OGRANIČENJA PRIMENE BRZIH SEROLOŠKIH TESTOVA NA SARS-KOV-2 KOD NEVAKCINISANIH PACIJENATA U URGENTNIM KARDIOŠKIM STANJIMA

PRIKAZ SLUČAJA

CASE REPORT

LIMITATIONS OF RAPID SEROLOGICAL TESTING FOR SARS-COV-2 IN NON-VACCINATED PATIENTS IN ACUTE CARDIAC CARE

Mihajlo Viduljević[1], Marija Polovina[1,2], Milika Ašanin[1,2], Igor Mrdović[1,2]

[1] Klinika za kardiologiju, Univerzitetski Klinički centar Srbije, Beograd, Srbija

[2] Medicinski fakultet Univerziteta u Beogradu, Beograd, Srbija

[1] *Cardiology Department, University Clinical Center of Serbia, Belgrade, Serbia*

[2] *Faculty of Medicine, Belgrade University, Belgrade, Serbia*

SAŽETAK

U decembru 2019. godine, u gradu Vuhanu, u Kini, registrovani su prvi slučajevi infekcije izazvane novim SARS-KoV-2 virusom. U daljem toku je bolest izazvana ovim virusom nazvana KOVID-19. Ubrzo nakon toga, 11. marta 2020. godine, Svetska zdravstvena organizacija proglasila je pandemiju KOVID-19 infekcije. KOVID-19 i akutna kardiološka stanja (npr. srčana insuficijencija, embolija pluća ili ishemija miokarda) često imaju sličnu kliničku prezentaciju, što predstavlja izazov u postavljanju adekvatne dijagnoze u kliničkoj praksi. Osnovna namena brzih seroloških testova za detekciju IgM i IgG antitela na SARS-KoV-2 je procena imunološkog odgovora na ovaj virus. Međutim, brzi serološki testovi se često upotrebljavaju van okvira osnovne namene, za trijažu potencijalno inficiranih, nevakcinisanih pacijenata, zbog brzog dobijanja rezultata, što je naročito važno u urgentnim stanjima. Ukoliko se serološko testiranje koristi za trijažu nevakcinisanih pacijenata, u smislu prijema u izolacionu jedinicu za suspektne KOVID-19 pozitivne bolesnike ili na odeljenja gde su smešteni KOVID-19 negativni bolesnici, potrebno je poznavati njihova ograničenja da bi se redukovao rizik lažno pozitivnih i lažno negativnih rezultata. Neophodna je adekvatna selekcija pacijenata i obazriva interpretacija rezultata kako bi se izbegla pogrešna dijagnoza. Cilj ovog rada je da opiše kako se serološki testovi mogu upotrebiti za trijažu kardioloških pacijenata, navakcinisanih protiv SARS-KoV-2 virusa, koji zahtevaju urgentnu hospitalizaciju. Radi ilustracije, prikazujemo dva bolesnika kod kojih je primena ovih testova dala svrsishodne rezultate.

Ključne reči: KOVID-19, trijaža, lažno pozitivni, lažno negativni, pogrešna dijagnoza

ABSTRACT

In December 2019 in the city of Wuhan, in China, the first cases of infection caused by the new SARS-CoV-2 virus appeared, and later on, the disease caused by this virus was named COVID-19. Shortly after this, on March 11th, 2020, the WHO characterized COVID-19 as a global pandemic. The symptoms of COVID-19 and acute cardiovascular disorders (e.g., heart failure, pulmonary embolism or myocardial ischemia) frequently overlap, which poses a challenge for the establishing of a differential diagnosis in clinical practice. Rapid serological tests, which detect IgM and IgG classes of antibodies for SARS-CoV 2, have been developed with the primary purpose of screening the population's immunological response to the SARS-CoV-2 virus. However, rapid serological tests are often used outside their original purpose, i.e., for the triage of possibly infected, non-vaccinated individuals, because they offer quick results, which may be particularly relevant in emergency settings. If serological testing is used to guide the admission of non-vaccinated patients with acute cardiovascular disorders to either an isolation unit for suspected COVID-19 positive individuals, or to hospital facilities for non-infected patients, it is important to recognize its limitations, in order to reduce the risk of false-positive or false-negative results. Hence, appropriate patient selection and cautious test interpretation is necessary to avoid misdiagnosis. The aim of this paper is to illustrate how serological testing may be used as a screening tool to inform the management of non-vaccinated patients with acute cardiovascular disorders requiring urgent hospital admission. As an illustration, we describe two clinical situations, in which serological testing produced meaningful results.

