeks) participants will take every week one (1) Study pill.
- You will receive a message via email or text message, according to your indicated preference, to remind you
  - Once a week to take the Study supplement
  - Once every two weeks to take the Study supplement, and fill out the brief electronic questionnaire (duration of 3-5 minutes) regarding your health and work status in the previous 2 weeks.
  - At any point in time if you have symptoms, to complete the daily symptoms diary (duration of 1-2 minutes) until 48 hours after the resolution of symptoms. If you have symptoms that should prompt testing for COVID-19, as listed on the cv19quebec.ca website, we will ask you to contact your health office for this purpose.
- If there is no response from you within a few days of sending the electronic questionnaire, we will contact you; if there is still no response from you within 7 days of us sending the electronic questionnaire, we will contact your next-of-kin indicated by you.

If you are infected during the study

If we obtain a positive COVID result from one of your saliva (or oro-nasopharyngeal) samples, you will be notified by a Study team member, by the preferred communication means you have indicated (phone, text message or email). As all positive results will be reported to the public health authorities you will be contacted as soon as possible by them for an assessment and instructions.

If you receive a positive COVID result from a test done outside this study (i.e., for clinical reasons), we ask that you inform us immediately and to indicate it in the follow-up questionnaire.

At the reception of a positive COVID test result,

- We will ask you to complete the daily diary of symptoms (duration of 1-2 minutes)
  - Until 48 hours after resolution of symptoms
  - Or if you remain asymptomatic, for a minimum of 14 days;
- If symptoms reappear, we will ask you to restart documenting them in the daily diary of symptoms
  - Until 48 hours after resolution of symptoms;
- If your symptoms continue beyond 14 days, we will ask you to complete the weekly diary of symptoms (duration of 1-2 minutes), once a week, until resolution of symptoms
- We will ask you to continue taking your weekly supplement and completing the follow-up questionnaire once every two weeks.

In case of an imminent vaccination against COVID-19

- If you expect to receive a vaccine against COVID-19 in the next few weeks, we will ask you to notify us immediately or via the questionnaire every two weeks.
  - We will rapidly organise a visit in person or remotely, before the scheduled date of the vaccination, to obtain a saliva sample to test for COVID-19 infection and a blood sample either in your vein (9 mL) or with the TASSO device at home to look for COVID-19 antibodies and level of vitamin D prior to the vaccination.
  - Just before, and about 1 month after, your second vaccine dose against COVID-19, we will ask you for another blood sample either in your vein (4.5 mL) or with the TASSO device at home to look for COVID-19 antibodies.
- We will ask you to continue
  - Once a week to take the Study supplement
  - Once every two weeks to fill out the brief electronic questionnaire (duration of 3-5 minutes) regarding your health and work status in the previous 2 weeks.
  - At any point in time if you have symptoms, to complete the daily symptoms diary (duration of 1-2 minutes) until 48 hours after the resolution of symptoms. If you have symptoms that should prompt testing for COVID-19, as listed on the cv19quebec.ca website, we will ask you to contact your health office for this purpose, whether you have been vaccinated or not.

End-of-study – Week 16 (or later if the study is prolonged)

At the end of the study, you will be invited to a last in-person or remote visit. This research visit will take approximately 30 minutes and involve the following:

- You will be asked to return the Study supplement bottle containing the unused pills.

- A venous (or capillary blood if done remotely) sample and, if you have not tested positive at COVID-19 before, a saliva (or oro-nasopharyngeal) sample will be collected.
- A rapid COVID-19 antibody test (NADAL®) may also be done on a blood drop (finger-prick or from the venous puncture) for validation purposes. If done remotely, this test may be done on blood sampled by finger-prick under our guidance by videoconference.
- You will complete the last few short study questionnaires and any missing information in the previous ones, if applicable.
- If conducted remotely, the samples, Study supplement bottle and unused material should also be shipped to the Coordinating Center.

