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Clinical and Laboratory Findings in Patients With Potential Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2) Reinfection, May–July 2020

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(See the Editorial Commentary by Piantadosi on pages 2226–7.)

Background. We investigated patients with potential severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) reinfection in the United States during May–July 2020.

Methods. We conducted case finding for patients with potential SARS-CoV-2 reinfection through the Emerging Infections Network. Cases reported were screened for laboratory and clinical findings of potential reinfection followed by requests for medical records and laboratory specimens. Available medical records were abstracted to characterize patient demographics, comorbidities, clinical course, and laboratory test results. Submitted specimens underwent further testing, including reverse transcription polymerase chain reaction (RT-PCR), viral culture, whole genome sequencing, subgenomic RNA PCR, and testing for anti-SARS-CoV-2 total antibody.

Results. Among 73 potential reinfection patients with available records, 30 patients had recurrent coronavirus disease 2019 (COVID-19) symptoms explained by alternative diagnoses with concurrent SARS-CoV-2 positive RT-PCR, 24 patients remained asymptomatic after recovery but had recurrent or persistent RT-PCR, and 19 patients had recurrent COVID-19 symptoms with concurrent SARS-CoV-2 positive RT-PCR but no alternative diagnoses. These 19 patients had symptom recurrence a median of 57 days after initial symptom onset (interquartile range: 47–76). Six of these patients had paired specimens available for further testing, but none had laboratory findings confirming reinfections. Testing of an additional 3 patients with recurrent symptoms and alternative diagnoses also did not confirm reinfection.

Conclusions. We did not confirm SARS-CoV-2 reinfection within 90 days of the initial infection based on the clinical and laboratory characteristics of cases in this investigation. Our findings support current Centers for Disease Control and Prevention (CDC) guidance around quarantine and testing for patients who have recovered from COVID-19.

Keywords. SARS-CoV-2; COVID-19; coronavirus; reinfection.

As of 3 January 2021, more than 83 million cases of coronavirus disease 2019 (COVID-19) have been confirmed worldwide, including 20 million cases in the United States [1]. For most diagnosed cases, the US Centers for Disease Control and Prevention (CDC) recommends completing isolation at 10 days after symptom onset with resolution of fever for at least 24 hours [2]. This recommendation is based on the absence of replication-competent virus for 10 days following symptom onset in mild to moderately severe cases. Retesting is not recommended for 90 days among persons who remain asymptomatic after recovery because severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) RNA can be detected in their upper respiratory specimens for up to 12 weeks [2–5]. Patients who do develop new symptoms within 90 days of recovery can be considered for retesting after investigation of alternative diagnoses. However, a better understanding of the duration and robustness of immunity to SARS-CoV-2 and the potential for reinfection would guide public health actions [6].

Reinfection has been documented among other species of human coronaviruses (HCoV), both experimentally [Callow] and in surveillance cohorts of community members [7, 8]. However, reinfection with SARS-associated coronavirus 1 (SARS-CoV-1) and Middle East respiratory syndrome coronavirus (MERS-CoV), 2 coronaviruses that can cause severe disease, has not been demonstrated, possibly related to the limited scope of these outbreaks [10, 11]. Recent case reports [12, 13]

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demonstrating phylogenic differences in SARS-CoV-2 genomes isolated from initial and recurrent episodes of COVID-19 in the same patients raise concern for reinfection; however, questions remain about the frequency and timing of such cases.

During March–April 2020, several reports described cases of recurrent or prolonged SARS-CoV-2 reverse transcription polymerase chain reaction (RT-PCR) positivity among individuals who had recovered from COVID-19 [14, 15]. An investigation in South Korea reported the absence of both viable virus and secondary transmission from investigated cases of recurrent SARS-CoV-2 RT-PCR positivity [4]. To better understand the clinical and public health implications of similar cases in the United States, in May 2020, CDC initiated an investigation of cases of potential reinfection. Here we summarize the clinical characteristics and available laboratory findings of cases of potential reinfection reported to CDC by clinicians and public health officials.

