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Coabitantes aumentam deteção persistente de RNA de SARS-CoV-2 em profissionais de saúde com COVID-19

Cohabitants increase the persistence of SARS-CoV-2 RNA detection in healthcare professionals with COVID-19

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/ Resumo

Introdução: Os profissionais de saúde, pela sua exposição, estão sujeitos a um risco superior de desenvolver COVID-19. Inicialmente, aqueles que tiveram resultado positivo para SARS-CoV-2, foram colocados em isolamento e só retornaram ao trabalho após dois testes consecutivos de PCR negativos, com um intervalo mínimo de 24 horas. A deteção prolongada de RNA viral tem sido associada a vários fatores, nomeadamente idade, género e resposta imune.

Objetivos: Analisar o papel das medidas de isolamento na deteção prolongada de RNA de SARS-CoV-2. Procurámos ainda analisar outras características dos profissionais de saúde.

Métodos: Realizámos uma análise retrospetiva baseada nos registos clínicos dos profissionais de saúde de um grande hospital da Região Centro de Portugal, com teste positivo para SARS-CoV-2. Recolhemos dados demográficos, comorbilidades, categoria profissional, existência de coabitantes e medidas de isolamento.

Resultados: A mediana do período de isolamento foi 35 dias. As mulheres tiveram períodos de isolamento mais longos que os homens. Verificámos ainda que os profissionais de saúde que não praticavam medidas de isolamento na habitação tiveram períodos de isolamento mais prolongados que aqueles que se isolaram ou viviam sozinhos.

Conclusões: Os nossos resultados sugerem que as medidas de isolamento podem ter um impacto importante na redução do período de deteção de RNA.

Palavras-chave: deteção de RNA prolongado, profissionais de saúde, medidas de isolamento

/ Abstract

Background: *Healthcare workers are at increased risk of infection with COVID-19, due to their greater exposure. In the beginning, those who tested positive, were asked to stay in isolation, and were only allowed to return to work after two consecutive negative RT-PCR tests from a respiratory sample with a minimum 24 hours interval. Prolonged RNA detection has been associated with disease severity, age gender and immune response.*

Objective: *We aim to evaluate the impact of isolation measures in prolonged RNA detection. We also aim to study health care workers characteristics.*

Methods: *We performed a retrospective analysis based on the healthcare workers records, from a large hospital in the Center Region of Portugal, who tested positive for COVID-19. During the follow up we collected demographic data, comorbidities, professional category, the existence of cohabitants and isolation measurements.*

Results: *We found an average isolation period of 35 days. Women took longer than men to be discharged. We also found that healthcare workers who did not isolate themselves from their cohabitants took longer than those who did and those who lived alone.*

Conclusions: *Isolation measurements may have an important impact on the reduction of RNA detection period.*

Keywords: *prolonged RNA detection, health care workers, isolation measures*

/ Introduction

Severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) is a single-stranded RNA virus that caused the outbreak of coronavirus disease 2019 (COVID-19) in humans.[1] It is responsible for an acute infection, with a broad clinical spectrum, that includes an asymptomatic infection, mild upper respiratory tract illness, a severe viral pneumonia with respiratory failure and even death.[2,3] The average incubation period is 5 days, with an interquartile range (IQR) of 2-7 days. Most patients develop symptoms within 12 days.[3]

Viral RNA levels are detectable in the respiratory tract 2-3 days before symptoms appear, peak at symptoms' onset[4–6] and decline over the next 7-8 days in most patients.[3–6] SARS-Cov-2 RNA can be detected for 20 days or longer.[4,7,8]

Healthcare workers (HCW) are potentially at increased risk of infection with COVID-19 and may transmit the disease to vulnerable patients.[9] Those who test positive for SARS-CoV-2 via reverse transcriptase PCR (RT-PCR) of nasopharyngeal and/or oropharyngeal swab specimens are asked to stay in isolation provided that they don´t need specialized care.

Return to work and discharge strategies can be symptom or test based.[10] Center for Disease Control and Prevention (CDC) recommends a 10-day isolation from symptom onset (including >24 hours since resolution of fever and improvement of symptoms) for mild-moderately ill patients without severely immunocompromising conditions. Moreover, CDC recommends up to 20 days for patients with severe illness or severely immunocompromising conditions.[11] HCW who were asymptomatic throughout their infection may return to work when at least 10 days have passed since the date of their first positive viral diagnostic test or two consecutive negative RT-PCR tests.[11] A test based strategy requires resolution of fever and symptoms improvement and at least two consecutive negative results from respiratory specimens collected ≥24 hours apart for SARS-CoV-2 RNA.[10,11] World Health Organization (WHO) allows countries to choose either symptom or a test based strategy.[12]

