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MAJOR ARTICLE

Viral Determinants of Acute COVID-19 Symptoms in a Nonhospitalized Adult Population in the Pre-Omicron Era

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Background. The influence of severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) RNA level and presence of infectious virus on symptom occurrence is poorly understood, particularly among nonhospitalized individuals.

Methods. The study included 85 nonhospitalized, symptomatic adults, who were enrolled from September 2020 to November 2021. Data from a longitudinal cohort studied over 28 days was used to analyze the association of individual symptoms with SARS-CoV-2 viral RNA load, or the presence or level of infectious (culturable) virus. Presence of infectious virus and viral RNA load were assessed daily, depending on specimen availability, and amount of infectious virus was assessed on the day of maximum RNA load. Participants were surveyed for the start and end dates of 31 symptoms at enrollment and at days 9, 14, 21, and 28; daily symptom presence was determined analytically. We describe symptoms and investigate their possible association with viral determinants through a series of single or pooled (multiple days across acute period) cross-sectional analyses.

Results. There was an association between viral RNA load and the same-day presence of many individual symptoms. Additionally, individuals with infectious virus were more than three times as likely to have a concurrent fever than individuals without infectious virus, and more than two times as likely to have concurrent myalgia, chills, headache, or sore throat.

Conclusions. We found evidence to support the association of viral RNA load and infectious virus on some, but not all symptoms. Fever was most strongly associated with the presence of infectious virus; this may support the potential for symptom-based isolation guidance for COVID-19.

Understanding the association between the biological markers of viral infection and symptoms is key to disease mitigation, pathogenesis, and treatment targets for severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2). There is a spectrum of disease presentation for SARS-CoV-2. After infection, individuals may experience a presymptomatic period, followed by symptoms leading to mild to critical cases, or they may remain asymptomatic [1]. Early case reports of coronavirus disease 2019 (COVID-19) observed that symptoms began around the time of peak viral load in some individuals, while others remained asymptomatic [2–5]. A proportion with severe illness developed a cytokine storm as the viral load declined [6–9]. Varying clinical observations in the setting of viral dynamics have led to questions about viral pathogenesis of symptoms.

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This association has been investigated for a variety of infectious viral diseases, including influenza and SARS-CoV-1, and prompted a biological hypothesis for SARS-CoV-2 that increased viral load could impact symptoms directly, by cellular damage, or indirectly, via the immune response [10–15]. Evidence from both influenza and SARS-CoV-1 support this hypothesis. While there is a growing body of literature supporting the relationship between SARS-CoV-2 viral load and symptoms [2, 5, 16], many of these studies lack viral dynamics, frequent measurement of symptom data, and inclusion of infectious virus data. In addition, most studies were conducted in hospitalized patients with increased potential for cytokine storm, complicating assessments of the direct effect of SARS-CoV-2 on symptoms [17]. Studies of nonhospitalized patients generally have been characterized by mild illness without cytokine storm, allowing for direct assessment of viral dynamics on COVID-19 symptoms. In one such study detailing individual symptoms, a decline in viral load was correlated with a decrease in symptom count [18].

Our objectives were to investigate (1) if viral load is associated with symptoms, including individual symptom presence and

daily symptom count, and (2) if specific symptoms are associated with the presence of infectious virus.

METHODS

Patient Consent Statement

The study was reviewed by the University of California, San Francisco (UCSF) Institutional Review Board and given a designation of public health surveillance according to federal regulations as summarized in 45 Code of Federal Regulations (C.F.R.) 46.102(d)(1)(2). This activity was reviewed by the Centers for Disease Control and Prevention (CDC) and was conducted consistent with applicable federal law and CDC policy (see, eg, 45 C.F.R. part 46.102(l)(2), 21 C.F.R. part 56; 42 United States Code [U.S.C.] §241(d); 5 U.S.C. §552a; 44 U.S.C. §3501 et seq). Written informed consent was obtained from all participants.

Study Design

This study uses data from an observational, longitudinal cohort study of the natural history of acute SARS-CoV-2 infection in San Francisco, California. Study procedures have been published elsewhere [19, 20].

Procedures

In brief, from September 2020 to November 2021, adults who tested positive for SARS-CoV-2 were identified via molecular testing at UCSF-affiliated testing sites. Eligibility screening was done via review of available data or by telephone interview. Index cases were eligible for inclusion if they were nonhospitalized, resided with at least 1 other individual, and lived in noncongregate settings in the San Francisco Bay Area. Index cases and household contacts were eligible for inclusion if they could be enrolled within 5 days of overall symptom onset. This analysis was restricted to adults who were symptomatic and tested positive for SARS-CoV-2 by reverse-transcription polymerase chain reaction (RT-PCR).

