Structured Study Protocol of PreVitaCOV

Title

PreVitaCOV: Prednisolone and vitamin B1, B6, and B12 in patients with Post-COVID-19-Syndrome (PC19S) – a randomised controlled trial in primary care.

Names and affiliations of protocol contributors

C. Förster[1], Y. Kaußner[2], C. Tengelmann[2], U. Malzahn[3], L. Lunden[4], C. Eidenschink[2], K. Erdmann[2], E. Feil[1], S. Sellmann[4], G. Borgulya[2], K. Häusler[5], S. Joos[1], H. Kaduszkiewicz[4], K. Hügen[3], K. Hartung, O[3]. Zolk[6], P.U. Heuschmann[3], I. Gágyor[2]

[1] Institute of General Practice and Interprofessional Care, University Hospital Tübingen

[2] Department of General Practice, University Hospital Wuerzburg

[3] Clinical Trial Centre, University Hospital Würzburg (CTCW)

[4] Institute of General Practice, University of Kiel

[5] Department of Neurology, University Hospital Würzburg

[6] Medizinische Hochschule Brandenburg CAMPUS GmbH

Abstract

Background: A relevant proportion of patients complains ongoing symptoms after infection with SARS-CoV-2, a condition termed Post-COVID-19-syndrome (PC19S). Evidence on therapeutic options is still sparse. Neurotropic vitamins and drugs with anti-inflammatory properties were suggested to alleviate symptoms. The aim of this trial is to assess the feasibility, effectiveness, and safety of treating patients with PC19S in primary care with prednisolone and/or a fixed combination of vitamin B1, B6, and B12.

Methods: Double blind, randomised controlled trial, factorial design, four parallel groups, placebo-controlled, phase IIIb

Discussion: The trial is designed as a two-step approach that will first prove the feasibility of recruitment and retention of patients with PC19S in a primary care setting, and second, investigate the effectiveness and safety of the treatment drugs.

Trial registration: This trial has been registered under the EudraCT number 2022-001041-20.

Keywords

Post-COVID-19-Syndrome, Prednisolone, Vitamin B, primary care, RCT.

Note: the numbers in curly brackets in this protocol refer to SPIRIT checklist item numbers. The order of the items has been modified to group similar items (see http://www.equator-network.org/reporting-guidelines/spirit-2013-statement-defining-standard-protocol-items-for-clinical-trials/).

Introduction

Background and rationale {6a}

Scientific Background

Post-COVID-19 syndrome (PC19S) is a new pathologic entity affecting a considerable portion of patients (dependent on the population studied and the defined duration) after an infection with SARS-CoV-2.[1-5] A population-based survey from Germany that included 1,459 patients reports a prevalence of 49% for persisting symptoms after COVID-19.[3] PC19S is characterised by a broad range of persistent disabling symptoms and signs.[6] The most frequent symptoms include fatigue, post-exertional malaise, cognitive dysfunction, impaired attention, dry cough, shortness of breath, headache, muscle ache, chest tightness, and sore throat.[7] Most patients seek help in ambulatory care; and 76% exclusively consult their General Practitioner.[8] The German S1-Guidelines summarise the current recommendations and options in PC19S care. So far, there is no evidence on specific medical treatment options for these patients. The lack of swiftly initiated trials that explicitly take place in primary care in Germany and generate valid results reflects the lack of infrastructure and preparation.

The exact pathophysiological mechanism of PC19S is unknown. Several mechanisms have been proposed as pathophysiological explanations, including long-term tissue damage and chronic auto-inflammation.[9] Preliminary findings suggest that cytokine signature differs between PC19S and convalescent COVID-19 patients without persistent symptoms.[10] Thus, an Italian study with 551 patients using a multiplex immunoassay found an abnormal hyperinflammatory cytokine profile in patients with persisting symptoms compared to a control.[10] Furthermore, the varied symptoms of PC19S might be induced by an increased demand for methyl-groups in the organism caused by SARS-CoV2 while impairing their supply due to the viral-induced cytokine storm and oxidative stress.[11] Accordingly, patients with PC19S may benefit from methyl-group support e.g. by therapy with vitamin B12. Other authors suggest vitamins B1, B6, and B12 due to their neurotropic effects considering that most patients suffer from fatigue or other neurologic symptoms.[12] For the present trial PreVitaCOV, we chose vitamin B1, B6, and B12 due to the above-mentioned possible effects.

Drugs with anti-inflammatory properties such as corticosteroids, among others, were also suggested to alleviate symptoms.[9] This approach is based on the idea of chronic inflammation, as has already been postulated for various autoimmune diseases[13] and the PC19S.[14] An observational study including 30 patients showed that treatment with corticosteroids at high initial dose and a rapid wean over a few weeks was well tolerated and associated with improvement of post-COVID-19 pulmonary symptoms.[15] In another study, but with significant limitations in study design, 24 patients diagnosed with Long-COVID were treated with corticosteroids in tapering doses for 8 to 10 weeks. The study reports improvement in fatigue and breathlessness, among other, but did not include a control group.[16] A Spanish study treated eight patients with 30 mg prednisolone for a total of four days. While the focus was on normalisation of immunologic laboratory findings, also the clinical parameters were reported to have improved.[17] Therefore, we chose corticosteroids as a further therapeutic approach for PC19S.

Based on the above-mentioned rationale and the urgent need for effective, evidence-based treatment options in PC19S, this trial will investigate two drugs that have been approved for years and that are successfully used in many clinical conditions:

Prednisolone

Like all corticosteroids, Prednisolone is a potent anti-inflammatory drug used in a variety of autoinflammatory or rheumatoid conditions. As stated above, chronic (auto-)inflammation is discussed to be a potential aetiology of PC19C.[7, 14]

The most effective regimen is a pulse therapy with high initial doses like it is used in many (auto-)inflammatory conditions. A first observational study showed that treatment with corticosteroids at a high initial dosage and a rapid wean over a few weeks was well tolerated and associated with improvement of post-COVID-19 pulmonary symptoms.[15] In order to balance therapy efficiency and potential side effects, we will therefore administer Prednisolone in a dosage of 20 mg during the first five days, a dosage that is considered to be a medium dosage.[18] Given its rapid anti-inflammatory effect, changes in clinical manifestations of (auto)immune diseases can usually be observed within a few days. The regimen will continue with a dose of 5mg after day five in order to maintain the anti-inflammatory effect after the initial induction.

A second rationale is the potential effect on patients with low blood levels of cortisol since low cortisol concentrations were recently reported in patients with PC19S.[19]

Vitamin B1, B6, and B12

According to the above-mentioned rationale, we choose vitamins B1 (thiamine), B6 (pyridoxine), and B12 (cyanocobalamin) because of their potential to alleviate viral-induced inflammation, cytokine storm, and oxidative stress[11] as well as because of their positive neurotropic effects.[12]

The dosage used in this trial will be same as used for supplementation of the respective vitamin deficiencies[20]: thiamine 100 mg, pyridoxine 50 mg, and Cyanocobalamin 500 µg. While the European Food Safety Authority (EFSA) defined no upper limits for Vitamin B1 and B12, the upper limit for Vitamin B6 was set to 25 mg per day.[21] This is because of possible neurotoxic side effects. However, these were observed after long-term use of vitamin supplementation only. Therefore, a short-term use over four weeks is considered to be safe with a dosage of 50 mg. In addition, the National Institute of Health sets the upper limit for pyridoxine to 100 mg per day.[22]

Objectives {7}

The study is divided into two parts: The pilot study aims to demonstrate feasibility of a pragmatic randomised controlled trial (RCT) with vitamin B1, B6, and B12 and prednisolone in primary care patients with PC19S. Specifically, it will inform on feasibility of recruitment and retention of patients in primary care, and on data collection (including bio samples).

