



Rapid Immune Tests SARS-COV-2 – An Experience in Beira Baixa

Testes Imunológicos Rápidos SARS-COV-2 -Uma Experiência na Beira Baixa

DOI:10.56238/isevjhv1n2-001

Recebimento dos originais: 01/07/2022

Aceitação para publicação: 25/07/2022

Francisco Rodrigues

Qualidade de Vida no Mundo Rural (QRural)
Sport Health & Exercise Unit (SHERU)
Instituto Politécnico de Castelo Branco, PhD
E-mail: franciscobrodrigues@ipcb.pt

Patrícia Coelho

Sport, Health & Exercise Unit (SHERU)
Qualidade de Vida no Mundo Rural (QRural)
Instituto Politécnico de Castelo Branco, PhD

Joana Liberal

Qualidade de Vida no Mundo Rural (QRural)
Instituto Politécnico de Castelo Branco, PhD

Manuel Martins

Qualidade de Vida no Mundo Rural (QRural)
Instituto Politécnico de Castelo Branco, PhD

Catarina Gavinhos

Qualidade de Vida no Mundo Rural (QRural)
Instituto Politécnico de Castelo Branco, PhD

Adriana Santos

BsC – Instituto Politécnico de Castelo Branco
E-mail: franciscobrodrigues@ipcb.pt

Inês Ribeiro

BsC – Instituto Politécnico de Castelo Branco

Cristina Carrondo

Instituto Politécnico de Castelo Branco, PhD

Elsa Alves

BsC – Instituto Politécnico de Castelo Branco

ABSTRACT

Introduction: SARS-CoV-2 affects the epithelial cells of the respiratory tract, causing severe infections. Since it is a pathology with a high level of transmissibility, it becomes central to mass testing. In addition, there was also a need to monitor the epidemic through serological tests. Objective: Evaluate the presence of antibodies against SARS-CoV-2 in the community residing in Beira Baixa through immunological screening tests. Materials and methods: Analytical, cross-sectional, and observational study, whose sample consists of 206 individuals. Data collection took place between February and April 2021, in the laboratories of the Dr. Lopes Dias Higher School of Health. Verbal informed consent, a questionnaire to collect sociodemographic data and the



serological test were applied. Results: Of the total number of participants, 15.5% admitted to having had COVID-19, of which 0.5% suspected they had been infected and 84% said they had never been infected. Regarding the presence of antibodies, 2.9% of the tests performed were positive for the presence of IgM's while 30.1% were positive for the presence of IgG's. Regarding vaccination, at the time of the investigation only 10.2% of the participants were vaccinated, of which 9.7% had IgG antibodies. Conclusion: Rapid serological tests can provide information about the presence of antibodies to SARS-CoV-2, thus being a very advantageous tool for immunity studies.

Keywords: SARS-CoV-2, serological tests, immunity.

1 INTRODUCTION

Coronaviruses belong to the Coronaviridae family, constituting etiological agents of human infections, which mainly affect the epithelial cells of the upper respiratory tract. In 2019, SARS-CoV-2 (Severe Acute Respiratory Syndrome – Coronavirus – 2) was identified for the first time in the city of Wuhan, responsible for COVID-19 (Coronavirus disease) that spread rapidly throughout the world, causing a global pandemic (Gautret, Million, Jarrot, Camoin-Jau, Colson, Fenollar, Leone, La Scola, Devaux, Gaubert, Mege, Vitte, Melenotte, Rolain, Parola, Lagier, Brouqui and Raoult, 2020).

Due to the rapid progress of the pandemic, testing for COVID-19 has become crucial for controlling transmission of the virus in the community. However, along with the development of diagnostic tests, of which real-time PCR (RT-PCR) stands out, there was a need to monitor the epidemic through serological tests (Zhengtu, Yongxiang, Xiaomei, Nian, Yang, Shaoqiang, Ruilin, Yanqun, Bicheng, Wei, Yongchen, Jing, Baofu, Ye, Jiasheng, Wensheng, Xuefeng, Jing, Zhiqiang, Kangjun, Weimin, Zhifei, Liyan and Feng, 2020).