Measurements

Questionnaire-Based Measurements

Interviewer-administered questionnaires collected data on demographic and socioeconomic characteristics, medical history, symptom status and overall symptom onset, and clinical course of acute COVID-19. For all participants, questionnaires were administered on day of enrollment and at days 9, 14, 21, and 28 after the index case's overall symptom onset. Symptoms that were new since infection (or worsened since infection for preexisting symptoms) were assessed using a 32-symptom checklist derived from the US CDC list of COVID-19 symptoms [21] and the Patient Health Questionnaire Somatic Symptom Scale [22]. During each survey, participants were asked if they were experiencing or had experienced each individual symptom, and, if yes, were asked to report the start and end date of each symptom. The daily presence of symptoms was subsequently determined analytically, assuming that the symptom was present for all days between (and inclusive of) the start and end date. Subjective and objective fever were combined into 1 "fever" symptom, resulting in 31 symptoms assessed (see Supplementary Table 1 for list of symptoms). The overall symptom onset (herein referred to as symptom onset) for the infection was defined as the day on which the participant first reports their symptoms starting, and all symptom data were analyzed relative to the given individual's symptom onset. SARS-CoV-2 infection and vaccination status documentation (including vaccine boosters) was reviewed by interviewers during study visits.

Laboratory-Based Measurements

Anterior nasal specimens were self-collected daily from day of enrollment to day 14, and on days 17, 19, 21, and 28. Specimens were collected relative to the index case's symptom onset but were analyzed relative to the symptom onset for the given individual.

Anterior nasal specimens were used to detect and quantify SARS-CoV-2 RNA and infectious virus. SARS-CoV-2 RNA was quantified using quantitative RT-PCR (qRT-PCR) targeting the nucleocapsid (N) and envelope (E) genes. Infectious virus was detected both qualitatively (yes/no infectious virus) and quantitatively. Qualitative infectious virus was characterized by the presence of cytopathic effect (CPE) in tissue culture; all positive CPE results were confirmed as SARS-CoV-2 by an additional qRT-PCR step. Quantitative infectious virus was characterized with infectious viral titers (expressed as plaque-forming units [PFU]/mL) determined by viral plaque assays on the specimen with maximum RNA load for each participant. Details of these laboratory tests have been previously reported [19].

Viral lineage was determined based on timing of the predominant variant circulating when an individual started their infection. Individuals whose infection started prior to May 2021 were considered to have a pre-Delta lineage; those whose infection started during or after May 2021 were considered to have the Delta lineage.

Statistical Analyses

For symptoms, the daily and overall (day 28 or earlier) presence or absence (binary) of all 31 individual symptoms was assessed. Any symptom with an overall prevalence of <20% was excluded. For each of the 18 remaining included symptoms, the following outcome variables were assessed: daily presence of each symptom (binary variable) and daily number of symptoms reported (count variable; possible range, 0–18). Descriptive analyses of these symptom outcomes were performed, including duration and overall prevalence.

The viral determinants used in these analyses included: RNA viral load over time (log copies/mL), maximum RNA viral

load (log copies/mL), duration of RNA detection, maximum infectious viral load (log PFU/mL), and duration of infectious viral shedding (presence of CPE, days post-symptom onset). We assigned maximum and duration of RNA viral positivity to each participant. Maximum RNA viral load was defined as the RNA value on the day with the highest RNA viral load for each participant. Duration of RNA detection was defined as the days between symptom onset and the last day of RNA positivity for each participant. Among those who had the presence of infectious virus, we determined their maximum and duration of infectious viral load and shedding. Maximum infectious viral load was defined as the titer value of the plaque assay performed on the day of maximum viral RNA load. Duration of infectious viral shedding was defined as days between symptom onset and the last day of CPE positivity for each participant. Participants who were CPE negative were assigned a zero, and we analyzed the infectious virus variables with and without those individuals who were CPE negative.

