Research Paper

Clinical features, diagnostics, and outcomes of patients presenting with acute respiratory illness: A retrospective cohort study of patients with and without COVID-19

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ARTICLE INFO

ABSTRACT

Background: Most data on the clinical presentation, diagnostics, and outcomes of patients with COVID-19 have been presented as case series without comparison to patients with other acute respiratory illnesses.
Methods: We examined emergency department patients between February 3 and March 31, 2020 with an acute respiratory illness who were tested for SARS-CoV-2. We determined COVID-19 status by PCR and metagenomic next generation sequencing (mNGS). We compared clinical presentation, diagnostics, treatment, and outcomes.
Findings: Among 316 patients, 33 tested positive for SARS-CoV-2; 31 without COVID-19 tested positive for another respiratory virus. Among patients with additional viral testing (27/33), no SARS-CoV-2 co-infections were identified. Compared to those who tested negative, patients with COVID-19 reported longer symptoms duration (median 7d vs. 3d, $p < 0.001$). Patients with COVID-19 were more often hospitalized (79% vs. 56%, $p = 0.014$). When hospitalized, patients with COVID-19 had longer hospitalizations (median 10.7d vs. 4.7d, $p < 0.001$) and more often developed ARDS (23% vs. 3%, $p < 0.001$). Most comorbidities, medications, symptoms, vital signs, laboratories, treatments, and outcomes did not differ by COVID-19 status.
Interpretation: While we found differences in clinical features of COVID-19 compared to other acute respiratory illnesses, there was significant overlap in presentation and comorbidities. Patients with COVID-19 were more likely to be admitted to the hospital, have longer hospitalizations and develop ARDS, and were unlikely to have co-existent viral infections.
Funding: National Center for Advancing Translational Sciences, National Heart Lung Blood Institute, National Institute of Allergy and Infectious Diseases, Chan Zuckerberg Biohub, Chan Zuckerberg Initiative.

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Research in context

Evidence before this study

Emerging data on the clinical presentation, diagnostics, and outcomes of patients with COVID-19 have commonly been presented as case series. Without control patients, it is not clear whether and how the clinical features, diagnostics, and outcomes differ from other respiratory infections.

Added value of this study

When compared to other patients with acute respiratory illness not caused by COVID-19, many of the clinical features and outcomes occur at similar rates. Notably different, patients with COVID-19 had a longer duration of symptoms, particularly fatigue, fever, and myalgias, were more likely to be admitted to the hospital and for a longer duration, and more likely to develop ARDS compared to those without COVID-19. Those infected with SARS-CoV-2 were unlikely to have co-existent viral infections when examined by PCR and metagenomic next generation sequencing.

Implications of all the available evidence

Given the considerable overlap in clinical features and outcomes, studies seeking to describe features unique to COVID-19 should employ a control group. Viral co-infection rates are variable and may be context specific.

1. Introduction

The severe acute respiratory coronavirus 2 (SARS-CoV-2) and its associated clinical disease, COVID-19, led to a global pandemic in early 2020, with more than 3 million cases and more than 200,000 deaths as of April 2020. [1] The initial published reports of COVID-19 describe the most common presenting symptoms as fever, cough, and dyspnea. [2−6] While many people recovered, reports from China, Italy, and the United States showed that approximately 5% of patients required intensive care, and 1.7 to 7.2% died. [1,7,8] The majority of clinical and outcomes data on COVID-19 have been from Asia and Europe, [4,6,7,9−14] although data are now emerging from the United States. In particular, studies have reported the clinical features and outcomes of hospitalized patients in Seattle, New York City, and Northern California. [15−19] However, reports have predominantly focused on patients diagnosed with COVID-19 and have not described in detail the presentation of patients with acute respiratory illness who did not have COVID-19. Without control patients, it is uncertain whether COVID-19 presents differently from other respiratory infections.

The prevalence of viral co-infections in patients with COVID-19 appears to be low in most but not all studies. [15−18,20−23] However, these studies used conventional microbiological techniques to evaluate for co-infections that are limited in their ability to diagnose respiratory infections. [24] Understanding the true scope of co-infections in patients with COVID-19 is critical to pursue appropriate diagnostics and management. Metagenomic next-generation sequencing (mNGS) offers a powerful alternative to test for viruses in a respiratory sample in an unbiased manner. [25]

Here we report the clinical characteristics, diagnostics, and outcomes of all patients presenting with respiratory illness to a tertiary academic medical center in San Francisco at the outset of the COVID-19 pandemic. We compare patients with COVID-19 disease to patients presenting during the same time period with an acute respiratory illness and report the prevalence of viral respiratory infections using both conventional microbiology and mNGS.

2. Methods

2.1. Setting and design

We conducted a retrospective cohort study to describe the characteristics, diagnostics, and outcomes of patients with respiratory illness presenting to the University of California, San Francisco (UCSF) Health Emergency Department (ED) during the COVID-19 outbreak, comparing patients with and without COVID-19 disease. We identified all patients 18 years or older who underwent testing for COVID-19 within 24 h of presentation to the ED between February 3 and March 31, 2020. Patients were tested for SARS-CoV-2 if they met U.S. Centers for Disease Control and Prevention (CDC) clinical testing criteria. [26]

Two physicians blinded to patients' COVID-19 status, independently reviewed the documented clinical presentation of all patients and included only those who presented with acute respiratory symptoms (e.g., cough, dyspnea) or influenza-like illness symptoms (e.g., fever, myalgias). Discordant results were re-reviewed together and a consensus decision was reached on all cases (Appendix Fig. 1). If patients had multiple encounters during the time period, the first encounter was examined. Patients who were discharged and readmitted within 48 h were considered a single clinical encounter and outcomes ascertained throughout the encounter.

2.2. Patient characteristics

Patient medical records were reviewed by trained physician chart reviewers and relevant data on initial presentation, radiology findings, and outcomes were abstracted using standardized case review forms. Additional information on patient demographics, vital signs, and laboratory results were obtained from the Epic-based electronic health record. We characterized patients' comorbidities and their presenting signs and symptoms based on the admission History & Physical and Emergency Department documentation. If a specific comorbidity was not mentioned in the admission documentation, it was considered not present. Records were also reviewed to obtain results of laboratory tests and chest imaging reports within the first 24 h after admission.

2.3. Clinical microbiological testing

Clinician-ordered testing for COVID-19 was carried out at the UCSF Clinical Microbiology Laboratory using an in-house Clinical Laboratory Improvements Amendments (CLIA)-validated reverse transcriptase polymerase chain reaction (PCR) assay. This assay was performed for 290/316 (92%) of patients on RNA extracted from oropharyngeal and/or nasopharyngeal swab specimens using primers targeting two regions of the SARS-CoV-2 N gene. The analytical sensitivity/specificity of the in-house assay compared to the US CDC assay performed at the CDC was 97% and 100%, respectively. Twenty-six (8%) of the patients had SARS-CoV-2 PCR testing ordered at the study site but performed at the Centers for Disease Control or other institutions using their clinically validated assays. At the time of the study,

PCR results were available at the earliest within 3 h, and the median time to result was 16 h.

Conventional PCR testing for other respiratory viruses was carried out at the discretion of treating clinicians for 270/316 (85%) of patients on pooled nasopharyngeal+oropharyngeal or nasopharyngeal swab specimens using two types of commercial assays as detailed in Appendix table 2. The first was a 12-target respiratory viral PCR assay (adenovirus, influenza AH1/AH3/B, human metapneumovirus, human rhinovirus, parainfluenza viruses 1−4, respiratory syncytial viruses A/B) manufactured by Luminex, Inc. The second was a 3-target (influenza A/B, respiratory syncytial virus) assay manufactured by Diasoren, Inc. Bacterial and fungal respiratory pathogens were assessed by semi-quantitative cultures. Patient blood cultures were performed via inoculation into BD Bactec Plus Aerobic and Lytic Anaerobic media (Becton Dickinson).