METHODS

Case Finding

We conducted case finding through the Emerging Infections Network (EIN), a provider-based sentinel network of over 1100 actively practicing infectious diseases professionals mainly from North America. EIN is administered by the Infectious Disease Society of America under a CDC cooperative agreement to help identify and understand emerging infectious diseases or clinical manifestations [16]. On 13 May 2020, we published a post on the EIN listserv soliciting reports about potential cases of reinfection from members. We also publicized this EIN posting to state, tribal, territorial, and local health departments. Cases of interest included patients with laboratory-confirmed COVID-19 who had any of the following after clinical recovery:

1) Recurrent COVID-19 symptoms and reverse transcription PCR (RT-PCR) positive result for SARS-CoV-2;
2) Two documented negative RT-PCR results followed by a positive RT-PCR result for SARS-CoV-2;
3) Persistently positive RT-PCR for SARS-CoV-2 results for >30 days.

Recovery was further defined as occurring at least 10 days after symptom onset and accompanied by resolution of fever for at least 24 hours, without the use of fever-reducing medications, and with improvement of other symptoms [17]. Clinicians and public health personnel could describe patients who met these criteria by completing a brief web-based form on the EIN website. We reviewed entries to assess if cases met potential reinfection criteria and contacted submitters via phone or email for any needed clarifications and to respond to questions.

During 13 May 2020 to 19 June 2020, we included patients for further clinical and laboratory characterization if they met the criteria listed in the EIN post. During 20 June 2020 to 17 July 2020, we narrowed the inclusion criteria to patients with recurrent COVID-19 symptoms [18] and a concurrent positive SARS-CoV-2 RT-PCR result without an alternative diagnosis. We excluded patients who were aged ≤18 years, not meeting the definition of recovery, or without sufficient clinical information to characterize the COVID-19 clinical course.

This investigation was determined to be nonresearch and was exempt from further institutional review board (IRB) review at CDC, and the University of Iowa IRB determined the investigation to be nonresearch. This activity was conducted consistent with applicable federal law and CDC policy (eg, 45 C.F.R. part 46, 21 C.F.R. part 56; 42 U.S.C. §241(d); 5 U.S.C. §552a; 44 U.S.C. et seq.).

Chart Abstraction and Case Review

For included cases, we requested deidentified medical records pertaining to COVID-19 care and SARS CoV-2 test results (RT-PCR testing laboratory, platform, and cycle threshold [Ct] values; serology). Data on demographics, comorbidities, clinical course, and laboratory testing were abstracted from the medical records and entered into a secured, electronic database (REDCap [Research Electronic Data Capture]) [19]. In addition to data validation checks, 3 clinician authors reviewed each case to ensure agreement on abstracted data fields, with a focus on SARS-CoV-2 test results and dates of onset and recovery of each clinical episode.

After chart abstraction, 3 clinician authors classified all patients into 4 categories based on their clinical course after recovery from an initial episode:

1) Recurrent COVID-19 symptoms without an alternative diagnosis, with concurrent positive SARS-CoV-2 RT-PCR result;
2) Recurrent COVID-19 symptoms with an alternative diagnosis (including potential complications following COVID-19), with concurrent positive SARS-CoV-2 RT-PCR result;
3) Asymptomatic with recurrent positive SARS-CoV-2 RT-PCR after 2 negative results in specimens collected 24 hours apart;
4) Asymptomatic with recurrent positive SARS-CoV-2 RT-PCR at ≥30 days after recovery, without 2 negative results in specimens collected 24 hours apart.

Data Analysis

We report descriptive statistics to characterize cases according to the 4 case categories. We expressed continuous variables as medians and interquartile ranges and summarized categorical variables as counts and percentages. All data were analyzed using R software, version 3.61 [20].

Laboratory Testing

We requested available respiratory specimens that tested RT-PCR positive for SARS-CoV-2 from 2 time points—the

initial diagnosis and at the recurrence of COVID-19 symptoms or recurrent test positivity. Respiratory specimens first underwent RT-PCR testing using the SARS-CoV-2 CDC assay protocol; Ct values were reported for the N1 and N2 viral nucleocapsid protein gene regions (CDC 2019-Novel Coronavirus [2019-nCoV] Real-Time RT-PCR Diagnostic Panel [21]). If the Ct value on the respiratory specimen was ≤34, we attempted: 1) viral culture using Vero-CCL-81 cells; 2) whole genome sequencing (WGS) of extracted nucleic acid [22]; and 3) detection of subgenomic viral RNA transcripts by RT-PCR (ie, Ct <40 for both subgenomic spike and nucleocapsid RNA). Specimens also underwent further testing at higher Ct values if laboratory capacity was available. Available serum specimens from after the initial diagnosis were analyzed by chemi-immunoluminiscent assay (CIA) to detect total anti-SARS-CoV-2 spike receptor-binding domain. Detailed laboratory methods are described in Supplementary materials.