Key words: COVID-19, triage, false-positive, false-negative, misdiagnosis

Autor za korespondenciju:
Marija Polovina
Klinika za kardiologiju, Univerzitetski klinički centar Srbije
Medicinski fakultet Univerziteta u Beogradu,
Koste Todorovića 8, 11000 Beograd, Srbija
E-mail: maki.marijapolovina@gmail.com

Corresponding author:
Marija Polovina
Cardiology Department, University Clinical Center of Serbia
Faculty of Medicine, University of Belgrade
8 Koste Todorovića Street, 11000 Belgrade, Serbia
E-mail: maki.marijapolovina@gmail.com

Primljeno • *Received*: June 28, 2021; **Revidirano • *Revised*:** July 24, 2021; **Prihvaćeno • *Accepted*:** July 28, 2021; ***Online first*:** September 30, 2021.

DOI: 10.5937/smclk2-32938

UVOD

U decembru 2019. godine, u gradu Vuhanu, u Kini, registrovani su prvi slučajevi infekcije izazvane novim SARS-KoV-2 virusom. U daljem toku je bolest izazvana ovim virusom nazvana KOVID-19 [1]. Ubrzo nakon toga, 11. marta 2020. godine, Svetska zdravstvena organizacija proglasila je pandemiju KOVID-19 infekcije. Karakteristična klinička slika uključuje obostranu intersticijumsku upalu pluća; no, kod mnogih pacijenta takođe dolazi i do pojave akutnog respiratornog distres sindroma (ARDS) ili do multiorganske disfunkcije. Konkretno, osobe koje pate od kardiovaskularnih oboljenja imaju dva do tri puta veći rizik od nepovoljnog ishoda, u poređenju sa pacijentima bez pridruženih bolesti [2].

Simptomi KOVID-19 oboljenja i akutnih kardioloških stanja (npr. srčane insuficijencije, embolije pluća ili ishemije miokarda) često imaju sličnu kliničku prezentaciju, što predstavlja izazov u postavljanju adekvatne dijagnoze u kliničkoj praksi, pogotovo kod urgentnih stanja. Potvrda KOVID-19 infekcije iziskuje pozitivan antigenski test ili pozitivan test reverzne transkripcije lančane reakcije polimeraze (*RT-PCR*) nazofaringealnog sekreta, što su standardne dijagnostičke metode [3]. Brzi antigenski test nije bio dostupan u većini zdravstvenih sistema tokom najvećeg dela 2020. godine. Kada je u pitanju, *RT-PCR* test, može biti potrebno dosta vremena da bi se dobili rezultati. Povremeno, pacijenti sa negativnim rezultatom mogu biti nosioci SARS-KoV-2 virusa, sa kasnijim razvojem infektivnosti (lažno negativni test), čime predstavljaju opasnost za dalje širenje virusa. Brzi serološki testovi, koji detektuju IgM i IgG klase antitela na virus SARS-KoV-2, razvijeni su sa primarnim ciljem da se pomoću njih proceni imunološki odgovor populacije na virus SARS-KoV-2 [4]. Međutim, brzi serološki testovi su često, tokom 2020. godine, korišćeni van okvira osnovne namene, za trijažu potencijalno inficiranih pacijenata, zbog brzog dobijanja rezultata, što je naročito važno u urgentnim stanjima. Cilj ovog rada je da opiše kako se serološki testovi mogu upotrebiti za trijažu pacijenata, navakcinisanih protiv SARS-KoV-2 virusa, koji zahtevaju urgentnu hospitalizaciju, usled akutnih kardiovaskularnih oboljenja.

Radi ilustracije, prikazujemo dve kliničke situacije, kod kojih je primena seroloških testova dala svrsishodne rezultate.