The association between viral RNA and symptoms was examined cross-sectionally and pooled cross-sectionally. Given that the strong correlation between N- and E-specific RNA viral load has already been shown [20], analyses were conducted using only N-specific RNA viral loads (log_{10} transformed). The association between same-day viral RNA load (log copies/mL, continuous) and symptom presence (binary) and the association between same-day viral RNA load (log copies/mL, continuous) and symptom count (count) were investigated over the entire acute period (pooled cross-sectional). The association between same-day maximum viral RNA load (log copies/mL, continuous) and symptom presence (binary) was investigated on the day of maximum viral load (cross-sectional).

Infectivity was also examined both cross-sectionally and pooled cross-sectionally. The association between same-day maximum infectious viral load (log PFU/mL, continuous) and symptom presence (binary) and the association between same-day maximum infectious viral load (log PFU/mL, continuous) and symptom count (count) were both investigated on the day of maximum viral load (cross-sectional). Last, two different exposures were used to investigate the association between the qualitative presence of infectious virus (binary) and daily symptom presence over the entire acute period (pooled cross-sectional). The first exposure was the same-day presence of CPE (binary), meaning that a person was considered infectious if they had a positive CPE on the same day that symptom presence was being examined. The second exposure was the 1-day prior presence of CPE (binary), meaning that a person was considered infectious if they had a positive CPE on either the day prior to or the same day (2-day interval) that symptom presence was being examined. As a pooled cross-sectional analysis, this examination was repeated for every possible 2-day interval (prior day and same day) across the acute period.

All analyses were adjusted for potential confounding informed by a directed acyclic graph (Supplementary Figure 1). See the Supplementary Material for analytical model details. A series of additional sensitivity analyses were conducted by investigating the association between symptoms and viral load accounting for the interaction between viral load and time since onset (binned by weeks) and the association between symptom presence and the trajectory (same-day and 1-day prior slope) of viral load (see the Supplementary Material for more details). All analyses were performed using STATA/SE 17.0 software (StataCorp, College Station, Texas).

RESULTS

Characteristics of Participants at Baseline

The analysis cohort included 85 nonhospitalized, SARS-CoV-2–infected adult participants enrolled from 19 September 2020 through 27 November 2021; all participants experienced at least 1 of the 18 symptoms included in the main analysis (ie, had an overall prevalence of >20%). The median age was 38 years, 47 (55%) were female, and 41 (48%) were non-Hispanic White. Among these participants, 49 (58%) were unvaccinated and 36 (42%) had received their full initial series of COVID-19 vaccination. Participants were infected with pre-Delta (n = 51 [60%]) and Delta (n = 34 [40%]) lineages (Table 1).

Symptom Prevalence and Duration

The median maximum daily symptom count was 8 symptoms (interquartile range [IQR], 5–10; absolute range, 1–15) (Supplementary Table 1).

The most prevalent symptoms during the first 28 days post–symptom onset were fatigue (84%), cough (79%), rhinorrhea (76%), subjective/objective fever (65%), and headache (65%) (Supplementary Table 1). The symptoms with the highest median duration during the first 28 days post–symptom onset of COVID-19 were fatigue and concentration problems (11 days each), followed by cough, shortness of breath, rhinorrhea, and anosmia/dysgeusia (8 days each); median duration of fever was 4 days (Supplementary Figure 2).

Viral RNA and Infectious Dynamics

The median copies of viral RNA in the nasal specimens collected across the full acute time period was 5.3 log copies/mL (IQR, 3.6–7.0 log copies/mL) (Table 2; Figure 1); a median of 7 specimens were collected per participant. Maximum viral load occurred a median of 5 days post–symptom onset (IQR, 4–7 days). Among the maximum viral load nasal samples, the median was 7.5 log copies/mL (IQR, 5.8–9.0 log copies/mL). The median duration of detection of viral RNA shedding was 10 days post–symptom onset (IQR, 8–12 days). On day 5 post–symptom onset, the proportion of individuals who were viral RNA positive was 94.1%; by day 10, the proportion had dropped to 55.3% (Figure 1).