It is known that after contact with the virus, the immune system induces the production of antibodies by B lymphocytes, having as main target the Spike (S) protein, as well as other nucleocapsid and envelope proteins. (Kang, Huang, Ouyang, Du, Yang, Chi, He, Ying, Chen and Wang, 2021).

Antibodies of the IgM class are the first to be produced, constituting the first line of defense against viral infections. The IgM antibody titer peaks during the first days of the acute phase of infection, remaining in circulation for approximately 5 to 7 days. Later, the immune system starts producing IgGs, which are maintained for several weeks after the primary infection and in cases of reinfection. (Deeks, Dinnes, Takwoingi, Davenport, Spijker, Taylor-Phillips, Adriano, Beese,



Dretzke, Ferrante di Ruffano, Harris, Price, Dittrich, Emperador, Hooft, Leeflang and Van den Bruel, 2020).

The assessment of immunity against SARS-CoV-2 is extremely important, as it allows the scientific community to understand the process of the immune response during and after infection by the same. Knowledge of the interaction of SARS-CoV-2 with the immune system has become one of the main allies for understanding the progression of infection and the protective effects of antibodies in the long term. (Figueiredo-Campos et al., 2020) Thus, serological tests are the most suitable method for this purpose, since they are based on the detection of circulating antibodies that arise when exposed to the coronavirus, through immunochromatography.

The main objective of this investigation was to evaluate the presence of antibodies against SARS-CoV-2 in the community residing in Beira Baixa, using rapid immunological tests.

2 MATERIALS AND METHODS

2.1 SAMPLE DESCRIPTION

The present study is an observational and cross-sectional analytical type. Sample collection was carried out during the months of February, March and April 2021, in the laboratories of Escola Superior de Saúde Dr. Lopes Dias, from the Polytechnic Institute of Castelo Branco.

2.2 STUDY PROTOCOL

After publicizing the possibility of carrying out rapid serological tests to the community, all data used for this investigation were recorded in a database.

Verbal informed consent and a questionnaire were applied to each participant. Data were coded in order to obtain an ID that did not refer to any data that could identify the participant, in order to guarantee confidentiality.

2.3 COLLECTED VARIABLES

In order to carry out the evaluation and identification of individuals with immunity to COVID-19, the following nominal qualitative variables were collected: sex, IgM (Immunoglobulins M) and IgG (Immunoglobulins G), presence of the disease prior to this evaluation, and vaccination. for the same.



3 PROCEDURE

The DIASource Immunoassays® kit was used for the qualitative detection of 2019-nCoV-specific IgG and IgM in whole blood. Obtaining whole blood samples was performed by collecting capillary blood.

3.1 STATISTICAL ANALYSIS:

The variables were coded (obtained through the survey) to be analyzed using the SPSS® statistical analysis program. Normality was assessed using the Kolmogorov – Smirnov test and the Mann Whitney test (parametric) was used for association between variables. A confidence interval of 95% was established and as a criterion of statistical significance a value of $p \leq 0.05$ was established.

3.2 ETHICAL ISSUES

The research team respected and complied with the principles mentioned in the Helsinki declaration, ensuring that there are no conflicts of interest.

The data provided by the participants is of exclusive access to the research team. The data collected were used for statistical analysis and this work is of academic interest only, with no economic interest whatsoever.

4 RESULTS

Data from 206 participants were analyzed, of which 115 (55.8%) were female and 91 (44.2%) were male, with a mean age of 45.51 ± 16.877 years, with the minimum age being 4 years and a maximum of 83 years. It could also be seen that the most prevalent age class was 51 to 60 years old, corresponding to a total of 48 (23.3%) participants. Of the total number of participants, 15.5% (n=32) admitted having had COVID-19, 0.5% (n=1) suspected having been infected and 84% (n=173) said they had never been infected.

As for the presence of antibodies, 2.9% of the tests performed were positive for the presence of IgM's and IgG's simultaneously, while 30.1% were positive only for the presence of IgG's.

