Review Article

COVID-19 reinfection: Linked Possibilities and future outlook

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Abstract

SARS-CoV-2 is the third major coronavirus epidemic to affect humans. There had been multiple instances of patients turning positive after recovering from SARS-2-CoV infection. Though many different theories emerge, false positive RT-PCR is logically the foremost cause and there is a general consensus that during quarantine re-infection from outside seems unlikely when strictly adhered to. As many new strains emerge worldwide during the course of on-going pandemic, the chances of re-infection cannot be ignored as it may contribute to false negative RT-PCR test results. SARS-2-CoV though a novel virus, is phylogenetically similar to SARS-like CoV with around 79% similarity. Studies on immunological response to these infections suggest that antibodies formed after infection confers immunity only for a short period of time before it starts to wane. Also studies on SARS-CoV-2 suggest that antibody formation and longevity of immunity in an individual is dependent on the strain of coronavirus, its severity and age of the person infected. All these considerations demand reviewing the treatment duration, discharge criteria, appropriate use of imaging techniques and importance of risk communication and health education to those recovered.

Keywords: Antibody, Covid, immunity, pandemic, radiological investigation, re-infection, SARS

Introduction

Corona viruses (CoV) are the largest group of known positive-sense RNA viruses.[1] until now, with a wide range of natural hosts. The newly evolving corona viruses are turning out to be global threats for public health in the past few decades.[2] SARS-CoV-2 is the third major coronavirus epidemic to affect humans in recent times, after the SARS (Severe acute respiratory syndrome) outbreak of 2002-03 and the MERS (Middle East respiratory syndrome) outbreak that started in 2012.[3] The SARS-CoV-2 epidemic started in late December 2019 in Wuhan, China.[4] On 7 January 2020, the virus was identified as a novel coronavirus and the WHO named the virus officially as 2019 novel coronavirus (2019-nCoV), the new coronavirus in 2019, later renamed Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2).[1]

Since February, there has been multiple scenarios, in which, the discharged patients were tested positive again. Whether these are re-infections or relapses are yet to be explored. Scenarios of reinfection during past corona virus epidemics can steer us to the right direction giving an in-depth understanding of the current situation. This needs immediate attention because it has huge implications on understanding transmission dynamics, vaccine development and the practice/preparedness of primary healthcare. As a person's first and most regular point of contact with the health system, primary health care is key to effectively diagnosing, tracing and reporting cases, helping to slow the spread of the outbreak. Primary health care (PHC), which cater more than 70 percent of people's health needs at every age and

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every stage of life, in India must be central to these efforts. Strengthening primary health care with Covid testing, screening activities and basic treatment now will not only reduce the impact of COVID-19 on the health and wellbeing of majority of population but also will limit the unnecessary movements from primary to tertiary centre.

COVID-19 Reported cases of reinfection

First case of reinfection after recovery came from Japan, a female tour bus guide in Japan who tested positive for the virus after recovering from a COVID-19 infection. She first tested positive on January 29, 2020 and discharged according to the hospital criteria on February 1, 2020 when her symptoms relieved. The woman tested negative on February 6, 2020 during the monitoring period, a few days after her release. She again became positive with RT-PCR on February 26, 2020 after developing sore throat and chest pain during her follow up visit.[5]

Yuan *et al.* conducted a study to understand the risk of reinfection in COVID-19 for which they followed up a total of 172 patients who were discharged from Shenzhen Third People's Hospital during the time period of January 23, 2020 to February 21, 2020. Since then every third day two swabs (cloacal and nasopharyngeal) were collected from the discharged patients for RT-PCR detection. Among them, 25 was identified with positive results after an average of 7.32 ± 3.86 days from their last negative RT-PCR.[6]

Lan *et al.* reported that four medical professionals who were treated from January 1, 2020 to February 15, 2020 and who met the discharging criteria for the hospital i.e., absence of clinical symptoms and radiological abnormalities and 2 negative RT-PCR test results, turned RT PCR positive 5 to 13 days later. These findings suggest that at least a proportion of those patients who tested negative after their treatment was over, still harbor virus in them or getting reinfected with the same viral infection.[7]

In a study by Kenneth et.al from China, 20 patients who were discharged with negative RT-PCR tests from First Affiliated Hospital of Wenzhou Medical University and Wenzhou Central Hospital, were followed up for 2 weeks. Three among them were tested positive again after 7 days of release. As mentioned by the author, since all discharged patients followed a strict protocol for self-isolation, RNA positivity at follow up is unlikely to be due to reinfection.[8] Here, one possibility could be a relapse due to residual infection. According to WHO, sensitivity of RT- PCR in terms of 95% hit rate is about 100 copies of RNA genome equivalent per reaction. which suggests another possibility of a false positive test.[9]