RESULTS

Patient Characteristics

From 13 May 2020, through 17 July 2020, 296 potential cases of reinfection were submitted through the EIN. After initial review, 75 cases did not meet our initial investigation criteria, and another 51 did not meet our narrowed investigation criteria in place after 19 June. We requested records for 170 cases and received complete records for 93 cases; upon review of these records, another 20 did not meet investigation criteria (Supplementary Figure 1). In our report, 73 cases were ultimately included. Among these cases, the most common symptoms of the initial COVID-19 episode were respiratory followed by constitutional, and the most common underlying medical condition category was cardiovascular (Table 1).

Of the 73 patients, 49 (67.1%) developed recurrent COVID-19 symptoms after recovery, including 30 patients whose symptoms were explained by an alternative diagnosis, identified either clinically, through laboratory evaluation, or based on treatment response. Of these 30 patients, 8 (26.6%) had cardiac/circulatory (eg, congestive heart failure leading to shortness of breath) diagnoses at their subsequent episode, 7 (23.3%) had a bacterial infection (eg, pneumonia improved with antibiotics), 5 (16.7%) had noninfectious pulmonary diagnoses (eg, asthma exacerbation), 3 (10.0%) had gastrointestinal diagnoses, and 3 (10.0%) had neurological dysfunction; each of the remaining 4 (13.3%) patients had diagnoses related to either autoimmune, endocrine, urological disorders, or fever of unknown origin (case #24, Table 2). During the recurrent versus initial episode, a lower proportion of patients had respiratory (60.0% vs 90.0%) and constitutional symptoms (40.0% vs 73.3%), and a higher proportion had other symptoms (eg, headache and chest pain) (70.0% vs 46.7%) (Table 1). The recurrent episode developed a median of 49.5 days (interquartile range [IQR]: 32.3–61.3 days) after the start of the initial episode (Figure 1a).

Nineteen patients had recurrent COVID-19 symptoms unexplained by an alternative diagnosis. Relative to the 30 patients with alternative diagnosis, these 19 patients were younger (median age 32 vs 63 years), had a higher proportion of healthcare workers (68.4% vs 10.0%), and had fewer underlying conditions (cardiovascular disease 21.1% vs 73.3%; diabetes 5.3% vs 53.3%; lung disease 21.1% vs 36.7%; and immunocompromised 21.1% vs 36.7%). Further, they were hospitalized less often at both the initial and recurrent COVID-19 episodes (0.0% vs 56.7%, 5.3% vs 66.7%, respectively). Their recurrent symptoms developed a median of 57 days after initial symptom onset (IQR: 47–76 days; Figure 1b). At the recurrent episode, the majority of patients had respiratory signs and symptoms (16/19 [84.2%]) such as cough and shortness of breath, and constitutional signs and symptoms (14/19 [73.7%]) such as fever and fatigue. One patient required inpatient care, and an additional 13 patients received evaluation at an emergency department, urgent care, or outpatient setting. None of the 19 patients required admission to the intensive care unit or mechanical ventilation at the initial or subsequent episode of illness.

A total of 24 patients displayed no COVID-19 symptoms after recovery. Among 14 patients who remained asymptomatic but had recurrent positive SARS-CoV-2 PCR result after 2 negatives, 12 (85.7%) were long-term care facility residents tested during routine surveillance or facility contact investigations. These patients received their second negative test a median of 27 days (IQR: 21–33.3) after symptom onset and tested positive again a median of 14 days (IQR: 10.3–19) after receiving their second negative test (Figure 1c). Among the remaining 10 asymptomatic patients who remained PCR positive without the intervening two negative results, 3 were healthcare workers, whereas others were tested prior to elective medical procedures or as part of a test-based strategy to discontinue isolation. These patients tested positive a median of 56.5 days (IQR: 47.3–66.5) and up to 71 days after their initial symptom onset date (Figure 1d).

