**BUSATTO ET AL:** **Post-acute sequelae of SARS-CoV-2 infection: relationship of central nervous system manifestations with physical disability and systemic inflammation - Supplementary material 1**

Methods S1. Further details on study design and procedures……………………………….….…02

Methods S2. Further details on the assessment of persistent symptoms and objective indices of functioning ………………………………………………………………………………..………….….02

Methods S3. Details on blood laboratory indices assessed at the follow-up …………………….02

Methods S4. Further details on the exploratory factor analysis and missing values……..…......03

Methods S5. Further details on the Item Response Theory analyses………………………….…03

Methods S6. Further details on the sensitivity analyses evaluating the Item Response Theory model……………………………………………………………………………………………….….....03

Methods S7. Further details on the analyses evaluating relationships between PASC symptom severity, objective signs of organ system dysfunction and laboratory test results………..….....04

Methods S8. Further details on the sensitivity analyses evaluating the influence of demographic variables on the relationship between the latent variable of PASC symptoms and levels of C-reactive protein or D-dimer …………………………………………………………………………….04

Results S1. Comparisons between the group of patients who attended the in-person assessments *versus* the remaining surviving individuals who did not participate………….…….04

Results S2. Further results of the exploratory factor analysis.……………………………….........04

Results S3. Results of the sensitivity analyses investigating relationships between the latent variable of PASC and blood test results after exclusion of subgroups stratified by demographic variables.…………………………………………………………………………………………...........05

Discussion D1. Interpretation of findings from the sensitivity analyses using subgroups stratified by demographic variables.…………………………………………………………………….............05

Table S1. Symptoms enquired at the follow-up visit, instruments and cutoffs used……………..06

Table S2. Objective indices of organ system dysfunction ……..……………………………….….08

Table S3. Frequency of positive answers to each symptom enquired at the follow-up visit and missing values…………………………………………………………………………………………...09

Table S4. Frequency of objective signs of organ system dysfunction and new-onset diagnoses at the follow-up visit and missing values………………………………………………………….…..10

Table S5. Comparison between surviving patients who attended the follow-up assessments *versus* subjects who did not take part in the study………………………………………………11

Table S6. One-factor solution from the exploratory factor analysis of persistent symptoms evaluated in COVID-19 patients six to eleven months after hospitalization……………..……12

Table S7. Discrimination of psychiatric and cognitive symptoms in the six Item Response Theory sensitivity analyses that included only one of those variables ……………………………13

Table S8. Symptoms included in the Item Response Theory sensitivity analysis excluding subjects with comorbidities that may be associated with central nervous system symptoms….14

Table S9. Relationships between the latent variable of PASC and blood test results in subgroups stratified by demographic variables ……………………………………………………..15

Table S10. Sensitivity analyses investigating relationships between the latent variable of PASC and blood test results after exclusion of subgroups presenting confounding comorbidities………………………………………………………………………………………...…..16

References …………………………………………………………………………………………..….17

**Methods S1. Further details on study design and procedures**

From 30 March 2020 through August 2020, over 3,500 patients with suspected moderate to severe COVID-19 were admitted as inpatients to *Hospital das Clínicas, Faculdade de Medicina, Universidade de São Paulo* (HCFMUSP) for at least 24 hours. We consecutively invited for the present study all adult individuals (≥18 years) who survived hospital treatment over the above period and who had their etiological diagnosis confirmed by reverse-transcriptase PCR (RT-PCR) on swab-collected nasopharyngeal and/or oropharyngeal samples, or by ELISA to detect serum antibodies (in subjects for whom an RT-PCR test collected up to the 10th day of symptom onset was not available).

All individuals who fulfilled criteria for the study and accepted to undergo the follow-up assessments were evaluated on the same day by multidisciplinary teams who were joined together to collect data in an integrated fashion, minimizing patient inconvenience and optimizing use of resources (Busatto *et al.*, 2021). Data collection occurred during October 2020 until April 2021.

Data from interviews, scales and complementary examinations were captured and stored at real-time using web-based case report forms developed on

A Research Electronic Data Capture (REDCap) system hosted at HCFMUSP (Busatto *et al.*, 2021) provided prospectively collected information on subjects´ in-hospital admissions on comorbidities, acute COVID-19 symptoms, indices of disease severity and events during hospitalization (Busatto *et al.*, 2021).

Additional data were acquired at the time of the follow-up assessment on: socio-economic status classified in six categories (A, B1, B2, C1, C2 and D-E) (ABEP, 2020); self-declared race (IBGE, 2021); years of education; and complementary information regarding comorbidities (Busatto *et al.*, 2021).

**Methods S2. Further details on the assessment of persistent symptoms and objective indices of functioning**

Details on the assessments of the thirty symptoms and scale cutoffs used to generate categorical ‘yes-no’ variables are provided in Table S1.