Understanding immunology of Corona Viruses

SARS-CoV-2 is a lipid enveloped positive single-stranded RNA (ssRNA) coronavirus.[10] Both T and B cell responses against SARS-CoV-2 are detected one week after the onset of COVID-19 symptoms in the blood.[11,12]

By the second week neutralizing antibody responses against the S protein can be seen and, in most patients, it can be detected by third week.[13] IgM is the first antibody to develop during acute phase which has a shorter life span (approximately 7-21 days).[14] followed by the long-lasting neutralising antibody i.e., IgG. Past studies have confirmed that these antibodies confer protection against reinfection.[15]

In SARS-CoV-2, two-thirds of viral RNA, mainly located in the first open reading frame (ORF 1a/b), encodes 16 non-structure proteins (NSPs). The rest of the virus genome encodes for four essential structural proteins, including spike (S) glycoprotein, small envelope (E) protein, matrix (M) protein, and nucleocapsid (N) protein, and also several accessory proteins.[16] The spike (S) and the nucleocapsid (N) are the main proteins (antigens) which trigger an antibody response in humans.[17]

SARS-CoV-2 is a completely new virus with a high mutating capacity.[18] Phylogenetically, it is most similar to severe acute respiratory syndrome (SARS)-like CoV with around 79% similarity.[19] Since the evolutionary ancestor of SARS-CoV, MERS CoV, and SARS-CoV-2 is bat,[20] their immunology can be seen side by side to develop better understanding and understand their tendency.

Seasonal coronavirus is the most common corona virus known. It comes second to rhinovirus in causing common cold. In a study by Callow *et al.* on 15 volunteers inoculated with seasonal coronavirus 229E,10 got infected as indicated by virus shedding, again in them only 8 developed cold. The infected group had immediate rise in antibody, reached its peak on 14 days then it slowly fallen down to lower side. After 1-year, all volunteers were tested for IgG, and all 10 in the infected group were found to have a raised IgG. Out of all the volunteers in the infected group, 6 were re-challenged with coronavirus strains, none of them developed cold. After 2 years, they were followed up, and there was a significant decline in the specific IgG noticed.[21]

Taisheng *et al.* observed 30 recovered SARS patients over a 2-year period. They showed a persistence of neutralising antibody along with T-cell responses against virus, with a significant decline in the titre after one year.[22] Two other studies on SARS showed T-cell exhaustion during acute phase and the development of specific memory T-cells against SARS-CoV, which persisted for 2 years after recovery. This suggest the persistence of SARS antibody for a period of 1-2 years after recovery.[23]

Park WB *et al.* followed a cohort of 17 patients from the outbreak of MERS-CoV at South Korea in 2015, to know about the acute phase rising antibody response. The results of this study suggested that in patients with non-severe disease, a robust serologic antibody titre did not develop during the acute phase of the illness, and the patients remained seronegative or with marginal antibody titres at 1 year after infection. On the contrary, higher antibody titres continued to persist for at least 1 year in those with severe disease. The antibody waned during the first 6

months after infection but then stabilized in next 6 months.[24] In a study by Alshukairi *et al.* on health care workers, who were MERS survivors, he reported that, antibody was detected for greater than 18 months after infection, in those who had experienced severe pneumonia.[25]

In a study on MERS-CoV outbreak in Jordan during 2012, Payne *et al.* found that, antibodies against MERS-CoV, including neutralizing antibodies, persisted in 6 (86%) of 7 persons for 34 months[26] while in another study (Choe *et al.*) on 11 patients found neutralising antibody remained for less than 1 year and antibody titres on 4-6 patients who had mild illness found with undetectable amount of antibodies.[27]

Wu F *et al.* screened 175 COVID-19 recovered patients with mild symptoms and found that SARS-CoV-2 specific neutralising antibodies were formed in patients from day 10-15 after the onset of the disease and remained stable till the period of study. Neutralising and spike antibodies were formed, with elderly and middle-aged patients having a higher antibody titre compared to young people.[28]

We conclude that,

1. Longevity of immunity for different strains of coronavirus differs from each other.
2. Immunity against coronavirus depends on severity of infection.
3. Immunity against coronavirus depends on age of presentation of primary infection
4. The possibility of waning immunity against SARS-CoV-2 cannot be overlooked.

Mystery behind reinfections

Studies on reinfection have suggested that with strict adherence to quarantine, chances of reinfections from outside seems unlikely. So, of the many other possibilities, false positive RT-PCR results top the list.