Laboratory Findings

We received paired specimens from the initial and recurrent COVID-19 symptomatic episodes for 9 cases, all of which had tested RT-PCR positive at outside labs under different assays and protocols. Of these, 6 patients had recurrent COVID-19 symptoms without an alternative diagnosis, and 3 patients had an alternative diagnosis. Table 2 shows the demographic, clinical presentation, and laboratory test results of these 9 cases. All 9 initial episode specimens tested positive for SARS CoV-2 on repeat RT-PCR using the CDC assay. Five initial episode specimens underwent additional testing, and all were positive for subgenomic SARS-CoV-2 RNA PCR and viral culture.

Table 1. Demographic and Clinical Characteristics of Potential Cases of Reinfection

Characteristics	All cases (n = 73)	Cases with recurrent symptoms and positive RT-PCR after recovery (n = 49)		Cases who remained asymptomatic with positive RT-PCR after recovery (n = 24)	
		Recurrent symptoms with alternative diagnosis (n = 30)	Recurrent symptoms without alternative diagnosis (n = 19)	Positive RT-PCR after 2 negative results (n = 14)	Positive RT-PCR > 30 days after recovery[a] (n = 10)
Female sex—no. (%)	38 (52.1%)	13 (43.3%)	13 (68.4%)	6 (42.9%)	6 (60.0%)
Median age (interquartile range)—yr	57.0 (37–69)	63 (52–76)	32 (29–40)	66 (62–73)	46 (37–59)
Healthcare worker	19 (26.0%)	3 (10.0%)	13 (68.4%)	0 (0.0%)	3 (30.0%)
Long-term care facility resident	26 (35.6%)	14 (46.7%)	0 (0.0%)	12 (85.7%)	0 (0.0%)
Underlying conditions					
Cardiovascular disease—no. (%)	39 (53.4%)	22 (73.3%)	4 (21.1%)	11 (78.6%)	2 (20.0%)
Diabetes—no. (%)	23 (31.5%)	16 (53.3%)	1 (5.3%)	5 (35.7%)	1 (10.0%)
Lung disease—no. (%)	22 (30.1%)	11 (36.7%)	4 (21.1%)	5 (35.7%)	2 (20.0%)
Immunocompromise—no. (%)	17 (23.3%)	11 (36.7%)	4 (21.1%)	2 (14.3%)	0 (0.0%)
Symptoms present at initial episode					
Respiratory—no. (%)	61 (83.6%)	27 (90.0%)	18 (94.7%)	11 (78.6%)	5 (50.0%)
Gastrointestinal—no. (%)	22 (30.1%)	8 (26.7%)	8 (42.1%)	4 (28.6%)	2 (20.0%)
Constitutional—no. (%)	51 (69.9%)	22 (73.3%)	17 (89.5%)	6 (42.9%)	6 (60.0%)
Other—no. (%)	36 (49.3%)	14 (46.7%)	15 (78.9%)	5 (35.7%)	2 (20.0%)
Symptoms present at subsequent episode					
Respiratory—no. (%)	34 (69.4%)	18 (60.0%)	16 (84.2%)	N/A	N/A
Gastrointestinal—no. (%)	16 (32.7%)	9 (30.0%)	7 (36.8%)	N/A	N/A
Constitutional—no. (%)	26 (53.1%)	12 (40.0%)	14 (73.7%)	N/A	N/A
Other—no. (%)	29 (59.2%)	21 (70.0%)	8 (42.1%)	N/A	N/A
Level of care at initial episode					
Inpatient hospital care—no. (%)	25 (34.2%)	17 (56.7%)	0 (0.0%)	7 (50.0%)	1 (10.0%)
Emergency department or urgent care—no. (%)	6 (8.2%)	2 (6.7%)	3 (15.8%)	0 (0.0%)	1 (10.0%)
Outpatient care—no. (%)	26 (35.6%)	8 (26.7%)	8 (42.1%)	7 (50.0%)	3 (30.0%)
Self-care—no. (%)	13 (17.8%)	2 (6.7%)	7 (36.8%)	0 (0.0%)	4 (40.0%)
Missing—no. (%)	3 (4.1%)	1 (3.3%)	1 (5.3%)	0 (0.0%)	1 (10.0%)
Level of care at subsequent episode					
Inpatient hospital care—no. (%)	21 (42.9%)	20 (66.7%)	1 (5.3%)	N/A	N/A
Emergency department or urgent care—no. (%)	8 (16.3%)	5 (26.3%)	3 (15.8%)	N/A	N/A
Outpatient care—no. (%)	13 (26.5%)	3 (10.0%)	10 (52.6%)	N/A	N/A
Self-care—no. (%)	5 (10.2%)	1 (3.3%)	4 (21.1%)	N/A	N/A
Missing—no. (%)	2 (4.1%)	1 (3.3%)	1 (3.3%)	N/A	N/A
Pattern for test results					
Positive RT-PCR occurring after two negative results	26 (35.6%)	11 (36.7%)	1 (5.3%)	14 (100.0%)	N/A
Positive RT-PCR occurring 30 days after recovery	54 (74.0%)	22 (73.3%)	14 (73.7%)	8 (57.1%)	10 (100.0%)