2.4. Respiratory virus detection by metagenomic sequencing

To further screen for the presence of other respiratory viral pathogens, metagenomic next generation sequencing (mNGS) of RNA was performed on available residual RNA initially extracted for COVID-19 clinical PCR testing. At our institution, during the time period of the study, SARS-CoV-2 PCR was performed using in-house CLIA validated PCR tests for the majority of samples. This in-house PCR test involved first extracting RNA from patient swab samples and then carrying out reverse transcriptase PCR as described in the Methods. Of the 316 PCR tests performed, leftover RNA was available for mNGS analysis on 178 patients. To balance the need for timely turnaround with the desire to assess a sufficiently large fraction of the cohort, we performed mNGS on 60% (N = 107) of these 178 samples, which included as many SARS-CoV-2 positive samples as possible (mNGS data generated on 14) plus an arbitrary selection of SARS-CoV-2 negative samples. SARS-CoV-2 negative samples were distributed as evenly as possible throughout the study timeframe and were selected blinded to patient characteristics and outcomes.

After DNase treatment, human ribosomal RNA depletion was carried out using FastSelect (Qiagen). To control for background contamination, we included negative controls (water and HeLa cell RNA) as well as positive controls (spike-in dilution series of RNA standards from the External RNA Controls Consortium [ERCC]). [27] The latter enabled subsequent bioinformatic assessment of the total RNA mass input in each sample. [28]

RNA was then fragmented and subjected to a modified metagenomic spiked sequencing primer enrichment (MSSPE) library preparation method. [29] Briefly, a 1:1 mixture of the NEBNext Ultra II RNAseq Library Prep (New England Biolabs) random primer stock and a pool of SARS-CoV-2 primers at 100 μM was used at the first strand synthesis step of the standard RNAseq library preparation protocol to enrich for the recovery of reads spanning the length of the SARS-CoV-2 genome sequence in the context of mNGS analysis. [30] RNA-seq libraries underwent 146 nucleotide paired-end Illumina sequencing on an Illumina NovaSeq 6000.

2.5. mNGS bioinformatic and phylogenetic analysis

Following demultiplexing, reads were host- and quality-filtered and then subjected to viral reference based alignment at both the nucleotide and amino acid level against sequences in the National Center for Biotechnology Information (NCBI) nucleotide (NT) and non-redundant (NR) databases, followed by assembly using previously validated bioinformatics pipelines. [31,32] We used spike-in positive control ERCC RNA standards to bioinformatically calculate the input RNA for the mNGS assay. Ten samples had insufficient (<25 pg) input RNA for accurate analysis and so were considered invalid, leaving 97 subjects available for analysis.

Negative control (water and HeLa cell RNA) samples enabled estimating the number of background reads to each virus, which were normalized by input mass determined based on the ratio of sample reads to spike-in positive control ERCC RNA standards. [28] Viruses with sequencing reads significantly greater compared to negative controls (adjusted p value < 0.05 using a Holm-Bonferroni correction within each sample) were identified by modeling the number of background reads as a negative binomial distribution with mean and dispersion fitted on the negative controls. For phylogenetic analysis of SARS-CoV-2 viruses, we constructed genomes using minimap2 [33] to align reads to the reference MN908947.3 and iVar [34] to trim primers and call variants, then restricted to samples with at least 10-fold coverage of at least 97% (2929 kgbases) of the genome (n = 10), and utilized the Nextstrain [35] pipeline to build a phylogenetic tree using iqtree. [36] Viral genomic data is publicly accessible via gisaid.org (Global Initiative on Sharing All Influenza Data) [37] and Genbank (MT385414 - MT385497).

2.6. Treatment and outcomes

Clinical treatment and outcomes were ascertained through a combination of chart review and extraction of structured fields from the electronic health record. Medication records were reviewed to identify the administration of relevant antibiotics. We determined if patients required respiratory support at any point during their hospitalization: nasal cannula, high flow nasal cannula, noninvasive ventilation (bilevel or continuous positive airway pressure), or endotracheal intubation. Patients were considered to have new-onset cardiomyopathy if a treating physician documented the diagnosis. Acute respiratory distress syndrome (ARDS) was defined according to the Berlin definition by two physicians. [38] Acute kidney injury was defined using the Kidney Disease: Improving Global Outcomes definition. [39] Outcome ascertainment was censored on April 25, 2020.

2.7. Statistical analysis

We used descriptive statistics to characterize the features of patients grouped by COVID infection. Where clinically relevant, we dichotomized continuous variables. For normally distributed continuous variables, we calculated the mean and standard deviation and tested for differences using t-tests. For non-normally distributed continuous variables, we calculated the median and interquartile range and tested for differences using the Wilcoxon rank sum test. For categorical and dichotomous variables, we evaluated differences between groups using the chi-square test or Fisher's exact test. The analyses were not adjusted for multiple comparisons and should be interpreted as descriptive and exploratory. The Human Research Protection Program Institutional Review Board at the University of California, San Francisco, approved this study (IRB# 16−20,956). We used Stata version 14.2 (College Station, TX) and SAS version 9.4 (Cary, NC) to conduct all analyses.

2.8. Role of the funding source

The funders had no role in study design, data collection and analysis, decision to publish, or preparation of the manuscript.

3. Results

3.1. Demographic characteristics and comorbidities

Out of 316 patients who presented with acute respiratory illness and underwent testing for COVID-19, 33 (10%) tested positive for SARS-CoV-2 by PCR. Patients with a positive COVID-19 test result were more likely to have traveled to an area of community transmission in the past 21 days or to have had contact with someone with

COVID-19 (46% vs 11%, $p < 0.001$), to be married (64% vs. 36%, $p = 0.02$), or to identify as Asian (42% vs. 24%, $p = 0.010$) (Table 1). Patients who tested positive were also more likely to report never smoking tobacco (61% vs. 40%, $p = 0.001$) and to have undergone solid organ transplantation (12% vs. 3%, $p = 0.027$). The prevalence of hypertension and diabetes did not differ significantly between COVID-19 positive and negative patients. There was no significant difference by COVID-19 status of the proportion of patients taking an angiotensin-converting enzyme inhibitor or angiotensin II receptor blocker.

3.2. Signs, symptoms and vital signs

Patients with COVID-19 reported a longer duration of symptoms prior to ED presentation (median 7 vs. 3 days, $p < 0.001$) (Table 1). COVID-19 patients reported fever (82% vs. 44%, $p < 0.001$), fatigue (85% vs. 50%, $p < 0.001$), and myalgias (61% vs 27%, $p < 0.001$), at a higher rate than COVID-19 negative patients. The presence and characteristics of cough, dyspnea, and chest pain did not differ based on COVID-19 infection. Gastrointestinal symptoms − nausea, vomiting, diarrhea, and abdominal pain − were present at similar rates in the two groups. With respect to vital sign abnormalities, tachycardia, hypotension, oxygen requirement, and tachypnea did not differ by COVID-19 status. However, patients with COVID-19 were more likely to present with a measured fever (46% vs 24%, $p = 0.010$).

3.3. Laboratory studies and imaging upon presentation

Lymphopenia was more common in patients with COVID-19 at the time of presentation (55% vs 34%, $p = 0.018$) (Table 2). Aspartate transaminase but not alanine transaminase was more often elevated in patients with COVID-19 (36% vs. 18% $p = 0.022$ and 11% vs. 10% $p = 1.000$, respectively). Patients with COVID-19 were less often acidemic (0% vs. 15%, $p = 0.031$) and less often found to be hypercarbic (4% vs. 28%, $p = 0.002$) by venous blood gas. Of the patients tested on presentation, neither troponin nor procalcitonin elevation differed by COVID-19 status. Chest X-rays were performed on all but 6 patients. Radiographs from patients with COVID-19 were more likely to reveal bilateral patchy or hazy opacities (55% vs. 24%, $p = 0.001$). Focal consolidations, interstitial abnormalities, and pleural effusions were observed at similar proportions.

3.4. Pathogen diagnostics

Clinicians ordered Influenza/Respiratory syncytial virus PCR testing for 99/316 (31%) patients and 12-target respiratory virus PCR for 171/316 (54%) patients; testing rates did not differ by COVID-19 status (Table 3). Orthogonal mNGS analysis was performed on swab specimens from 97/316 (31%) of patients to provide additional broad range screening of both common and uncommon viral pathogens. By PCR, SARS-CoV-2 was the most prevalent respiratory virus detected in 33/316 patients (10%). No co-infections with SARS-CoV-2 and other viruses were identified. Other respiratory viruses were identified in 31/194 (16%) of patients without COVID-19. Independent mNGS analyses corroborated 13/14 (93%) of SARS-CoV-2 infections and 11/11 (100%) of other respiratory viral infections detected by clinical PCR assays. Respiratory bacterial co-infection was not more common in patients with COVID-19 (11% vs. 18%, $p = 1.000$) and no cases of ventilator associated pneumonia were identified in COVID-19 patients. Bacteremia or fungemia was also not more common in patients with COVID-19 disease (5% vs. 7%, $p = 1.00$).