Collecting information on COVID-19 tests made for clinical reasons

The results for a COVID-19 test performed for clinical reasons outside this study will be documented by you in the follow-up questionnaire (*faster means to inform us*) as well as in your institution's (Pandemic) or provincial database of COVID-19 cases, namely Trajectoire Santé publique (TSP) including all individuals who tested positive and all healthcare workers who tested positive under the supervision of the Ministère de la santé et des services sociaux (MSSS). If you are unable to answer the follow-up questionnaires, the information documented in these databases would ensure that we have complete information on the primary outcome of the study and thus allow us to determine accurately the impact of the intervention on the risk of infection with COVID-19.

Collecting information on healthcare services

The date, diagnosis, type of professional and of health care services which you have received during medical visits and hospitalisations will be obtained from the administrative databases of the Régie de l'assurance maladie du Québec (RAMQ) and Quebec hospital discharges (MED-ECHO). This will allow us to accurately determine the impact of the intervention on the severity of COVID-19 infection and other concomitant illnesses.

Collecting information of work absence

The number of days of work absence, overall, by type (i.e., holiday, illness, etc.), and specifically due to COVID-19, including absences due to an infection acquired at work or outside of work, preventive withdrawal due to pregnancy or other health conditions, awaiting test results/investigation, or other reason for quarantine will be collected from you via the follow-up questionnaire (*faster and most detailed means*), as well as from your institution's *Direction of Health Resources* or, if you are an attending physician, from the *Direction of professional services*. If you are unable to answer the follow-up questionnaires, the information documented in these databases would ensure that we accurately ascertain the impact of the intervention on work absences.

BIOBANK

For the purposes of this study, we will keep the biological samples collected (blood, saliva and/or oro-nasopharyngeal) in a biobank as well as the clinical and administrative data collected during the course of this study in order to complete the study's objectives, and to

conduct research on vitamin D, COVID-19 and its treatments and other related diseases. We would like to quantify specific cellular receptors which allow entry of COVID-19 into cells (for example, the angiotensin converting enzyme-ACE2) and inflammatory markers (such as the C-reactive protein). The collected samples will be kept in a biobank in the Research Center of CHU Sainte-Justine under the supervision of Dr Francine M. Ducharme. The samples will be kept as long as the research team can guarantee their proper management. Confidentiality of the identity of the samples will be guaranteed by assigning them a specific code. Your sample will not be identified by your name and cannot be used to identify you directly. After 5 years, the code key will be destroyed, and the samples will become completely anonymous. Your samples could possibly be shared with other researchers in other institutions. However, the access to data will only be allowed for approved projects by an independent research ethics board.

GENETIC ANALYSIS (optional)

Each person has their own set of unique genes or "genome". Genetic research aims to determine if there are genetic predispositions which make you more susceptible to a COVID-19 infection, to respond to vitamin D, to modulate disease severity and the interaction of these factors.

If you accept to participate in the genetic analysis, these analyses will be done on a small part (4 mL) of the venous blood sample provided during the first visit. If you decide to participate remotely, we will ask you to provide a saliva sample in a small tube.

We would like to sequence your entire genome and conduct gene expression analyses. We would also like to share your genetic data as well as other collected clinical data during the PROTECT study with the Canadian database Hostseq COVID-19 for use for COVID-19 related research and other aspects of human health. This biobank will serve as a centralized resource in Canada for COVID-19 research and other health-related studies. The data in the HostSeq database are under the supervision of CGen, a national Canadian platform financed by the federal government for sequencing and genome analysis. The principal investigators of the PROTECT study as well as the administrators of the Hostseq biobank COVID-19 will share your genetic and clinical information with other Canadian and international researchers whom are approved by CGen (the sponsor). The data could also be used for commercial use. However, your data will not be shared with until after an examination by a data access committee. This committee will verify that the use of the proposed research is in line with the objectives of the database HostSeq and that the research team which requests access has already been granted the required approval in accordance in terms of research ethics requirements. Approved researchers will sign agreements. These agreements will control how the data will be used. Individual results of any research conducted using your samples or any individual incidental findings will not be shared with you, as the research conducted on your data will have no individual diagnostic or therapeutic significance to you.

WHAT ARE THE BENEFITS AND RISKS OF THIS STUDY?