Objective indices recorded at the follow-up visit and cutoffs are provided in Table S2.

A frontal and lateral chest X-ray (searching for signs suggestive of COVID-related lesions such as ground-glass opacities, consolidation, and linear and reticular opacities) was acquired (Litmanovich *et al.*, 2020). Subjects also underwent a spirometry test as described elsewhere (Pereira et al, 2007); pulmonary capacity was rated as deficient when the forced vital capacity was rated as lower than 80% of predicted.

Physical capacity was assessed by the handgrip test and the 1-minute sit-to-stand test, using age- and sex-corrected cutoffs for impaired performance (Bohannon, 2015; Strassmann *et al.*, 2013; Vianna et al, 2007). Oximetry measurements and the Borg Dyspnea Scale (BORG 1982.pdf, 1982) were undertaken immediately before and after the sit-to-stand test.

In order to ascertain the presence of impaired kidney function at the follow-up assessment, diagnostic blood measurements of creatinine were obtained (see Table S2).

**Methods S3. Details on blood laboratory indices assessed at the follow-up**

Serum levels of C‐reactive protein were measured using a Cobas c702 analyzer (Roche Diagnostics, Basel, Switzerland), based on the latex‐enhanced immunoturbidimetric assay. D-dimer levels were measured in 3.2% citrate plasma by standard turbidimetric method using an ACL TOP 750 automated coagulation analyzer (IL, Werfen, MA, USA). Data were available for most subjects of the sample (n=748 for C-reactive protein, and n=745 for D-dimer).

**Methods S4. Further details on the exploratory factor analysis and missing values**

The adequacy of the data for conducting factor analysis was tested using the Kaiser–Meyer–Olkin measure of sampling adequacy. Factors were extracted using the iterated principal axis method (principal factors) based on an eigenvalue greater than one, with no rotation. We used the likelihood ratio test of independence against the saturated model to verify the meaningfulness of the factor analysis and the inter-correlation of items.

A total of 203 patients had to be excluded from the EFA analysis (27.1% of the total sample) due to missing values for at least one symptom (see Table S4). A proportion of cases lacked data on psychiatric symptoms due to the absence of specialized research teams on circumscribed dates during the overall period of data collection (see Table S4). Missing values for these and other symptoms assessed in the study were also present at random, representing enrollees who occasionally skipped questions (see Table S4).

**Methods S5. Further details on the Item Response Theory analyses**

In the 2-parameter logistic IRT analysis assessing the characteristics of all thirty symptoms of PASC, four iterations were used for fitting the fixed-effects model, and five for fitting the full model.

IRT is a set of mathematical models that attempts to explain the relationship between items (i.e., PASC symptoms – observed) and an underlying latent trait (i.e., PASC syndrome – unobserved) (Saha *et al.,* 2010). In IRT, each binary item included in the model can be assessed regarding its properties of discrimination and severity in an Item Characteristic Curve graph. Both properties are described by their coefficient of the two-parameter logistic IRT model, z score, p value and 95% confidence interval (Krueger *et al.*, 2004). In the present study, our measure of interest was the property of discrimination of each item (i.e. symptom), which refers to the increment in the probability of a given manifestation being scored as positive as the latent dimension score increases; the higher the discrimination of a manifestation, the steeper its curve slope on the graph (maximal discrimination = vertical angle) (Krueger *et al.*, 2004). The property of discrimination for each symptom was determined by the rate at which the probability of presenting the manifestation changed given the latent trait score (at a *P*-value threshold of 0.05) (Krueger *et al.*, 2004); this would allow the ranking of symptoms with basis on how well they distinguished between those subjects who were higher on the continuum and those who were lower on the continuum (Hays *et al*., 2000).

**Methods S6. Further details on the sensitivity analyses evaluating the Item Response Theory model**

We conducted a sensitivity analysis re-assessing the properties of all symptoms included in the model after leaving out subjects with the following comorbidities: chronic lung, liver, kidney and heart diseases (except hypertension); active cancer; HIV; and organ transplantation (n=299, 39.9% of the sample). This was aimed to ascertain whether the discriminative role of psychiatric and cognitive symptoms initially included in the latent variable could have been determined by the presence of those comorbid conditions, which are known to be themselves associated with central nervous system (CNS) manifestations.

Given the high level of comorbidity among psychiatric symptoms and cognitive complaints, there could be a general tendency of inclusion of each of the self-reported manifestations referring to psychiatry and cognition tested in the overall IRT model (as several other psychiatric features would probably be included in the same latent trait). Thus, we conducted six additional sensitivity analyses, each including all multi-organ symptoms but only one psychiatric or cognitive manifestation at a time.