3.5. Genomic epidemiology of SARS-CoV-2

To understand the genomic epidemiology of SARS-CoV-2 in the cohort, phylogenetic analysis was performed. SARS-CoV-2 genomes with at least 97% coverage at 10-fold sequencing depth could be recovered from 10 of the 13 mNGS-positive subjects. These 10 genomes originate from several parts of the global SARS-CoV-2 phylogeny, with clades A2a ($n = 3$, widely prevalent in New York) and B1 ($n = 3$, detected in Washington State in February 2020) representing slightly more than half of the lineages we identified (Appendix Fig. 2). The SARS-CoV-2 isolated from patients who required ICU care were not associated with any single clade.

3.6. Hospitalization treatment and outcomes

In all, 186 patients were hospitalized and patients with COVID-19 were more likely to be admitted (79% vs. 56%, $p = 0.014$) and have longer lengths of stay (median 10.7 vs. 4.7 days, $p < 0.001$). Among hospitalized patients, antibiotics and oseltamivir were used in similar proportions (Table 4). Hydroxychloroquine was more often used in patients with COVID-19 (22% vs. < 1%, $p < 0.001$); however, azithromycin and corticosteroids use did not differ by COVID-19 status. Six of 26 inpatients with COVID-19 were enrolled in a randomized trial of remdesivir. Respiratory support was provided in similar proportions of patients and, when respiratory support was needed, the level of support did not differ by COVID-19 status.

Numerically, more patients with COVID-19 required ICU care compared to non-COVID-19 patients, although the difference was not statistically significant (42% vs. 26%, $p = 0.092$) (Table 5). When transferred to the ICU, there was no observed difference in the use of ICU interventions; however, patients with COVID-19 had a longer ICU length of stay (median 8.8 vs. 2.9 days, $p = 0.005$). Those diagnosed with COVID-19 were more likely to develop ARDS (23% vs. 4%, $p < 0.001$) but were no more likely to develop cardiomyopathy or acute kidney injury when compared to non-COVID-19 patients. Among those tested, patients diagnosed with COVID-19 were no more often observed to have abnormal coagulation tests or elevated troponin. Treatment administered to patients not admitted to the hospital are presented in Appendix Table 1.

4. Discussion

While a number of studies describe the clinical features of patients with COVID-19, few have directly compared the clinical presentation and outcomes of COVID-19 to other respiratory illnesses. [23,40−44] Without a control group, and in settings of restricted COVID-19 test availability, we cannot ascertain whether COVID-19 presents differently from other forms of respiratory illnesses. In our study comparing acutely ill patients with and without COVID-19 presenting for emergency care, we found that patients with COVID-19 had a longer duration of symptoms, were more likely to be admitted to the hospital, had longer hospitalizations and were more likely to develop ARDS. Using standard laboratory PCR testing, and mNGS, we found a 16% prevalence of other respiratory viruses in the COVID-19 negative patients, and a lack of detectable viral co-infections in the COVID-19 positive patients.

Patients diagnosed with COVID-19 were more likely to be Asian (44%), which likely reflects differences in the dynamics of COVID-19 transmission early in the pandemic in San Francisco, where the proportion of people who self-identify as Asian is high (36%). [5] Although Asians were overrepresented in the initial COVID-19 cases at our institution, this is not indicative of the current situation in San Francisco, where Asians make up only 13% of the total number of COVID-19 cases. [45] COVID-19 patients were more likely to be never smokers, in line with other studies showing no link between tobacco use and increased COVID-19 risk. [4] [46,47] Largely similar comorbidity profiles were observed between COVID-19 positive and negative patients, aside from a higher proportion of chronic kidney disease and history of solid organ transplantation in COVID-19 patients.

Table 1

Characteristics of 316 patients presenting with acute respiratory illness and tested for COVID-19.

	COVID-19 positive (n = 33)	COVID-19 negative (n = 283)	P value
Demographics			
Age, median (IQR), yr	63 (50, 75)	62 (43, 72)	0.243
Female sex	12 (36%)	140 (50%)	0.154
Marital status			
Married or partnered	21 (64%)	103 (36%)	0.019
Single	7 (21%)	136 (48%)	
Divorced	2 (6%)	18 (6%)	
Widowed	2 (6%)	19 (7%)	
Housing insecure	1 (3%)	44 (16%)	0.063
Race			
White	8 (24%)	124 (44%)	0.010
Black or African American	2 (6%)	50 (18%)	
Asian	14 (42%)	69 (24%)	
Hispanic or Latino ethnicity	5 (15%)	21 (8%)	0.128
Required interpreter	6 (18%)	46 (16%)	0.777
Travel to an area with known community transmission in last 21 days or known COVID exposure	15 (46%)	31 (11%)	<0.001
Comorbidities			
Tobacco use			
Current smoker	0 (0%)	52 (18%)	0.001
Former smoker	9 (27%)	47 (17%)	
Never smoker	20 (61%)	113 (40%)	
Unknown	4 (12%)	71 (25%)	
Hypertension	16 (49%)	119 (42%)	0.479
Coronary artery disease	5 (15%)	38 (13%)	0.785
Diabetes	9 (27%)	50 (18%)	0.180
Cancer, active (excluding non-melanoma skin cancer)	5 (15%)	42 (15%)	0.962
Cancer, in remission (excluding non-melanoma skin cancer)	5 (15%)	19 (7%)	0.090
Prior stroke	0 (0%)	25 (9%)	0.090
Chronic kidney disease	7 (21%)	28 (10%)	0.049
Liver disease	0 (0%)	13 (5%)	0.375
Human immunodeficiency virus	0 (0%)	15 (5%)	0.382
Chronic obstructive pulmonary disease/emphysema	1 (3%)	41 (15%)	0.098
Asthma	4 (12%)	38 (13%)	1.000
Chronic bronchitis	0 (0%)	5 (2%)	1.000
Congestive heart failure	4 (12%)	43 (15%)	0.798
Solid organ transplant	4 (12%)	8 (3%)	0.027
Other immunosuppressive condition	5 (15%)	33 (12%)	0.560
Home medications			
Steroids	5 (15%)	26 (9%)	0.275
Immunosuppression medications (aside from steroids)	6 (18%)	35 (13%)	0.347
ACE inhibitors or ARB	6 (18%)	43 (15%)	0.654
Signs and Symptoms			
Onset of symptoms relative to presentation, d (IQR)	7 (5, 9)	3 (2,7)	<0.001
Fever, patient reported	27 (82%)	125 (44%)	<0.001
Fatigue/malaise	28 (85%)	140 (50%)	<0.001
Cough	28 (85%)	208 (74%)	0.156
Dry	12 (43%)	62 (30%)	0.298
Productive	10 (36%)	77 (37%)	
Unspecified	6 (21%)	69 (33%)	
Myalgia	20 (61%)	77 (27%)	<0.001
Dyspnea	23 (70%)	171 (60%)	0.301
Chest pain	5 (15%)	81 (29%)	0.100
Sore throat	9 (27%)	73 (26%)	0.855
Congestion/Rhinorrhea	10 (30%)	74 (26%)	0.610
Diarrhea	9 (27%)	45 (16%)	0.101
Nausea	8 (24%)	48 (17%)	0.300
Vomiting	5 (15%)	28 (10%)	0.350
Abdominal pain	4 (12%)	26 (9%)	0.535
Headache	7 (21%)	47 (17%)	0.506
Altered mentation	2 (6%)	39 (14%)	0.280
Presenting vital signs			
Tachycardia (HR > 100 beats/min)	16 (49%)	164 (58%)	0.299
Low mean arterial pressure (<60 mmHg)	0 (0%)	2 (1%)	1.00
Tachypnea (RR > 20 breaths/min)	13 (39%)	124 (44%)	0.616
Fever ($T_{max} \geq 100.4°F$)	15 (46%)	69 (24%)	0.010
Highest level of respiratory support in the first 24 h			
Nasal cannula	10 (30%)	64 (23%)	0.864
High flow nasal cannula	2 (6%)	23 (8%)	
CPAP or BiPAP	0 (0%)	10 (4%)	
Mechanical ventilation	1 (3%)	12 (4%)	

Legend:
COVID-19 - Coronavirus Disease 2019; IQR - interquartile range; ACE - angiotensin-converting enzyme; ARB - Angiotensin II receptor blockers; HR - heart rate; CPAP - continuous positive airway pressure; BiPAP - bilevel positive airway pressure; RR - respiratory rate.

