SARS-CoV-2 Viremia is Associated With Inflammatory, But Not Cardiovascular Biomarkers, in Patients Hospitalized for COVID-19

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BACKGROUND: COVID-19 may present with a variety of cardiovascular manifestations, and elevations of biomarkers reflecting myocardial injury and stress are prevalent. SARS-CoV-2 has been found in cardiac tissue, and myocardial dysfunction post-COVID-19 may occur. However, the association between SARS-CoV-2 RNA in plasma and cardiovascular biomarkers remains unknown.

METHODS AND RESULTS: COVID MECH (COVID-19 Mechanisms) was a prospective, observational study enrolling consecutive, hospitalized patients with laboratory-confirmed infection with SARS-CoV-2 and symptoms of COVID-19. Biobank plasma samples used to measure SARS-CoV-2 RNA and cardiovascular and inflammatory biomarkers were collected in 123 patients at baseline, and in 96 patients (78%) at day 3. Patients were aged 60±15 (mean ± SD) years, 71 (58%) were men, 68 (55%) were White, and 31 (25%) received mechanical ventilation during hospitalization. SARS-CoV-2 RNA was detected in plasma from 48 (39%) patients at baseline. Patients with viremia were more frequently men, had more diabetes mellitus, and lower oxygen saturation. Patients with viremia had higher concentrations of interleukin-6, C-reactive protein, procalcitonin, and ferritin (all <0.001), but comparable levels of cTnT (cardiac troponin T; P=0.09), NT-proBNP (N-terminal pro-B-type natriuretic peptide; P=0.27) and D-dimer (P=0.67) to patients without viremia. SARS-CoV-2 RNA was present in plasma at either baseline or day 3 in 50 (52%) patients, and these patients experienced increase from baseline to day 3 in NT-proBNP and D-dimer concentrations, while there was no change in cTnT.

CONCLUSIONS: SARS-CoV-2 viremia was associated with increased concentrations of inflammatory, but not cardiovascular biomarkers. NT-proBNP and D-dimer, but not cTnT, increased from baseline to day 3 in patients with viremia.

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and dysfunction, remains unknown. We hypothesized that presence of SARS-CoV-2 RNA in plasma (viremia) is associated with higher concentrations of cardiovascular biomarkers reflecting thromboembolism, myocardial injury and dysfunction.

METHODS

The prospective, observational COVID MECH (COVID-19 Mechanisms; NCT04314232) study enrolled consecutive adult patients hospitalized with COVID-19 from March 18 to May 4, 2020 at Akershus University Hospital, Norway. Patients with laboratory-confirmed COVID-19 as the reason for admission were eligible for inclusion. This was defined by SARS-CoV-2 in combined nasopharyngeal and oropharyngeal swabs determined by real-time polymerase chain reaction test with cycle threshold as described by Corman et al. (2020). Using the ABI7500 Systems (Thermo Fisher Scientific), and COVID-19 symptoms (ie, cough, fever, dyspnea, or flu-like symptoms) as the main reason for admission. Data from the COVID MECH study cannot be publicly shared because of risk of violating privacy, as regulated by the institutional Data Protection Officer.

Study-specific consent forms were signed by participants, or next-of-kin for patients unable to consent. The study was approved by the Regional Ethics Committee of Norway (REK South-East C, Ref. no. 117589) and by the institutional Data Protection Officer (Ref.no. 20/02873). Clinical information was extracted from medical records by the investigators. All patients receiving mechanical ventilation were treated in the intensive care unit (ICU). Some additional patients were briefly evaluated at the ICU and thereafter returned to the bed ward, if the intensive care physicians decided there was no emergent need for mechanical ventilation.

Blood samples were drawn by venipuncture at baseline (emergency room admission or early at the ICU) and 2–5 days later (target day 3). D-dimer and C-reactive protein were analyzed immediately at the hospital central laboratory. Biobank serum samples were temporarily stored at 4°C, centrifuged and transferred into aliquots that were stored at −80°C. Interleukin-6, procalcitonin, ferritin, cTnT (cardiac troponin T) and NT-proBNP (N-terminal pro-B-type natriuretic peptide) were analyzed from never previously thawed anonymized serum samples from 10 people without SARS-CoV2 and demonstrated no false positive test results. Patients were classified with viremia if SARS-CoV-2 RNA was detected (above the limit of detection).