If feasibility is confirmed, the trial will be transformed into a confirmatory RCT to prove the effectiveness of the treatment approaches.

By involving general practices and health care professionals in university hospitals, the study team will establish proceedings for future RCTs and provide a use case for existing infrastructures such as practice-based research networks and the NAPKON(23).

Trial design {8}

Multicentre, randomised, placebo controlled, double-blind phase III trial with four parallel groups.

Methods: Participants, interventions and outcomes

Study setting {9}

The trial is conducted with a sample of patients recruited in primary care. This will facilitate transferability of the findings to patients with PC19S. Most patients with suspected PC19S seek help from their general practitioners (GPs). Therefore, it is necessary to investigate which treatment options are effective and feasible and that can be easily guided by GPs and implemented in primary care.

Eligibility criteria {10}

Inclusion criteria

1. adult patient
2. history of documented SARS-CoV-2 infection (documented by either positive PCR or Antibody-Test or both) at least 12 weeks ago (if patients had more than one infection with SARS-CoV-2 the 12 weeks refer to the infection causing the Post-COVID-19 symptoms)
3. symptoms concerning at least one of the following domains: fatigue, dyspnea, cognition, anxiety, depression
4. symptoms that developed during or after the SARS-CoV-2 infection, that persist until study inclusion and that are associated with COVID-19 (the latter will be assessed by the patient's GP or the local investigator)

Exclusion criteria

1. acute COVID-19 at baseline visit (rapid SARS-CoV-2 antigen test)
2. patients who were treated in the intensive care unit because of COVID-19
3. pregnancy/breastfeeding
4. diabetes mellitus
5. hypertension
6. PC19S symptoms can be explained by an alternative diagnose (e.g., chronic fatigue syndrome, depression, active or preceding cancer therapy, severe anemia, sleep apnea syndrome) as assessed by the patients' GP or the investigator (responsible physician)

7. History of severe medical conditions such as

- concomitant acute infectious disease
- gastrointestinal ulcer
- liver disease/ liver cirrhosis
- malabsorption or condition after bariatric surgery
- chronic airway disease (e.g., asthma, COPD)
- chronic heart failure (NYHA III or IV)
- neurologic disorders (e.g., multiple sclerosis, motoneuron disease)
- untreated hypothyroidism
- significantly impaired glucuronidation (e.g., Gilbert-Meulengracht, ROTOR, or Crigler-Najjar syndrome)
- immunodeficiency or a chronically weakened immune system (e.g., HIV, AIDS, lymphoma, chemo-radio-therapy, immunosuppressive pathology)
- mental disorders (e.g., depression, psychosis, dementia)
- active cancer
- any other severe medical conditions that preclude participation as determined by responsible physician

8. current use of

- immunosuppressive drugs
- non-steroidal anti-inflammatory drugs (NSAID)
- fluoroquinolones
- anticoagulation: phenprocoumon or other cumarin derivates, direct oral anticoagulants, ASS
- any other drug with a possible interaction with the study medication

9. current or previous treatment with any of the trial drugs for at least seven days since COVID-19 or any parenteral application (includes vitamin supplements containing vitamin B1, B6, or B12)

10. known allergy and contraindications to the intervention drugs

11. need of care and/or peer dependency

12. nursing home residents

13. inability to understand the scope of the study, to follow study procedures and to give informed consent or to attend the study sites

14. participation in another interventional trial at the same time or within the past three months before enrolment

15. Female patients considering to get pregnant during the trial and within one week after the last dose of study drug(s).

Who will take informed consent? {26a}

Before the trial starts, every patient participating in the trial must give the investigator written consent after being fully informed of the nature, significance and scope of the clinical trial verbally and in writing in a manner that they are able to understand. The contents of this information will be documented on the informed consent form.

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

The informed consent applies to biological specimens as well. For ancillary studies a separate informed consent is necessary.

Interventions

Explanation for the choice of comparators {6b}

Using placebo as a comparator will not only overcome context effects of the trial but also balance for placebo effects. Yet, the use of placebo as a comparator will result in some participants not receiving the active treatment. However, as long as effective evidence-based treatment options are lacking, participants randomised to placebo will not be deprived of the standard care.

PC19S is a new entity and therefore only few placebo-controlled trials are available yet. Based on the current literature and evidence from similar conditions, the placebo response rate in patients suffering from PC19S is expected to be between 20% and 40%. Thus, a recent study involving patients with PC19S and olfactory impairment reported that 42% of participants in the placebo group improved.[24] Other studies of patients with long-term conditions like Lyme disease found a placebo response rate of 36%.[25] A systematic review and meta-analysis on placebo responses in chronic fatigue syndrome which shares some similarities with PC19S reports a placebo response rate of 19.6%.[26]

Intervention description {11a}

Trial drug 1: Prednisolone 20 mg for five days followed by 5 mg for 23 days

Trial drug 2: Vitamin B1 (100 mg), B6 (50 mg), and B12 (500 µg) for 28 days

Trial drug 3: Trial drugs 1 and 2

Trial drug 4 (comparator): placebo

Patients will be instructed on how to take the medication. The tablets are swallowed in whole once daily with plenty of liquid in the morning.

Table 1 summarises the treatment arms together with the duration and the dosage of the intake of the trial drugs.

Table 1: Treatment arms, duration and dosage of intake.

	Day 0	**Day 1-5**	**Day 6-28**
1st arm (prednisolone and placebo)	enrolment	20 mg prednisolone 1x1 and placebo 1x1	5 mg prednisolone 1x1 and placebo 1x1
2nd arm (placebo and Vitamin B compound)	enrolment	placebo 1x1 and Vitamin B compound (100 mg B1, 50 mg B6, 500 µg B12) 1x1	placebo 1x1 and Vitamin B compound (100 mg B1, 50 mg B6, 500 µg B12) 1x1
3rd arm (prednisolone and Vitamin B compound)	enrolment	20 mg prednisolone 1x1 and Vitamin B compound (100 mg B1, 50 mg B6, 500 µg B12) 1x1	5 mg prednisolone 1x1 and Vitamin B compound (100 mg B1, 50 mg B6, 500 µg B12) 1x1
4th arm (placebo and placebo)	enrolment	placebo 1x1 and placebo 1x1	placebo 1x1 and placebo 1x1

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

Patients will be advised to avoid the intake of drugs that may interfere with the trial medication:

- NSAIDs, i.e., painkillers like ibuprofen or diclofenac; ASS even in prophylactic anticoagulatory dosage. Patients will be made aware of a potential risk associated with NSAID and ASS, given alternatives for analgesic medication, and instructed to contact his or her GP if they wish to take NSAR or ASS, for example as an analgesic. This information is also provided to the patient in form of a leaflet. If the use of NSARs or ASS becomes necessary for patients, the treating general practitioners are sensitised to adjust the medication. We will inform them of their patients' possible corticosteroid use at the start of the study. If patients present for newly initiated therapy with pain medication, GPs will decide on the necessity of therapy with PPI according to individual risk.
- fluoroquinolones
- antacids (magnesium-/aluminiumhydroxid) for at least two hours after taking the trial medication
- any other drug with a possible interaction with the study medication (as described in Fachinformation Prednisolon STADA®, Predni H Tablinen® Zentiva or Fachinformation Vitamin B Komplex forte Hevert).