Abbreviations: N/A, not applicable; RT-PCR, reverse transcription polymerase chain reaction.
[a]Positive RT-PCR results >30 days after recovery without intervening 2 negative PCR results.

Table 2. Clinical Course and Laboratory Findings of Clinician-Suspected Cases of Reinfection With Laboratory Specimens for Testing

Case Patient ID	#7	#9	#10	#11	#15	#19
Demographics	27M pharmacist, otherwise healthy.	30F RN with a history of asthma and tachycardia	61M physician, otherwise healthy	30F RN with a history of asthma	25F nursing assistant, otherwise healthy	37F HCW, with a history of T2DM and psoriasis, not on immunomodulating drugs
1st episode symptom onset to recovery	16 March–30 March	March 23–April 2	March 11–March 22	March 22–April 2	April 14–April 24	May 3–May 13
1st episode primary symptoms	Fevers, cough, sore throat, shortness of breath	Fevers, cough	Fevers, cough, fatigue, rash on back and chest	Fevers, cough, nausea, vomiting, diarrhea	Fevers, cough, runny nose, change in taste/smell	Runny nose, change in taste/smell
Clinical course and treatment	Outpatient evaluation followed by home care, no steroid or antiviral use	Outpatient evaluation followed by home care, no steroid or antiviral use	Outpatient evaluation followed by home care, no steroid or antiviral use	Outpatient evaluation followed by home care, no steroid or antiviral use	Outpatient evaluation followed by home care, no steroid or antiviral use	Outpatient evaluation followed by home care, no steroid or antiviral use
1st episode specimen tested at CDC (collection date)	NP Swab (19 March)	NP Swab (24 March)	NP Swab (12 March)	NP Swab (24 March)	NP Swab (16 April)	NP Swab (3 May)
RT-PCR (N1 Ct)	Positive (19.7)	Positive (27.0)	Positive (18.6)	Positive (17.1)	Positive (16.3)	Positive (30.6)
Viral culture[a]	Not attempted[b]	Positive	Positive	Positive	Positive	Negative
WGS[a]	Whole genome obtained	Whole genome obtained	Whole genome obtained	Whole genome obtained	Whole genome obtained	Whole genome obtained
sgRNA PCR[a]	Not attempted[b]	Positive	Positive	Positive	Positive	Negative
2nd episode symptom onset	30 April	7 May	29 April	18 May	12 June	8 July
2nd episode primary symptoms	Fevers, chills, shortness of breath, change in taste/smell, diarrhea	Fever, cough, body aches, diarrhea	Fever	Nausea, diarrhea, myalgia	Fevers, chills, shortness of breath, change in taste/smell, diarrhea	Fevers, fatigue, wheezing
2nd episode specimen tested at CDC (collection date)	NP Swab (4 May)	NP Swab (8 May)	NP Swab (29 April)	NP Swab (18 May)	NP Swab (14 June)	NP Swab (8 July)
RT-PCR (N1 Ct)	Positive (32.4)	Positive (33.0)	Positive (37.0)	Inconclusive	Positive (37.7)	Negative
Viral culture[a]	Not attempted[b]	Negative	Not attempted	Not attempted	Negative	Not attempted
WGS[a]	Unsuccessful	Partial genome obtained	Unsuccessful	Not attempted	Unsuccessful	Not attempted
sgRNA PCR[a]	Not attempted[b]	Negative	Negative	Not attempted	Negative	Not attempted
SARS-CoV-2 antibody	No serum available	Reactive (7 May)	No serum available	Reactive (13 May)	No serum available	No serum available
Additional etiologies identified for recurrent symptoms	None	None	None	None	None	None