**Methods S7. Further details on the analyses evaluating relationships between PASC symptom severity, objective signs of organ system dysfunction and laboratory test results**

Two-parameter logistic regression analyses were conducted to investigate significant associations between the latent dimension of PASC symptoms and the following eight objective signs of organ system dysfunction: reduced BMI; decreased oxygen saturation (at rest and during effort); abnormal chest X-ray; spirometry findings of forced vital capacity lower than 80% of predicted; low estimated glomerular filtration rate; abnormal sit-to-stand performance; and reduced handgrip strength.

In the Differential Item Functioning (DIF) analyses, we aimed to test whether the relationship between abnormal results in the blood tests and the latent dimension of PASC symptoms were due to different probabilities of endorsing any of the thirty symptoms included in the syndrome (Saha *et al.*, 2010). Initially, we investigated the relationship between blood test results and the value of each subject for the latent dimension of PASC (i.e, syndrome level), with statistical significance assessed using logistic coefficients and 95% confidence intervals. If any significant relationship with the severity of the latent PASC trait was detected, we then tested differential effects at the level of the individual PASC symptoms (i.e., manifestation level) through Mantel-Haenszel tests, calculating adjusted odds ratios, and 95% confidence intervals for each manifestation, as well as chi-square values to verify if any odds ratios were different from unity (p<0.05) (Penfield and Camilli, 2007) 13. If the observed significant differential effect at the manifestation level is found in the same direction as at the syndrome level (+/+ or -/-), it can be established that there is DIF (Saha *et al.*, 2010).

**Methods S8. Further details on the sensitivity analyses evaluating the influence of demographic variables on the relationship between the latent variable of PASC symptoms and levels of C-reactive protein or D-dimer**

In order to verify the degree to which associations between blood test results and the latent variable of PASC symptoms were affected by the influence of demographic variables on levels of C-reactive protein or D-dimer, sensitivity analyses (logistic regressions) were conducted to investigate relationships between the latent variable of PASC and blood test results after leaving out subgroups of subjects stratified by the following demographic variables: sex; age (five strata); race (black, white, or mixed); and socio-economic status (six strata, A-B1-B2-C1-C2-D/E) (ABEP, 2020).

**Results S1. Comparisons between the group of patients who attended the in-person assessments *versus* the remaining surviving individuals who did not participate**

In Table S5, the results of comparisons between the group of patients who attended the in-person assessments *versus* the remaining surviving individuals who did not attend those assessments are displayed.

The two groups did not differ regarding age or sex distribution, but the patients who agreed to undergo the in-person assessments had a significantly greater duration of hospitalization, need for intensive care unit (ICU) stay, need for intubation, and use of renal replacement therapy (Table S5).

Rates of smoking, hypertension, and rheumatological disease were also significantly higher in the patients who attended the follow-up visits (Table S5).

**Results S2. Further results of the exploratory factor analysis**

The data compiled on the thirty symptoms for the subjects who did not present missing values (n=546) and who were selected for the initial EFA were associated with an overall Kaiser–Meyer–Olkin value greater than 0.8 (kmo= 0.8305), indicating adequacy for conducting factor analysis. The likelihood ratio test of independence against the saturated model was significant (chi2(435)=2883.67, p=<0.0001), indicating that the EFA was meaningful and the items inter-correlated.

**Results S3. Results of the sensitivity analyses investigating relationships between the latent variable of PASC and blood test results after exclusion of subgroups stratified by demographic variables**

Results remained significant in all sensitivity analyses investigating relationships between the latent variable of PASC and blood levels of C-reactive protein after exclusion of each subgroup stratified by demographic variables, including: sex (coefficients>0.270, p≤0.040); age (coefficients>0.335, p<0.001); race (coefficients>0.345, p≤0.004); and socio-economic status (coefficients>0.370, p<0.001). Regarding D-dimer, results remained significant in the following sensitivity analyses: female sex excluded (coefficient=0.644, p<0.001); exclusion of each age stratum except for the range between 60 and 69 years (coefficients>0.395, p≤0.040); black or mixed races excluded (coefficients>0.435, p≤0.007); and exclusion of each socio-economic stratum except for the C1 stratum (coefficients>0.320, p≤0.040).

Associations between the latent trait of PASC symptoms and blood biomarkers in each subgroup stratified by demographic variables are detailed in Table S9. Results are not shown for the A, B1 and D-E socio-economic subgroups, since each of those categories included less than 10% of the overall sample.These results show that the direct association between C-reactive protein levels and the latent trait of PASC symptoms was significant in the following subgroups: female sex; male sex; B2, C1 and C2 socio-economic strata (which accounted for 82.4% of the overall sample); and the subgroups aged 40-49 years, 50-59 years, and 60-69 years (which accounted for 69% of the overall sample) (Table S9). Regarding race, the direct association between C-reactive protein levels and the latent PASC trait remained significant in the mixed and white subgroups, but not in the black subgroup (which accounted for 13.6% of the overall sample). The direct association between D-dimer levels and the latent PASC trait was significant in the following subgroups: males but not females; and the subgroups aged 50-59 years and 60-69 years (Table S9). Regarding race and socio-economic status, the direct association between D-dimer levels and the latent PASC trait was significant only in the white subgroup and the C1 socio-economic subgroup (Table S9).