Table 2
Laboratory and imaging findings within 24 h of presentation among 316 patients presenting with acute respiratory illness and tested for COVID-19.

	Lab normal values	COVID-19 positive (n = 33)	COVID-19 negative (n = 283)	P value
Complete blood count				
White blood cell count				
Leukopenia*	3.4−10.0 × 10⁹/L	3/33 (9%)	10/279 (4%)	0.148
Leukocytosis†		0/33 (0%)	110/279 (39%)	<0.001
Neutrophil count	1.8−6.8 × 10⁹/L			
Neutropenia*		2/33 (6%)	7/274 (3%)	0.250
Neutrophilia†		4/33 (12%)	126/274 (46%)	<0.001
Lymphocyte count	1.0−3.4 × 10⁹/L			
Lymphopenia*		18/33 (55%)	92/274 (34%)	0.018
Lymphocytosis†		0/33 (0%)	15/274 (6%)	0.384
Platelet count	140−450 × 10⁹/L			
Thrombocytopenia*		7/33 (21%)	31/279 (11%)	0.093
Thrombocytosis†		0/33 (0%)	14/279 (5%)	0.377
Hemoglobin	13.6−17.5 g/dL			
Anemic*		19/33 (58%)	176/280 (63%)	0.554
Chemistry				
Hyponatremia*	135−145 mmol/L	11/32 (34%)	56/274 (20%)	0.071
Hypernatremia†		1/32 (3%)	12/274 (4%)	v
Creatinine, elevated† (%)	0.73−1.18 mg/dL	11/32 (34%)	71/274 (26%)	0.306
Aspartate transaminase, elevated†	5−44 U/L	10/28 (36%)	38/217 (18%)	0.022
Alanine transaminase, elevated†	10−61 U/L	3/28 (11%)	22/217 (10%)	1.000
Troponin I, elevated	<0.05 ug/L	2/13 (15%)	37/161 (23%)	0.735
Procalcitonin, elevated	<0.26 ug/L	4/25 (16%)	44/125 (35%)	0.065
Venous blood gas				
pH	7.31−7.41			
Acidemic*		0/29 (0%)	28/192 (15%)	0.031
Alkalemic†		11/29 (38%)	46/192 (24%)	0.116
Hypercarbic†	41−51 mmHg	1/29 (4%)	54/192 (28%)	0.002
Elevated lactate†	0.5−2.0 mmol/L	5/29 (17%)	51/194 (26%)	0.295
Chest X-ray findings				
X-ray within first 24 h		33/33 (100%)	277/283 (98%)	1.000
Patchy/hazy opacities				
Unilateral		4/33 (12%)	37/277 (13%)	0.001
Bilateral		18/33 (55%)	67/277 (24%) 173/277 (63%)	
Not present		12/33 (33%)	37/277 (13%)	
Focal consolidation				
Unilateral		1/33 (3%)	29/277 (11%)	0.368
Bilateral		2/33 (6%)	13/277 (5%)	
Not Present		30/33 (91%)	235/277 (85%)	
Interstitial abnormalities				
Unilateral		0/33 (0%)	7/277 (3%)	0.561
Bilateral		4/33 (12%)	52/277 (19%)	
Not Present		29/33 (88%)	218/277 (79%)	
Pleural effusion				
Unilateral		1/33 (3%)	18/277 (7%)	0.031
Bilateral		0/33 (0%)	18/277 (7%)	
Not Present		32/33 (97%)	241/277 (87%)	

Legend
Results reflect lab tests and imaging tests performed within 24 h of presentation.
COVID-19 - Coronavirus Disease 2019.
* lower than the lower limit of normal.
† greater than the upper limit of normal.

Patients diagnosed with COVID-19 had a longer duration of symptoms prior to presentation and were more likely than control patients to report fever, fatigue and myalgias. It is notable, however, that 44% of COVID-19 negative patients reported fevers and systemic symptoms were common. In contrast to other reports, [4,6,7] COVID-19 positive patients in this cohort had relatively high rates of upper respiratory symptoms (21% with headache, 27% with sore throat, and 30% with congestion/rhinorrhea) and gastrointestinal symptoms. In terms of laboratory values, patients with COVID-19 were significantly more likely to have lymphopenia and no patient with COVID-19 had leukocytosis.

Determining rates of co-infection in patients with COVID-19 has significance given that SARS-CoV-2 testing may be deferred if an alternative respiratory pathogen is identified, especially in settings with limited test availability. In this cohort, no patients with COVID-19 had evidence of viral co-infection by either clinical PCR testing or by mNGS analysis. Only one COVID-19 positive patient had evidence of co-infection with a bacterial respiratory pathogen, and no difference in the prevalence of bacterial co-infection was identified based on COVID-19 status. These results are distinct from those reported in a recent study of COVID-positive patients that found a 21% rate of viral co-infections [23] but consistent with data from several other institutions demonstrating very low rates (≤6%) of viral or bacterial co-infection in hospitalized COVID-19 positive patients, including two recent large studies from New York City. [15−18,20−23] Given the consistency in the low rate of co-infections across studies, it may be that there is an inherently low rate of viral and bacterial co-infection in COVID-19 patients. Alternatively, it is possible that early social distancing initiatives and school closures in San Francisco may have concomitantly reduced rates of other circulating respiratory viruses in our population. Similarly, the high rate of antibiotic use in our cohort may have contributed to a lower recovery rate of bacterial co-infection. Further investigation of co-infections in COVID-19 positive patients, and assessment of their potential impact on disease severity

Table 3
Results of infectious disease testing among 316 patients presenting with acute respiratory illness and tested for COVID-19.

	COVID-19 positive (n = 33)	COVID-19 negative (n = 283)	P value
Other viral testing performed	82% (27/33)	69% (194/283)	0.116
Influenza/Respiratory syncytial virus PCR	27% (9/33)	32% (90/283)	0.596
12-target respiratory virus PCR panel	55% (18/33)	54% (153/283)	0.958
Metagenomic next generation sequencing	42% (14/33)	29% (83/283)	0.123
Positive identification of virus other than SARS-CoV-2*	0% (0/27)	16% (31/194)	0.025
Influenza A[†]	0/27	5/194	
Influenza B[†]	0/27	2/194	
Respiratory syncytial virus[†]	0/27	3/194	
Rhinovirus[‡]	0/26	9/188	
Metapneumovirus[‡]	0/26	8/188	
Parainfluenza[‡]	0/26	1/188	
Coronavirus-229E[§]	0/14	2/83	
Coronavirus-NL63[§]	0/14	1/83	
Bocavirus[§]	0/14	1/83	
Blood culture ordered	19/33 (58%)	139/283 (49%)	0.358
Blood culture positive	1/19 (5%)	10/139 (7%)	1.000
Enterococcus faecalis	0/19	1/139	
Enterococcus faecium	1/19	1/139	
E. coli	0/19	1/139	
Group A Streptococcus	0/19	2/139	
Group C Streptococcus	0/19	1/139	
Group G Streptococcus	0/19	1/139	
Klebsiella pneumoniae	0/19	1/139	
Staphylococcus aureus	0/19	1/139	
Candida glabrata	0/19	1/139	
Sputum or lower respiratory culture ordered	9/33 (27%)	33/283 (12%)	0.012
Sputum or lower respiratory culture positive[ǁ]	1/9 (11%)	6/33 (18%)	1.000
Enterobacter cloacae complex	0/9	1/33	
H. parainfluenzae	0/9	3/33	
Staphylococcus aureus	0/9	1/33	
Pseudomonas aeruginosa	0/9	2/33	
Stenotrophomonas maltophilia	1/9	0/33	

Legend: COVID-19 - Coronavirus Disease 2019; PCR - polymerase chain reaction.
  * One case of viral co-infection identified (i.e., 32 pathogenic viruses in 31 patients).
  † ascertained by Influenza/RSV PCR or 12-target respiratory viral PCR panel or metagenomic next generation sequencing; 194 patients without COVID-19 and 27 with COVID-19 had any additional viral testing done.
  ‡ ascertained by 12-target respiratory viral PCR panel or metagenomic next generation sequencing; 188 patients without COVID-19 and 26 with COVID-19 had either test performed.
  § ascertained by mNGS only; 83 patients without COVID-19 and 14 with COVID-19 had mNGS testing performed.
  ǁ One case of multiple bacterial pathogens identified by sputum culture (i.e., 7 pathogenic bacteria in 6 patients).