Patients will also be advised to avoid the intake of the trial medication available as over the counter drugs:

- vitamin supplementation containing vitamin B1, B6, or B12
- prednisolone (not including local application)

Long-term medication will be documented on the eCRF; ingestion should continue as usual. Patients will record in the follow-up survey whether they received any new medical treatment since enrollment in the trial.
Furthermore, apart from oral instruction of the patients, a patient leaflet containing further information concerning the intake of concomitant medication, especially NSAIDs because auf their increased ulcerogenic potential, will be handed out on the first visit at the study centre.

Provisions for post-trial care {30}

For any harm caused by participation in this study there is a patient insurance. This provides insurance for trial-related injuries to health with a maximum sum covered of 500.000 € per patient. This insurance covers any injuries, which the patient suffers directly or indirectly as a result of the trial product or interventions connected with the clinical trial as well as securing travel accident insurance for all patients travelling to and from the site.

Outcomes {12}

The trial is divided into two phases:

In a pilot study we will evaluate feasibility and safety of treating PC19S with Prednisolone and/or Vitamin B1, B6, and B12 in comparison to placebo in patients recruited by their GPs.

> The primary outcome of the pilot study will be feasibility and acceptance of screening and recruitment in primary care, as assessed by the retention rate at day 28.

If the pilot phase shows feasibility, the study will be transferred into a confirmatory study to determine the effectiveness of the interventions.

> The primary outcome of the confirmatory study will be the change in symptom severity from baseline to day 28 as assessed by a specifically tailored total score based on the patient reported outcome measurement information system (PROMIS).

Secondary endpoints will be:

1. Severity of each PC19 symptom (PROMIS[27] total and subscores in the domains included in the total score, MYMOP[28]; PC19S functional status[29]; PC19 symptom list)
2. Health related quality of life (EQ-5D-5L[30] and visual analogue scale)
3. Depression (PHQ 8)[31]
4. Fatigue (Chalder Scale)[32]
5. Pain (numeric rating scale for pain)
6. Cognitive function: Alertness, distractibility, divided attention, sustained attention (TAP)[33]
7. Physical exercise (1minute Sit-to-Stand-Test)[34]
8. Use of on-demand medication and change in concomitant medication (patient diary)
9. feasibility and acceptance (qualitative interviews with subgroup sample; questionnaire).

Participant timeline {13}

The study intervention per patient will take 28 days followed by an observation period of five months after the end of study treatment. The individual patient study duration is six months. Data are collected at baseline (day 0; visit 1), at day 5(+3) (phone call 1), at day 14 (patient diary 1), at day 21 (patient diary 2), at day 28(+3) (visit 2), at follow-up after 60(+/- 5) days (phone call 2) and 180 (+/-7) days (phone call 3).

Sample size {14}

Pilot study

The aim of the sample size calculation for the pilot study was to ensure that we could estimate the retention rate, i.e., the proportion of patients who completed the 4-week intervention, with sufficient precision. Sufficient precision here means that the width of the corresponding 95% confidence interval doesn't exceed 0.15 (15%). Conservatively we base the calculation on the exact (Clopper-Paerson) method. Furthermore, because the target retention rate in this trial is 0.85 (85%), we assume for the calculation an observed sample proportion of 0.85. Then, 100 patients will be needed to yield a confidence interval with the postulated precision.

According to the study design, four equal sized treatment groups are planned. Therefore 4*25 = 100 patients are to be randomised in a 1:1:1:1 ratio at the beginning.

Confirmatory study:

Summary of sample size calculation:

The aim is the detection of an underlying main effect corresponding to at least 3-points (T-score) on the outcome scale. The outcome scale is the scale for the changes in our primary endpoint from baseline to day 28. The primary outcome measure is a tailored weighted mean T-score from five PROMIS subscales.

The range of values for the primary outcome measure is 16.7 – 57.7.

In the case that this 3-point difference is the smaller one of the two underlying main treatment effects (vitamin B compound and prednisolone) the sample size will ensure that both effects will be detected with sufficient power (80%). In the case that this 3-point difference is the stronger one of the two underlying main treatment effects the sample size will ensure with sufficient power that at least this stronger main effect will be detected.

Within the primary analysis, four treatment groups are compared: patients with neither intake of vitamin B compound nor prednisolone, patients with intake of vitamin B compound only, patients with intake of prednisolone only and patients with intake of both vitamin B compound and prednisolone combined.

We base the sample size calculation of the primary analysis for the primary endpoint by a two-factor analysis of variance (ANOVA). The dependent variable within the ANOVA model is our primary endpoint, operationalised as the change in the tailored PROMIS score from baseline to day 28 (visit 2). Furthermore, the two-factor ANOVA model contains the two main factors vitamin B compound and prednisolone, both with two factor levels: intake no vs. yes. Finally, the model contains the interaction factor with four levels. Within this model, we will estimate the main- and interaction effects and test the two null hypotheses of no difference between the two-factor level means in each case (for the main factors vitamin B compound and prednisolone) and test the null hypothesis of no interaction between the factors vitamin B compound and prednisolone by the corresponding F-tests with one nominator degree of freedom in each case.

Assumptions for sample size calculation: K. Kroenke[27] et al. reported a difference in T-score of 3 to 4 points as a **reasonable minimally important difference** for the PROMIS depression scales. Considering this for our primary outcome measure, as a weighted mean of T-scores from five PROMIS short form instruments, we aim to detect such a difference of 3 points as a significant effect. The same seems to be reasonable for change scores, i.e. for our primary endpoint. A 3-point difference between the effects of the two level (*no intake* versus *intake yes*) for a main factor within the linear model for a two-factor experiment corresponds to a standard deviation of means (SM) of 1.5.

Conservatively, we claim that the sample size has to be adequate in this sense that **any** underlying main effect corresponding at least to a standard deviation of means (SM) of 1.25 can be detected as a significant deviation from the null hypothesis of no effect with a power of 80% by the corresponding F-test at a significance level $\alpha = 0.05$. Furthermore, we assume a standard deviation (SD) of 7.5 points for the primary endpoint variable (see section Outcomes {12}). With this expressed in terms of effect sizes, the assumptions above correspond to an effect size about 0.167 (precisely 1.25/ 7.5).

A sample size of 4*72 = 288 ensures a power of 80% to detect any main effect with an effect size of at least 0.167 as significant deviation from the null hypothesis of no effect.

Assuming a lost to follow up rate of 15%, **4*85 = 340** patients are to be included in the study.