Table 2. Continued

Case ID	#43	#30	#24
Demographics	67M LTCF resident with heart failure, chronic kidney disease, bed bound from bilateral above knee amputations from peripheral artery disease	60F LTCF resident with morbid obesity, asthma, obstructive apnea, and hypertension, chronically bed bound with cerebral palsy	20F student and part time boat attendant, with a history of morbid obesity, hyperlipidemia, and mood disorders
1st episode symptom onset to recovery	April 3–April 21	April 1–April 11	April 15–April 25
1st episode primary symptoms	Fevers, wheezing, hypoxemia	Sore throat, cough, hypoxemia	Cough, runny nose, myalgias, shortness of breath
Clinical course and treatment	Outpatient care based at LTCF, no steroid or antiviral use	Outpatient care based at LTCF, no steroid or antiviral use	Outpatient evaluation followed by home care, no steroid or antiviral use
1st episode specimen tested at CDC (collection date)	NP Swab (4 April)	NP Swab (7 April)	NP Swab (15 April)
RT-PCR(N1 Ct)	Positive (19.3)	Positive (34.5)	Positive (36.0)
Viral culture	Positive	Not attempted	Not attempted
WGS	Whole genome obtained	Unsuccessful	Not attempted
sgRNA PCR	Positive	Not attempted	Not attempted
2nd episode symptom onset	1 June	2 June	12 June
2nd episode primary symptoms	Hypoxemia, shortness of breath, chest tightness	Fevers, sore throat	Fevers, myalgia, change in taste/smell, shortness of breath, nausea/vomiting
2nd episode specimen tested at CDC (collection date)	NP swab (1 June)	NP Swab (2 June)	NP Swab (15 June)
RT-PCR(N1 Ct)	Negative	Negative	Negative
Viral culture	Not attempted	Not attempted	Not attempted
WGS	Not attempted	Unsuccessful	Not attempted
sgRNA PCR	Not attempted	Not attempted	Not attempted
SARS-CoV-2 antibody (collection date)	Reactive (1 June)	Reactive (2 June)	No serum available
Additional etiologies identified for recurrent symptoms	Upon evaluation, diagnosed with heart failure exacerbation, symptoms improved after dieresis	Transiently hypoxemic in the context of altered mental status, evaluated briefly at emergency department and returned to LTCF.	Hospitalized and evaluated for fever of unknown origin given prolonged presentation, suspected to have mononucleosis vs Lyme disease.

Abbreviations: CDC, Centers for Disease Control and Prevention; HCW, healthcare worker; LTCF, long-term care facility; NP, nasopharyngeal swab; RN, registered nurse; RT-PCR, reverse transcription polymerase chain reaction; SARS-CoV-2, severe acute respiratory syndrome coronavirus 2.

[a]Respiratory specimens with Ct ≥34 values were not to undergo further testing by protocol but could have been tested if laboratory capacity were available.

[b]Further testing was not attempted on these specimens preexisting in the CDC lab before the start of our investigation.