Table 4
Treatment of 186 hospitalized patients with acute respiratory illness and tested for COVID-19.

	COVID-19 positive (n = 26)	COVID-19 negative (n = 160)	P value
Antibiotics administered	17/26 (65%)	134/160 (84%)	0.054
Vancomycin	8/26 (31%)	72/160 (45%)	0.126
Piperacillin/tazobactam	5/26 (19%)	55/160 (35%)	0.107
Cefepime	4/26 (15%)	17/160 (11%)	0.504
Ceftriaxone`	10/26 (39%)	74/160 (46%)	0.459
Carbapenems	3/26 (12%)	19/160 (12%)	1.000
Azithromycin	8/26 (31%)	44/160 (28%)	0.731
Doxycycline	7/26 (29%)	70/160 (44%)	0.106
Fluoroquinolones	4/26 (15%)	32/160 (20%)	0.581
Other antibiotics	4/26 (15%)	43/160 (27%)	0.329
Oseltamivir	3/26 (12%)	15/160 (9%)	0.729
Remdesivir clinical trial*	6/26 (23%)	0/160 (0%)	<0.001
Chloroquine	0/26 (0%)	0/160 (0%)	—
Hydroxychloroquine	6/26 (22%)	1/160 (<1%)	<0.001
Steroids	3/26 (12%)	23/160 (14%)	1.000
No respiratory support	6/26 (23%)	55/160 (34%)	0.255
Respiratory support			
Supplemental oxygen	10/20 (50%)	61/105 (58%)	0.711
High flow oxygen	5/20 (25%)	21/105 (20%)	
Noninvasive positive-pressure ventilation or invasive mechanical ventilation	5/20 (25%)	23/105 (22%)	

Legend
COVID-19 - Coronavirus Disease 2019.
  * Rows are not mutually exclusive, 1 patient received hydroxychloroquine and was enrolled in a blinded remdesivir trial.

Table 5
Outcomes of 186 hospitalized patients with acute respiratory illness and tested for COVID-19.

	COVID-19 Positive (n = 26)	COVID-19 Negative (n = 160)	Difference in proportions (95% CI)	P value
ICU admission				
ICU stay during hospitalization	11/26 (42%)	42/160 (26%)	16% (4%, 36%)	0.092
Time to ICU, median days (IQR)	3.1 (0.4, 4.77)	0.3 (0.2, 0.4)		0.027
ICU days, median days (IQR)*	8.8 (2.7, 17.8)	2.9 (1.6, 5.7)		0.005
Intensive care unit interventions				
Endotracheal intubation	6/11 (55%)	21/42 (50%)	5% (−28%, 38%)	0.788
Paralytics	2/11 (18%)	3/42 (7%)	11% (−0.7%, 15%)	0.275
Prone positioning	1/11 (9%)	0/42 (0%)	9% (−8%, 26%)	0.208
Vasopressors	6/11 (55%)	21/42 (50%)	5% (−28%, 38%)	0.788
Extracorporeal membrane oxygenation	0/11 (0%)	0/42 (0%)	−	−
Renal replacement therapy	1/11 (9%)	5/42 (12%)	−3% (−23%, 17%)	1.000
Acute respiratory distress syndrome[†]	6/26 (23%)	7/160 (4%)	20% (3%, 36%)	<0.001
Acquired cardiomyopathy[‡]	0/26 (0%)	5/160 (3%)	−3% (−6%, −0%)	1.000
Troponin tested	14/26 (54%)	113/160 (71%)	−17% (−37%, 3%)	0.088
Any troponin elevation	5/14 (36%)	37/113 (33%)	3% (−24%, 30%)	0.824
Acute kidney injury[§]	10/26 (39%)	56/160 (35%)	4% (−16%, 24%)	0.732
AKI First day	7/10 (70%)	37/56 (66%)	4% (−27%, 35%)	0.808
Abnormal coagulation test				
Elevated INR	4/19 (21%)	30/107 (28%)	−7% (−27%, 13%)	0.779
Elevated aPTT	5/10 (50%)	15/63 (24%)	26% (−7%, 59%)	0.085
Elevated D-dimer	4/4 (100%)	14/16 (88%)	12% (−4%, 28%)	1.000
Elevated fibrinogen	8/9 (89%)	12/20 (60%)	29% (−0%, 59%)	0.201
Final diagnosis				
Pulmonary - infectious	26/26 (100%)	63/160 (39%)	61% (53%, 69%)	<0.001
Pulmonary - non-infectious	0/26 (0%)	27/160 (17%)		
Other infectious	0/26 (0%)	24/160 (15%)		
Cardiac	0/26 (0%)	19/160 (12%)		
Malignancy	0/26 (0%)	6/160 (4%)		
Renal	0/26 (0%)	3/160 (2%)		
Other	0/26 (0%)	18/160 (11%)		
Discharge disposition				0.523
Died	1/26 (4%)	16/160 (10%)	−6% (−15%, 3%)	
Home	13/26 (50%)	78/160 (49%)	1% (−20%, 22%)	
Home hospice	0/26 (0%)	3/160 (2%)	−2% (−4%,0%)	
Home with services	10/26 (39%)	37/160 (23%)	16% (−4%, 36%)	
Skilled nursing facility	2/26 (8%)	25/160 (16%)	7% (−35%, 18%)	
Still admitted	0/26 (0%)	1/160 (1%)	−1% (−3%, 0%)	
Length of stay, median days (IQR)*	10.7 (7.9, 22.7)	4.7 (2.9, 7.0)		<0.001

Legend
All outcomes assessed through April 25, 2020.
COVID-19 - Coronavirus Disease 2019; ICU - intensive care unit; INR - international normalised ratio; aPTT - activated partial thromboplastin time,.
* censored at April 25; length of stay for those still admitted, calculated.
† ARDS defined using Berlin definition[37].
‡ based on treating physician diagnosis.
§ based on KDIGO definition[38].

and outcomes is needed, especially if SARS-CoV-2 circulation extends to overlap with other highly prevalent seasonal respiratory pathogens.

Although patients with COVID-19 were more likely to be diagnosed with ARDS, there were no differences in their need for ICU care or mechanical ventilation. We also did not find significant differences in terms of acquired cardiomyopathy or troponin elevation during the hospitalization. Despite concerns for cardiac complications in COVID-19 positive patients, our findings highlight the importance of comparisons to control groups of hospitalized patients. [16,48,59] Large proportions of patients in both groups received broad-spectrum antibiotics, despite all of the COVID-19 positive patients having a confirmed viral etiology. This has important implications for antibiotic stewardship in the COVID-19 era and likely reflects clinical uncertainty about the true rate of bacterial co-infection early in the pandemic. COVID-19 was associated with longer hospital lengths of stay. While the duration of hospitalization may reflect the severity of illness, it could also be a marker of concern for late decompensation in these patients [50] or difficulties with hospital discharge due to requirements for isolation and infection control.

Prior studies describing the clinical presentation of patients with COVID-19, have for the most part, identified non-specific features that characterize respiratory infections in general. To our knowledge, this is the first U.S. study to identify characteristics distinguishing patients with COVID-19 from patients who underwent investigation for COVID-19 but were ultimately found to have an alternate diagnosis. Previous publications on this topic are primarily smaller in scope and are all outside of the US. [40,40,43] The clinical, laboratory, and imaging data we highlight have important implications for front line providers making decisions in real-time regarding the pre-test probability of COVID-19, especially in settings with limited access to rapid COVID-19 diagnostics.

In contrast to other areas in the United States, the Bay Area has not yet experienced a large surge in cases of COVID-19. The fact that resources were not strained may have affected the clinical course and outcomes observed. For example, while the sample size is not sufficient to evaluate differences in mortality, only one of the 33 with COVID-19 died (3%), which is lower than in other studies of hospitalized U.S. patients. [17,18] There is speculation that variations in circulating SARS-CoV-2 strains may affect pathogenicity and contribute to

geographic differences in case fatality rates. [51,52] Exploratory phylogenetic analysis presented here demonstrated a diversity of strains among the COVID-19 patients requiring ICU care without a predominant clade; larger studies are needed to assess any potential relationship.