Baseline characteristics are presented as mean±SD and n (%) and compared by Student’s t-test and chi-square test. Biomarker concentrations are reported as median (quartile 1, quartile 3 [Q1, Q3]) and compared by the non-parametric Kruskal–Wallis test. Changes in biomarkers concentrations from baseline to day 3 in patients with and without viremia were assessed using Wilcoxon matched-pairs signed-rank test. Using viremia as the exposure variable, log-transformed biomarker concentrations at baseline and log-transformed delta values from baseline to day 3 were analyzed in univariable and multivariable linear regression models, adjusting for age, sex, race, and a history of cardiovascular disease.

RESULTS

Baseline measurements of plasma SARS-CoV-2 RNA and cardiovascular and inflammatory biomarkers were available in 123 of 135 patients from the COVID MECH study. Missing blood samples were attributable to logistic challenges related to blood sampling in the emergency room or ICU. In total, 201 patients with a positive SARS-CoV-2 test were admitted to the hospital until May 4, and 66 of these were not included in the study as they were discharged immediately from the emergency room, admitted before study initiation or did not have COVID-19 symptoms. Among eligible patients, <10 declined study participation or were not recruited because of logistic challenges. Mean (±SD) age in the final study population (n=123) was 60±15 years (range, 25–87 years), 71 (58%) were men, 68 (55%) were White patients, and the mean body mass index was 28.3±5.5 kg/m². Thirty-one (25%) received mechanical ventilation at the ICU during
SARS-CoV-2 RNA was detected in plasma from 48 (39%) patients at baseline, and among these, 24 (41%) had concentrations between the limit of quantification and limit of detection. Patients with viremia were more frequently men (69% versus 51%, P=0.048) and had more diabetes mellitus (39% versus 9%, P=0.004) (Table 1). Age, race, body mass index, smoking, hypertension, cardiovascular disease, pulmonary disease, renal disease, and the use of renin-angiotensin-aldosterone-system inhibitors did not differ by viremia. Mean symptom duration was 9.3±4.6 days, with no difference by presence of SARS-CoV-2 viremia (P=0.28). Patients with viremia had lower oxygen saturation at baseline (91% versus 94%, P<0.001), but comparable temperature, heart rate, respiratory rate, and blood pressure to patients without viremia.

Concentrations of inflammatory biomarkers were elevated in the total population (median [Q1, Q3]: interleukin-6, 49 [20, 75] pg/L; C-reactive protein, 60 (30, 130) mg/L; procalcitonin, 0.14 (0.7, 0.24) µg/L; and ferritin, 477 [238, 985] µg/L. Patients with viremia had higher concentrations of inflammatory markers, also after adjusting for age, sex, race, and cardiovascular disease (Table 2). Cardiovascular biomarker concentrations in the total population (median [Q1, Q3]) were: cTnT, 9 (5, 16) ng/L; NT-proBNP, 100 (36, 285) ng/L; and D-dimer, 0.5 (0.3, 1.0) mg/L. cTnT and NT-proBNP were above upper-reference limits in 45 (37%) and 45 (37%) patients, respectively, indicating myocardial injury and stress. Baseline concentrations of cTnT, NT-proBNP, and D-dimer did not differ between patients with and without viremia, and these findings were consistent in adjusted models (Table 2). More patients with viremia had NT-proBNP above upper-reference limits (50% versus 28%, P=0.013), while no such difference was seen for cTnT (42% versus 33%, P=0.35). Among patients with viremia, there was no association between levels of plasma SARS-CoV-2 RNA and concentrations of cTnT (P=0.82), NT-proBNP (P=0.36), or D-dimer (P=0.71).