Summary

A total of 100 patients will be enrolled for the pilot study. The aim of the sample size calculation for the pilot study is to ensure that we could estimate the retention rate - that means proportion of patients who completed the 4-week intervention, with sufficient precision. Sufficient precision here means that the width of the corresponding 95% confidence interval does not exceed 0.15 (15%). Conservatively we base the calculation on the exact (Clopper-Paerson) method. Furthermore, because the target retention rate in this trial is 0.85 (85%) we assume for the calculation an observed sample proportion of 0.85. Then 100 patients will be needed to yield a confidence interval with the postulated precision.

In accordance with the extension of the “CONSORT 2010 statement to randomised pilot and feasibility trials”(35) the primary endpoint of the confirmatory study is defined as severity of PC19S measured by the tailored PROMIS total score. This score is composed of subscores for five typical symptom domains. The corresponding primary outcome measure is the change in this score from day 0 to day 28. Therefore, the calculation of the total sample size needed for both phases, 4*72 = 288 for the primary analysis, was based on the analysis of this outcome measure. Accordingly, 340 are to be allocated to the trial and 700 to be assessed for eligibility in total, whereas the patients of the pilot study are to be included.

Figure 1 illustrates the two-step-approach of the study and summarises timelines per patients. Table 2 summarises visits, assessments as well as data and outcomes to be collected in a chronological order.

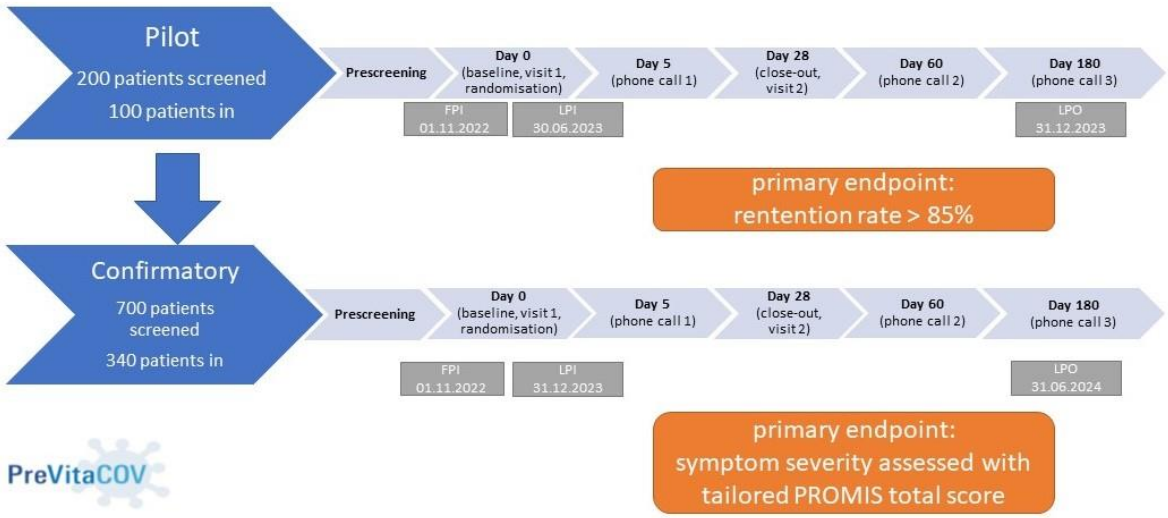

Figure 1: Flowchart on study timelines.

Table 2: Visit schedule, assessments as well as data and outcomes to be collected.

Patient contact	**Baseline**									**Follow-up**		
Day	**0**	**1-4**	**5***	**6-13**	**14**	**15-20**	**21**	**22-27**	**28***	**32-35**	**60****	**180*****
Visit number	1								2			
Phone call			1								2	3
Trial drug intake		X	X	X	X	X	X	X	X			
information, verification of in-/exclusion criteria, informed consent	X											
Demographic and baselinedata, Medical history, MoCA	X											
biosamples, vital signs	X								X			
2nd pregnancy test										X		
Outcomes												
- tailored PROMIS total score and subscores	X		X		X		X		X		X	X
- MYMOP	X		X						X		X	X
- PC19 Symptom list	X		X						X		X	X
- PCFS - Scale	X		X						X		X	X
- EQ-5D-5L and visual analogue scale	X		X						X		X	X
- PHQ 8	X		X						X		X	X
- Chalder Fatigue Scale	X		X						X		X	X
- Visual analogue scale for pain	X		X						X		X	X
- Tests Alertness, divided attention, distractibility, optional sustained attention (TAP)	X								X			
- 1min-Sit-to-Stand-Test, physical examination	X								X			
- Patient diary (on-demand medication)		X	X	X	X	X	X	X	X			
- Feasibility & acceptance questionnaire												X
- Feasibility & Acceptance Interviews (subgroup of patients)	X											
Safety outcomes												
- adverse events			X						X		X	X
- serious adverse events	as necessary											
Drug accountability			X						X			

Recruitment {15}

A two-step-approach will be pursued for recruiting the patients.

a) First, patients who visit their GP with symptoms of PC19S will be prescreened for eligibility by GPs in their practices. In addition, GPs will screen patients based on the electronic health records. To support screening, we will provide diagnostic guidance for GPs. Eligible patients will receive a letter of invitation from their GP inviting them to participate and providing the contact address of the trial sites.

b) Second, patients will present at the trial sites upon appointment, equipped with their medical history and concomitant medication provided by their GP on paper. Again, in- and exclusion criteria will be proven carefully by clinicians at the trial site at day 0 (baseline visit) to minimise the risk of false patient inclusion. Further trial management including patients' enrolment, treatment and management will be carried out at the three trial sites. If recruitment rates are low, further recruitment strategies (e.g., social media, press releases) will be considered.

Patient prescreening by GPs follows a pragmatic approach using their experience in patient assessment in term of eligibility for our trial. These considerations justify that the trial puts the diagnostic decision into the hands of GPs, mimicking a real-life situation.

We expect the guidelines on the diagnostics of PC19S to evolve, and we will provide the GPs working in the trial with an up-to-date guideline on how to diagnose PC19S and how to exclude patients with other diagnoses than PC19S in the GP setting.

Patients who contact one of the sites directly may also be admitted. Ideally, however, a presentation should be made to one of the participating GPs in advance, so that they can collect important medical data (such as medical history, concomitant medication) and forward them to the site. Basically, recruitment via the GPs is to be preferred for reasons of standardisation.

Assignment of interventions: allocation

Sequence generation {16a}

Patients will be allocated to the four treatment groups via stratified block randomisation with varying block size. The single stratification factor is the study centre. With this method of random treatment assignment, we try to avoid predictability, selection bias and full or partial unblinding still accomplish an acceptable balance.

Concealment mechanism {16b}

Computerised central randomisation and generation of the randomisation list recording the code numbers and assignment to the treatments will take place at the CTCW; the process will be overseen by the trial statistician. The randomisation lists will be generated outside the secuTrial® and then imported to secuTrial®.

Based on the randomisation list, the trial medication will be labelled with code numbers by the Hospital Pharmacy of the Charité. On entry to the study, the patients will receive the code number marked on their medication.

Implementation {16c}

Computerised central randomisation and generation of the randomisation list recording the code numbers and assignment to the treatments will be carried out by the clinical trial centre in Würzburg. The process will be overseen by the trial statistician. The randomisation code will be marked on his/her medication and linked with the patient identifier (Pat-ID) within secuTrial®.