There are several limitations inherent to the study design and data available that should be considered when interpreting the results of this study. As a retrospective study based in a single academic medical center and focusing on patients presenting for emergency care, it may not generalize to other institutions with different patient populations or patients with milder forms of the disease. The study design relies on review of the medical record and thus variation in clinician assessment and documentation, particularly absent mention of symptoms and comorbidites, may result in misestimation of the prevalence of these clinical features. Although all patients in the COVID-19 negative group presented with respiratory complaints and/or influenza-like illness, only 56% of patients were given a final diagnosis of respiratory infection, which may affect the generalizability of our outcomes data. The low co-infection rate may have been influenced by incomplete testing for respiratory viral PCR and metagenomics though this is unlikely to have accounted for the full difference when compared to other cohorts. Additionally, as community-transmission increased, CDC clinical criteria for testing changed during the study period; this temporal change could bias the estimate of presenting clinical features. Finally, this study was undertaken at the end of the influenza season and during a period of social distancing, both of which likely impacted the prevalence of circulating viruses and the rate of co-infections.

In summary, while many clinical features of COVID-19 overlap with those of other acute respiratory illnesses, several unique characteristics were identified. Patients with COVID-19 had a longer duration of symptoms, particularly fatigue, fever, and myalgias, were more likely to be admitted to the hospital and for a longer duration, were unlikely to have co-existent viral infections, and were more likely to develop ARDS. Though this health system has not experienced a surge in COVID-19 cases, these key clinical characteristics may, in part, explain the observed differences in the propensity of COVID-19 to strain health systems. While we did find meaningful differences that may inform one's clinical suspicion for COVID-19, we did not find significant differences in cardiopulmonary comorbidities, ACE inhibitor/ARB use, or mortality rate. These findings enhance understanding of the clinical characteristics of COVID-19 in comparison to other acute respiratory illnesses.

Author contributions

Drs. Shah and Langelier had full access to all of the data and take responsibility for the integrity of the data and the accuracy of the data analysis.

Concept and design: Shah, Barish, Prasad, Kistler, Babik, Fang, Kangelaris, Langelier

Acquisition, analysis, or interpretation of data: Shah, Barish, Prasad, Kistler, Kamm, Li, Chiu, Babik, Fang, Kangelaris, Langelier, Abe-Jones, Alipanah, Alvarez, Botvinnik, Castaneda, The CZB CLIAhub Consortium, Dadasovich, Davis, Deng, Detweiler, Federman, Haliburton, Hao, Kerkhoff, Kumar, Malcolm, Mann, Martinez, Marya, Mick, Mwakibete, Najafi, Peluso, Phelps, Pisco, Ratnasiri, Rubio, Sellas, Sherwood, Spottiswoode, Tan, Yu

Drafting of the manuscript: Shah, Barish, Kistler, Kamm, Babik, Fang, Kangelaris, Langelier,

Critical revision of the manuscript for important intellectual content: All authors

Statistical analysis: Shah, Prasad, Li, Kamm, Hao, Martinez

Obtained funding: Shah, Chiu, Fang, Kangelaris, Langelier, DeRisi,

Supervision: Shah, Kistler, Chiu, Kangelaris, Langelier, DeRisi

Declaration of interests

Dr. Prasad reports personal fees from EpiExcellence, LLC, outside the submitted work. Dr. Chiu reports grants from National Institutes of Health/NHLBI, grants from National Institutes of Health/NIAID, during the conduct of the study. Dr. Peluso reports grants from Gilead Sciences, outside the submitted work. Dr. Deng has a patent 62/667,344 pending. All other authors have nothing to disclose.

Funding

Data sharing

Data used to complete this analysis used protected health elements. Researchers can contact the corresponding authors to request access to the study data.

Appendix

Table A1
Treatment of Emergency department and observation patients with COVID19 infection.

	COVID positive ($n = 7$)	COVID negative ($n = 123$)	P Value
Treatment			
Doxycycline	2/7 (29%)	13/123 (11%)	0.186
Fluoroquinolones	0/7 (0%)	3/123 (2%)	1.00
Azithromycin	2/7 (29%)	4/123 (3%)	0.033
Cephalosporin	1/7 (14%)	4/123 (3%)	0.245
TMP-SMX	0/7 (0%)	2/123 (2%)	1.00
Oseltamivir	0/7 (0%)	4/123 (3%)	1.00
No antimicrobials given on dc	3/7(43%)	100/123 (80%)	0.041
Respiratory support			
Supplemental oxygen	0/7 (0%)	3/123 (3%)	1.00
High Flow	0/7 (0%)	0/123(0%)	−
Crystalloid bolus volume within first 24 h (mean, SD)	1000 (0) $n = 3$	1351.4 (716) $n = 37$	0.406

Table A2
Complete microbiological test results for each patient.

Patient	COVID-19 PCR	Respiratory Viral PCR	mNGS	Respiratory Culture Pathogen	Blood Culture Pathogen	Multiplex Viral PCR Ordered	RSV/Flu PCR Ordered
1	negative		n/a			yes	yes
2	negative		negative			yes	yes
3	negative		n/a			yes	yes
4	negative		n/a			yes	no
5	negative		negative			no	no
6	negative		negative		*Candida glabrata*	no	no
7	negative		n/a			yes	no
8	negative		n/a			yes	no
9	negative		n/a			no	no
10	negative		n/a			yes	no
11	negative		n/a			no	no
12	negative	Human Metapneumovirus	n/a			yes	yes
13	negative	Rhinovirus + RSV	n/a			yes	yes
14	negative	Rhinovirus	Rhinovirus A			yes	no
15	negative		negative			yes	yes
16	negative		n/a			yes	yes
17	negative		n/a			no	no
18	negative		n/a			no	no
19	negative		n/a			yes	no
20	negative		n/a			yes	no
21	SARS-CoV-2		n/a			yes	yes
22	negative		n/a			yes	no
23	negative		n/a			no	no
24	negative		n/a			no	no
25	SARS-CoV-2		SARS-CoV-2			yes	no
26	negative		n/a			yes	yes
27	SARS-CoV-2		SARS-CoV-2			no	no
28	negative		n/a			no	no
29	SARS-CoV-2		invalid			yes	yes
30	negative	Human metapneumovirus	n/a			yes	no
31	negative		n/a			no	no
32	negative		negative			yes	yes
33	SARS-CoV-2		SARS-CoV-2			no	no
34	SARS-CoV-2		SARS-CoV-2			no	no
35	negative		n/a			yes	no
36	negative		n/a	*Staphylococcus aureus*		no	no
37	negative		negative			yes	yes
38	negative		n/a			no	no
39	negative		negative			yes	yes
40	negative		negative			no	no
41	negative		n/a			yes	no
42	negative		n/a			yes	no
43	negative		n/a		*Group A Streptococcus*	yes	no
44	negative		n/a			yes	no
45	negative		n/a			yes	yes
46	negative		n/a			no	no
47	negative		n/a			yes	no
48	negative	Rhinovirus	Rhinovirus C			yes	no
49	negative		negative			yes	yes
50	negative		n/a		*Group G Streptococcus*	yes	no
51	negative		Human CoV 229E			yes	yes
52	negative		n/a			no	no
53	negative		n/a			yes	no
54	negative		negative			yes	yes