Table 1. Characteristics of Nonhospitalized, Symptomatic Cohort at Baseline

Characteristic	All (N = 85)
Age, y, median	38
IQR	30–45
Absolute range	19–73
Female birth sex	47 (55)
Gender identity	
Female	47 (55)
Male	38 (45)
Transgender male	0 (0)
Transgender female	0 (0)
Prefer not to answer	0 (0)
Race/ethnicity[a]	
Hispanic/Latino	18 (21)
Hawaiian/Pacific Islander	2 (2)
White	41 (48)
Black/African American	3 (4)
Asian	16 (19)
American Indian or Alaska Native	2 (2)
Education[a]	
Any high school or less	16 (19)
Any college	43 (51)
Any graduate school	25 (29)
Sexual orientation[a]	
Asexual	0 (0)
Bisexual	1 (1)
Gay/lesbian	4 (5)
Straight/heterosexual	79 (93)
Annual household income[a]	
$50 000 or less	11 (13)
$50 001 to $100 000	12 (14)
$100 001 to $300 000	28 (33)
More than $300 000	7 (8)
BMI[a], kg/m^2	
≤24.9	33 (39)
25–29.9	26 (31)
≥30	23 (27)
Self-reported comorbid conditions	
Any	20 (24)
Autoimmune disease[a]	1 (1)
Cancer[a,b]	4 (5)
Diabetes[a]	3 (4)
HIV[a]	0 (0)
Heart attack or heart failure[a]	0 (0)
Hypertension[a]	10 (12)
Lung problems[a,c]	13 (15)
Kidney disease[a]	1 (1)
Ever used tobacco[a,d]	13 (15)
Fully vaccinated at baseline[e]	36 (42)
SARS-CoV-2 variant	
Pre-Delta	51 (60)
Delta	34 (40)

Data are presented as No. (%) unless otherwise indicated.

Abbreviations: BMI, body mass index; HIV, human immunodeficiency virus; IQR, interquartile range; SARS-CoV-2, severe acute respiratory syndrome coronavirus 2.

[a]Missing and nonresponse. Race/ethnicity: 3 missing; education: 1 missing; sexual orientation: 1 prefer not to answer; income: 27 prefer not to answer; BMI: 3 missing; autoimmune: 1 missing; cancer: 2 missing; diabetes: 2 missing; HIV: 3 missing; heart attack: 1 missing; hypertension: 2 missing; lung problems: 1 missing; kidney disease: 3 missing; ever used tobacco: 3 missing.

[b]Cancer requiring treatment within the 2 years before coronavirus disease 2019 (COVID-19).

[c]Asthma, chronic obstructive pulmonary disease, emphysema, or bronchitis experienced in the 5 years before COVID-19.

[d]Cigarettes, cigars, or any product containing tobacco in a hookah.

[e]Participant who completed a primary series of a COVID-19 vaccine >14 days prior to enrollment; complete primary series was defined as 2 doses for messenger RNA vaccines or 1 dose of Johnson & Johnson.

Table 2. Description of Virologic Characteristics of Total Infected Cohort During Acute Phase of SARS-CoV-2 (First 28 Days)

Virologic Characteristic	(N = 85)
RNA viral load, log copies/mL	5.3 (3.6–7.0)
Maximum RNA viral load, log copies/mL	7.5 (5.8–9.0)
Duration of RNA detection[a], days post–symptom onset	10 (8–12)
Maximum infectious viral load, log PFU/mL	5.2 (3.2–6.1)
Duration of infectious viral shedding among participants with ≥1 nasal sample positive for viral culture (presence of CPE, days post–symptom onset)[b]	7 (5–8)
Duration of infectious viral shedding among all participants (presence of CPE, days post–symptom onset)	6 (2–8)

All data are presented as median (interquartile range).

Abbreviations: CPE, cytopathic effect; PFU, plaque-forming units.