Assignment of interventions: Blinding

Who will be blinded {17a}

This study is designed as a double-blind trial. Therefore, neither patients nor clinicians will know the study arm they are in. This will be achieved by an identical appearance of the tablets containing Vitamin B or placebo and identically encapsulated tablets for prednisolone or placebo (double dummy design).

Procedure for unblinding if needed {17b}

Early unblinding is performed only in case of medical emergency when it is essential to know the trial medication, i.e., when the physician believes that clinical management depends importantly upon knowledge of whether the patient received prednisolone and/or Vitamin B compound. In this case, the responsible study centre has to be informed immediately by telefax or phone.

The emergency and/or the SAE as well as the reason for the early unblinding has to be documented on both the case report form and on the opaque envelope. The trial medication will be stopped but the patient will be monitored by the end of the follow-up.

In cases where urgent unblinding is necessary, there is a digital randomisation list accessible from all trial sites for emergency unblinding by a physician who is not part of the study team during and outside of the trial site office hours. There, the physician providing emergency care can arrange for unblinding at any time. (This information is provided on the patient emergency card).

Data collection and management

Data management {19}

A computerised data entry and management system will be developed by the CTCW where all study data relating to the described main study can be entered.

A closed and password protected data entry system has been designed to ensure that only the responsible data entry person and the supervisor can enter and/or edit data and this can be done only by using the programs and/or utilities available on the menu system. An audit trail will be created by data and user stamping. Range checks, review screens, and various error trapping routines are built into the system as quality control procedures. The data will be managed and processed by the Centre for Clinical Trials of the University Hospital Würzburg.

Implementation and programming of the study database will be performed by data management of the CTCW. The system provides the capability to perform the major data management activities within a consistent, auditable and integrated electronic environment (data security, data entry, data validation). All data will be entered directly in the eCRFs and saved in the study database. Range, validity and consistency checks will be implemented in the system for application during data entry. Data entries will be recorded routinely via audit trail, as well as data modifications and data corrections. In case of necessary corrections or existing data inconsistencies, data queries will be generated consecutively by the data manager. Queries will be sent electronically to the study team of the respective trial site. Afterwards, data in the database will be corrected according to the answered and resolved queries. After database lock data are imported into standard statistical software systems and analysed by biostatisticians at the CTCW.

The Principal Investigators (PIs) or other delegated participating site staff will use electronic case report forms (eCRF) to capture all relevant data pertaining to patients enrolled in this study in a timely manner.

It is the investigator’s responsibility for data collected as part of the clinical trial to be entered correctly and completely in the database created specifically for this trial. It is possible to use worksheet as a documentary aid.

The PI is responsible for the correct and complete entry of data collected at their trial centre. Data entry at the trial sites must be carried out only by authorised individuals.

Corrections to the eCRF must be made only by authorised individuals or by the responsible investigator and must be justified. Corrections are made in a way that the old entry can still be retrieved. All data and corrections will automatically be logged with the date, time and person making the entry.

Confidentiality {27}

The patient identification list will be under strict control of the PIs and will not be transferred to the data management centre CTCW. Data recorded in the eCRF will be pseudonymised. Any personal data (such as medical history) will be identifiable only by a unique, private patient identification code (PreVitaCOV patient ID).

In accordance with applicable legal provisions, individuals who have direct access to source data shall take all the necessary precautions to ensure the confidentiality of information relating to IMP, research, patients and particularly their identity, as well as the results obtained. These individuals, like the investigators, are subject to the conditions of professional secrecy.

The CTCW will ensure that no unauthorised access to the computer system takes place and that no data loss occurs according to adequate data backup procedures. Only specified persons involved in the study will be given authorisation to enter or access data in the study databases based on predefined roles. There will be a complete audit trail of all transactions.

All employees of the CTCW involved in the study are required to actively protect confidential data against access by third parties in accordance with the prevailing guidelines.

All patient-related data will be recorded after pseudonymisation. The PreVitaCOV patient ID is defined according to the following standardised procedure:

All patients are numbered by the participating site code consisting of one numerical digit for the specific participating site number and four numerical digits for consecutive numbering of study patients per participating site. The patient number in the order of study enrolment is starting from ‘0001’ in every participating site. All subsequent patients are assigned consecutive numbers in chronologic sequence (e.g., second patient is assigned patient number 0002 and the third patient is assigned patient number 0003, etc.). Consequently, each patient included into the study will be uniquely identified in the study by a combination of his/her participating site and patient number which is their PreVitaCOV patient ID.

All patients eligible for the study will be documented in the screening log. In case a patient will not be included, this will be documented with the specific reason for non-inclusion.

The numbering will be performed by the eCRF. Additionally, the patient will be randomised by the CTCW via the eCRF with the deposited randomisation list (by problems with the availability of the eCRF by the printed randomisation list through the study team). The PIs will be maintaining a patient identification list linking the identification number with the personal data of the patient. This list will be kept strictly confidential at the participating site under control of the PIs. After termination of the study, this list has to be archived at the participating site following local guidelines.

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

n/a

Statistical methods

Statistical methods for primary and secondary outcomes {20a}

Primary outcome (primary endpoint of trial)

Pilot study

The primary outcome for the pilot study is the retention rate at day 28 of the trial. The proportion of patients who completed the 4-week intervention, point and interval estimations (95% CI, confidence interval) will be given. This will be done combining all four treatment groups.

Confirmatory study

The intention for the primary endpoint of the confirmatory study was to construct a tailored measure for the disease burden of patients with Post-COVID-19-Syndrome. This should be a patient reported outcome not too burdensome for patients. We operationalised this as a mean of the T-scores from five PROMIS® Short Form instruments:

- PROMIS Short Form v1.0 – Fatigue 4a
- PROMIS Short Form v1.0 – Dyspnoea Severity 10a
- PROMIS Short Form v2.0 – Cognitive Function 4a
- PROMIS Short Form v1.0 – Emotional Distress-Anxiety 4a
- PROMIS Short Form v1.0 – Emotional Distress-Depression 4a

At this, we have to distinguish between negatively-worded concepts (like Fatigue, Dyspnoea, Anxiety and Depression) and positively worded concepts like Cognitive Function. Therefore, the outcome measure isn't a simple mean but a linear combination adding up the 4 T-scores for the 4 PROMIS Short Forms Fatigue, Dyspnoea Severity, Anxiety, Depression and subtracting the T-score for Cognitive Function.

Mean and SD for all T-scores of the five validated instruments are 50 and 10 within the reference population from the population 2000 General US census. Therefore, by construction above the mean of our primary outcome measure in the reference population will be 30. But we don't know the variance and with this don't know the SD in the reference population. For this, we would need to know all ten pairwise covariances resp. all ten pairwise correlation coefficients in the reference population between the five T-scores. But we can get a very rough approximation: if we assume for all ten pairs of the five T-scores a medium strength of correlation, quantified by a Pearson correlation coefficient between 0.4 and 0.5, the SD of our transformed Post-COVID-19 tailored PROMIS total score will be between 7 and 8 (rather conservative).

Our primary outcome measure in the confirmatory study is defined as a change score, the difference in our specifically tailored outcome measure from baseline (visit 1) to visit 2 after 28 days:

Tailored-score (V2) – Tailored-score (V1).