Table 1. Baseline Characteristics of Patients With and Without SARS-CoV-2 Viremia

|                      | COVID-19 Without Viremia (n=75) | COVID-19 With Viremia (n=48) | P Value |
|----------------------|--------------------------------|------------------------------|---------|
| Age, y               | 58.4±16.3                      | 61.5±13.2                    | 0.26    |
| Male sex             | 38 (50.7%)                     | 33 (68.8%)                   | 0.048   |
| White                | 46 (61.3%)                     | 22 (45.8%)                   | 0.09    |
| Body mass index, kg/m² | 27.9±4.0                      | 29.0±7.2                    | 0.28    |
| Diabetes mellitus    | 7 (9.3 %)                      | 14 (29.2%)                   | 0.004   |
| Hypertension         | 21 (28.4%)                     | 18 (38.3%)                   | 0.26    |
| Cardiovascular disease | 11 (14.7%)                  | 7 (14.6%)                    | 0.99    |
| Pulmonary disease    | 2 (2.7 %)                      | 4 (8.3 %)                    | 0.15    |
| Chronic kidney disease | 6 (8.0 %)                     | 3 (6.3 %)                    | 0.72    |
| RAAS-inhibitors use  | 18 (24.0%)                     | 17 (35.4%)                   | 0.17    |
| Smoking              | 5 (6.7 %)                      | 1 (2.1 %)                    | 0.26    |
| Symptom duration, d  | 9.7±5.0                        | 8.8±3.6                     | 0.28    |
| Fever                | 59 (78.7%)                     | 41 (85.4%)                   | 0.35    |
| Cough                | 58 (77.3%)                     | 40 (83.3%)                   | 0.42    |
| Dyspnea              | 48 (64.0%)                     | 38 (79.2%)                   | 0.07    |
| Temperature (°C)     | 38.1±0.9                       | 38.2±0.9                    | 0.54    |
| Heart rate, /min     | 90.0±18.4                      | 90.3±13.6                   | 0.91    |
| Respiratory rate, /min | 25.7±8.2                     | 28.4±9.0                    | 0.09    |
| Blood pressure, mmHg | 134.5±19.2                     | 128.2±17.2                  | 0.06    |
| Oxygen saturation, % | 94.2±3.0                       | 90.7±7.8                    | <0.001  |

RAAS indicates renin-angiotensin-aldosterone-system.

Table 2. SARS-CoV-2 RNA in Plasma (Viremia) and Baseline Concentrations of Inflammatory and Cardiovascular Biomarkers in Unselected Patients Hospitalized for COVID-19 (n=123)

|                    | COVID-19 Without Viremia (n=75) | COVID-19 With Viremia (n=48) | Unadjusted Linear Regression | Linear Regression Adjusted for Age, Sex, Race, and CVD |
|--------------------|--------------------------------|------------------------------|-----------------------------|--------------------------------------------------------|
| IL-6, pg/mL        | 27.2 [14.8, 51.9]               | 57.9 [39.5, 115.0]           | 0.90 (0.45–1.35)            | <0.001                                                  |
| CRP, mg/L          | 49 [24, 100]                   | 105 [55, 200]                | 0.87 (0.45–1.30)            | <0.001                                                  |
| Procalcitonin , µg/L | 0.10 [0.06, 0.18]          | 0.19 [0.09, 0.49]            | 0.63 (0.23–1.02)            | <0.001                                                  |
| Ferritin, µg/L     | 376 [185, 776]                 | 691 [37, 1588]              | 0.72 (0.34–1.10)            | <0.001                                                  |
| D-dimer, mg/L      | 0.5 [0.4, 1.0]                 | 0.6 [0.3, 1.0]               | 0.07 (−0.26 to 0.40)        | 0.67                                                   |
| cTnT, ng/L         | 7.0 [4.0, 15.0]                | 11.5 [7.0, 20.5]             | 0.35 (−0.06 to 0.76)        | 0.09                                                   |
| NT-proBNP, ng/L    | 86 [34, 213]                  | 161 [65, 317]               | 0.34 (−0.26 to 0.96)        | 0.27                                                   |

