

Prolonged SARS-CoV-2 RNA detection in anal/rectal swabs and stool specimens in COVID-19 patients after negative conversion in nasopharyngeal RT-PCR test

To the Editor,

Since the initial cases in late 2019 in Wuhan, China, the coronavirus disease 2019 (COVID-19) has spread rapidly across the world. As of 6 May 2020, there had been more than 3 588 773 confirmed cases, with more than 247 503 fatalities.¹ Current evidence suggests that COVID-19 is mainly spread through respiratory droplets and by fomites.² Recently, the severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2), the causative agent of COVID-19, has also been isolated from anal/rectal swabs and stool specimens,^{3,4} raising concerns of potential alternative routes of viral transmission. In previous epidemics such as the Zika and Ebola virus, the persistence of the virus in various body fluids was noted to occur in convalescing patients.^{5,6} It is however unclear as to whether this phenomenon is also present in the COVID-19 pandemic. We, therefore, systematically analyzed current literature to identify any evidence of prolonged SARS-CoV-2 detection in anal/rectal swabs and stool specimens in COVID-19 patients after negative conversion in nasopharyngeal reverse transcriptase-polymerase chain reaction (RT-PCR) test.

A systematic electronic search was carried out in Medline (PubMed interface) and China National Knowledge Infrastructure using the search strategy (a) "COVID-19" or "SARS-CoV-2" or "2019-nCoV"; (b) "Feces" or "Rectal Swab" or "Anal Swab"; (c) a and b, between 2019 and present time (ie, 8 May 2020), without applying language restrictions. The title, abstract, and full text of all documents identified with these search criteria were assessed to identify eligible studies. Studies were included if they reported paired rectal/anal/fecal and nasopharyngeal RT-PCR test results in COVID-19 patients, and had data on rectal/anal/stool test results after negative conversion of nasopharyngeal RT-PCR test result (at least two negative tests). The reference list of all retrieved documents was also hand-searched (through forward and backward citation tracking) for identifying additional eligible studies. Data were extracted by two independent reviewers (VK and IC). The synthesis of results was carried out in two steps. First, findings on all eligible studies reporting prolonged SARS-CoV-2 RNA detection in stool samples were presented in the form of a summary of findings table (Table 1). Thereafter, a pooled analysis incorporating only cohort studies or case series with sample ≥ 10 patients was conducted to calculate pooled prevalence estimates (PPE) of prolonged SARS-CoV-2 RNA detection in anal/rectal swabs and stool samples after negative conversion in nasopharyngeal RT-PCR using the MetaXL (software Version 5.3;

EpiGear International Pty Ltd, Sunrise Beach, Australia). A random effects model was applied due to the heterogeneity displayed by the data. The magnitude of heterogeneity among the included studies was assessed using the χ^2 test and I^2 statistic. For the χ^2 test, a Cochrane's Q P value of less than .10 was considered significant. The study was carried out in accordance with the declaration of Helsinki. As data were publicly available, no ethical approval was required.

The initial search produced 49 potentially relevant articles. Following primary screening and assessment by full text for eligibility in the meta-analysis, 37 articles were excluded since they were review articles ($n = 11$), commentaries, or other editorial materials ($n = 8$), or did not contain data on prolonged viral detection in anal/rectal swabs or stool specimens in COVID-19 patients ($n = 18$).

A total of 12 studies ($n = 324$ patients) were included in the final analysis.^{3,4,7,8-16} All studies were from China. Of the included studies, six were cohort studies, five were case series, while one was a case report. Four studies investigated prolonged viral shedding in pediatric patients only, three in adults, while the rest recruited both pediatric and adult subjects. The included studies compared the duration of SARS-CoV-2 positivity between nasopharyngeal swabs and stool samples (nine studies), anal swabs (two studies), and rectal swabs (one study) (Table 1). Overall, 107 had a positive rectal/anal/stool SARS-CoV-2 test after the negative conversion of the nasopharyngeal RT-PCR test. The reported duration of test positivity ranged from 5 to 35 days (Table 1).

In the pooled analysis of eight cohort studies/case series ($n = 315$), the PPE for prolonged rectal/anal/stool SARS-CoV-2 RNA was 32% (95% confidence interval, 22-44) (Figure 1). There was a high level of inter-study heterogeneity ($I^2 = 75\%$). There was insufficient data to perform meta-regression analysis to identify any moderators for the pooled prevalence. Nonetheless, no significant difference was observed in the leave-one-out sensitivity analysis. Therefore, there is some evidence of the persistence of SARS-CoV-2 in body secretion in convalescing COVID-19 patients. It is noteworthy that a significant proportion of these patients are within the pediatric age-group (Table 1). Furthermore, the study by Jiang and colleagues¹⁷ highlight the potential of gastrointestinal shedding of the virus even in asymptomatic patients.