Prvi je slučaj sedamdesetšestogodišnjeg muškarca koji je u periodu od 10 dana pre prijema imao visoku temperaturu (38,6 °C) i dispneju. Pacijent je imao istoriju kardiomiopatije i znakove srčane insuficijencije (S3 galop, bilateralni kasnoinspirijumski pukoti na plućima, hepatomegaliju, edem donjih ekstremiteta), ali je takođe, 12 dana pre prijema, bio u kontaktu sa članom porodice pozitivnim na KOVID-19, a nije bio vakcinisan

INTRODUCTION

In December 2019 in the city of Wuhan, in China, the first cases of infection caused by the new SARS-CoV-2 virus were registered, and later on, the disease caused by this virus was named COVID-19 [1]. Shortly after this, on March 11th, 2020, the WHO characterized COVID-19 as a global pandemic. The typical clinical presentation includes bilateral interstitial pneumonia; however, many patients also develop acute respiratory distress syndrome (ARDS) or multiorgan involvement. In particular, individuals with cardiovascular diseases have a twice to threefold higher risk of adverse outcomes, as compared to patients without comorbidities [2].

The symptoms of COVID-19 and acute cardiovascular disorders (e.g., heart failure, pulmonary embolism or myocardial ischemia) frequently overlap, which poses a challenge for the establishing of a differential diagnosis in clinical practice, especially in emergency settings. The confirmation of COVID-19 infection requires a positive antigen test or a positive reverse transcriptase polymerase chain reaction (RT-PCR) test of nasopharyngeal secretions, as standard diagnostic methods [3]. Rapid antigen testing was not available in most healthcare settings during the best part of 2020. With regard to RT-PCR testing, it may take a considerable amount of time to obtain the results. Occasionally, patients with a negative test result may be SARS-CoV-2 carriers, with delayed development of viral shedding (false- negative test), thus posing a threat for the spreading of the virus. Rapid serological tests, which detect IgM and IgG classes of antibodies for SARS-CoV-2, have been developed with the main purpose of screening the population for immunological response to SARS-CoV-2 [4]. However, rapid serological tests have often been used during 2020 outside their original purpose, for the triage of possibly infected individuals, because they offer quick results, which may be particularly relevant in emergency settings. The aim of this paper is to illustrate how serological testing may be used as a screening tool to inform the management of non-vaccinated patients with acute cardiovascular disorders requiring urgent hospital admission.

As an illustration, we describe two clinical situations, in which serological testing produced meaningful results.

The first case is a 76-year-old male presenting with high fever (38.6 °C) and dyspnea for 10 days before admission. The patient had a history of cardiomyopathy and signs of heart failure (S3 gallop, bilateral lung rales, hepatomegaly, lower extremity edema), but also, he had been in contact with a COVID-19 positive family member 12 days before admission, and he had not been vaccinated for SARS-CoV 2. Although he was

protiv virusa SARS-KoV 2. Iako je pacijentu dijagnostikovana dekompenzovana srčana insuficijencija, pozitivan test na IgM antitela za SARS-KoV-2 indikovao je prijem u izolacionu jedinicu, a KOVID-19 je potom potvrđen pozitivnim *RT-PCR* testom.

Drugi je slučaj sedamdesetdevetogodišnjeg muškarca sa dispnejom, subfebrilnom temperaturom (37,4 °C), oštrim bolovima u grudima, u trajanju od tri dana pre prijema, i negativnim epidemiološkim upitnikom na KOVID-19, koji je takođe bio nevakcinisan. Dijagnostikovana mu je plućna embolija, ali je nalaz kompjuterizovane tomografije (CT) takođe otkrio i ekstenzivne obostrane promene denziteta „mlečnog stakla", suspektne na KOVID-19 pneumoniju. Test ovog pacijenta na SARS-KoV-2 IgM antitela je bio negativan, ali je on ipak primljen u izolacionu jedinicu, s obzirom na to da je bio visoko klinički suspektan na KOVID-19 oboljenje, koje mu je u daljem toku potvrđeno pozitivnim *RT-PCR* testom.

DISKUSIJA

Ukoliko se serološko testiranje primenjuje (kada brzi antigenski test nije dostupan) kako bi se donela odluka o tome da li da se osobe nevakcinisane protiv KOVID-19 infekcije sa akutnim kardiovaskularnim oboljenjima prime u jedinicu za izolaciju pacijenata za koje postoji sumnja da su pozitivni na KOVID-19 ili u kapacitete bolnice za neinficirane pacijente, važno je razumeti ograničenja ovog testa u pogledu smanjenja rizika od lažno pozitivnih ili lažno negativnih rezultata. Stoga, u ovu svrhu, treba primenjivati samo testove sa visokom senzitivnošću i specifičnošću. Test koji se koristi u našoj ustanovi ima prijavljenu senzitivnost od 87% i specifičnost od 100% za detektovanje IgM klase antitela, dok je senzitivnost za otkrivanje IgG klase antitela 96%, a specifičnost 98%.