Assuming a medium strength for the correlation between baseline measurements and measurements 28 days after baseline, we assume for this change score roughly a SD between 7 and 8 points, too, justified by experience gained from earlier change score analyses.

Data analysis for the primary endpoint of **the confirmatory study**: the specifically tailored PROMIS total score, derived from five Short Form instruments:

- Summarising descriptive statistics at baseline and day 28 (visit 2) for the tailored score within the four treatment groups
- Mean change in the tailored score from baseline to day 28 (visit) with 95% CI within the four treatment groups
- Factorial ANOVA for the dependent variable change in the tailored score from baseline to day 28 (visit 2)
- Problems investigated by corresponding F-tests within a model of a 2-factor ANOVA (factorial design)
    - Are there differences in the mean change score among patients treated with and without vitamin B compound?

    Null hypothesis: there is no effect for administration of vitamin B compound

    - Are there differences in the mean change score among patients treated with and without prednisolone?

    Null hypothesis: there is no effect for administration of prednisolone

    - Are there any vitamin-B-compound-prednisolone combinations that exhibit a mean change score that cannot be explained by administration of vitamin B compound and/or administration of prednisolone alone?

    Null hypothesis: there is no such interaction effect.

Further analysis within the 2-factor ANOVA:

- Estimation of marginal means for the primary outcome measure for the two factors intake of vitamin B compound (no/yes) and intake of prednisolone (no/yes) with 95%CI
- 95% CI for the mean change score within all four factor combinations of vitamin B compound and prednisolone.

Secondary outcome (secondary endpoint of trial)

For all secondary endpoints the corresponding total sum scores and if applicable subscores will be calculated for the different visits together with the corresponding change scores from baseline to day 28. Where appropriate, convenient transformations will be made.

Generally, all study variables will be presented using the appropriate descriptive statistics according to the measurement scale of data and for interval and ratio scaled data, respectively, according to the properties of the observed distributions (e.g., outlier values, skewness). Measures of frequency, of central tendency and dispersion or variation will be given as appropriate.

That means, for continuous variables the number of missing and of non-missing observations, arithmetic mean and/or median, range, SD or interquartile range, minimum, maximum. Frequency tables will be used to summarise categorical variables for the four treatment groups at different points in time. If appropriate, shift (change from baseline) tables will be given. For binary variables, numbers of missing and non-missing observations will be given together with the corresponding proportions. Shift tables (change from baseline) will be given if possible and appropriate.

In exploratory analyses, factors (such as trial centre, age, symptom duration or laboratory parameters) potentially influencing the primary outcome will be investigated using appropriate multivariable modelling.

Analysis for the secondary endpoint use of on-demand medication (patient diary): to compare the counts of on-demand medications within the period from baseline to day 28 between the four treatments negative binomial regression (NBR) models resp. zero-inflated negative binomial regression (ZINBR) models will be used. Within these models the count of using additional on-demand medication is the dependent variable, furthermore the models include the two binary main factors treatment with vitamin B compound, treatment with prednisolone as well the interaction factor. Using NBR models we can account for possible over-dispersion and using ZINBR models we can account for a possible elevated frequency of zero accounts in addition.

All secondary outcomes will be analysed in exploratory analyses. Further details on the statistical analyses will be provided in a detailed statistical analysis plan.

Safety outcomes will be analysed by reporting frequencies (percentages) of (S)AEs as well as the number of patients with worsening symptoms for each group.

Planned statistical analyses will be done using the current SAS version (SAS Institute Inc., Cary, NC, USA) as well as the current IBM SPSS version.

For selected secondary endpoints (specified within the detailed statistical analysis plan) secondary analyses will be carried out for the corresponding change scores from baseline to day 28 (where appropriate after a convenient transformation) in the same manner as for the primary endpoint by a 2x2 factorial analysis of variance.

Statistical methods for the analyses of changes from baseline to day 28 in outcome variables from physical examination (auscultation of chest and heart, orientating neurological examination) depend on measurement scales (interval, ordinal, nominal, dichotomous) and data types (categorical, counts, numerical, continuous, ratios) in each case. Descriptive analysis may comprise appropriate summarising statistics and shift tables. Confirmatory analysis resp. model-based analysis may comprise variance and covariance analysis as well as hierarchical models like hierarchical binary or ordinal logistic regression. Details for this as well as for all other statistical analyses will be given within the SAP.

Interim analyses {21b}

An important condition for a subsequent confirmatory study (successfully assessing and comparing the effectiveness of treating PC19S patients with prednisolone and/or vitamin B1, B6 and B12 vitamins, is a sufficient underlying retention rate. For this we assess 0.85 (85%) as a sufficient retention rate. Therefore, an actually observed retention rate based on 100 patients which is not compatible to this requirement would lead to a stop and not passing to the confirmatory study. We assess an observed retention rate < 0.80 (80%) as such an incompatible retention rate. A reasoning for this argumentation is that assuming an observed retention rate of 0.80 the upper limit of the corresponding one-sided upper-limit 90% confidence interval, calculated according to the Wilson-score method, is 0.846 (84.6%). That means this one-sided upper-limit 90% confidence interval would not include the target value 0.85 and with this indicate insufficient retention. If the retention rate is not achieved, the DSMB may recommend to continue based on the progression of retention (e.g., if retention has been improved in the course of the study, but has formally fallen the 85%).

If the criteria to continue (retention rate or recommendation DSMB) are met, there will be no analysis of secondary endpoints and the study will be transformed into the confirmatory study. **Patients enrolled in the pilot study will be included in the analyses of the confirmatory study.**

According to paragraph 10, section 1, of the German GCP Ordinance, the sponsor will inform both the ethics committee and the competent federal higher authority (BfArM) if any changes to the protocol should be necessary, especially after phase 1 of the study. In addition to that, the sponsor will inform both the ethics committee and the BfArM within 15 days if, during or after phase 1 of the study, the sponsor becomes aware of facts that must be assumed to have changed the benefit-risk-assessment (paragraph 13, section 4, of the German GCP Ordinance). This regards in particular a) reports of expected and severe side effects with unexpected outcome, b) an accumulation of expected and severe side effects that are considered clinically relevant, c) suspected cases of unexpected and severe side effects occurring after the participant had ended the trial, and d) any event related to the trial of preparation of the trial medication that is possibly endangering the safety of participants.

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

For the calculation of the primary endpoint the free Health Measuring Scoring Service (HMSS) will be used to obtain T-scores for the 5 PROMIS Short Forms (Fatigue, Dyspnea Severity, Cognitive Function, Anxiety, Depression). To indicate when a patient did not answer an item, whether because it was skipped or not applicable, either the word “SKIP” or a blank field will be listed and with this the HMSS algorithm will deal with missing data at the calculation of T-scores.

Generally, multiple imputation at the score level will be used for the patient reported outcome measures in this study.

Oversight and monitoring

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

A 4-member team of independent experts has been established as a Data Safety Monitoring Board (DSMB) in line with EMEA/CHMP/EWP/5872/03 Corr. to assess the safety aspects of the clinical trial regularly. The DSMB receives confidential information on the progress of the study regularly and is responsible for the following tasks:

- Approval of the trial protocol with regard to patient safety
- Assessment and discussion of safety aspects (SAEs, SUSARs and trial-related hospital referrals)
- Issue of a recommendation to the sponsor about how to proceed (to continue or discontinue the clinical trial) according to the frequency and severity of safety-relevant events.