Patient	COVID-19 PCR	Respiratory Viral PCR	mNGS	Respiratory Culture Pathogen	Blood Culture Pathogen	Multiplex Viral PCR Ordered	RSV/Flu PCR Ordered
55	negative	RSV	RSV			yes	yes
56	negative		n/a			no	no
57	negative		n/a			yes	no
58	negative		n/a			yes	yes
59	negative		n/a		*Klebsiella pneumoniae*	yes	yes
60	negative		n/a			yes	no
61	negative		negative			yes	yes
62	negative		n/a			no	no
63	negative		n/a			no	no
64	negative		negative			no	no
65	negative		n/a			no	no
66	negative		n/a			no	no
67	SARS-CoV-2		n/a			no	no
68	negative		n/a			no	no
69	negative		n/a			yes	no
70	SARS-CoV-2		SARS-CoV-2			yes	no
71	negative		n/a			no	no
72	negative		n/a			yes	yes
73	negative		negative			no	no
74	negative		n/a			no	no
75	negative		n/a			yes	no
76	negative	n/a	yes	no			
77	negative	negative	yes	yes			
78	negative	negative	yes	yes			
79	negative	Influenza A	n/a	no	yes		
80	negative	n/a	no	no			
81	negative	n/a	yes	no			
82	negative	invalid	no	no			
83	negative	n/a	no	no			
84	negative	n/a	yes	yes			
85	negative	negative	yes	yes			
86	SARS-CoV-2	SARS-CoV-2	yes	no			
87	SARS-CoV-2	SARS-CoV-2	yes	no			
88	negative	negative	no	no			
89	negative	negative	*H. parainfluenzae*	no	no		
90	negative	n/a	no	no			
91	negative	n/a	no	no			
92	negative	n/a	yes	yes			
93	negative	n/a	no	no			
94	SARS-CoV-2	n/a	yes	yes			
95	SARS-CoV-2	n/a	yes	yes			
96	SARS-CoV-2	n/a	no	no			
97	negative	negative	yes	no			
98	negative	negative	no	no			
99	negative	Human metapneumovirus	n/a	yes	yes		
100	negative	n/a	no	no			
101	negative	Human metapneumovirus	Human Metapneumovirus	yes	yes		
102	negative	n/a	yes	no			
103	negative	n/a	yes	no			
104	negative	n/a	no	no			
105	negative	n/a	yes	no			
106	SARS-CoV-2	n/a	yes	no			
107	SARS-CoV-2	n/a	no	yes			
108	SARS-CoV-2	invalid	yes	yes			
109	negative	n/a	yes	yes			

Table A2 (Continued)

Patient	COVID-19 PCR	Respiratory Viral PCR	mNGS	Respiratory Culture Pathogen	Blood Culture Pathogen	Multiplex Viral PCR Ordered	RSV/Flu PCR Ordered
110	SARS-CoV-2	SARS-CoV-2	*Stenotrophomonas maltophilia*	*Enterococcus faecium*	yes	yes	
111	negative	n/a	yes	no			
112	negative	RSV	RSV	yes	yes		
113	negative	negative	yes	yes			
114	negative	n/a	no	no			
115	negative	negative	*Staphylococcus aureus*	yes	yes		
116	negative	negative	no	yes			
117	negative	Human CoV NL63	no	no			
118	negative	n/a	*Group A Streptococcus*	yes	no		
119	negative	n/a	yes	yes			
120	negative	n/a	yes	yes			
121	negative	Rhinovirus	Rhinovirus A	yes	no		
122	SARS-CoV-2	n/a	no	no			
123	negative	negative	yes	yes			
124	negative	negative	no	yes			
125	negative	negative	yes	yes			
126	negative	n/a	yes	no			
127	negative	n/a	no	no			
128	negative	n/a	yes	yes			
129	SARS-CoV-2	n/a	yes	no			
130	negative	n/a	yes	no			
131	SARS-CoV-2	SARS-CoV-2	no	no			
132	negative	negative	*Enterobacter cloacae complex*	yes	yes		
133	negative	n/a	no	no			
134	negative	n/a	no	no			
135	negative	Influenza A virus	no	no			
136	negative	n/a	no	no			
137	negative	n/a	*Group C Streptococcus*	yes	yes		
138	negative	n/a	yes	yes			
139	negative	n/a	no	no			
140	negative	negative	no	no			
141	negative	Influenza B	n/a	no	yes		
142	negative	n/a	no	no			
143	negative	n/a	no	no			
144	negative	n/a	no	yes			
145	negative	negative	*Enterococcus faecium*	yes	no		
146	negative	n/a	yes	no			
147	negative	n/a	yes	no			
148	negative	invalid	yes	yes			
149	negative	n/a	no	yes			
150	negative	n/a	yes	no			
151	negative	n/a	no	no			
152	negative	negative	no	yes			
153	negative	negative	no	no			
154	negative	n/a	yes	yes			
155	negative	Human metapneumovirus	Human Metapneumovirus	yes	yes		
156	negative	n/a	yes	yes			
157	negative	negative	no	no			
158	negative	invalid	no	yes			
159	negative	n/a	yes	yes			
160	negative	Parainfluenza virus 4	no	no			
161	negative	negative	no	no			
162	negative	n/a	*H. parainfluenzae*	no	no		
163	negative	n/a	no	no			
164	negative	n/a	no	no			

Table A2 (*Continued*)

Patient	COVID-19 PCR	Respiratory Viral PCR	mNGS	Respiratory Culture Pathogen	Blood Culture Pathogen	Multiplex Viral PCR Ordered	RSV/Flu PCR Ordered
165	negative	n/a	yes	yes			
166	negative	n/a	yes	no			
167	negative	n/a	no	no			
168	negative	Influenza A	Influenza A virus	yes	yes		
169	negative	n/a	yes	yes			
170	negative	negative	yes	yes			
171	negative	n/a	*Enterococcus faecalis*	no	no		
172	negative	n/a	yes	yes			
173	SARS-CoV-2	n/a	no	no			
174	negative	n/a	yes	yes			
175	SARS-CoV-2	n/a	no	no			
176	negative	Rhinovirus	Rhinovirus A	*H. parainfluenzae*	yes	no	
177	negative	n/a	no	no			
178	SARS-CoV-2	SARS-CoV-2	no	no			
179	negative	n/a	yes	no			
180	negative	n/a	no	no			
181	negative	n/a	no	no			
182	negative	negative	no	no			
183	negative	Human metapneumovirus	invalid	yes	no		
184	negative	n/a	no	no			
185	negative	negative	yes	no			
186	negative	n/a	yes	yes			
187	negative	n/a	no	no			
188	negative	negative	yes	yes			
189	negative	n/a	yes	no			
190	negative	Human Metapneumovirus	no	no			
191	negative	n/a	no	no			
192	negative	n/a	no	no			
193	negative	n/a	no	no			
194	negative	negative	yes	no			
195	negative	n/a	no	no			
196	negative	invalid	yes	no			
197	negative	n/a	yes	no			
198	negative	n/a	no	no			
199	negative	n/a	no	no			
200	negative	n/a	no	no			
201	SARS-CoV-2	n/a	yes	no			
202	negative	n/a	yes	no			
203	negative	n/a	yes	no			
204	negative	n/a	yes	yes			
205	negative	negative	yes	yes			
206	negative	negative	no	no			
207	negative	n/a	no	no			
208	negative	n/a	no	no			
209	negative	negative	no	no			
210	negative	n/a	no	no			
211	negative	Human CoV 229E	no	no			
212	negative	n/a	no	no			
213	negative	n/a	no	no			
214	negative	n/a	yes	yes			
215	negative	negative	yes	yes			
216	SARS-CoV-2	SARS-CoV-2	no	no			
217	negative	n/a	no	no			
218	negative	invalid	yes	no			
219	negative	n/a	yes	no			

Table A2 (*Continued*)

Patient	COVID-19 PCR	Respiratory Viral PCR	mNGS	Respiratory Culture Pathogen	Blood Culture Pathogen	Multiplex Viral PCR Ordered	RSV/Flu PCR Ordered
220	negative	n/a	no	no			
221	negative	negative	yes	yes			
222	negative	n/a	no	no			
223	negative	negative	no	yes			
224	negative	n/a	no	no			
225	negative	invalid	yes	no			
226	negative	negative	no	no			
227	negative	n/a	no	yes			
228	negative	n/a	no	no			
229	negative	negative	yes	yes			
230	negative	Rhinovirus C	no	no			
231	negative	n/a	no	no			
232	negative	negative	yes	yes			
233	negative	n/a	yes	no			
234	negative	n/a	no	no			
235	negative	n/a	yes	no			
236	negative	n/a	no	no			
237	negative	Influenza A virus	no	no			
238	negative	Influenza A virus	no	no			
239	negative	n/a	yes	yes			
240	negative	negative	no	no			
241	negative	n/a	yes	yes			
242	negative	negative	yes	yes			
243	negative	n/a	no	no			
244	negative	n/a	yes	no			
245	negative	Rhinovirus	Rhinovirus A	yes	no		
246	negative	n/a	yes	no			
247	negative	n/a	no	no			
248	negative	n/a	yes	yes			
249	negative	n/a	no	no			
250	negative	n/a	no	no			
251	negative	n/a	no	no			
252	negative	n/a	yes	no			
253	negative	Rhinovirus C	no	no			
254	negative	negative	yes	no			
255	negative	n/a	*E. coli*	yes	no		
256	negative	n/a	yes	no			
257	negative	negative	yes	yes			
258	negative	n/a	yes	no			
259	negative	n/a	no	no			
260	negative	Human Bocavirus	yes	yes			
261	SARS-CoV-2	n/a	yes	no			
262	negative	n/a	yes	no			
263	SARS-CoV-2	SARS-CoV-2	no	no			
264	SARS-CoV-2	n/a	yes	no			
265	negative	n/a	yes	no			
266	negative	n/a	no	no			
267	negative	n/a	no	no			
268	negative	Human metapneumovirus	n/a	yes	yes		
269	negative	n/a	yes	no			
270	negative	n/a	yes	yes			
271	negative	negative	*Pseudomonas aeruginosa*	yes	yes		
272	negative	negative	yes	yes			
273	negative	n/a	yes	yes			
274	negative	Rhinovirus A	yes	yes			