In Portugal, a test-based strategy was in place until recently.[13] In the general population, clinically recovered COVID-19 patients who have one negative test where discharged,[14] while HCW could only return to work after two consecutive negative RT-PCR tests from respiratory specimens with a minimum 24 hours interval, as previously suggested by CDC.[10]

This approach, could be overestimating the length of infectious spreading by detecting non-infectious viral shedding,[3,15] since the ability of SARS-CoV-2 to replicate in cultured cells is a better surrogate for infectivity.[15]

Prolonged RNA detection has been associated with disease severity, age, gender and immune response.[6,7,15] We hypothesize that there could also be an association with the fact that there are cohabitants who are also potentially infected and the isolation measures at home. Until this moment, there are no studies in the literature exploring this specific topic. This study aims to evaluate the HCW characteristics and its potential association with prolonged RNA detection, in order to provide a better understanding of SARS-CoV-2's viral clearance profile.

/ Methods

We performed a retrospective analysis based on HCW records. The data was collected during the COVID-19 follow up from a large hospital in the Center Region of Portugal.

The diagnosis was established after a positive detection of SARS-CoV-2 through a semi quantitative RT-PCR targeting SARS-CoV-2 on nasopharyngeal and/or oropharyngeal swab samples. The ones who tested positive were accompanied by the Occupational Health Department (OHD) until the moment of discharge and return to work, which was established by achieving two consecutive negative tests from respiratory specimens with a minimum 24-hour interval.

During this period, data regarding age, gender, comorbidities, professional category, the existence of cohabitants, and isolation measures were collected. Professional categories were divided into physicians, nurses, health care assistants and others, which includes technical assistants, diagnosis and therapeutic technicians and pharmaceuticals. We analyzed data collected between 14 of March and 15 of June of 2020.

The statistical analysis was performed using SPSS 26.0 (IBM, USA). Quantitative variables were tested for normality using Shapiro Wilk test. Comparisons between groups were performed using t-student, Mann-Whitney or Kruskal-Wallis. Correlation between quantitative variables was analyzed with Spearman's Correlation Coefficient. The significance level was established at 0.05.

/ Results

Between March 14 and June 15 of 2020, 208 out of 8037 HCW from tested positive for SARS-CoV-2. From those, 172 were included in the analysis, since 8 were still positive at the time of the analysis. Four were possible false positive results, as they repeated the test right after and were negative, and therefore were excluded. Finally, 24 did not have enough information on their clinical file and, consequently, were excluded. There were only two HCW that required specialized care and were hospitalized, but happily none in an intensive care unit.

The average age of our sample was 43.34 ± 10.56 years, ranging from 19 to 65 years old. Mostly women (n=135; 78.45%). We found that the most affected group were nurses (n= 88; 51.16%), followed by health care assistants (n=42; 24.42 %), physicians (n=28; 16.28%) and finally others (n=14; 8.14%), as shown in Table I. The isolation period lasted 37.08 ± 14.63 days on average, ranging from 11 to 78 days.

Interestingly, we found significantly less time to discharge men than women (P=0.03). In women, it took 36 days (IQR 26 – 48) to achieve two negative test results, while in men it took 31 days (IQR 21.4 – 42.5). There was no correlation between age groups and the isolation period (Spearman's rho=0.067, P>0.05).

Most workers had cohabitants during the time of disease (n=149; 86.63% versus n=23; 13.37%). Those with (36 days, IQR 26-47.5) or without (32 days, IQR 21 - 42) cohabitants took a similar amount of time to achieve two negative consecutive tests, P>0.05, but that ignores the role of isolation. Regarding those with cohabitants, 124 (83.22%) were isolated from them and 25 (16.78%) were not. The ones who didn't perform any isolation measures at home took 47 days (IQR 32-55) to discharge, which was significantly longer than those who did (34.5 days IQR 26-45; P=0.042) and those who lived alone (32 days IQR 21-42; 0.015). No differences were found between the group that lived alone and the ones who performed isolation measures, P>0.05.

Most HCW did not have cohabitants with a diagnosis for COVID-19 (n=120, 69.77% vs n=29, 16.86%). Furthermore, no differences were found between the ones who had a known positive inhabitant (41 days IQR 32-52.5), and those who did not (34.5 days IQR 26-47) and those who lived alone (32 days IQR 21-42). The group with a positive inhabitant took longer than the other groups to achieve 2 consecutive negative tests, but the difference was not statistically significant, P>0.05.

No differences were found in HCW with comorbidities, as represented in Table II. Moreover, we also did not find differences between workers without any comorbidity (35 days IQR 23-48), having one (35 days IQR 26-46.5) or two or more (36 days IQR 25.5-44.25), P>0.05. The same happened between smokers (36 days IQR 24-44) and non-smokers (35 days IQR 26-47), P>0.05.