**Discussion D1. Interpretation of findings from the sensitivity analyses using subgroups stratified by demographic variables**

Statistical significance was retained in all sensitivity analyses investigating relationships between the latent variable of PASC and blood levels of C-reactive protein after exclusion of each subgroup stratified by demographic variables, strongly suggesting that the relationship between C-reactive protein levels and PASC symptoms reported in the present study was not determined by confounding influences of these factors. Significant associations between C-reactive protein levels and the latent trait of PASC symptoms were found in most subgroups stratified by demographic variables (Table S9), except for the black race (which accounted for only 13.6% of the overall sample).

Conversely, demographic variables were found to affect the association between D-dimer levels and PASC symptoms to a greater degree, with significant findings restricted to the male sex, white race and subjects at the age strata from 50 to 59 years and 60 to 69 years.

**Table S1. Symptoms enquired at the follow-up visit, instruments and cutoffs used**

|  |  |  |  |
| --- | --- | --- | --- |
| **Symptom** | **Assessment instrument** | **Cutoff** | **Procedure to ascertain if symptoms emerged before or after COVID-19** |
| Muscle / joint pain | Visual-analogue scale a | Score ≥ 65 a | Yes-no questioning  |
| Fatigue | Functional Assessment of Chronic Illness Therapy - Fatigue Scale b | Score ≤ 39 b | CIS-R interview c |
| Post traumatic stress | PTSD Checklist d (enquiring about COVID-related symptoms)  | Score ≥ 45 d | n.a. |
| Memory loss | Memory Complaint Scale e (adapted for COVID-related complaints)  | Score ≥ 7 e | Memory Complaint Scale e |
| Insomnia | Insomnia Severity Index f | Score ≥ 8 f | CIS-R interview c |
| Dyspnea | Medical Research Council Dyspnea Scale g | Score ≥ 2 g | Yes-no questioning |
| Anxiety | Hospital Anxiety and Depression Scale h | Sub-score > 8 h | CIS-R interview c |
| Loss of taste  | Visual-analogue scale i | Score ≤80 i | Yes-no questioning |
| Depression | Hospital Anxiety and Depression Scale h | Sub-score > 8 h | CIS-R interview c |
| Loss of smell | Visual-analogue scale i | Score ≤80 i | Yes-no questioning |
| Dizziness | Direct "yes-no" questioning | n.a. | Yes-no questioning |
| Body pain | Direct "yes-no" questioning | n.a. | Yes-no questioning |
| Lack of concentration | CIS-R interview c | n.a. | CIS-R interview c |
| Nocturia | Direct "yes-no" questioning | n.a. | Yes-no questioning |
| Chest pain | Direct "yes-no" questioning | n.a. | Yes-no questioning |
| Cough | Direct "yes-no" questioning | n.a. | Yes-no questioning |
| Edema | Direct "yes-no" questioning | n.a. | Yes-no questioning |
| Nasal obstruction | Nasal obstruction symptom evaluation scale j | Positive response to any of the scale questions | Yes-no questioning |
| Paresthesia | WHO screening tool for neuroepidemiology investigations in LMIC k | n.a. | Yes-no questioning |
| Skin problems | Direct "yes-no" questioning | n.a. | Yes-no questioning |
| Tinnitus | Direct "yes-no" questioning | n.a. | Yes-no questioning |
| Hearing loss | Direct "yes-no" questioning | n.a. | Yes-no questioning |
| Abdominal pain | Direct "yes-no" questioning | n.a. | Yes-no questioning |
| Weakness | WHO screening tool for neuroepidemiology investigations in LMIC k | n.a. | Yes-no questioning |
| Loss of appetite | Direct "yes-no" questioning | n.a. | CIS-R interview c |
| Gait impairment | WHO screening tool for neuroepidemiology investigations in LMIC k | n.a. | Yes-no questioning |
| Headache | WHO screening tool for neuroepidemiology investigations in LMIC k | n.a. | Yes-no questioning |
| Diarrhea | Direct "yes-no" questioning | n.a. | Yes-no questioning |
| Episodes of loss of consciousness | WHO screening tool for neuroepidemiology investigations in LMIC k | n.a. | Yes-no questioning |
| Nausea / vomiting | Direct "yes-no" questioning | n.a. | Yes-no questioning |

Abbreviations: CIS-R, Clinical Interview Schedule- Revised version; PTSD, post-traumatic stress disorder; n.a., not applicable; WHO, World Health Organization; LMIC, Lower middle-income countries.

a: Boonstra *et al.*, 2014; b: Webster *et al.*, 2003; c: Lewis *et al.*, 1992; d: Weathers *et al*, 1993; e: Vale et al, 1999; f: Bastien *et al.*, 2001; g: Bestall *et al.*, 1999; h: Zigmond and Snalth, 1983; i: Brandão Neto *et al.*, 2021; j: Stewart *et al*, 2004; k: WHO working group, 2020.