Table A2 (Continued)

Patient	COVID-19 PCR	Respiratory Viral PCR	mNGS	Respiratory Culture Pathogen	Blood Culture Pathogen	Multiplex Viral PCR Ordered	RSV/Flu PCR Ordered
275	negative	negative	yes	yes			
276	SARS-CoV-2	n/a	no	no			
277	negative	negative	yes	yes			
278	negative	n/a	yes	yes			
279	negative	Influenza B	n/a	yes	yes		
280	negative	n/a	no	no			
281	negative	n/a	no	no			
282	negative	negative	no	no			
283	negative	negative	no	no			
284	negative	n/a	no	no			
285	negative	n/a	no	no			
286	negative	negative	no	no			
287	negative	negative	yes	yes			
288	negative	negative	no	no			
289	negative	invalid	no	no			
290	SARS-CoV-2	n/a	yes	yes			
291	negative	n/a	yes	no			
292	negative	n/a	Pseudomonas aeruginosa	yes	no		
293	negative	n/a	yes	no			
294	negative	n/a	no	no			
295	SARS-CoV-2	negative	no	no			
296	negative	n/a	no	no			
297	negative	n/a	no	no			
298	negative	n/a	yes	no			
299	negative	n/a	no	no			
300	negative	n/a	no	no			
301	negative	negative	no	yes			
302	negative	n/a	yes	no			
303	negative	n/a	yes	yes			
304	negative	n/a	yes	no			
305	negative	n/a	yes	yes			
306	negative	n/a	no	no			
307	negative	n/a	yes	no			
308	negative	n/a	no	no			
309	SARS-CoV-2	SARS-CoV-2	yes	no			
310	negative	n/a	yes	yes			
311	negative	n/a	yes	no			
312	negative	n/a	yes	no			
313	negative	n/a	yes	no			
314	SARS-CoV-2	n/a	yes	yes			
315	negative	n/a	no	no			
316	negative	n/a	no	no			

Legend: Respiratory culture: sputum, endotracheal aspirate or bronchoalveolar lavage; negative: not detected; n/a = not applicable because RNA from patient sample unavailable for testing; invalid = sample unable to be analyzed by mNGS due to insufficient (<25 pg) RNA.
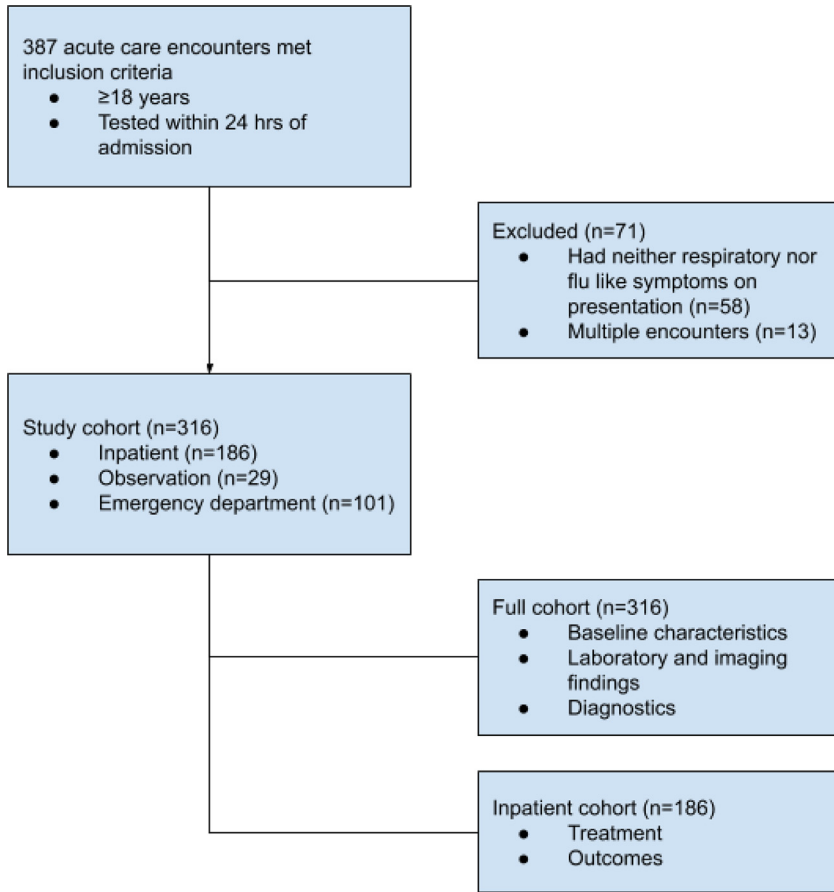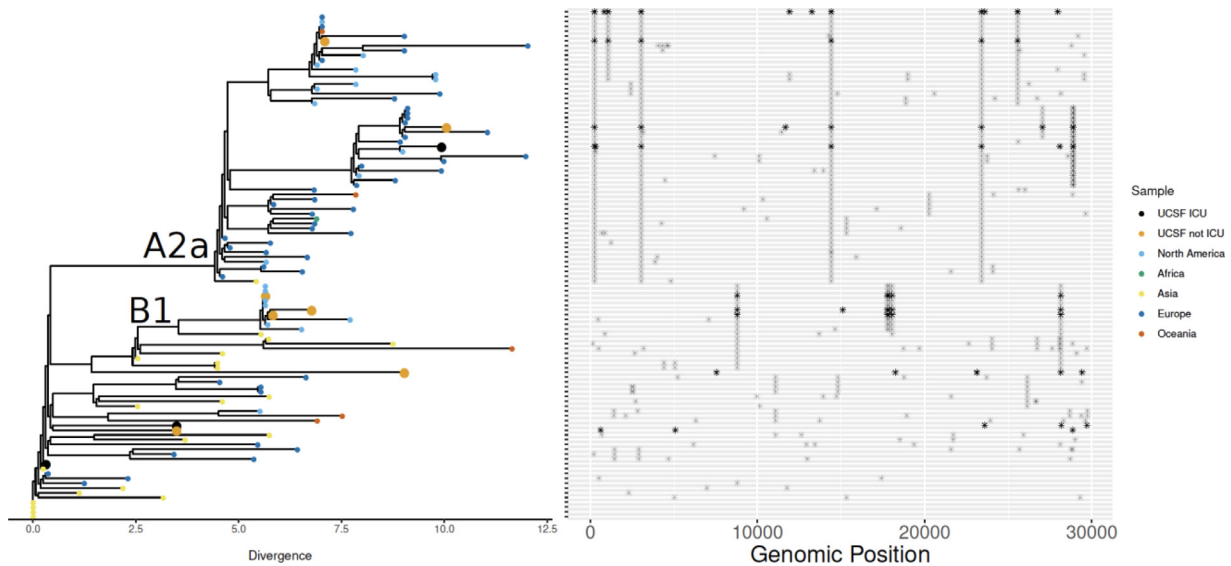
Fig. A1. Cohort flow diagram.

Fig. A2. Genomic epidemiology of SARS-CoV-2 in study population. Phylogenetic analysis of 10 SARS-CoV-2 genomes from patients in the cohort indicated strains originating from a diversity of geographic locations. Single nucleotide polymorphisms are plotted in the panel adjacent to the phylogenetic tree. Most samples fell into the Nextstrain.org clades A2a (widely prevalent in New York) and B1 (detected in Washington State in February 2020). The SARS-CoV-2 from patients who required ICU care were not associated with any single clade.

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