At the time of the investigation, only 21 participants (10.2%) had started the process of vaccination against COVID-19. However, only 20 individuals (9.7%) had positive IgG class antibodies and were vaccinated.

The Kolmogorov - Smirnov test showed that the sample was normally distributed, using the Mann Whitney parametric test, obtaining the following significance when comparing the aforementioned variables and the presence of immunoglobulin G by SARS-CoV-2.

Table 1 - Statistical significance between the presence of Immunoglobulins and the variables mentioned below.

Variable	<i>p value</i>	<i>Statistical significance</i>
Age classes	0,798	No
COVID-19	<0.05	Yes
Sex	0.623	No
Vaccination	<0.05	Yes

5 DISCUSSION

After analyzing the results, the presence of antibodies was confirmed in individuals who had already tested positive for infection. Of the 15.5%, 2.4% had IgM antibodies and 14.6% had IgG antibodies, and in the total number of individuals, the seroprevalence found was 2.9% and 30.1%, respectively. It should be noted that all individuals who presented IgM antibodies also showed IgG antibodies. These results demonstrated that the majority of subjects had only IgG class antibodies, consistent with older infections. (Chansaenroj, Yorsaeng, Posuwan, Puenpa, Sudhinaraset, Chirathaworn and Poovorawan, 2021).

As for the distribution by sex, contrary to what was mentioned in the “National Serological Survey COVID-19 (ISN COVID-19)” (Ana Paula Rodrigues, Ana Cristina Garcia, 2021), no statistically significant association was found between sex and the presence of antibodies against SARS-CoV-2, with even a slightly higher prevalence of antibodies in females (17.9% vs. 15.1%). There was also a higher prevalence of antibodies in the age classes from 41 to 50 years old and from 51 to 60 years old (IgM – 1.5% in both; IgG – 8.5% vs. 9.5%), as observed in the “National Serological Survey COVID-19 (ISN COVID-19)” (Ana Paula Rodrigues, Ana Cristina Garcia, 2021). Even so, there are some discrepancies in the results obtained when compared with the results of the “National Serological Survey COVID-19 (ISN COVID-19)”, justified, for example, by the type of sampling, which was a sampling by quotas, while the present study presents a convenience sample.

The high emergency in the control of the pandemic, determined that the scientific community was dedicated to the production of vaccines effective in the immunization against SARS-CoV-2. Currently, it is known that the main class of antibodies produced after vaccination



is IgG. (Dai & Gao, 2021) The results of this study confirm these data, verifying that the vast majority of vaccinated individuals had IgG class antibodies against SARS-CoV-2. The difference between all vaccinated individuals and those who had IgG antibodies may be due to the fact that only the first dose of vaccine was administered to the individual who tested negative for the presence of IgG's. (Guo, Mi and Nie, 2020) It should also be noted that many of the respondents had been vaccinated for less than 14 days, so, according to some studies, sufficient time for a physiological response might not have elapsed. (Mahajan and Manchikanti, 2020) Still, most vaccinated individuals had antibodies against the virus.

As has been much debated, despite the importance of these analyses, this type of tests does not indicate when the individual was infected, nor the period of time of infection, which is always one of the main points addressed by most works in the area. (Li et al., 2020)

Many commercial companies have invested in serological antibody tests, being widely accepted in the scientific community, however, always keeping in mind their particular characteristics, as well as the evolution that they have been experiencing since their initial phase. (de Jong, Rosing, Vermunt, Huitema and Beijnen, 2021). Effectively, there is an increase in the levels of sensitivity and specificity that these rapid tests have acquired (Deeks et al, 2020). However, these tests have a lower sensitivity than other reference methods, so they may not be sufficient for the detection of low antibody titers, which may result in a higher percentage of false negatives, and are not suitable for clinical diagnostic procedures (Cassaniti, Novazzi, Giardina, Salinaro, Sachs, Perlini, Bruno, Mojoli, Baldanti; Members of the San Matteo Pavia COVID-19 Task Force, 2020), although some works have pointed in the opposite direction (Indenbaum, Koren, Katz-Likvornik, Yitzchaki, Halpern, Regev-Yochay, Cohen, Biber, Feferman, Cohen Saban, Dhan, Levin, Gozlan, Weil, Mor, Mandelboim, Sofer, Mendelson and Lustig, 2020). In addition, the presence of false negatives may be related to a longer and less effective immune response, since it varies from individual to individual. (Li et al., 2020)