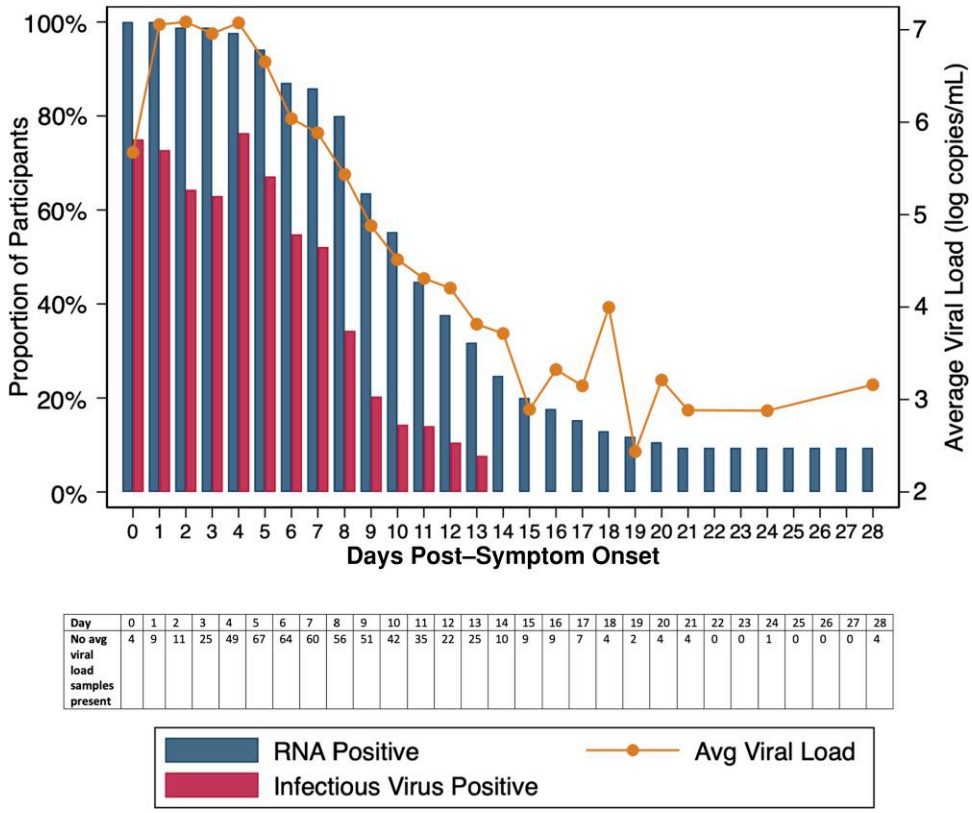
[a]Eight individuals were unable to have their duration determined.

[b]n = 65.

The median duration for infectious viral shedding was 7 days post–symptom onset (IQR, 5–8 days) (Table 2). On day 5 post–symptom onset, the proportion of individuals who were shedding infectious virus was 69.4%; by day 10, the proportion dropped to 11.8% (Figure 1). Among the nasal specimens reflecting the maximum RNA load per participant, the median infectious viral titer was 5.2 log PFU/mL (IQR, 3.2–6.1 log PFU/mL).

Association of Viral RNA Dynamics With Symptoms

In adjusted pooled cross-sectional analyses, participants with higher viral load were more likely to experience same-day fever, chills, fatigue, cough, shortness of breath, rhinorrhea, sore throat, myalgia, loss of appetite, diarrhea, concentration problems, headache, joint pain, back pain, and trouble sleeping; for example, for every 10-fold increase in N-viral load, fever was 1.3 times more likely to be reported (Figure 2). When this association was assessed adjusting for the interaction between log-transformed RNA levels and time since onset (binned by weeks), there was evidence of interaction among the following symptoms: fever, rhinorrhea, loss of appetite, headache, and dizziness. For these symptoms, these associations were more significant in the first week of illness compared to the second week (Supplementary Figure 3A). Same-day viral RNA load and symptom count were also strongly associated. Every 10-fold increase in log N-viral load was associated with a



Day	0	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26	27	28
No avg viral load samples present	4	9	11	25	49	67	64	60	56	51	42	35	22	25	10	9	9	7	4	2	4	4	0	0	1	0	0	0	4

Figure 1. Virologic characteristics by day. For the proportion of SARS-CoV-2 RNA–positive participants, if participants did not provide a specimen on a given day, they were assumed to be positive until they stopped testing positive. For the proportion of infectious virus–positive participants, individuals' infectivity status was determined on days when specimens were available. Average viral load was the average of all the log-transformed viral load values available on a given day.

mean increase in symptom count of 0.65 (95% confidence interval [CI], .44–.86).

Maximum viral load was only statistically significantly associated with a few same-day symptoms. For every 10-fold increase in log maximum N-viral load, an individual was more likely to have fatigue, rhinorrhea, back pain, and trouble sleeping on the day of maximum N-viral load (Supplementary Figure 4). In additional analyses evaluating the association between the trajectory of viral RNA load and same day symptoms, there was not a significant association (Supplementary Figures 3*B* and 3*C*).

Association of Culturable Virus With Symptoms

When assessing the relationship between same-day infectivity and symptoms, individuals with positive CPE were statistically significantly more likely to have most symptoms on that current day, except dizziness and joint pain, compared to individuals who were not infectious on that day (Figure 3). Compared to individuals without infectious virus detected, individuals with infectious virus detected were more than three times as likely to report fever on that same day (prevalence ratio [PR], 3.29) and were more than twice as likely to report same-day myalgia (PR, 2.88), chills (PR, 2.68), headache (PR, 2.36), or sore throat (PR, 2.08).