/ Discussion

Several studies, using a test based strategy, showed that viral SARS-CoV-2 RNA can be detected in nasopharynx for 20 days or longer, [4,7,8] but we found a higher period of time, with HCW taking 35 days (IQR 26 – 46.75) between the moment of diagnosis and achieving 2 consecutive negative tests at least 24 hours apart.

Women took longer to achieve two consecutive negative tests than men. Our results are in accordance with Zheng et al.[6] but in disagreement with Vaz et al.[7]. There is also evidence suggesting a higher proportion of males among severe cases[16–18], even though this difference seems to be attenuated by adjustments for age and comorbidities.[18]

We did not find differences between age groups or correlation between age and time needed to test negative. There is some

TABLE I – GENERAL CHARACTERISTICS OF HCW WITH CONFIRMED COVID-19						
		Physicians	Nurses	Healthcare assistants	Others	Total
Gender N (%)	Female	20 (71.43)	68 (77.27)	34 (80.95)	13 (93.86)	135 (78.45)
	Male	8 (28.57)	20 (22.72)	8 (19.05)	1 (7.14)	37 (21.51)
Age (years) Average ± SD		41.86 ± 14.46	40.97 ± 8.96	48.43 ± 9.93	46 ± 8.65	43.34 ± 10.56
Total N (%)		28 (16.28)	88 (51.16)	42 (24.42)	14 (8.14)	172 (100)

Abbreviations: HCW, healthcare workers; COVID-19, coronavirus disease 2019; SD, standard deviation.

TABLE II – CHARACTERISTICS OF HCW CONFIRMED WITH COVID-19 AND TIME NEEDED TO ACHIEVE 2 CONSECUTIVE TESTS PERFORMED AT LEAST 24 HOURS APART				
		N (%)	Isolation period, Median (IQR)	P value*
Gender	F : M	135 (78.45) : 37 (21.51)	36 (26 - 48) : 31 (21.4 – 42.5)	**0.03**
Age (years)	< 20	1 (0.58)	34 (-)	0.272
	20 – 29	19 (11.05)	33 (21 - 43)	
	30 – 39	46 (26.74)	35 (23 – 44.35)	
	40 – 49	50 (29.10)	39.5 (27.75 - 53)	
	50 – 59	43 (25.0)	33 (26 - 46)	
	≥ 60	13 (7.56)	33 (28 - 45)	
With comorbidities: Without comorbidities	Hypertension	22 (13.02) : 147 (86.98)	33.3 (26.75 – 43.5) : 35 (26 – 47)	0.556
	Diabetes	3 (1.78) : 166 (98.22)	35 (-): 35 (26 - 47)	0.564
	Other cardiovascular disease	7 (4.14) : 162 (95.86)	30 (27 - 52) : 35 (26 – 46.25)	0.972
	Immunosuppression	2 (1.18) :167 (98.82)	26.5 (-) : 35 (26 - 47)	0.285
	Chronic respiratory disease	35 (20.71) : 134 (79.29)	35 (24 - 43) : 35.3 (26 – 47.25)	0.279
	Obesity	27 (15.98) : 145 (85.80)	36 (27 - 46) : 30 (25 - 47)	0.427
Number of comorbidities	None	82 (48.52)	35 (23 - 48)	0.956
	1	69 (40.83)	35 (26 – 46.5)	
	≥2	18 (10.65)	36 (25.5 – 44.25)	
Smoker	Yes	21 (12.21)	36 (24 - 44)	0.905
	No	151 (87.79)	35 (26 - 47)	
Cohabitant	Yes	149 (86.63)	36 (26 - 47.5)	0.103
	No	23 (13.37)	32 (21 - 42)	
Isolation measures	Isolated from cohabitants	124 (72.09)	34.5 (26 - 45)	**0.013**
	Not isolated from cohabitants	25 (14.53)	47 (32 - 55)	
	Without cohabitant	23 (13.37)	32 (21 - 42)	
Cohabitant with COVID-19	Yes	29 (16.86)	41 (32 - 52.5)	0.07
	No	120 (69.77)	34.5 (26 - 47)	
	Without cohabitant	23 (13.37)	32 (21 - 42)	
Total		172(100)	35 (26 - 46.75)	-

Abbreviations: F, female; M, Male; COVID-19, Coronavirus disease 2019; IQR, Inter-Quartile range.
* Non-parametric tests comparing the isolation period between groups. The isolation period corresponds to the number of days between the diagnosis and the achievement of two consecutive negative tests results, performed at least 24 hours apart.
The bold values represent the level of significance is 0.05 (5%).