Additionally, these tests do not allow the origin of the antibodies to be determined, since, as already mentioned, the IgGs may come from a previous infection or from vaccination (Pieri, Nuccetelli, Nicolai, Sarubbi, Grelli and Bernardini, 2021). In this way, they should be complemented with other methods to obtain a more accurate diagnosis of COVID-19, namely in cases where only IgM class antibodies are detected. (Mekonnen, Mengist, Derby, Nibret, Munshea, He, Li and Jin, 2021).

There is a huge effort to validate more and more tests, using the most capable methodologies and thus increasing the capacity for screening and evaluating the immunity of the



population. This type of studies can significantly help health authorities to estimate the number of infected in the epidemic, predict future spread, determine the effectiveness of vaccines and prioritize which people should be vaccinated/received a booster vaccine. Thus, there are several works that are being developed in this direction, highlighting for now the project “Beira Baixa Com Vida”, to be developed at the Instituto Politécnico de Castelo Branco/Escola Superior de Saúde Dr. Lopes Dias and who wants to know the immunological scenario of the population of Beira Baixa in relation to SARS-COV-2, which will present preliminary results soon.

6 CONCLUSION

With the completion of the present study, it was realized that rapid serological tests are an important tool to obtain knowledge about acquired immunity during and after infection by SARS-CoV-2. These tests proved to be quite promising, having several advantages. However, they reveal some limitations, which, in order to be overcome, these tests should be complemented with other methods.