PREVENCIJA LAŽNO NEGATIVNIH REZULTATA TESTA

Prioritet treba dati nevakcinisanim pacijentima sa najvišom verovatnoćom pre testiranja, odnosno visokom verovatnoćom da su zaraženi infekcijom KOVID-19 i da produkuju broj antitela koje je moguće detektovati, za šta je potrebno oko jedne do tri sedmice od inficiranja [5]. Ovo je dobro ilustrovano prvom opisanom kliničkom situacijom, gde su epidemiološki podaci bili ubedljivi i gde je prošlo dovoljno vremena da bi se mogla detektovati antitela.

Za pacijente sa niskom verovatnoćom pre testiranja, kao što je u drugom slučaju opisanom u ovom radu (javljanje lekaru u prvih 7 do 10 dana nakon pojave simptoma, uz nejasne podatke o izloženosti virusu), serološko testiranje može biti nesvrsishodno i može dati lažno negativne rezultate. U takvoj situaciji, treba primeniti ortogonalni algoritam testiranja. Ovo iziskuje

diagnosed with decompensated heart failure, positive IgM antibodies for SARS-CoV-2 indicated admission to an isolation unit, and COVID-19 was subsequently confirmed with a positive RT-PCR test.

The second case is a 79-year-old male presenting with dyspnea, low-grade fever (37.4 °C), stabbing chest pain, which had begun three days before admission, and a negative epidemiological survey for COVID-19, also non-vaccinated. He was diagnosed with pulmonary embolism, but computed tomography (CT) scanning also revealed extensive bilateral ground-glass opacities, suspicious of COVID-19 pneumonia. The patient tested negative for SARS-CoV-2 IgM antibodies but was, nevertheless, admitted to the isolation unit, due to high clinical suspicion of COVID-19, which was subsequently confirmed with a positive RT-PCR test.

DISCUSSION

If serological testing is used (when rapid antigen testing is not available) to guide the admission of non-vaccinated patients with acute cardiovascular disorders to either an isolation unit for suspected COVID-19 positive individuals, or to hospital facilities for non-infected patients, it is important to recognize its limitations in reducing the risk of false-positive or false-negative results. Hence, only tests with high sensitivity and specificity should be used for this purpose. The test used in our institution has a reported sensitivity of 87% and a specificity of 100% for detecting the IgM class of antibodies, whilst the sensitivity and specificity for detecting the IgG class of antibodies is 96% and 98%, respectively.

PREVENTING FALSE-NEGATIVE TEST RESULTS

Priority should be given to non-vaccinated patients with high pretest probability, i.e., high likelihood of having the COVID-19 infection and of producing a detectable number of antibodies, which takes approximately one to three weeks from infection [5]. This point is well illustrated by the first clinical situation described above, where the epidemiological data were compelling, and sufficient time had elapsed to allow antibody detection.

For patients with low pretest probability, as in the second case described above (i.e., presenting within the first 7-10 days after symptom onset, with unclear exposure data), serological testing may be futile, yielding false-negative results. In this situation, an orthogonal testing algorithm should be employed. This implies the sequential use of two independent tests (SARS-CoV-2 antigen detection in nasopharyngeal secretions or RT-PCR testing) to validate serological findings. In our case, the patient was admitted to an isolation unit

sekvencijalnu primenu dva nezavisna testa (detektovanje SARS-KoV-2 antigena u nazofaringealnom sekretu ili *RT-PCR* test) kako bi se potvrdio serološki nalaz. U našem slučaju, pacijent je primljen u izolacionu jedinicu, na osnovu kliničke sumnje, i sprovedeno je ortogonalno testiranje primenom *RT-PCR* testa kako bi se potvrdila KOVID-19 infekcija. Važno je napomenuti da je ovakav pristup omogućio i odgovarajuće kliničko zbrinjavanje i prevenciju širenja SARS-KoV-2 virusne infekcije unutar bolnice.