**Table S2. Objective indices of organ system dysfunction**

|  |  |
| --- | --- |
| **Variable** | **Index of dysfunction** |
| Weight loss | ≤ 5% of BMI value prior to COVID-19 a |
| Weight increase | > 5% of BMI value prior to COVID-19 a |
| Brachial perimeter, cm | ≤23 for men, ≤ 22 for women |
| Reduced resting oxygen saturation (≤ 90%) b | SpO2 at rest ≤ 90%  |
| Reduced resting oxygen saturation (≤ 92%) b | SpO2 at rest ≤ 92% |
| Reduced resting oxygen saturation (≤ 95%) b | SpO2 at rest ≤ 95% |
| Decreased oxygen saturation during effort c | SpO2 reduction of ≥4 points during exercising (sit-to-stand test) |
| Reduced exercise capacity | Reduced number of repetitions during sit-to-stand test (cutoff corrected for age and gender) d |
| Reduced muscle strength | Reduced handgrip strength (cutoff corrected for age and gender) e |
| Pulmonary abnormalities suggestive of COVID-19 | COVID-related changes at X-Ray f |
| Reduced lung functional capacity | FVC <80% of predicted at spirometry test  |
| Reduced kidney functional capacity g  | eGFR lower than 60 mL/min/1.73 m² (in subjects with no previous history of kidney disease)  |

Abbreviations: BMI, Body mass index; SpO2, Oxygen saturation; FVC, Forced vital capacity; eGFR, Estimated glomerular filtration rate.

a Difference between current BMI measurement and calculated BMI based on self-reported estimates of body weight prior to COVID-19.

b Three different cutoffs were tested.

c Not undertaken in subjects presenting resting pulse oximetry ratings lower than 90%.

d Given the lack of studies evaluating 1-minute sit-to-stand test performance in representative Brazilian samples, a conservative cutoff strategy was applied, using as guidance data from a population-based investigation in Europe (n= 6,926, age range 20-79 years)8. Male subjects were rated as presenting reduced sit-to-stand performance when they had a number of repetitions lower than the 2.5 percentile rating reported for each 10-year age interval by Strassman et al (2013), with a three-point lower value at each 10-year age interval applied for females; due to the lack of normative data for subjects from 80 to 89 years, a cutoff value three points lower than the cutoff applied for the interval between 70-79 years was used for this age interval.

e Vianna *et al*, 2007

f Litmanovich *et al.*, 2020

g Estimated glomerular filtration rate calculated according to CKD EPI30.

**Table S3. Frequency of objective signs of organ system dysfunction at the follow-up visit and missing values**

|  |  |  |
| --- | --- | --- |
| **Dysfunction**  | **n (% of the overall sample)** | **Missing values** |
| Reduced handgrip strength | 462 (63.6%) | 23 |
| Reduced sit-to-stand performance | 375 (55.6%) | 73 |
| Low FVC 80% predicted at spirometry test | 205 (32%) | 108 |
| Abnormal X-ray | 182 (29%) | 122 |
| Weight loss | 200 (27%) | 9 |
| Weight increase | 185 (25%) | 9 |
| Reduced resting oxygen saturation (≤ 95%, ≤ 92%, ≤ 90%) | 172 (23%), 52(7%), 23 (3%) | 3 |
| Decreased eGFR  | 138 (18.5%) | 7 |
| Decreased oxygen saturation during effort | 68 (10%) | 73  |
| Reduced brachial perimeter | 8 (1%) | 2 |

Abbreviations: BMI, Body mass index; FVC, Forced vital capacity; eGFR, Estimated glomerular filtration rate.