REFERENCES

- Cassaniti I, Novazzi F, Giardina F, Salinaro F, Sachs M, Perlini S, Bruno R, Mojoli F, Baldanti F; Membros da Força-Tarefa San Matteo Pavia COVID-19. O desempenho do Teste Rápido igm/igg do vivadiag COVID-19 é inadequado para o diagnóstico de COVID-19 em pacientes agudos referentes ao pronto-socorro. *J Med Virol.* 2020 ;92(10):1724-1727. Doi: 10.1002/jmv.25800. Epub 2020 Abr 8. PMID: 32227490; PMCID: PMC7228409.
- Chansaenroj J, Yorsaeng R, Posuwan N, Puenpa J, Sudhinaraset N, Chirathaworn C, Poovorawan Y. Detecção de anticorpos sars-cov-2 específicos através de imunoenaios diagnósticos rápidos em pacientes COVID-19. *Virol J.* 2021; 9;18(1):52. Doi: 10.1186/s12985-021-01530-2. PMID: 33750394; PMCID: PMC7942515.
- Dai L, Gao GF. Metas virais para vacinas contra o COVID-19. *Nat Rev Immunol.* 2021 Fev;21(2):73-82. Doi: 10.1038/s41577-020-00480-0. Epub 2020 Dez 18. PMID: 33340022; PMCID: PMC7747004.
- De Jong KAM, Rosing H, Vermunt M, Huitema ADR, Beijnen JH. Quantificação de anticorpos anti-SARS-cov-2 em soro humano com LC-QTOF-MS. *J Pharm Biomed Anal.* 2021;205:114319. Doi: 10.1016/j.jpba.2021.114319. Epub à frente da impressão. PMID: 34416552; PMCID: PMC8354797.
- Deeks JJ, Dinnes J, Takwoingi Y, Davenport C, Spijker R, Taylor-Phillips S, Adriano A, Beese S, Dretzke J, Ferrante di Ruffano L, Harris IM, Price MJ, Ditttrich S, Emperador D, Hooft L, Leeftang MM, Van den Bruel A; Cochrane COVID-19 Grupo de Precisão de Teste de Diagnóstico. Testes de anticorpos para identificação de infecção atual e passada com SARS-cov-2. Banco de dados Cochrane Syst Rev. 2020;6(6):CD013652. Doi: 10.1002/14651858.CD013652. PMID: 32584464; PMCID: PMC7387103.
- Gautret P, Million M, Jarrot PA, Camoin-Jau L, Colson P, Fenollar F, Leone M, La Scola B, Devaux C, Gaubert JY, Mege JL, Vitte J, Melenotte C, Rolain JM, Parola P, Lagier JC, Brouqui P, Raoult D. História natural do COVID-19 e opções terapêuticas. *Especialista Rev Clin Immunol.* 2020;16(12):1159-1184. Doi: 10.1080/1744666X.2021.1847640. Epub 2020 Dez 24. 33356661.
- Guo CC, Mi JQ, Nie H. Taxa de soropositividade e precisão diagnóstica de testes sorológicos em casos de 2019-ncov: uma análise agrupada de estudos individuais. *Eur Rev Med Pharmacol Sci.* 2020 Out;24(19):10208-10218. Doi: 10.26355/eurrev_202010_23243. 33090430.
- Indenbaum V, Koren R, Katz-Likvornik S, Yitzchaki M, Halpern O, Regev-Yochay G, Cohen C, Biber A, Feferman T, Cohen Saban N, Dhan R, Levin T, Gozlan Y, Weil M, Mor O, Mandelboim M, Sofer D, Mendelson E, Lustig Y. Testing igg antibodies contra o RBD de SARS-cov-2 é suficiente e necessário para o diagnóstico CO-19. *Plos 1.* 2020;15(11):e0241164. Doi: 10.1371/journal.pone.0241164. PMID: 33227020; PMCID: PMC7682882.
- Kang K, Huang L, Ouyang C, Du J, Yang B, Chi Y, He S, Ying L, Chen G, Wang J. Development, performance evaluation, e aplicação clínica de um Rapid SARS-cov-2 igm e igg Test Kit baseado em imunoenasiao de fluorescência automatizada. *J Med Virol.* 2021;93(5):2838-2847. Doi: 10.1002/jmv.26696. Epub 2021 Mar 1. PMID: 33231312; PMCID: PMC7753814.
- Mahajan A, Manchikanti L. Value and Validade do Teste de Anticorpos Coronavirus. *Pain Medical.* 2020; 23(4S):S381-S390. 32942795.



Mekonnen D, Mengist HM, Derbie A, Nibret E, Munshea A, He H, Li B, Jin T. Precisão diagnóstica de testes sorológicos e cinética de síndrome respiratória aguda grave coronavírus 2 anticorpo: Uma revisão sistemática e meta-análise. *Reverendo Med Virol.* 2021;31(3):e2181. Doi: 10.1002/rmv.2181. Epub 2020 Nov 5. 33152146.

Pieri M, Nuccetelli M, Nicolai E, Sarubbi S, Grelli S, Bernardini S. Validação clínica de uma imunoenensaio automatizada de imbâmbulos anti-SARS-cov-2 igg e igm. *J Med Virol.* 2021 Abr;93(4):2523-2528. Doi: 10.1002/jmv.26809. Epub 2021 Jan 26. PMID: 33463719; PMCID: PMC8013349.

Zhengtu L , Yongxiang Y , Xiaomei L , Nian X, Yang L, Shaoqiang L, Ruilin S, Yanqun W, Bicheng H, Wei C, Yongchen Z, Jing W, Baofu H, Ye L, Jiasheng Y, Wensheng C, Xuefeng W, Jing C, Zhiqiang C, Kangjun S, Weimin P, Zhifei Z, Liyan C, Feng Y. Desenvolvimento e aplicação clínica de um teste rápido de anticorpos combinados igm-igg para diagnóstico de infecção sars-cov-2. (2020) *J Med Virol*;92(9):1518-1524. Doi: 10.1002/jmv.25727. Epub 2020 Abr 13.