In a clinical study by Xiao *et al.* on 70 COVID-19 patients, 15 (21.4%) patients experienced a "turn positive" of nucleic acid detection by RT-PCR test, after two consecutive negative results. False positivity of RT-PCR test and prolonged nucleic acid conversion were the two possible reasons proposed by them.[29] As, RT-PCR results are influenced by many external factors like specimen source (upper and lower respiratory swab), low patient viral load, sampling time, sampling technique, disease progression, performance of detection kits, so results of RT-PCR can't be 100% accurate.[30]

Tao *et al.* done detailed case study on two COVID-19 patients who tested positive again during the quarantine after hospital discharge and suggested the possibility of residual infection in lower respiratory tract which later transferred to upper respiratory tract leading to reappearance of cough giving RT-PCR test positive.[31] We can consider this situation as a case of "false negative test results as minimum viral load to get detected by RT-PCR was absent in previous samples. Though these cases can be minimized by taking lower respiratory swab which is again due to cost issue and technical difficulties is not feasible everywhere.

Most followed guidelines worldwide for discharge criteria of COVID-19 patients are two consecutive negative RT-PCR tests 24 hour apart, along with resolution of clinical symptoms.

Fang *et al.* found that, the sensitivity of CT scan for COVID-19 infection was 98% while RT-PCR sensitivity was only 71%.[30] In a large study on 1014 patients, 59% (601/1014) had positive RT-PCR results, and 88% (888/1014) had positive chest CT scans. In the same study, 60% to 93% of cases had initial positive CT consistent with COVID-19 prior to the initial positive RT-PCR results.[32] Further, in a Meta-Analysis study pooled sensitivity of CT- scan was found to be 94% and 89% for RT-PCR.[33] Even though sensitivity of CT-scan is higher, because of its high cost it cannot be included in discharge criteria especially in low-middle income countries. In those whom, symptoms reappear after two negative RT-PCR or unresolved symptoms present with negative RT-PCR or unhealed radiologic findings coexist with negative RT-PCR, they should be followed up with CT-scan in addition to RT-PCR. This would also help in addressing false negative cases.

Multiple studies have thrown light on re-emergence of new subtype of coronavirus. Tang *et al.* conducted population genetic analyses of 103 SARS-CoV-2 genomes and found two types of SARS-CoV-2, L type and S type. The L type is more prevalent (70%) and aggressive than the S type (30%) and also has a cap ability to spread more quickly than S type.[34] In a research it was found that, 27 isolates of SARS-CoV-2 can be divided into 6 genotypes, indicating that the SARS-CoV-2 has mutated in different patients.[35] Another group of scientists has discovered novel recurrent mutations of SARS-CoV-2 with their exact location. Four types of mutation have been observed in Europe while other 3 has been detected in England. RdRp is seen to be most common hotspot for mutation in European viral genomes.[36] Thus, all these mutations can affect primer and probe target giving "false negative" results.[37,38] There is chance that these cases remain undetected with high potential to become a cause of unknown death. Other possibility is this they can reinfect those who have already recovered from SARS-2-CoV especially when the severity and the duration of infection was less in them.

Recommendations

From above it is very obvious that recovered patients acquire some antibody through infection. The amount of antibody persisting in a patient will depend on severity of infection, age of individual, and also geographical region as new mutant varieties of novel coronavirus has been emerging in different parts of the world. In the present scenario with handful of positive RT-PCR on retesting, the false negative tests on discharge will hinder the efforts to control the pandemic. But, at this point where we are deficient in knowledge about reinfection, it cannot be completely denied.

Today many governments are approving the proposals of giving "Immunity passports" to recovered people after detecting the antibody titre in their blood. On the basis of presence of coronal antibody, they will be allowed to go back to work or travel.[38,39] Till we get strong evidence suggesting that the immunity developed after the infection confers protection for some period, allowing recovered persons to freely move about in the community would be dangerous to the community as well as the individual. More research is needed in this direction to find out the cut-off point to label someone 'immune' to the disease. Since, the knowledge about novel CoV is inadequate, our preparedness to tackle any emergency situation should always be one step forward We recommend following measures that should be followed at different levels to prevent any unknown community transmission because of patients discharged and still positive to transmit infection-

1. Those patients with unresolved symptoms with negative RT-PCR should be regularly followed up for extended period till two negative RT-PCR results is obtained with radiological clearance of infection.
2. Following patients can be followed up with CT-Scan along with RT-PCR for better management of patients-

a. Unresolved symptoms with negative RT- PCR results
b. Those who retested positive with RT-PCR after giving two negative RT-PCR
c. Radiological changes persisting but RT-PCR test came out negative

3. Patients even after recovering should follow all precautionary measures i.e., social distancing, wearing a mask, hand hygiene, etc., as recommended for COVID-19. Avoiding intimate contacts like sharing utensil and food with family members, having sex with partner during extended period of quarantine.
4. Recovered patients along with their family members should be properly educated regarding the nature of disease and possibility of re infection.
5. Where ever possible, use of CT-scan should be done when any hidden lower respiratory tract infection is suspected.

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Conflicts of interest

There are no conflicts of interest.

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