A SINGLE NEGATIVE SARS-COV2 RT-PCR TEST DOES NOT EXCLUDE COVID-19 IN PATIENT WITH SEVERE ARDS, CASE STUDY

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INTRODUCTION

Controlling the spread of SARS-CoV2 is currently the most urgent issue facing healthcare systems worldwide. According to this, the highest priority should be given to early identification and subsequent isolation of COVID19 suspected cases. RT-PCR is considered nowadays the gold standard method to confirm the active infection

THE AIM

The aim of this case study is to demonstrate that singular negative RT-PCR test for SARS-CoV-2 in symptomatic patient, should not be considered as conclusive

CASE STUDY

The 89-year-old resident of a nursing home was admitted to the Emergency Department with acute, severe dyspnea and hypotension. Due to fast respiratory deterioration rapid sequence intubation was performed in the ambulance with full personal protection regimen in place. At admission patient was fully conscious with excessive respiratory effort. Despite the fluid resuscitation and high doses of catecholamines, patient's hemodynamic was not stable, with mean artery pressure (MAP)<60mmHg and heart rate of 90/min generated by implantable cardiac pacemaker. Apart from severe hypoxemia refractory to 100% oxygen therapy and increased body temperature (37.4°C), laboratory tests revealed significant leukopenia (1.96 K/ul), lymphocytopenia (0.42 K/ul), high lactate dehydrogenase (371 U/L), high AST (55 U/L), elevated CRP (50.2 mg/L) and procalcitonin (10.5 ng/mL).

Due to the strong suspicion of COVID19 a single nasopharyngeal swab for SARS-CoV2 RT-PCR was obtained. Chest Computed Tomography (CT) demonstrated bilateral infiltrates more severe in the right lung with small pleural effusion, massive consolidations in the lower lobe and peribronchial consolidations with patchy ground-glass opacities in the middle lobe. The left lung was less affected with consolidations mainly in the lower lobe. Mediastinal and hilar lymphadenopathy was not observed [Figure 1]. Consequently, the estimated probability of COVID19 was below 70% as per British Society of Thoracic Imaging guidelines (BSTI).

According to administrational procedures patient was transferred to the Department of Pneumonology designated for COVID19 screening. Blood samples for standard microbiology cultures were collected. Despite aggressive treatment with meropenem, azithromycin and hydroxychloroquine as well as invasive mechanical ventilation patient's condition remained unstable meeting criteria of severe Acute Respiratory Distress Syndrome (ARDS) with FiO₂ 1,0, PEEP 12 cmH₂O and calculated PaO₂/FiO₂ ratio 50.1 [1]. Due to resistant hypotonia patient was maintained on norepinephrine and dobutamine infusion with and positive fluid balance. Laboratory tests repeated within 24 hours showed low WBC count (0.82 K/ uL), a two-fold increase in creatinine, hypoalbuminemia 32 g/L and severe acid-base balance disorders with mixed respiratory-metabolic acidosis pH 7.25.

The first RT-PCR test for SARS-CoV2 (nasopharyngeal swab) proved negative, therefore patient was transferred to the ICU and no longer isolated in accordance with the hospital safety protocol.

In the following hours patient's condition deteriorated. Blind bronchoalveolar lavage (BAL) samples for standard microbiological diagnosis were collected, while echocardiography showed no obvious heart pathology with estimated left ventricular ejection fraction of 45%. In order to maintain MAP 65mmHg, in addition to norepinephrine and dobutamine adrenaline infusion was started. As soon as hemodynamic stabilized, lung recrutation was performed according to the ARDS-net protocol, but no significant improvement in oxygenation was achieved. Maximal SpO₂ value with FiO₂ =1.0 after recrutation maneuver was 88-90%.

During four-day ICU stay we observed only a slight increase in WBC count (maximal 3.19 K/ μ L), persistent anuria and severe mixed acid-base disorders. All microbiological samples were found negative. Significant deterioration of patient's condition prompted retesting for COVID19 and sample from the blind BAL was harvested on day 6. In the next few hours patient died because of the multiorgan failure. CPR was not attended.



Fig 1. Computed Tomography images on admission. A – lung apices, B – ground-glass opacity in the right middle lobe, C – left lower lobe consolidations, D right-sided pleural effusion and right lower lobe consolidations



Fig. 2. Diagnostic protocol for suspected COVID-19 infection.

The result of repeated RT-PCR testing was received after 48h and proved to be positive. This led to an in immediate exclusion from active clinical work and mandatory 7-day quarantine of 11 physicians, 42 nurses, and 4 other hospital employees exposed to the direct unprotected contact with the patient. Two RT-PCR testing for SARS-CoV-2 separated by 7 days proved negative in all personnel.

DISCUSSION

Here, we present a case of a COVID-19 patient hospitalized due to ARDS who was found negative at first SARS-CoV2 molecular testing, but eventually proved positive when repeatedly sampled for RT-PCR after six days.

This case is of crucial importance both individually for the patient but also in respect to the in-hospital care organization and medical personnel safety. All should be carefully addressed.

Reliability of molecular testing for SARS-CoV2 is of ultimate importance since RT-PCR is considered nowadays the gold standard method to confirm the active infection. Yet, in this particular case the false negativity of the first RT-PCR analysis seems highly probable.