Benefits:

You may not benefit directly from the study intervention if it is not efficacious or if you have been assigned in the placebo group. However, the screening may identify earlier an active or past COVID-19 infection that was not apparent. Vour participation will help advance our knowledge on vitamin D and on the prevention of COVID-19 infection in healthcare workers and other individuals at risk of infection.

Each positive COVID-19 result from the saliva (or oro-nasopharyngeal) or blood sample will be shared with you according to your preferred way of communication: telephone, text message or email. All positive saliva (or oro-nasopharyngeal) results will also be shared by the Microbiology Laboratory of CHUM with the Public health authorities and will be added into your file at the CHUM and Dossier Santé Québec. No other research result will be provided to you. Research findings resulting from your participation to this study could potentially contribute to creating commercial products from which you would not be able to claim any financial benefit.

Risks:

• Related to study medication:

The vitamin D dose used in this study has been shown to be safe in adults. This dose is approved by Health Canada for the purpose of this study only, but not for clinical use yet. It is unlikely that you will have any side effects because of the amount of vitamin D used in this study as when combining the first and weekly doses, the total remains below the maximum amount allowed.

However, we will ask you to notify us immediately if you have any of the following, as they could be signs of an acute excess intake of vitamin D: mainly, a marked increase in thirst or an increase in the volume and frequency of urination (with or without fatigue, loss of appetite, nausea or vomiting, headaches, drowsiness, cardiac arrhythmias, constipation, muscle or bone or chest pain, mouth dryness or a metallic taste).

Later signs and symptoms that may indicate a chronic excess intake of vitamin D are: a marked increase in thirst, an increase in the volume and frequency of urination including during the night, loss of appetite, weight loss, red eye or conjunctivitis, inflammation of the pancreas, light sensitivity, runny nose, itching, fever, reduced libido, kidney stones, increased concentration of some analytes in the blood (BUN, AST, ALT, cholesterol), or in urine (albumin), ectopic calcification, hypertension, cardiac arrhythmias and rarely, a psychosis.

It's possible that other currently unknown risks are associated with Vitamin D intake.

One of the reasons we collect a blood sample is to measure the concentration of vitamin D in the blood at the start and end of the study. this will allow us to see if the vitamin D blood level is linked to the number and severity of COVID-19 confirmed cases.

• Related to study procedures:

The salivary collection sample is painless. If done, an oro-nasopharyngeal swab may cause slight discomfort during collection that will subside after its removal. The side effects of having blood collected by venous puncture or TASSO can include bleeding, bruising, discomfort and pain at the sample site. It is possible that the NADAL COVID-19 IgG/IgM Test may give false positive or false negative results. In case of divergence of results, we will communicate to you the results of the approved IgG test when available.

• Related to confidentiality:

There is always a small risk that your data could one day be re-identified. The genetic information is unique to each person, just as your fingerprint. This means that theoretically, you could be identified using your genetic code; however, this is not easy to do. Considering the advances in technology, there could be new ways to link you to data that we have not foreseen today, despite the strict confidentiality measures in place. Possible re-identification or unintentional disclosure of your genetic and clinical research data could lead to a loss in confidentiality and a possible future discrimination against yourself or your biological parents. But all security measures will be put in place to protect your privacy.

WHAT ARE THE COSTS OF TAKING PART IN THIS STUDY?

The Study supplements are provided free of charge by the manufacturer, Laboratoire RIVA.

WHAT ARE THE OTHER FINANCIAL ASPECTS?

For each completed visit (0 and 16 weeks), you will receive a $25 check by mail to compensate for your time. The check may arrive at your home between 4 and 8 weeks after the visit.

HOW IS PRIVACY INSURED?

During your participation in this research study, the investigators responsible for this study as well as the members of their research team will collect, in a research file, the required personal information to answer the scientific objectives of this research project.

These information could include your demographic data (name, sex, date of birth, ethnic origin, weight and height), your past and present health status, your health-related habits, medication you take, your work absences, and the results of all tests, exams, and procedures which you will participate in. Your personal file will include your address, email, telephone numbers, RAMQ number, and employee or practice number be kept in a separate file with restricted access; this information is required to create a medical and pharmacy file at the CHUM and for communication purposes during the study.