evidence suggesting that patients older than 60-65 years' experience more severe[16] and longer disease[6,7], but since our sample is taken from a working-age population that difference may not be as evident. We did not perform an analysis for severity of illness, since only two of the 172 HCW required inpatient care, although there is evidence suggesting that patients with severe disease took longer to test negative than those with mild disease.[7]

We compared the time needed to achieve two negative consecutive tests with the isolation measures at home. We found that HCW who did not isolate themselves from their cohabitants took longer than those who did and those who lived alone. We did not find differences between the group that lived alone and the ones who performed isolation measures. This suggests that the isolation measures may be an epidemiologic factor with relevance in the time needed to achieve two consecutive negative tests, with potential impact on planning of end of isolation and return to work.

We also found that the group with a positive inhabitant took longer than the other groups to achieve two consecutive negative tests, but the difference was not statistically significant. Since we do not have direct control over the screening of relatives who are not CHUC HCW, we cannot confirm if the testing was performed. More studies would be needed to exclude the importance of this factor in the disease duration and virus detection.

Previous studies found evidence suggesting that hypertension[16,19–21] (particularly in patients over 60 years old[19]), diabetes[20,21], other cardiovascular diseases[20,21], chronic obstructive pulmonary disease[21,22] and obesity[18,23–25] have been associated with worse clinical outcome. However, we didn't find any differences on the time needed to achieve two consecutive negative tests in HCW with or without co-morbidities like hypertension, diabetes, other cardiovascular diseases, immunosuppression, chronic respiratory disease, or obesity. We also did not find differences between workers without comorbidities, with one comorbidity, or two or more. The same happened between smokers and non-smokers. Accordingly, results regarding smoking have been contradictory.[22,26] It is important to consider that the population we analyzed has been associated with a higher proportion of non-severe cases.[16] It may be due to the younger nature of our population, as they are all active people. The prevalence and severity of these comorbidities might be different from the general population, and therefore their effect may be understated.

It is also important to consider that our study reflects a very specific population, of working age, and with sociodemographic characteristics that may limit the generalization of its conclusions to the general population.

In conclusion, our study suggests that SARS-CoV-2 viral RNA may persist for a long period in respiratory samples, and that isolation measures may have an important impact on the duration of that period.

Our study has several limitations. Firstly, polymerase chain reaction (PCR) cannot distinguish between viable and non-viable virus and does not reflect the replication level of the virus. Secondly, isolation measures at home, based on the report of the HCW, were not verified by the investigator. Therefore, there could be other factors regarding housing conditions with impact on duration of test positivity, such as number of inhabitants, ventilation conditions, and so on.

Further investigation is needed to better understand how isolation measures influence temporal dynamics in viral shedding and if there is any impact on transmissibility of COVID-19.

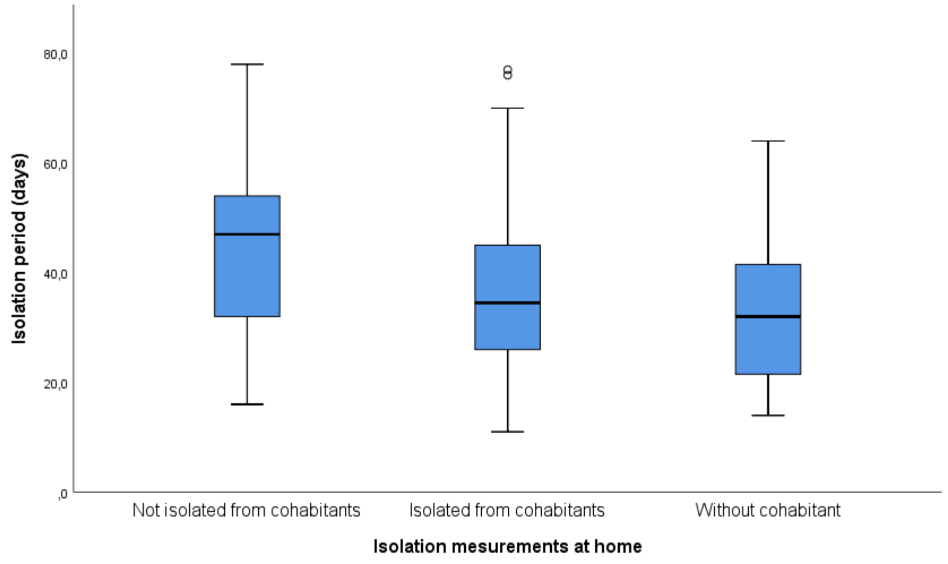

Fig. 1 – Boxplot representing the time needed to achieve 2 consecutive negative tests at least 24 hours apart according to the isolation measures performed.

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