Furthermore, when assessing the relationship between 1-day prior and same-day infectivity with symptoms, we observed a similar pattern of findings as seen with just same-day infectivity, highlighted by strong associations with most symptoms (Figure 3). Compared to individuals without infectious virus detected, individuals with infectious virus detected the same day or the day prior were more than three times as likely to report fever, chills, or myalgia.

However, higher infectious virus titers on the day of maximum RNA load were not typically associated with symptom presence (Supplementary Figure 5) or same-day symptom count (mean difference, 0.36 [95% CI, −.23 to .96]; the exception was an increase in reporting of same-day myalgia.

DISCUSSION

In this observational, longitudinal cohort of symptomatic nonhospitalized adults, we found an association between multiple individual symptoms and both viral RNA load and infectious (culturable) virus. The burden of viral RNA in the nose was

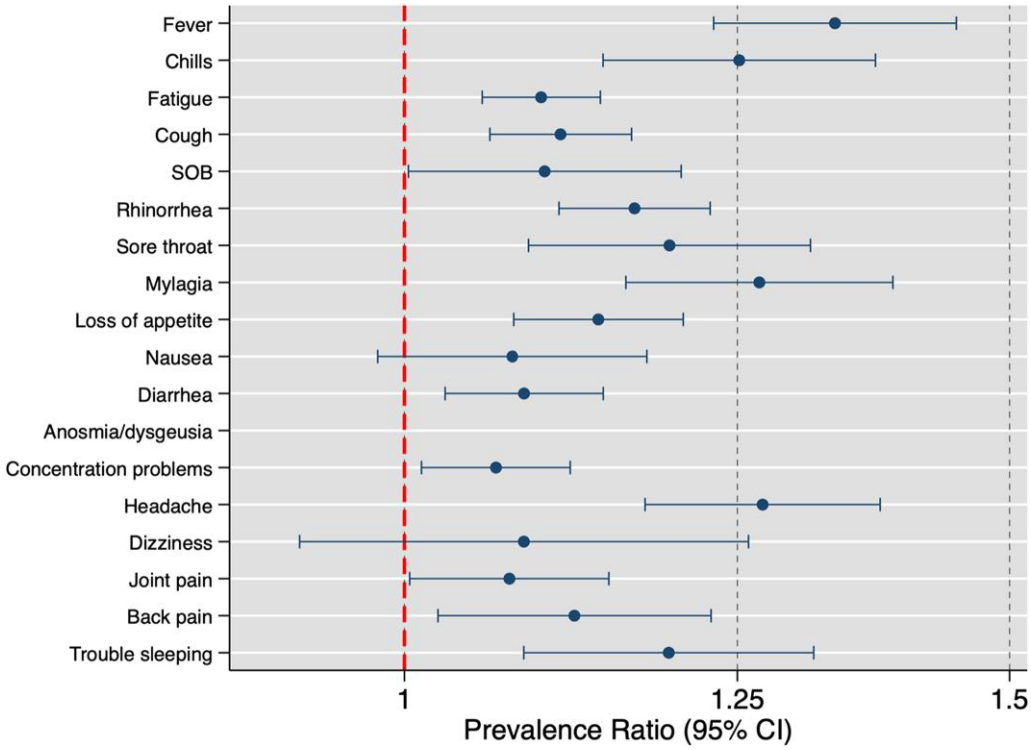


Figure 2. Association between same-day SARS-CoV-2 log-transformed RNA levels and symptom presence. Log-transformed RNA levels were continuous and symptom presence was binary. This association was estimated with pooled cross-sectional generalized estimating equation–modified Poisson regression models (log link), that clustered on participant and adjusted for confounders. The measurement of association was prevalence ratios (PRs) with 95% confidence intervals (CIs) (plotted on the above graph), which indicates the likelihood of experiencing a given symptom on the same day for every 1-unit change in nucleocapsid viral load. If the PR is >1, for each 10-fold increase in viral load, individuals were more likely to experience a given symptom on the same day (n = 82). Abbreviation: SOB, shortness of breath.

associated with many symptoms. Fourteen of 18 symptoms were associated in separate models with both viral RNA load and presence of culturable virus. The lack of complete overlap between these two models furthers our study's previously published findings that cycle threshold values (and thus viral load) cannot be used as surrogate markers for infectiousness [23]. These findings provide evidence in support of the causal effect of SARS-CoV-2 virus on symptoms, suggesting a potential role of antiviral therapies in symptom alleviation. Furthermore, the presence of specific symptoms, especially fever, were associated with the presence of infectious virus and may indicate the potential for symptom-based community isolation guidance for COVID-19.