**Table S4. Frequency of positive answers to each symptom enquired at the follow-up visit and missing values**

|  |  |  |
| --- | --- | --- |
| **Symptom** | **Frequency of incident and persistent symptom, n (% of the overall sample)** | **Missing values** |
| Muscle / joint pain | 299 (40.7%) | 15 |
| Fatigue | 286 (38.3%) | 3 |
| Dizziness | 264 (35.7%) | 11 |
| Post traumatic stress | 102 (13.8%) | 14 |
| Memory loss | 239 (34.7%) | 60 |
| Body pain | 248 (33.6%) | 12 |
| Insomnia | 242 (32.4%) | 3 |
| Lack of concentration | 208 (31%) | 79 |
| Dyspnea | 218 (29.5%) | 8 |
| Anxiety | 176 (26.3%) | 78 |
| Nocturia | 176 (24.1%) | 19 |
| Loss of taste | 161 (22.5%) | 34 |
| Depression | 146 (21.7%) | 78 |
| Loss of smell | 150 (20.6%) | 22 |
| Chest pain | 143 (19.7%) | 23 |
| Cough | 139 (18.8%) | 14 |
| Edema | 129 (17.6%) | 18 |
| Nasal obstruction | 118 (16.2%) | 20 |
| Paresthesia | 116 (15.8%) | 14 |
| Skin problems | 113 (15.3%) | 11 |
| Tinnitus | 110 (14.9%) | 12 |
| Hearing loss | 106 (14.4%) | 12 |
| Abdominal pain | 101 (13.7%) | 13 |
| Weakness | 96 (13%) | 13 |
| Loss of appetite | 91 (12.3%) | 10 |
| Gait impairment | 83 (11.3%) | 13 |
| Headache | 80 (10.9%) | 13 |
| Diarrhea | 44 (6%) | 16 |
| Episodes of loss of consciousness | 27 (3.7%) | 18 |
| Nausea / vomiting | 24 (3.2%) | 13 |

**Table S5. Comparison between surviving patients who attended the follow-up assessments *versus* subjects who did not take part in the study**

|  |  |  |  |
| --- | --- | --- | --- |
| **Variable** | **Patients who attended (N= 749)** | **Non-participants (N=976)** | **Statistics** |
| **Age** - mean ± SD, years | 55 ± 14 | 54± 17 | t= 1.07, p=.28 |
| **Sex –** Male; Female | 397 (53%); 352 (47%) | 501 (51%); 475 (48.7%) | χ2= 0.48, p=.49 |
| **Comorbidities**  |   |  |  |
| Charlson comorbidity score – mean ± SD  | 3.0 ± 1.8 | 3.1 ± 2.0 | t= -1.18, p=.23 |
| Hypertension | 425 (56.7%) | 492 (50.4%) | χ2= 6.71, p=.01 |
| Chronic cardiovascular disease | 136 (18.2%) | 151 (15.5%) | χ2= 2.42, p=.29 |
| Diabetes |  261 (34.8%) | 313 (32%) | χ2= 1.44, p=.23 |
| Chronic respiratory disease | 58 (7.7%) | 91 (9.3%) | χ2= 1.34, p=.24 |
| Cerebrovascular disease | 40 (5.3%) | 47 (4.8%) | χ2= 0.25, p=.61 |
| Chronic kidney disease (dialytic) | 35 (4.7%) | 22 (2.2%) | χ2= 2.78, p=.09 |
| Chronic kidney disease (non-dialytic) | 49 (6.5%) | 68 (7%) | χ2= 0.12, p=.72 |
| Chronic liver disease | 26 (3.5%) | 22 (2.3%) | χ2= 2.32, p=.12 |
| Rheumatological disease | 31 (4.1%) | 21 (2.1%) | χ2= 5.73, p=.017 |
| HIV | 4 (0.5%) | 13 (1.3%) | χ2= 2.76, p=.097 |
| Cancer | 35 (4.7%) | 55 (5.6%) | χ2= 1.59, p=.20 |
| Organ transplantation | 35 (4.7%) | 30 (3%) | χ2= 2.99, p=.084 |
| **Smoking** | 284 (38%) | 180 (18.4%) | χ2= 82.06, p<.001 |
| **Duration of COVID-19 symptoms (in days)** - mean ± SD | 9.0 ± 6.5 | 8.8 ± 6.2 | t= 0.76, p=.44 |
| **Events during hospitalization** |  |  |  |
| WHO clinical progression scale – frequency in different categories a  |  |  | χ2= 46.05, p<.001 |
| 3-4 | 85 (11.3%) | 217 (22.2%) |  |
| 5 | 327 (43.6%) | 429 (44%) |  |
| 6 | 32 (4.3%) | 47 (4.8%) |  |
| 7+8+9 | 305 (40.7%) | 283 (29%) |  |
| Hospital stay, duration in days - mean ± SD | 18.6 ± 19.2  | 15.1 ± 16.9 | t= 4.02, p<.001 |
| Renal replacement therapy (YES/NO) | 96 (12.8%) | 68 (7%) | χ2= 16.86, p<.001 |
| ICU stay (YES/NO) | 445 (59.4%) | 492 (50.4%) | χ2= 13.84, p<.001 |
| Intubation (YES/NO) | 305 (40.7%) | 283 (30%) | χ2= 5.72, p=.017 |

Abbreviations: SD, standard-deviation; t, t-test value (unpaired, two=tailed); χ2, chi-square test; IQR, interquartile range; WHO, World Health Organization; ICU, intensive care unit.

a WHO scale categories (WHO working group, 2020): 3-4, no continuous supplemental oxygen needed; 5, continuous supplemental oxygen only; 6, Continuous Positive Airway Pressure ventilation, Bi-Level Positive Airway Pressure or High Flow Nasal Oxygen; 7-9, Invasive Mechanical Ventilation and/or Extra-Corporeal Membrane Oxygenation (ECMO).