That being said, RT-PCR does not usually distinguish between the infectious virus and noninfectious nucleic acid,¹⁸ and therefore viral isolation in the stool may not necessarily imply the potential for transmission or infectivity. However, Wang and colleagues¹⁹ recently

TABLE 1 Characteristics of the included studies

Study	Setting	Study design	Sample size	Study age group	Gastrointestinal symptoms	Specimens tested	Tests for SARS-CoV-2	Duration of pharyngeal swab positivity since index text	No. of patients with positive anal/rectal swab after negative conversion in nasopharyngeal swabs	Duration of positive anal/rectal swab after negative conversion in nasopharyngeal swabs
Hu et al ⁷	Zhejiang, China	Case series	2	Adults (female, 28 y and male, 25 y)	None	Nasopharyngeal and anal swabs	RT-PCR	Patient 1: 15 d Patient 2: 11 d	2	5-15 d
Fan et al ⁸	Yangtze, China	Case report	1	Pediatric (female, 3 mo)	Diarrhea	Nasopharyngeal and anal swabs	RT-PCR	14 d	1	14 d
Xu et al ⁹	Guangzhou, China	Case series	10	Pediatric (6 males, 4 females) Age range: 2 mo to 15 y	Diarrhea (3 patients)	Nasopharyngeal and rectal swabs	RT-PCR	2-10 d	8	6-20 d
Xing et al ¹⁰	Qingdao, China	Case series	3	Pediatric (2 males, 1 female)	Abdominal pain and diarrhea (1 patient)	Nasopharyngeal swab and fecal sample	RT-PCR	10-15 d	3	8-20 d
Xiao et al ³	Zhuhai, China	Cohort	73	Both pediatric and adult patients (10 mo to 78 y)	NA	Nasopharyngeal and fecal sample	RT-PCR	NA	17	...
Wu et al ⁴	Zhuhai, China	Cohort	74	NA	NA	Nasopharyngeal and fecal sample	RT-PCR	15.4 ± 6.7 d	41	11.2 ± 9.2 d
Chen et al ¹¹	Wuhan, China	Cohort	42	Adults (median age-51); 24 males, 27 females	Abdominal pain, diarrhea, vomiting (8 patients)	Nasopharyngeal and fecal sample	RT-PCR	6.5-13 d	18	7 (6-10) d
Zhang et al ¹²	Tianjin, China	Case series	3	Pediatric (males, 6-9 y)	Nausea and anorexia (2 patients)	Nasopharyngeal and fecal sample	RT-PCR	14, 11, and 7 d, respectively	3	16, 17, and 20 d, respectively

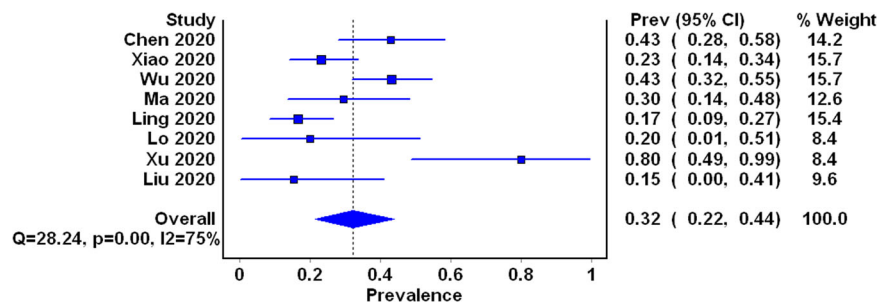
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TABLE 1 (Continued)

Study	Setting	Study design	Sample size	Study age group	Gastrointestinal symptoms	Specimens tested	Tests for SARS-CoV-2	Duration of pharyngeal swab positivity since index text	No. of patients with positive anal/rectal swab after conversion in nasopharyngeal swabs	Duration of positive anal/rectal swab after negative conversion in nasopharyngeal swabs
Ma et al ¹³	Shandong, China	Cohort	27	Both pediatric and adult patients	NA	Nasopharyngeal swab and fecal sample	RT-PCR	1-14 d	8	15-35 d
Lo et al ¹⁴	Macau, China	Case series	10	Both pediatric and adult patients; 3 males, 7 females	Abdominal pain, nausea, and diarrhea (8 patients)	Nasopharyngeal swab & anal fecal sample	RT-PCR	Mean: 18.2 d	2	6-10 d
Ling et al ¹⁵	Shanghai, China	Cohort	66	Both pediatric and adult patients	NA	Throat swab and fecal sample	RT-PCR	Median: 9.5 d	11	20 d
Li et al ¹⁶	Zhejiang, China	Cohort	13	Adults (6 males, 7 females)	NA	Nasal swab and fecal sample	RT-PCR	Median: 25 d	2	14 and 15 d, respectively

Abbreviations: NA, not applicable; RT-PCR, reverse transcriptase-polymerase chain reaction; SARS-CoV-2, severe acute respiratory syndrome coronavirus 2.

FIGURE 1 Forest plot for pooled prevalence estimate of prolonged severe acute respiratory coronavirus 2 RNA detection after negative conversion in the nasopharyngeal reverse transcriptase-polymerase chain reaction. CI, confidence interval




cultured four SARS-CoV-2-positive fecal specimens with high copy numbers, and successfully demonstrated live virus using electron microscopy in two specimens. These findings, however, need to be further investigated in larger cohorts of patients.

Our study was limited by the small number of studies, with small sample sizes. High-quality studies are urgently needed to better characterize the magnitude of SARS-CoV-2 viral persistence in body secretions, as well as its potential for disease transmission and infection, and possible implications for COVID-19 discharge and isolation policies.

CONFLICT OF INTERESTS

The authors declare that there are no conflict of interests.

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