

IMMUNITY TO COVID-19 AND ISSUES OF SCREENING FOR SARS-COV-2 ANTIBODIES


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This paper briefly presents the known data on the immune response to SARS-CoV-2, and also analyzes the possibilities and limitations of serological testing for antibodies that should be accounted for when planning population studies and interpreting their results.

Keywords: COVID-19, SARS-CoV-2, immunity, antibodies, screening

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ИММУНИТЕТ К COVID-19 И ВОПРОСЫ ПРОВЕДЕНИЯ СКРИНИНГОВЫХ ИССЛЕДОВАНИЙ АНТИТЕЛ К SARS-COV-2


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В работе кратко представлены известные данные об иммунном ответе на SARS-CoV-2, а также проанализированы возможности и ограничения серологического тестирования на антивирусные антитела, которые следует учитывать при планировании популяционных исследований и интерпретации их результатов.

Ключевые слова: COVID-19, SARS-CoV-2, иммунитет, антитела, скрининг

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The evolving COVID-19 pandemic caused by the SARS-CoV-2 coronavirus is unprecedented in modern history. SARS-CoV-2 quickly spread throughout the world, affecting over 5 million and causing death of more than 300 thousand people (WHO data as of May 25, 2020) [1]. It dramatically changed the way of life in many countries, threatening further economic shocks. The reaction of scientific community to the pandemic is also distinguished with speed and scope. As soon as they receive papers from authors, world leading journals publish the latest information about the pathogen and its effect on the body, approaches to treatment and principles of curbing the spread of the virus. Such papers allowed developing the tools to detect SARS-CoV-2 RNA and test for its antibodies within the shortest time. This paper briefly overviews the known data on the immune response to SARS-CoV-2, and also analyzes the possibilities and limitations of serological testing for antibodies that should be accounted for when planning population studies and interpreting their results.

SARS-CoV-2 infection immune response

The information accumulated to date allows a degree of certainty to the statement that the immune response to SARS-CoV-2 infection develops following a typical scenario. In most SARS-CoV-2 patients specific antibodies of various classes appear 1–2 weeks after manifestation of the symptoms [2]. One study reports 40–55% of patients admitted with COVID-19 developing antibodies by days 5 to 7, with their number reaching 100% by days 17 to 19 [3]. In these patients, the antibody titer increased 2–4 times throughout the observation period (up to 27 days from the onset of symptoms) with seroconversion, i.e. appearance of specific antibodies, in the background. Other researchers

report the average of 10–15 days as the term of seroconversion in symptomatic SARS-CoV-2 cases [4]. Another important factor is that most COVID-19 patients, including those that had it in a mild form, develop specific functional antibodies capable of neutralizing the virus that make them effectively immune. Two weeks after the onset of symptoms, such antibodies were found in 94% of COVID-19 patients [5].

Cellular immunity to SARS-CoV-2 is developing in parallel with antibodies thereto. Within 2–4 weeks after infection, the body generates a pool of virus-specific T-lymphocytes [7, 8]. The hypothesis is that CD4- and CD8- T-lymphocytes will reliably protect their carriers from SARS-CoV-2 reinfection if they had no antibodies detected.

At this stage of development of the pandemic, the data available disallow conclusions about the term of persistence of SARS-CoV-2 immunity. The earlier research into seasonal coronaviruses, MERS-CoV and SARS-CoV-1, which are related to SARS-CoV-2, suggests certain ideas. SARS-CoV-1 patients had the IgG concentration remaining high for 4–5 months, then it was gradually decreasing over the course of 2–3 years, although after 2.5 years up to 90% of those who had SARS-CoV-1 retained neutralizing antibodies. The antibody response followed the like path in MERS-CoV patients: those who recovered retained the antibodies for up to 34 months [4, 6].

The SARS-CoV-2 reinfection potential question remains open. So far, no confirmed SARS-CoV-2 reinfection cases have been described. A study on primates showed that having been infected with SARS-CoV-2, the body cannot be reinfected with this virus [6]. There are also no SARS-CoV-1 and MERS-CoV reinfection cases described. However, the cases of reinfection with seasonal coronaviruses are quite common, with the conditions brought by them typically being mild acute

respiratory diseases. Here, reinfection may be associated with a rapid decline of protective immunity or contracting a new serovariation of the virus [4, 6].

COVID-19 seroepidemiology

So far, most COVID-19 seroepidemiology studies have only considered cohorts of patients admitted to the hospitals and patients with symptoms of the infection. It is possible that the bodies of individuals that had the symptoms manifesting minimally or not manifesting at all generate antibodies at a different rate and their post-infection immunity has different properties [9]. Various media outlets report the frequency of asymptomatic (or subclinical) seroconversion in various populations sharing a territory or an occupation, such reports being of varying degrees of reliability. The scientific community has produced a very small amount of publications covering this topic. A serological examination of Los Angeles residents that aimed to assess the cumulative incidence of COVID-19 revealed IgG and/or IgM antibodies in 35 (4.65%) of 863 participants [10], with 10 (29%) of the 35 reporting no symptoms of acute respiratory viral infections in the last two months, which could point to seroconversion as a result of asymptomatic SARS-CoV-2 infection. The possibility of subclinical seroconversion was demonstrated in a small study showing 3 (23%) of 13 patients and 11 (44%) of 25 employees of an outpatient dialysis center in the USA generating anti-SARS-CoV-2 IgM and/or IgG 21 days after contact with a COVID-19 patient. In this study, the majority of participants with antibodies (2 of 3 patients and 9 of 11 employees) had no COVID-19-like symptoms [11].