Details of the tasks and responsibilities of the DSMB are specified in the DSMB charta.

Adverse event reporting and harms {22}

The Sponsor or its representative must document all the AEs of which it is notified in detail and transmit these on request to the competent federal higher authority (BfArM).

The investigator must inform the CTCW about the occurrence of a SAE without undue delay but not later than within 24 h of obtaining knowledge of it. The investigator should do so by using the SAE form in the eCRF. The investigator must then transmit a detailed written report on the SAE in question to the Sponsor or its representative. If an investigator learns of an (S)AE that can be reasonably related to study drug he should notify the sponsor even after the patient's study end.

Frequency and plans for auditing trial conduct {23}

Clinical monitoring, audits and (if requested by authorities) inspections will be conducted during the clinical trial for quality assurance purposes. Monitoring will be carried out by a monitor from the CTCW. An adaptive monitoring plan will be implemented following a specific risk assessment. The scope and contents will be laid down in a study-specific work instruction. Within the framework of trial sites audit, the investigators agree to allow an auditor delegated by the sponsor, the supervisory authorities or the registration authorities, or a member of the ethics committee responsible, to verify that the execution of the clinical trial is carried out in accordance with the trial protocol and GCP and to check all the relevant documents whereby random sample checks of the original documents (source data) can be undertaken. The clinical investigator shall allow this person, as well as the monitor, access to all premises used for trial execution and documentation of the clinical trial.

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

All aspects of the protocol are to be complied with during the trial. Should amendments become necessary, these are to be discussed immediately and in detail with the coordinating investigator resp. the Sponsor. The agreement reached is presented in writing in the form of a protocol amendment. The primary investigator either applies for the approval of the responsible ethics committee (“substantial” amendment) and authorities or informs the responsible ethics committee and authorities accordingly (“non-substantial amendment”).

Dissemination plans {31a}

Results of the trial will be published in national and international journals. In case of positive results, these will be introduced into guidelines and GP training/ quality circles, in order to implement them into medical care.

Also, the results of this trial will be fed directly into the Bavarian Practice Based Research Network (www.BayFoNet.de), the Research Practice Network Baden-Württemberg (www.forschungspraxennetz-bw.de) and throughout Germany via the Initiative of German Research Practice Networks (www.desam-fornet.de) through presentations, publication in newsletters.(36-38) Data will be provided in an open data repository and to existing infrastructures (NAPKON, www.napkon.de). A cooperation with NAPKON will further inform about recruitment findings.

Abbreviations

ANOVA	Analysis of variance
AE	Adverse event
AIDS	Acquired Immunodeficiency Syndrome
AMG	Arzneimittelgesetz (German Medicines Act)
BayFoNet	Bavarian Research Network in General Medicine
BfArM	Federal Institute for Drugs and Medical Devices
COPD	Chronic Obstructive Pulmonary Disease
COVID	Coronavirus Disease
CRO	Contract Research Organisation
CTCW	Clinical Trial Centre Würzburg
DEGAM	Deutsche Gesellschaft für Allgemeinmedizin und Familienmedizin (German College of General Practioners and Family Physicians)
DESAM-ForNet	Initiative Deutscher Forschungspraxennetze (The Initiative of German Research Practice Network)
DSGVO	Datenschutzgrundverordnung (German federal Data Protection Law)
DSMB	Data Safety Monitoring Board
eCRF	Elctronic case form
EQ-5D	European Quality of Life 5 Dimensions
EQ-VAS	European Quality of Life visual analogue scale
FoPraNet-BW	Forschungsnetzwerk in Baden-Württemberg
FOSA	Fach- und Organspezifischen Arbeitsgruppen
Gamma GT	Gamma-Glutamyltransferase
GCP	Good Clinical Practice
GCP-V	GCP-Verordnung
GM-CSF	Recombinant Human Granulocyte Macrophage Colony-Stimulating-Factor
GOT	Glutamat-Oxalacetat-Transaminase
GP	General Practitioner
GPT	Glutamat-Pyruvat-Transaminase
HMSS	Health Measuring Scoring Service
ICH	International Council for Harmonization
ID	Identity document

IMP	Investigational Medicinal Product
IMPD	Investigational medicinal product dossier
IIT	Investigator-initiated trial
ITT	Intention to treat
MoCA	Montreal Cognitive Assessment
MYMOB	Measure Yourself Medical Outcome Profile
NSAID	Nonsteroidal Anti-inflammatory Drugs
NAPKON	Nationales Pandemie Kohorten Netz (German National Pandemic Cohort Network)
NUM	Netzwerk Universitätsmedizin
NUM NUKLEUS	NUM Klinische Epidemiologie und Studienplattform
ECU	Epidemiology Cor Unit
NYHA	New York Heart Association
PC19S	Post-COVID-19-Syndrome
PCFS	Post-COVID-19 Functional Status Scale
PCI	Principal coordinating investigator
PCR	Polymerase chain reaction
PHQ8	Eight-item Patient Health Questionnaire depression scale
PI	Principal investigator
PP	Per protocol
PROMIS	Patient Reported Outcomes Measurement Information System
RCT	Randomised controlled trial
SAE	Serious adverse event
SAR	Serious adverse reaction
SARS	Severe acute respiratory syndrome
SD	Standard deviation
SM	Standard deviation of means
SmPC	Summary of product characteristics
SOP	Standard operating procedure
SUSAR	Suspected unexpected serious adverse reaction
TAP	Testbatterie zur Aufmerksamkeitsprüfung
TNF-α	Tumornekrosefaktor Alpha
TSC	Trial Steering Committee
TSH	Thyroid-stimulating-hormone
UADR	Unexpected adverse reaction

Funding {4}

The clinical trial is funded by the Federal Ministry of Education and Research.

Ethics approval and consent to participate {24}

The trial was approved by the ethics committee of the Medical Faculty; University Hospital Würzburg; Josef-Schneider-Str. 4/C12, 97080 Würzburg.