In a prior study that included asymptomatic individuals, no significant correlation was observed between viral load and symptoms [24]. However, by focusing on symptomatic individuals in this study, we identified that same-day higher viral load was associated with symptom presence. Previous studies only used overall symptom scores as an outcome when investigating this relationship, while our study investigated the presence of each individual symptom as separate outcomes [18, 24]. We did not see an effect of the change in viral load on symptoms. Further investigation is needed to fully understand the biological significance of this. Regardless, the association of the burden of viral RNA and symptoms provides some evidence that increased viral load may directly or indirectly impact symptoms. Our findings build upon this literature by illustrating that this relationship holds true among multiple variants of SARS-CoV-2, utilizing a more in-depth look at COVID-19's clinical presentation.

We also illustrated an association between same-day active replicating virus and current symptoms during the acute phase of COVID-19. This relationship has been investigated before in mild SARS-CoV-2 cases, where culturable virus was found to be more likely in symptomatic Delta cases than nonsymptomatic non-Delta cases [25]. However, their sample size was smaller and lacked high-resolution symptom data that our study utilized. Our findings further support that the association holds true when taking a more granular look at the data and among a larger sample of individuals.

There are several limitations in our study. First, we attempted to enroll individuals within 5 days of symptom onset, but we may have still missed the true peak of viral load. Second, because symptom data were not surveyed daily and symptom