The coded blood, saliva (and/or oro-nasopharyngeal) samples will be sent to the biobank located at the Research Center of CHU Sainte-Justine under the supervision of Dr Francine M. Ducharme. The coded results of completed analyses will be kept on a protected server with restricted access at DACIMA company during the study, and thereby transferred to a secure server with restricted access in the Research Center of CHU Sainte-Justine under the supervision of Dr Francine M. Ducharme. During the study, the personal information

used to arrange virtual and in-person study visit appointments will be kept on a protected server with restricted access at the company providing the appointment-making software. Following the conclusion of the study, this information of yours will be transferred to a secure server with restricted access in the Research Center of CHU Sainte-Justine under the supervision of Dr Francine M. Ducharme. The database of HostSeq will be kept on secure cloud servers (online) that are based in Canada and will be indefinitely kept or until they are not useful for research.

To ensure your privacy, a copy of the consent form as well as the results to the diagnostic tests required for conducting the research project, will be copied in the research and medical file of the CHUM. Therefore, each person or company which you authorize to consult your medical file, will have access to this information.

The research data will be kept for at least 25 years by the principle investigator. The data collected could be published or discussed during scientific meetings, but it would not be possible to identify you.

All collected information will remain confidential within the limits provided by law. You will only be identified by a code number. The key to the code linking your name to your research file will be kept by the investigator responsible for this research project.

To ensure your safety, a copy of the consent form as well as the results of the diagnostic tests required for research purposes will be placed in the research file and the medical file of the CHUM. Consequently, any person or company to whom you give access to your medical file will have access to this information.

Research data will be kept for at least 25 years by the investigator responsible for this research project. Research data may be published or be the subject of scientific discussion, but it will not be possible to identify you.

For the purposes of surveillance, control, safety and marketing of the Study drug, your research as well as your medical files could be consulted by a person mandated by a regulatory organization, in Canada or elsewhere, such as Health Canada, as well as sponsor representatives of the company manufacturing the vitamin D pills for this project (Laboratoire RIVA), the institution or research ethics committee. These people and organizations adhere to a strict confidentiality agreement.

You have the right to consult your research file to verify the collected data and to correct them, if needed. Moreover, access to certain information before the end of the study could mean your removal from this study in order to maintain the study's integrity.

IS YOUR PARTICIPATION VOLUNTARY?

Yes. Taking part in this study is voluntary. You may choose not to be in this study. You can decide to stop being in the study at any time, without needing to provide any reason, but simply informing the research team.

Your decision to refuse participation or to stop participating in the study at a later time, will have no effect on the quality of care or services to which you are entitled or on your relationship with the people that provide them.

The principal investigators of this study, the research ethics board, the funding agency or the sponsor could decide to end your participation in the study without your consent. This could happen if there are new information or findings that indicate your participation is no longer in the best of your interests, or if you have not been following the study instructions as explained, or if there are other administrative-related reasons to stop the project.

If you stop participating in the study or if you have been removed from it, the collected information and material already received will be kept (as well as the data pertaining to healthcare services and work absences will continue to be collected) and analysed to ensure the validity of this project, unless you specifically ask for them to be destroyed. If this is the case, these data and/or material will be removed from the biobank provided that the code key (linking between nominal data and the study code) is still available, that is, up to 5 years after the end of the study.

If you decide to drop out of the HostSeq database, your data will no longer be shared, and no new data will be collected. The data already in the HostSeq database will be destroyed once informed about this decision. However, it could be impossible to remove the results once they have been compiled with the results of other participants or if they have been published. Moreover, if certain data have been shared with other researchers, it could be possible not to be able to remove this part of the data. In such a case of unsuccessful withdrawal from the study, your identity will always be protected.

All new information acquired during the course of the study which could have an impact on your decision to continue participation will be shared with you rapidly, which is the reason why we would like to keep your personal information and have your approval to communicate with you after the end of the study (optional).

WHOM DO I CALL IF I HAVE QUESTIONS OR PROBLEMS?