PREVENCIJA LAŽNO POZITIVNIH REZULTATA TESTA

Iako se smatra da serološki testovi imaju visoku specifičnost i senzitivnost na SARS-KoV-2, izuzetno se mogu pojaviti lažno pozitivni rezultati testa kod nevakcinisanih pacijenata. Malo je dostupnih podataka, ali oni koji postoje govore u prilog tome da kod pacijenata sa visokim titrom reumatoidnog faktora, IgM antitela na SARS-KoV-2 mogu dati lažno pozitivan rezultat, pretpostavlja se zbog ukrštene reaktivnosti antitela [6]. Na isti način, opisani su slučajevi ukrštene reaktivnosti sa serološkim testovima za denga groznicu i Kavasakijevu bolest [7,8]. Ove retke, ali klinički relevantne situacije, moraju se uzeti u obzir prilikom tumačenja rezultata testova.

ZAKLJUČAK

Može se zaključiti da dijagnostička upotrebljivost seroloških testova kod nevakcinisanih pacijenata zavisi od verovatnoće pre testiranja i verovatnoće stvaranja antitela u dovoljnom broju da se ona mogu detektovati. Stoga, ukoliko se serološko testiranje koristi u svrhu trijaže pri prijemu u bolnicu, neophodan je odgovarajući odabir pacijenata i oprezno tumačenje rezultata testiranja kako bi se izbegla pogrešna dijagnoza. Nadalje, *RT-PCR* test nastavlja da bude zlatni standard za potvrđivanje KOVID-19 infekcije.

Sukob interesa: Nije prijavljen.

based on clinical suspicion, and orthogonal testing with the use of RT-PCR to confirm COVID-19 was employed. It is of note that this approach allowed both appropriate clinical management and the prevention of in-hospital spread of the SARS-CoV-2 infection.

PREVENTING FALSE-POSITIVE TEST RESULTS

Although serological tests are considered to have high sensitivity and specificity for SARS-CoV-2, exceptionally, false-positive results in non-vaccinated patients may occur. Available data are scarce, but the data that does exist suggest that, in patients with a moderate to high titer of the rheumatoid factor, IgM antibodies for SARS-CoV-2 can test false-positive, presumably due to antibody cross-reactivity [6]. In the same way, cases of cross-reactivity with serological tests for dengue fever and Kawasaki disease have been described [7,8]. These rare, but clinically relevant situations, need to be considered when interpreting test results.

CONCLUSION

In conclusion, the diagnostic validity of serological tests in non-vaccinated patients depends on the pre-test probability and the likelihood of producing a detectable number of antibodies. Hence, if serological testing is used to guide hospital admission, appropriate patient selection and cautious test interpretation is necessary to avoid misdiagnosis. Furthermore, RT-PCR remains the gold standard for confirming COVID-19.

Conflict of interest: None declared.

LITERATURA / *REFERENCES*

1. Zhu N, Zhang D, Wang W, Li X, Yang B, Song J, et al. A Novel Coronavirus from Patients with Pneumonia in China, 2019. The New England Journal of Medicine. 2020;382(8):727-33.
2. Li B, Yang J, Zhao F, Zhi L, Wang X, Liu L, et al. Prevalence and impact of cardiovascular metabolic diseases on COVID-19 in China. Clin Res Cardiol. 2020.
3. Sharfstein JM, Becker SJ, Mello MM. Diagnostic Testing for the Novel Coronavirus. JAMA. 2020;323(15):1437-8.
4. Caini S, Bellerba F, Corso F, Díaz-Basabe A, Natoli G, Paget J, et al. Meta-analysis of diagnostic performance of serological tests for SARS-CoV-2 antibodies up to 25 April 2020 and public health implications. Euro Surveill. 2020;25(23).
5. Deeks JJ, Dinnes J, Takwoingi Y, Davenport C, Spijker R, Taylor-Phillips S, et al. Antibody tests for identification of current and past infection with SARS-CoV-2. The Cochrane Database of Systematic Reviews. 2020;6(6):Cd013652.
6. Wang Q, Du Q, Guo B, Mu D, Lu X, Ma Q, et al. A Method to Prevent SARS-CoV-2 IgM False Positives in Gold Immunochromatography and Enzyme-Linked Immunosorbent Assays. J Clin Microbiol. 2020;58(6).
7. Yan G, Lee CK, Lam LTM, Yan B, Chua YX, Lim AYN, et al. Covert COVID-19 and false-positive dengue serology in Singapore. Lancet Infect Dis. 2020;20(5):536.
8. To KK, Chua GT, Kwok KL, Wong JS, Au DCY, Lam YY, et al. False-positive SARS-CoV-2 serology in 3 children with Kawasaki disease. Diagn Microbiol Infect Dis. 2020;98(3):115141.