Insufficient sample quality or laboratory error are the major sources of false negative RT-PCR results. While BAL provides the optimal material in terms of testing sensitivity (93%), the nasopharyngeal swab is considered the sample of choice in the everyday clinical practice (63% sensitivity) [2, 3]. Indeed, the bias caused by incorrect swabbing technique seems to be the most common and cannot be underestimated, but in this case, we consider it negligible while on admission the latter was collected by well experienced anesthesiologist re-trained in the swabbing technique as per the very recent recommendations [4]. Yet non-surprisingly, it was the bronchoalveolar lavage that provided positive outcome of SARS-CoV2 RT-PCR upon second sampling. SARS-CoV-2 hits primarily the ACE2+/TMPRSS2+ cells mainly pneumocytes II, but also nasal goblet secretory cells and ileac absorptive enterocytes [5]. Therefore, it is recommended to collect specimen directly from the lower respiratory tract if possible. In particular in severe, intubated patients, the targeted BAL of the most involved lobe should be considered alongside the potential risk of using bronchofiberoscopy in such difficult setting [6]. The patient did not expectorate and standard BAL was not feasible due to the severity of patient's condition. Therefore, blind bronchial sampling was performed as an optimal testing method [7].

The optimal period for sample harvesting in COVID-19 patients is between days 4th to 10th since the onset of symptoms due to the highest viral load in the upper respiratory tract [4]. In this case first swab was collected at the hospital admission, while the other at day six with concomitant signs of severe ARDS at both times. Importantly, there are no direct data to confirm that SARS-CoV-2 viral load in the respiratory tract is negatively correlated with age and disease severity. However, there are reports suggesting the higher risk of consecutive false negative testing in elderly patients with mild to moderate COVID-19 which might in part relate to the quantity of virus present in the airway [8].

Other potential source of insufficient reliability of SARS-CoV2 RT-PCR is the RNA stability, which depends on proper sample preservation and transport [9]. Yet, in this case the optimal shipment conditions were ensured. As for any diagnostic method, the individual variability of laboratory assay needs to be considered, however at the moment there are no published, head-to head reliable comparisons. Nevertheless, any diagnostic platform includes internal control enabling proper supervision. In addition, analyses should be performed by a laboratory experienced in molecular diagnostic of respiratory viral infections. The overall management of testing process, including sampling and laboratory standard operating procedures (SOPs) are crucial, as reliability of analyses depends on number of variables and consequently, the false-negative result rate ranges from 17% to 63% [10]. Therefore, current guidelines emphasize the need for the very careful and critical interpretation of any laboratory diagnostic for COVID-19, including RT-PCR [2]. Accordingly, symptomatic patient with suggestive chest CT could be considered negative only after a second sample collected at least after 48h have turned negative [2, 4]. Yet, vigilance and common sense should be always employed as there are case reports of patients fully symptomatic with typical chest CT, negative in consecutive RT-PCRs, who tested positive after more than 10 days since the COVID-19 onset [11]. The recommendations have not been followed in this case, as at that time the Polish guidelines for the diagnostic and management of patients with SARS-CoV2 infection have not yet been published. Indeed, the current document as well as international recommendations clearly state that high risk patients should undergo additional RT-PCR testing within 24-48h, as presented in figure 2 [4, 12,]. Thus, this case strongly attests to the need of clear and unified strategy for triage, diagnostic and treatment of patients with suspected COVID -19 in line with latest evidence and recommendation.

Finally, it could not be discounted that the primary RT-PCR test was truly negative. Symptomatology on admission fulfilled the criteria of health-care acquired pneumonia. Therefore, the primary reason for severe ARDS might be other than SARS-CoV2 any bacterial or viral pathogen. A hospital acquired SARS-CoV-2 coinfection, although possible, is highly unlikely. While the proportion of pre-symptomatic transmission (i.e. from healthcare professional to patients) is estimated between 48% and 62% [13], the medical personnel with confirmed direct contact with the patient has been tested and found negative. Also, studies suggest that coinfection with other respiratory pathogens is common in patients with COVID-19 (mainly Mycoplasma pneumonia, Pseudomonas aeruginosa and Haemophilus influenzae), but the differential diagnosis can be hard to perform. Thus, according to surviving sepsis protocol broad spectrum antibiotics should be administered immediately but ideally after blood cultures obtained. In our case the PCT concentration was high, but the respiratory tract and blood cultures found to be negative, either because of the early broad-spectrum antibiotics administration or presence of or non-bacterial cause of elevated PCT such as multiorgan failure [14,15]. In addition to recommended molecular testing, a wide variety of immunological tests are available. Despite their advantages, which include mainly the simplicity and short time to obtain results, there are some major limitations - rapid tests detecting virus antigen are not accessible in many European countries, and rapid antibodies tests have limited usefulness for early COVID-19 diagnosis due to ten days of window period. World Health Organization is now performing validation studies of these assays to determine their place in COVID-19 diagnosis [16].

The in-hospital standard operating procedures for COVID19 have been introduced early on in our center and two separate medical teams working in the weekly rounds regimen were organized to prevent in-hospital SARS-CoV-2 transmission as well as assure patients and personnel safety.

Unfortunately, in this particular case the safety protocol has failed twice rendering patients and staff undefended. Due to the single negative result of RT-PCR testing patient stopped being considered "COVID-19 suspect" within 24 hours after admission. Accordingly, personnel protection regimen relaxed allowing possibility of uncontrolled in-hospital virus transmission. As a consequence, the significant number of medical personnel and their families has been put in quarantine.

Despite an attempt to contact the Nursing Home, we did not obtain reliable information concerning other residents and healthcare professionals, and thus we cannot determine the probable chain of infection.

CONCLUSIONS

Currently, there are numerous unanswered concerns regarding COVID-19 diagnostic and management, however this case report clearly demonstrates that singular negative RT-PCR test for SARS-CoV-2 in symptomatic patient, in particular within ICU, should not be considered as conclusive. All precautions preventing potential in-hospital transmission should be in place and sustained until consecutive negative testing result is available.

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Conflict of interest

Authors declare no conflict of interest

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