If you have any questions about the research project or if you have any problems that you believe are related to your participation in the project, you can call the researchers responsible for the project:

- Dr. Francine M. Ducharme at 514 345 4931, extension 4398
- Dr. Cecile Tremblay at 514 890-8000, extension 14645

If you would like information about your rights related to your participation in the research, you may contact the Ombudsman - complaints and quality services of the CHU Sainte-Justine at 514 345-4749, of the CHUM at 514 890-8484 or your CISSS/CIUSSS:

- CIUSSS de l'Est-de-l'Île-de-Montréal : 514 252-3510
- CIUSSS de l'Ouest-de-l'Île-de-Montréal : 514-989-1885, extension 1010
- CIUSSS du Centre-Sud-de-l'Île-de-Montréal : 514 593-3600
- CISSS de la Montérégie-Est : 450-468-8447
- CISSS de la Montérégie-Centre : 450-466-5434

RESEARCH ETHICS COMMITTEE

The Research Ethics Board of CHU Sainte-Justine has approved this study and will continue to monitor it for all participating institutions of the Quebec Health and Social Services network.

LIABILITY

This research is not funded by a private industry. In case of side effects resulting from the study medication or from procedures required for this research project, you will receive all necessary medical care covered by the Quebec's provincial health insurance plan (RAMQ) or by your private drug insurance plan. You will be responsible for paying the portion of any costs not covered.

CONSENT FORM

Research project title: PRevention of COVID-19 with Oral Vitamin D supplemental Therapy in Essential healthCare Teams (PROTECT)

The nature and procedures of this research project were explained to me. I have read the information and consent forms and I kept a copy, or a copy has been provided to me. I was able to ask my questions and they were answered to my satisfaction. After consideration, I agree to participate in this research project.

I authorize the research team to consult the collected data about me in the COVID infection database (Pandemic) of my institution and/or the provincial TSP database, the medical and hospitalisation services database (RAMQ and MED-ECHO), and the workplace absenteeism database (Human Resources Directorate or Professional Services Directorate) to obtain information that is pertinent to this project.

By agreeing to participate in this study, you are not waiving any of my rights under the law. You are not releasing the investigators from their legal and professional liability.

Name of participant (Print)	Signature	Date

1. I consent to the analysis of gene expression and the sequencing of the whole genome of my coded biological material (blood, saliva, and/or oro-nasopharyngeal). The whole genome sequence could be hosted in the Canadian HostSeq COVID-19 biobank and linked to a database containing the viral genome. This would serve to explore any genetic predisposition to COVID-19, the severity of the disease and response to vaccine.

❒ Yes _____ (Initials) ❒ No_____ (Initials)

2. I consent to prolonging the access to my coded data on healthcare use, COVID-19 infections and work absenteeism for 12 months following the study end date, to explore the long-term impact of COVID-19 infection and vaccination.

❒ Yes _____ (Initials) ❒ No_____ (Initials)

3. I consent to being contacted to update my personal information, obtain additional information about my health or to be invited to participate in new research.

❒ Yes _____ (Initials) ❒ No_____ (Initials)

4. In case I receive a vaccine against COVID-19 during the study, I agree to do the blood samples before the first and second vaccine dose as well as 1 month after the 2nd vaccine dose, even if these samples were to be done after the end-of-study's visit planned at week 16 (or 24).

❒ Yes _____ (Initials) ❒ No_____ (Initials)

Participant's signature: _____

I have explained the research study and the terms of this information and consent form to the research participant, and I answered all his/her questions. I explained that participation in a research project is free and voluntary and could be stopped at any time they choose.

Name of person obtaining consent (Print)	Signature	Date

(FOR THE CHUM PARTICIPANTS ONLY)

COMMITMENT OF THE PRINCIPAL INVESTIGATOR AT THE CHUM

I certify that this information and consent form was explained to the research participant, and that the questions the participant had were answered.

I undertake, together with the research team, to respect what was agreed upon in the information and consent form, and to give a signed and dated copy of this form to the research participant.┼

Name (Print)	Signature of the principal investigator at the CHUM	Date