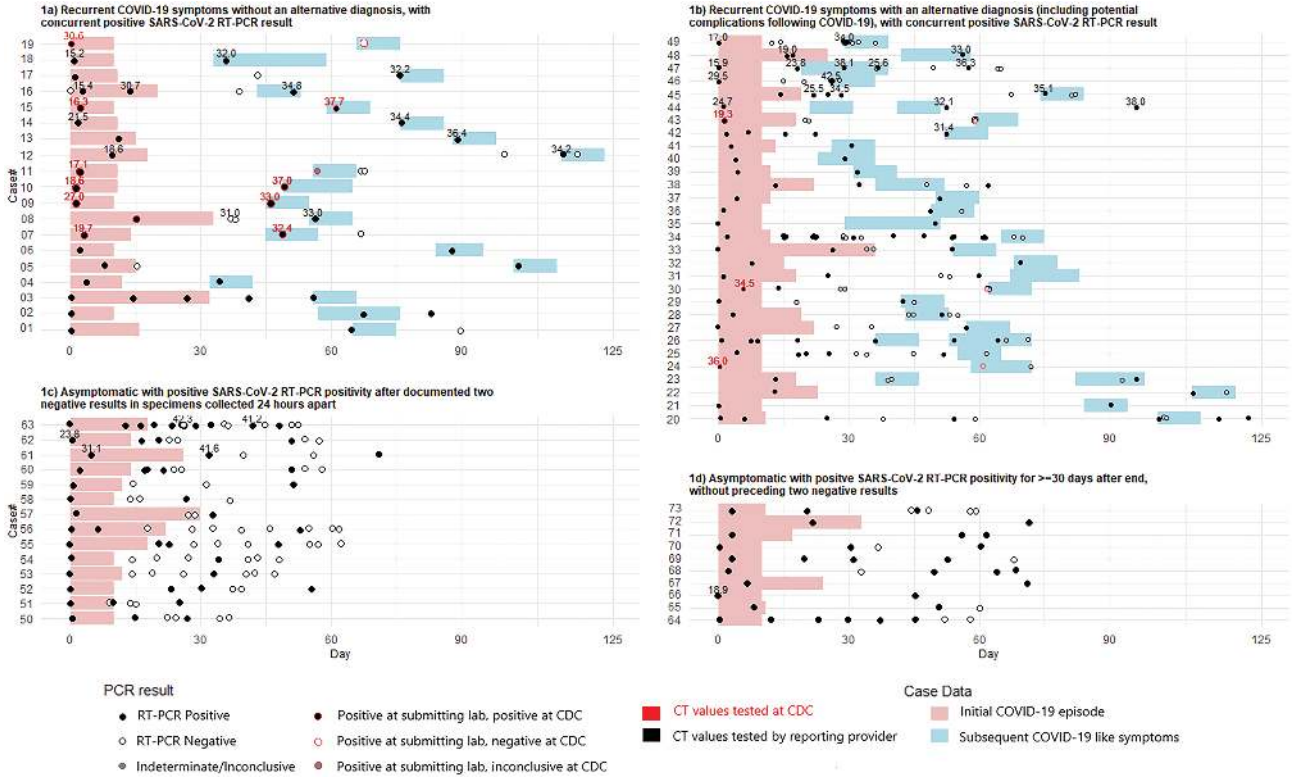

Figure 1. Onset of symptoms and RT-PCR results of clinician suspected cases of reinfection. Abbreviations: COVID-19, coronavirus disease 2019; RT-PCR, reverse transcription polymerase chain reaction; SARS-CoV-2, severe acute respiratory syndrome coronavirus 2.

The remaining 4 specimens did not undergo additional testing either due to high Ct values (3 cases) or because the specimen had been received prior to the start of our investigation (1 case).

Among 9 recurrent episode specimens, repeat RT-PCR at CDC was negative in 4 specimens and inconclusive for 1 specimen (Table 2). Among the 4 specimens with positive RT-PCR results, 2 specimens had Ct values >34 for N1 and N2 gene targets with viral cultures and whole genome sequencing not attempted or unsuccessful when attempted, and negative subgenomic RNA PCR. One specimen had a Ct value of 33.0 with negative viral culture, negative subgenomic RNA PCR, and partial genomic sequencing (<200 base pairs). A fourth specimen had a Ct value of 32.4, and whole genome sequencing was unsuccessful.

Serological specimens were available for 4 out of 9 cases—all were positive for antibodies against SARS-CoV-2 either immediately prior to or at the time of symptom onset for the recurrent episode.

Ct values were reported for an additional 16 cases using a variety of extraction techniques, platforms, and PCR targets applied at the diagnosing labs (Figure 1). Among these cases, only 1 case (case 48, Figure 1), a kidney transplant recipient with a complicated medical course, had a value <30 on the subsequent specimen.

DISCUSSION

In this investigation of 73 cases of potential SARS-CoV-2 reinfection reported from clinicians across the United States, we did not demonstrate reinfection within 90 days of the initial infection. Clinically, 70% of patients either had recurrent COVID-19 symptoms explained by alternative diagnoses or remained asymptomatic after recovery but were incidentally found to have recurrent or persistent RT-PCR positivity through surveillance and contact investigations. The remaining 19 patients, predominantly healthcare workers, were perhaps more concerning for reinfection because their recurrent COVID-19 symptoms, developing almost 2 months after recovery, had no alternative diagnoses. Further CDC laboratory investigations of 9 available paired specimens from these cases could not confirm reinfection, with an absence of culturable SARS- CoV-2, sub genomic RNA, or complete genome sequence from the recurrent episode specimen.