The problems of anti-SARS-CoV-2 serological testing

There is no doubt that serological tests for antibodies to SARS-CoV-2 can be extremely useful for diagnosing infection, studying population immunity, evaluating the response to vaccination etc. At the same time, such tests have specific limitations, which should be acknowledged on par with their advantages. A number of materials published in the respected scientific journals [6, 9, 12–14] recommend caution in the use of anti-SARS-CoV-2 serological tests, most of which are flawed.

Like any serological test, the anti-SARS-CoV-2 test will inevitably produce a certain percentage of erroneous (false-positive and false-negative) results. Obviously, their number will depend on the analytical capabilities of the test system, such as sensitivity (ability to detect antibody carriers; proportion of true-positive results) and specificity (selectivity of the test; proportion of true-negative results).

The sensitivity and specificity of 90–95% are often misleading; the figures are interpreted as a guaranteeing a low probability of error, 5–10%, respectively, which can be simply neglected. However, it should be remembered that the number of false results will vary depending on the prevalence of antibodies in a given population. This fact is often ignored, which leads to an inaccurate interpretation of the results. Below are some cases exemplifying the concern outlined above. Suppose there is a test system with a declared sensitivity and specificity of 95%. It is applied to 1) examine a population of former COVID-19 patients (the expected prevalence of antibodies is 90%); 2) examine a population not infected with COVID-19 and showing no symptoms of acute respiratory viral infections in the last 2 months (the expected prevalence of antibodies is 5%). The following formula allows assessing reliability of test result with the help of the positive prognostic value (PPV), i.e. the probability that a positive test result is true-positive [14]:

$$PPV = \frac{\text{Sensitivity} \times \text{Prevalence}}{\text{Sensitivity} \times \text{Prevalence} + (1 - \text{Specificity}) \times (1 - \text{Prevalence})}$$

For the first population (former COVID-19 patients) the PPV is 0.99, i.e., the probability that with a positive test result the individual actually has antibodies is 99%. However, the situation looks radically different in the second hypothetical population, for which the PPV value (with the same sensitivity and specificity of the test system) is only 50%. Thus, selectivity of the test system with a specificity of 95% is insufficient for a population with a low level of occurrence of antibodies; such a system produces false-positive results in half (!) of the cases.

Therefore, the degree of sensitivity and specificity of a test system should be evaluated depending on the characteristics of the population subject to examination, and interpretation of results of the tests should factor in the possible error. Simple calculations show that in order to have $PPV > 80\%$ in a population 5% of which has antibodies, the test system's specificity must be over 99%.

Currently, the market offers dozens of SARS-CoV-2 antibody test systems from various manufacturers. They are actively promoted, the campaigns' messages declaring accuracy and reliability of testing. Not all manufacturers openly report analytical characteristics of their products; probably, some of them simply have no knowledge of such since they did not set up the studies needed to acquire such information [9]. The use of such unreliable tests during a pandemic can be dangerous both for a specific person and for the population as a whole. The decisions about admission of medical personnel to work, making restrictive measures milder/stricter, as well as stigmatization of "people without antibodies" and, conversely, granting complete indulgence to individuals with SARS-CoV-2 antibodies down to giving them the so-called "immunological passports" based on a single study with uncertain level of confidence, can lead to serious consequences [9, 13].

In addition, the scale of serological testing should be reasonable. Testing the entire population is impractical and not necessary. For example, there are detailed guidelines issued by Rospotrebnadzor describing the procedure for organizing and conducting serological monitoring of the status of herd immunity to vaccine-controlled infections [15]. These guidelines describe indicator representative groups that should be tested, the frequency of their examination, etc. The SARS-CoV-2 antibodies screening activities should be clearly planned so that the results obtained in relatively small samples could allow reliable conclusions about herd immunity in the population as a whole and in individual high-risk (medical workers in particular) or vulnerable groups (elderly people, patients with chronic diseases, etc.).

In the view of the stated considerations, the gigantic scale of anti-SARS-CoV-2 population screening launched in May 2020 in Moscow, the effort involving 3 to 6 million people and costing 0.5–1 billion rubles [16], is bewildering, to say the least. The test system used for this effort has a specificity of 95.38%, which is clearly not enough for use in a population with a low prevalence of antibodies, as mentioned above. With a high degree of certainty, it can be said that Moscow residents, who have been in isolation for almost two months, constitute just such a population. Even if we assume that the real proportion of seropositive individuals is 10%, a test with the indicated specificity will give over 30% of false-positive results.

CONCLUSIONS

Mass-scale testing of individuals that did not have COVID-19 (i.e., populations with a low incidence of SARS-CoV-2

antibodies) can generate a large number of false-positive results, significantly exceeding the number of true-positive results. The sample size for such studies should be reasonably sufficient, and the data interpreted with the systems' analytical

characteristics factored in. These characteristics should be publicly available and verifiable. Otherwise, the widespread use of imperfect serological tests can be a source of serious errors in the medical and managerial decisions made.

¹ This value is indicated in the decision of the Moscow Department of Health COVID-19 Clinical Committee of 12.05.2020 (not published officially). Other sources of data on the analytical characteristics of this test system could not be found.

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