**Table S6. One-factor solution from the exploratory factor analysis of persistent symptoms evaluated in COVID-19 patients six to eleven months after hospitalization**

|  |  |  |  |
| --- | --- | --- | --- |
| **Symptom** | **Factor loading** | **Eingenvalue** | **Explained variance** |
|  |  |  |  |
| Memory loss | 0.6141  | 4.649 | 64.76% |
| Anxiety | 0.5941  |  |  |
| Depression | 0.5874  |  |  |
| Lack of concentration | 0.5620  |  |  |
| Post traumatic stress | 0.5575  |  |  |
| Insomnia | 0.5352  |  |  |
| Fatigue | 0.6389  |  |  |
| Dyspnea | 0.4597  |  |  |
| Body pain  | 0.4253  |  |  |
| Chest pain | 0.2615  |  |  |
| Muscle / joint pain | 0.4063  |  |  |
| Cough | 0.1953  |  |  |
| Abdominal pain | 0.2727  |  |  |
| Nausea / vomiting | 0.2595  |  |  |
| Diarrhea | 0.1770  |  |  |
| Loss of appetite  | 0.3974  |  |  |
| Loss of smell  | 0.3970  |  |  |
| Loss of taste | 0.4081  |  |  |
| Hearing loss | 0.3707  |  |  |
| Nasal obstruction | 0.2786  |  |  |
| Dizziness | 0.3109  |  |  |
| Tinnitus  | 0.3829  |  |  |
| Paresthesia | 0.1832  |  |  |
| Episodes of loss of consciousness  | 0.0990  |  |  |
| Gait impairment  | 0.2147  |  |  |
| Headache | 0.2143  |  |  |
| Weakness | 0.2835  |  |  |
| Nocturia | 0.3650  |  |  |
| Edema | 0.2862  |  |  |
| Skin problems | 0.2096  |  |  |

**Table S7. Discrimination of psychiatric and cognitive symptoms in the six Item Response Theory sensitivity analyses that included only one of those variables**

|  |  |
| --- | --- |
|  | **Discrimination** |
|  | Coefficient  | *P* | 95%CI |
| Memory loss | 1.57 |  | 1.23 | 1.92 |
| Post traumatic stress | 1.45 |  | 1.08 | 1.82 |
| Insomnia | 1.40 | 0.000  | 1.10 | 1.70 |
| Depression | 1.38 |  | 1.04 | 1.72 |
| Anxiety | 1.37 |  | 1.05 | 1.69 |
| Lack of concentration | 1.20 |  | .92 | 1.49 |

Abbreviations: CI, confidence interval.

**Table S8. Symptoms included in the Item Response Theory sensitivity analysis excluding subjects with comorbidities that may be associated with central nervous system symptoms**

|  |  |  |  |
| --- | --- | --- | --- |
|  |  |  |  |
|  | **Discrimination** |
|  | Coefficient | *P* | 95%CI |
| **Psychiatric/ cognitive symptoms** |  |  |  |  |
| Post traumatic stress | 2.101 |  | 1.592 | 2.610 |
| Depression | 2.047 |  | 1.581 | 2.513 |
| Memory loss | 2.026 | 0.000 | 1.600 | 2.452 |
| Anxiety | 1.953 |  | 1.526 | 2.380 |
| Lack of concentration | 1.667 |  | 1.308 | 2.027 |
| Insomnia | 1.524 |  | 1.206 | 1.842 |
| **Other symptoms** |  |  |  |  |
| Fatigue | 2.290 |  | 1.821 | 2.760 |
| Nausea / vomiting | 1.394 |  | .831 | 1.958 |
| Loss of appetite | 1.339 |  | .984 | 1.694 |
| Dyspnea | 1.298 |  | 1.012 | 1.584 |
| Body pain | 1.124 |  | .870 | 1.378 |
| Hearing loss | 1.097 |  | .793 | 1.401 |
| Muscle / joint pain | 1.081 |  | .837 | 1.325 |
| Diarrhea | 1.059 |  | .658 | 1.459 |
| Loss of smell | 1.058 |  | .785 | 1.332 |
| Loss of taste | 1.030 |  | .763 | 1.297 |
| Tinnitus | .967 |  | .682 | 1.251 |
| Abdominal pain | .949 | 0.000  | .661 | 1.237 |
| Weakness | .858 |  | .573 | 1.143 |
| Nocturia | .767 |  | .539 | .996 |
| Nasal obstruction | .757 |  | .501 | 1.014 |
| Gait impairment | .740 |  | .456 | 1.025 |
| Headache | .700 |  | .416 | .985 |
| Dizziness | .693 |  | .489 | .897 |
| Chest pain | .676  |  | .444 | .909 |
| Edema | .672 |  | .433 | .910 |
| Paresthesia | .671 |  | .423 | .920 |
| Episodes of loss of consciousness | .662 | 0.004 | .215 | 1.108 |
| Skin problems | .592 | 0.000  | .349 | .835 |
| Cough | .508 |  | .287 | .729 |