Results of paired RT-PCR Ct values for 16 cases, including 12 (of 19) cases most concerning for reinfection demonstrated Ct values >32 from the recurrent episode specimens, suggesting the absence of viable virus. Although Ct values are not standardized measures for viral burden, studies performed on diverse specimens, extraction techniques, and platforms found diminishing likelihood of viral isolation with Ct values >30 [3, 23, 24]. While low Ct values raise

suspicion for reinfection, the findings of higher Ct values cannot rule out reinfection but can provide context for the likelihood of replication-competent virus.

Our investigation and previous reports of SARS-CoV-2 reinfection highlight the need for a standardized approach in understanding reinfection. For instance, To et al demonstrated SARS-CoV-2 reinfection by showing phylogenetically distinct SARS-CoV-2 viruses from the first and second episodes of infection as well as Ct values and serological findings on the reinfection episode consistent with acute infection [9]. However, additional reports of potential cases of reinfection [24–26] did not present evidence of both distinct viral genomes and significant viral burden on reinfection.

To develop a common understanding of what constitutes reinfection, CDC has issued the Investigative Criteria for Suspected Cases of Reinfection [27], which provides guidance on prioritizing cases with a higher index of suspicion for reinfection and genomic testing of paired specimens, including quality criteria for testing and levels of evidence for reinfection. The highest priority for investigation is suggested for person with detection of SARS-CoV-2 RNA (RT PCR Ct value <33 if known) ≥90 days after the first detection, with or without symptoms, and if paired respiratory specimens are available. For persons with COVID-19–like symptoms and detection of SARS-CoV-2 RNA 45–89 days since first SARS-CoV-2 infection, additional criteria are applied, including absence of an obvious alternative etiology for COVID-19–like symptoms or having had close contact with a person with laboratory-confirmed COVID-19. CDC's guidance is expected to be updated as evidence regarding the duration and robustness of immunity to SARS-CoV-2 emerge. A Common Investigation Protocol (CIP) [28], has been available to support investigations into suspected SARS-CoV-2 reinfection cases.

This report has several limitations. This investigation was notable for difficulty in obtaining medical and laboratory reports, and specimens for further analysis at CDC, as most laboratories do not routinely retain positive specimens. These challenges contributed to the small sample of cases included in our investigation and tested at CDC. Further, our passive ascertainment of cases through EIN was subject to bias, with an over-representation of healthcare workers or long-term care facility residents, likely reflecting their increased access to SARS-CoV-2 testing. This investigation was focused on potential reinfection cases within 90 days of initial infection, thus we cannot generalize our findings to reinfection beyond 90 days, when waning immunity or divergent strains could increase susceptibility to reinfection.

Retesting of specimens collected in routine clinical care can have significant variations in specimen collection techniques, timing of repeat testing, and sample degradation that could reduce the likelihood of subsequent WGS, viral culture, and subgenomic RNA, and potentially introduce variability in Ct values. Although these issues with retesting of specimens at CDC could have played a role in the failure to culture and sequence recurrent episode specimens (almost all collected within 3 days of symptom onset), our laboratory successfully cultured and sequenced initial episode specimens. Because we did not collect patient identifiers, we were unable to collect exposure and secondary transmission information to support our laboratory findings. Finally, we were unable to rule out reinfection among patients who were asymptomatic with recurrent or persistently positive RT-PCR because we did not receive paired specimens from these cases.

In conclusion, this large public health investigation of potential reinfection contributed to our knowledge of the natural history of SARS-CoV-2 in the United States and informed a standard approach for assessing reinfection. We did not confirm SARS-CoV-2 reinfection within 90 days of the initial infection based on both clinical and laboratory characteristics of 73 cases. Our findings support CDC guidance around quarantine and testing for patients who have recovered from COVID-19 [2]. Additional systematic, prospective cohort investigations [29] are needed to better understand the clinical presentation, risk factors, and frequency of SARS-CoV-2 reinfection cases.

Supplementary Data

Supplementary materials are available at *Clinical 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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