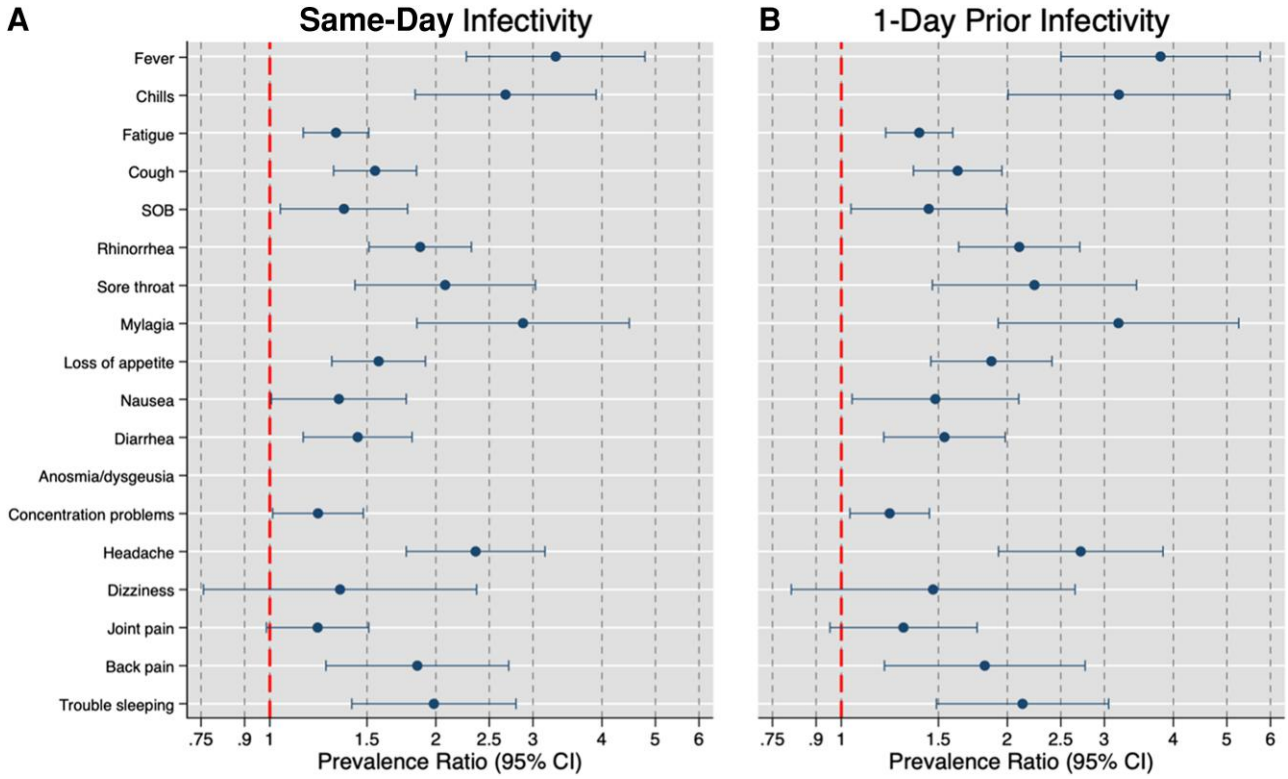


Figure 3. Association between the presence of infectious virus and symptom presence. For both graphs, infectivity and symptom presence were binary. *A*, For same-day infectivity, infectious individuals were those who had positive cytopathic effect (CPE) titers on the same day that symptom presence was investigated. This association was estimated with a pooled cross-sectional generalized estimating equation (GEE)–modified Poison regression model (log link) that clustered on participant and adjusted for confounders. The measurement of association was prevalence ratios (PRs) with 95% confidence intervals (CIs) (plotted on the graph on the left), which indicates the likelihood of experiencing a given symptom for individuals who were infectious on that day compared to noninfectious individuals. If the PR is >1, individuals who were infectious on that day were more likely to experience a given symptom on that same day (n = 81). *B*, For 1-day prior infectivity, infectious individuals were those who had positive CPE titer on the prior and/or same day that symptom presence was investigated. This association was estimated with a pooled cross-sectional GEE–modified Poison regression model (log link) that clustered on participant and adjusted for confounders. The measurement of association was PRs with 95% CIs (plotted on the graph on the right), which indicates the likelihood of experiencing a given symptom for individuals who were infectious on the prior day and/or that day compared to noninfectious individuals. If the PR is >1, individuals who were infectious on the day prior and/or that day were more likely to experience a given symptom on that current day (n = 81). Abbreviation: SOB, shortness of breath.

presence was assumed during the reported start and end dates, fluctuations in intermittent symptoms may not have been captured. Third, participants in this study experienced mild illness, which limits the generalizability of the study to nonhospitalized populations. Fourth, because immunological markers were unmeasured, we could not ascertain the relative contribution of early immune responses on symptoms. Fifth, more than 50% of our study population was unvaccinated, which limits the generalization of our results. Sixth, we were unable to determine the main biological driver of symptom presence given the inability to distinguish the relative contributions of burden of viral RNA and culturable virus on the outcome. Last, samples from Omicron-infected individuals were not included; further investigation is needed to see if these associations continue to hold with newer variants.

Our findings highlight the association of viral RNA burden and/or culturable virus with symptom presentation in mild COVID-19 cases, possibly supporting the biological hypothesis linking viral pathogenesis to symptoms. The presence of an association between many of the assessed symptoms and both burden of viral RNA and culturable virus, despite them having slightly different dynamics, provides further evidence that supports this hypothesis. To fully understand the complexity of COVID-19 symptom pathogenesis, it will be important to perform a mediation analysis of viral and inflammatory markers with symptoms during mild COVID-19 illness. Understanding the biological determinants of symptoms among individuals who receive antivirals (and other therapeutics that target viral load) in future studies may explain treatment effects on number or duration of symptoms and may reinforce the benefit of early treatment if symptomatic.

Supplementary Data

Supplementary materials are available at *Open Forum Infectious Diseases* online. Consisting of data provided by the authors to benefit the reader, the

posted materials are not copyedited and are the sole responsibility of the authors, so questions or comments should be addressed to the corresponding author.

Notes

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Author contributions. M. J. P., J. D. K., and J. N. M. designed and oversaw the cohort; M. J. P., J. D. K., S. L., M. C. D., and J. N. M. designed the study instruments with input from S. L. and K A.; K. A., S. L., and J. P. R. assisted with Spanish translation; K. A. and M. C. D. recruited participants and coordinated research visits; K. A., S. L., J. P.-R., M. C. D., and P. R. R. administered study questionnaires and collected clinical data; K. A., S. L., J. P.-R., and M. C. D. collected biospecimens; M. G.-K. and M. T. performed all laboratory tests on all collected biospecimens; S. L., S. A. G., M. C. D., S. M., C. A. F., K. C. D., K. A., P. R. R., and J. Y. C performed data entry and validation; J. N. M. designed and maintained the study database; S. A. G. and S. L. cleaned the data and performed the analyses, which were planned by S. A. G., J. D. K., and J. N. M.; S. A. G. drafted the manuscript, with extensive input from G. R. A., S. S., M. B.-H., C. M. M., D. V. D., M. J. P., J. D. K., and J. N. M. All authors reviewed, edited, and approved the manuscript.

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