Abbreviations: CI, confidence interval.

|  |  |
| --- | --- |
|  | **Table S9.** **Relationships between the latent variable of PASC and blood test results in subgroups stratified by demographic variables** |
|  |  | **C-reactive protein > 3.0 mg/L**  | **D- dimer > 2.000ng/ml**  |  |
| **Variables** | **N** | **Coefficient** | ***P*** | **95%CI** | **Coefficient** | ***P*** | **95%CI** |
| ***Sex*** |  |  |  |  |  |  |  |  |  |
| Female | 352 | 1.333 | <.001 | 0.895 | 1.772 | 0.293 | .320 | -0.285 | 0.870 |
| Male | 397 | 0.305 | .007 | 0.083 | 0.526 | 0.644 | <.001 | 0.327 | 0.960 |
| ***Age*** |  |  |  |  |  |  |  |  |  |
| 18-39 years | 119 | 0.209 | .279 | -0.163 | 0.567 | npc | npc | npc | npc |
| 40-49 years | 156 | 0.490 | .005 | 0.144 | 0.836 | 0.111 | .071 | -0.464 | 0.686 |
| 50-59 years | 173 | 0.518 | .012 | 0.112 | 0.924 | 0.517 | .006 | 0.147 | 0.887 |
| 60-69 years | 190 | 0.465 | .002 | 0.175 | 0.755 | 0.700 | .018 | 0.119 | 1.281 |
| ≥70 years | 111 | -0.005 | .985 | -0.477 | 0.469 | 0.194 | .640 | -0.620 | 1.088 |
| ***Race*** |  |  |  |  |  |  |  |  |  |
| Black | 102 | 0.172 | .507 | -.336 | 0.681 | -0.061 | .902 | -1.032 | 0.910 |
| Mixed | 273 | 0.414 | .003 | 0.141 | 0.687 | 0.225 | .089 | -0.030 | 0.484 |
| White | 342 | 0.393 | .001 | 0.162 | 0.623 | 0.661 | .039 | 0.032 | 1.289 |
| ***Socio-economic status*** |  |  |  |  |  |  |  |  |  |
| B2 | 137 | 0.407 | .038 | 0.023 | 0.791 | 0.464 | .119 | -0.119 | 1.047 |
| C1 | 243 | 0.387 | .005 | 1.088 | 2.399 | 0.422 | .046 | 0.007 | 0.837 |
| C2 | 227 | 0.391 | .016 | 0.074 | 0.708 | 0.366 | .105 | -0.077 | 0.810 |

Abbreviations: CI, confidence interval; npc, not possible to calculate coefficient due to insufficient observations in the subgroup of patients with D-dimer levels higher than 2.000ng/ml.

|  |
| --- |
| **Table S10. Sensitivity analyses investigating relationships between the latent variable of PASC and blood test results after exclusion of subgroups presenting confounding comorbidities** |
|  | **C-reactive protein > 3.0 mg/L**  | **D- dimer > 2.000ng/ml**  |  |
| **Excluded comorbidities** | **Coefficient** | ***P*** | **95%CI** | **Coefficient** | ***P*** | **95%CI** |
| Pro-inflammatory conditions | 0.340 | <.001 | 0.153 | 0.526 | 0.41 | .012 | 0.092 | 0.734 |
| Chronic pulmonary conditions | 0.378 |  | 0.208 | 0.548 | 0.44 | .006 | 0.122 | 0.748 |
| Diabetes mellitus | 0.280 | .006 | 0.082 | 0.479 | 0.54 | .013 | 0.114 | 0.967 |
| Chronic cardiological conditions | 0.389 |  | 0.209 | 0.568 | 0.41 | .014 | 0.084 | 0.734 |
| Chronic kidney failure | 0.417 | ≤.001 | 0.247 | 0.587 | 0.43 | .002 | 0.150 | 0.701 |
| Clinical conditions commonly associated with central nervous system manifestations | 0.346 |  | 0.138 | 0.555 | 0.40 | .046 | 0.074 | 0.799 |
| Obesity | 0.362 | .006 | 0.105 | 0.619 | 0.31 | .016 | 0.059 | 0.567 |

Abbreviations: CI, confidence interval.

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