References

1. Nehme M, Braillard O, Alcoba G, Aebischer Perone S, Courvoisier D, Chappuis F, et al. COVID-19 Symptoms: Longitudinal Evolution and Persistence in Outpatient Settings. Ann Intern Med. 2021;174(5):723-5.
2. Crook H, Raza S, Nowell J, Young M, Edison P. Long covid-mechanisms, risk factors, and management. Bmj. 2021;374:n1648.
3. Förster C, Colombo MG, Wetzel AJ, Martus P, Joos S. Persisting Symptoms After COVID-19. Dtsch Arztebl Int. 2022;119(10):167-74.
4. Al-Aly Z, Xie Y, Bowe B. High-dimensional characterization of post-acute sequelae of COVID-19. Nature. 2021;594(7862):259-64.
5. Nasserie T, Hittle M, Goodman SN. Assessment of the Frequency and Variety of Persistent Symptoms Among Patients With COVID-19: A Systematic Review. JAMA Netw Open. 2021;4(5):e2111417.
6. Davis HE, Assaf GS, McCorkell L, Wei H, Low RJ, Re'em Y, et al. Characterizing long COVID in an international cohort: 7 months of symptoms and their impact. EClinicalMedicine. 2021;38:101019.
7. Lund LC, Hallas J, Nielsen H, Koch A, Mogensen SH, Brun NC, et al. Post-acute effects of SARS-CoV-2 infection in individuals not requiring hospital admission: a Danish population-based cohort study. Lancet Infect Dis. 2021;21(10):1373-82.
8. Schulz MM, Sandra; Scherer, Martin; Karagiannidis, Christian; Czihal, Thomas. Post-Acute Sequelae of SARS-CoV-2 Infection

Characterization of Community-Treated Patients in a Case–Control Study Based on Nationwide Claims Data. Deutsches Ärzteblatt International. 2022;119(10):177-8.
9. Yong SJ. Long COVID or post-COVID-19 syndrome: putative pathophysiology, risk factors, and treatments. Infect Dis (Lond). 2021;53(10):737-54.
10. Montefusco L, Ben Nasr M, D'Addio F, Loretelli C, Rossi A, Pastore I, et al. Acute and long-term disruption of glycometabolic control after SARS-CoV-2 infection. Nat Metab. 2021;3(6):774-85.
11. McCaddon A, Regland B. COVID-19: A methyl-group assault? Med Hypotheses. 2021;149:110543.
12. Calderón-Ospina CA, Nava-Mesa MO. B Vitamins in the nervous system: Current knowledge of the biochemical modes of action and synergies of thiamine, pyridoxine, and cobalamin. CNS Neurosci Ther. 2020;26(1):5-13.

13. Elkon K, Casali P. Nature and functions of autoantibodies. Nature Clinical Practice Rheumatology. 2008;4(9):491-8.
14. Kazama I. Stabilizing mast cells by commonly used drugs: a novel therapeutic target to relieve post-COVID syndrome? Drug Discov Ther. 2020;14(5):259-61.
15. Myall KJ, Mukherjee B, Castanheira AM, Lam JL, Benedetti G, Mak SM, et al. Persistent Post-COVID-19 Interstitial Lung Disease. An Observational Study of Corticosteroid Treatment. Ann Am Thorac Soc. 2021;18(5):799-806.
16. Goel N, Goyal N, Nagaraja R, Kumar R. Systemic corticosteroids for management of 'long-COVID': an evaluation after 3 months of treatment. Monaldi Arch Chest Dis. 2021;92(2).
17. Utrero-Rico A, Ruiz-Ruigómez M, Laguna-Goya R, Arrieta-Ortubay E, Chivite-Lacaba M, González-Cuadrado C, et al. A Short Corticosteroid Course Reduces Symptoms and Immunological Alterations Underlying Long-COVID. Biomedicines. 2021;9(11).
18. Buttgereit F, da Silva JA, Boers M, Burmester GR, Cutolo M, Jacobs J, et al. Standardised nomenclature for glucocorticoid dosages and glucocorticoid treatment regimens: current questions and tentative answers in rheumatology. Ann Rheum Dis. 2002;61(8):718-22.
19. Klein J, Wood J, Jaycox J, Lu P, Dhodapkar RM, Gehlhausen JR, et al. Distinguishing features of Long COVID identified through immune profiling. medRxiv. 2022:2022.08.09.22278592.
20. Vitamin B-Komplex forte Hevert - Fachinformation, (2008).
21. Authority EFS. TOLERABLE UPPER INTAKE LEVELS FOR VITAMINS AND MINERALS. 2006.
22. Health NIo. Vitamin B6 - Fact Sheet for Health Professionals Office of Dietary Supplements2022 [updated 02-06-2022. Available from: https://ods.od.nih.gov/factsheets/VitaminB6-HealthProfessional/.
23. NAPKON. Nationales Pandemie Kohorten Netz, https://napkon.de/ [
24. Di Stadio A, D'Ascanio L, Vaira LA, Cantone E, De Luca P, Cingolani C, et al. Ultramicronized Palmitoylethanolamide and Luteolin Supplement Combined with Olfactory Training to Treat Post-COVID-19 Olfactory Impairment: A Multi-Center Double-Blinded Randomized Placebo- Controlled Clinical Trial. Curr Neuropharmacol. 2022;20(10):2001-12.
25. Klempner MS, Baker PJ, Shapiro ED, Marques A, Dattwyler RJ, Halperin JJ, et al. Treatment Trials for Post-Lyme Disease Symptoms Revisited. The American Journal of Medicine. 2013;126(8):665-9.
26. Cho HJ, Hotopf M, Wessely S. The Placebo Response in the Treatment of Chronic Fatigue Syndrome: A Systematic Review and Meta-Analysis. Psychosomatic Medicine. 2005;67(2):301-13.
27. Kroenke K, Stump TE, Chen CX, Kean J, Bair MJ, Damush TM, et al. Minimally important differences and severity thresholds are estimated for the PROMIS depression scales from three randomized clinical trials. J Affect Disord. 2020;266:100-8.
28. Hermann K, Kraus K, Herrmann K, Joos S. A brief patient-reported outcome instrument for primary care: German translation and validation of the Measure Yourself Medical Outcome Profile (MYMOP). Health and Quality of Life Outcomes. 2014;12(1):112.

29. Klok FA, Boon G, Barco S, Endres M, Geelhoed JJM, Knauss S, et al. The Post-COVID-19 Functional Status scale: a tool to measure functional status over time after COVID-19. Eur Respir J. 2020;56(1).
30. Herdman M, Gudex C, Lloyd A, Janssen M, Kind P, Parkin D, et al. Development and preliminary testing of the new five-level version of EQ-5D (EQ-5D-5L). Qual Life Res. 2011;20(10):1727-36.
31. Kroenke K, Strine TW, Spitzer RL, Williams JB, Berry JT, Mokdad AH. The PHQ-8 as a measure of current depression in the general population. J Affect Disord. 2009;114(1-3):163-73.
32. Chalder T, Berelowitz G, Pawlikowska T, Watts L, Wessely S, Wright D, et al. Development of a fatigue scale. J Psychosom Res. 1993;37(2):147-53.
33. Zimmermann P, Gondan, M. & Fimm, B. Testbatterie zur Aufmerksamkeitsprüfung für Kinder KITAP. . Herzogenrath: PSYTEST Verlag. 2002.
34. Meriem M, Cherif J, Toujani S, Ouahchi Y, Hmida AB, Beji M. Sit-to-stand test and 6-min walking test correlation in patients with chronic obstructive pulmonary disease. Ann Thorac Med. 2015;10(4):269-73.
35. Eldridge SM, Chan CL, Campbell MJ, Bond CM, Hopewell S, Thabane L, et al. CONSORT 2010 statement: extension to randomised pilot and feasibility trials. Pilot Feasibility Stud. 2016;2:64.
36. BayFoNet. Bayerisches Forschungsnetz in der Allgemeinmedizin, https://bayfonet.de/ [
37. FoPraNet-BW. Forschungspraxennetz Baden-Württemberg, https://www.forschungspraxennetz-bw.de [
38. DESAM-ForNet. Initiative Deutscher Forschungspraxennetze; https://www.desam-fornet.de/ [