Concentrations are median [quartile 1, quartile 3]. Biomarker values were log-transformed in the adjusted regression models. CRP indicates C-reactive protein; cTnT, cardiac troponin T; CVD, cardiovascular disease; IL-6, interleukin 6; and NT-proBNP, N-terminal pro-B-type natriuretic peptide.
On day 3, 96 (78%) patients had available blood samples. SARS-CoV-2 RNA was present in plasma at either baseline or day 3 in 50 (52%) patients. In patients without SARS-CoV-2 viremia at either time point there was no significant change in NT-proBNP (+14 [Q1, Q3 −21, 75] ng/L, P=0.14) and D-dimer (0 [−0.3, +0.1] mg/L, P=0.51) from baseline to day 3 (Figure). In contrast, patients with SARS-CoV-2 viremia experienced a significant increase from baseline to day 3 in NT-proBNP (median +79 [Q1, Q3 +2, +268] ng/L, P<0.001) and D-dimer (+0.1 [0, +0.4], P<0.001) mg/L (Figure). The presence of viremia was associated with significantly greater delta values for NT-proBNP and D-dimer (P=0.018 and P=0.023, respectively), and this persisted after adjustment for age, sex, and race (P=0.023 and 0.047, respectively). After additional adjustment for established cardiovascular disease, the association remained significant for NT-proBNP (P=0.016), but not for D-dimer (P=0.09). There was no significant change in cTnT among patients with viremia (0 [−1, +3] ng/L, P=0.14) or without viremia (0 [−1, +2] ng/L, P=0.26), and there were no differences in delta values between the groups (P=0.22).

**DISCUSSION**

In this prospective study of hospitalized patients with COVID-19, the presence of SARS-CoV-2 viremia was not cross-sectionally associated with concentrations of D-dimer, cTnT, and NT-proBNP. However, patients with SARS-CoV-2 viremia experienced a greater increase in D-dimer and NT-proBNP, but not cTnT, during hospitalization.

SARS-CoV-2 viremia is associated with disease severity and organ damage in patients hospitalized with COVID-19. SARS-CoV-2 viral RNA has previously been detected in cardiac tissue in a majority (62%) of patients who died of COVID-19. COVID-19 is associated with increased risk of myocardial infarction, acute heart failure, arrhythmias, myocarditis, Takotsubo cardiomyopathy, and thromboembolism. cTnT, NT-proBNP, and D-dimer are typically elevated in these situations, reflecting myocardial injury, stress, and activated hemostasis. Retrospective studies of patients with COVID-19 have found that higher concentrations of these established cardiovascular biomarkers associate with worse outcome. However, this was not confirmed in a prospective study of consecutive patients hospitalized for COVID-19.

The prevalence of detectable SARS-CoV-2 RNA in plasma was 39% among our unselected patients hospitalized for COVID-19. This is comparable with findings from hospitalized patients in Wuhan, China (29%); Cologne, Germany (44%); Northern California, USA (33%). Not unexpectedly, in the current study we found a strong association between SARS-CoV-2 viremia and the inflammatory biomarkers IL-6, C-reactive protein, procalcitonin, and ferritin. In contrast, there was no significant difference in cardiovascular biomarker concentrations between patients with and without...
SARS-CoV-2 viremia. However, patients with viremia showed a greater increase in D-dimer and NT-proBNP during the first days of the hospitalization, suggesting a potential link between SARS-CoV-2 viremia and development of thromboembolism and myocardial stress. In contrast, changes in cTnT, reflecting myocardial injury, did not differ according to the presence of viremia.

Strengths of this study include the prospective design and the standardized measurement of biomarkers in one batch from a dedicated biobank. The main limitation is the limited sample size. Clearly, the current results should be validated in further prospective studies.

In conclusion, patients with SARS-CoV-2 viremia did not have higher concentrations of cardiovascular biomarkers, but experienced a significant increase in D-dimer and NT-proBNP. There was no association between viremia and cTnT, suggesting that myocardial injury in COVID-19 is indirect, ie, caused by hypoxemia or inflammation and not by disseminated virus per se.

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