

Low frequency of SARS-CoV2 infection in daycare centers during the reopening of school activities in the Southeast's poor area of Brazil

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ABSTRACT

SARS-Cov2 has already infected over 482 million people and caused more than 6.1 million deaths. The beginning of the pandemic has led the health authorities of several countries to adopt non-pharmacological preventive measures such as daycare closures. The reopening took place when the country had the highest rates of infection and mortality (mainly due to the gamma variant (P.1) outbreak) and the beginning of the vaccination program. Therefore, we aimed to investigate the prevalence of SARS-CoV2 in daycare after educational activities resumed. The study was conducted in seven childcare facilities. Swab samples from the nasopharynx were collected from children and staff members. The viral RNA was obtained through PureLink RNA extraction kit purification and SARS-CoV2 presence was detected using the All plex SARS-CoV2 kit. The study population included 201 participants, including daycare workers and children. The average age of the workers and children is 40 and 3 years old, respectively. Among the children, 47.5% are female and among the workers, 91.4%. One (0.5%) test came out positive for the presence of SARS-CoV-2, which was from a sample of an asymptomatic childcare worker, and no secondary infections were detected. Considering that the return to daycare activities occurred during a period with a high number of deaths and a lack of vaccines throughout the country, the small number of cases indicates the effectiveness of the several preventive measures used by daycare centers in preventing SARS-CoV2 transmission.

KEYWORDS: SARS-CoV-2. Daycare centers. Epidemiology.

BACKGROUND

SARS-CoV-2 is the etiologic agent of the Coronavirus 19 Disease (COVID-19), the virus responsible for the current pandemic. It has already reached over 482 million people with more than 6.1 million deaths¹. SARS-CoV-2 belongs to the *Coronaviridae* family and its genome consists of a positive single-stranded RNA of approximately 26 to 32kb². The rapid viral spread combined with the lack of drugs or vaccines that could contain the novel virus led global authorities to implement non-pharmaceutical measures aimed to reduce the dissemination of SARS-CoV-2 and, consequently, reduce the burden on health services. Among the measures adopted, daycare closure has been implemented in many countries, including Brazil³.

Since the beginning of the current pandemic, several studies have been performed to better understand how COVID-19 may affect children⁴ and how the interruption

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of school activities could help reduce the spread of the virus at mass gathering places. To date, children and adolescents account for 1% to 2% of COVID-19 cases worldwide⁴. This finding suggests that the major problems of the SARS-CoV2 infection in children may not be transmission and hospitalization, unlike what happens with other respiratory viral infections. Despite this assumption, children are still susceptible to the virus, which can be alarming⁵.

Daycare centers and schools remained closed for an average of 115 school days in low-middle-income countries and 53 school days in high-income countries. To ensure a safe return to school, each country has implemented one or more types of measures to reduce the incidence of SARS-CoV-2 infections in schools, such as social distancing, classroom ventilation, frequent hand washing, use of masks, and scheduling classes in a hybrid teaching method, combining in-person classes with activities at home, regardless of the age group. Overall, low-income countries had more difficulty implementing some of these measures, and in-person classes took about 50% longer in these countries compared to high-income ones. The lack of basic hygiene products or infrastructure in many low-income countries has led to a further delay in the reopening of schools in these countries. Sanitary measures were applied in most schools holding in-person classes, even though cases of kids transmitting the virus to adults were thought to be rare. In addition, school closures were one of the most significant non-pharmaceutical measures adopted to decrease the transmission of SARS-CoV-2⁶. After school activities resumed, some studies were carried out to evaluate the rates of virus transmission in the school environment. A previous study showed that the transmission rate among students aged 11 to 18 years old was considerably higher than among children aged 0 to 5. In addition, they were not affected by any secondary infection⁷. In daycare facilities in the United States, SARS-CoV-2 outbreaks occurred in 5.8% of units and about 20% of cases were asymptomatic, most of them in children⁸.

Up to 2020, data regarding the role of children's transmission of SARS-CoV2 were limited. However, the rate of transmission for over a year had been considerably high worldwide. Although the rate of coronavirus mutation is lower than for other RNA viruses, important variants have emerged throughout the pandemic. Variants associated with greater transmissibility, mortality, or ability to evade host immunity and which still maintain their prevalence and spread rapidly across different countries are called "Variants of Concern" (VOC)⁹. To date, five VOCs have been identified, namely, B.1.1.7 or Alpha from the United Kingdom, B.1.351 or Beta from South Africa, P.1 or Gamma from Brazil, B.1.617.2 or Delta from India, and lastly Omicron, detected in multiple countries.

The Delta variant is 60% more transmissible than the variants that were already circulating, which had already caused high rates of infectivity. This increased the number of cases, hospitalizations, and deaths, even in countries with higher vaccination rates¹⁰. In the United States, the Delta variant quickly became prevalent, increasing the transmission rates like those seen prior to receiving an eligible vaccine. Furthermore, it is observed that even fully vaccinated people can still be infected¹¹. According to the American Academy of Pediatrics, there has been a significant increase of infected children, and the long-term effects that COVID-19 infection may cause in this population are not yet known.

As in other countries, Brazil chose to lift in-person school activities in March 2020. At the beginning of the 2021 school year, some cities gradually reopened their schools¹². In Sao Paulo city, the reopening took place at a critical moment when the health system was under pressure. Thus, the objective of this study was to assess the frequency of SARS-CoV2 infection in daycare attendants in a poor area of Sao Paulo city, Brazil.

MATERIALS AND METHODS

Study and population

A prospective observational study was conducted in daycare centers located in the South of Sao Paulo, Brazil. This study was approved by the Research Ethics Committee of Santo Amaro University (N° 4.090.860).

This study was conducted from March to September 2021. Seven daycare centers were selected to participate in this study. Sao Paulo's South region is considerably large and comprehends both wealthy and poor areas. Our study focused on daycare centers located in the poor region, which comprises the peripheral area of the city.

Sampling and SARS-CoV2 detection

Two swabs were collected from each participant, one from the nasopharynx and the second from the oropharynx. Nasopharynx samples were collected with flocked rayon (Laborclin, Vargem Grande, Parana, Brazil) introduced into the nostrils and when it reached a resistance point, the swab was rotated 2-3 times. The oropharynx samples were collected with rayon swabs and comprised the collection from the adenoids. The swabs were placed in a Falcon tube containing phosphate-buffered saline (PBS). The RNA was obtained by Nucleic acid extraction, using the Purelink RNA purification kit (Invitrogen, Waltham, Massachusetts, EUA). The viral diagnosis was performed in one step, with

Table 1 - Sociodemographic data of the population served by the participating daycare centers.

District	Daycare center ID	Children in the study (%)	Adults in the study (%)	Total population	Population from 0 to 3 years old (%)	Households in slums	Families in extreme poverty
Capela do Socorro	1,6,7	51	25	629.260	5.7	23.842	31.599
M'Boi Mirim	2	23	40	631.925	5.8	43.527	32.246
Campo Limpo	3,4,5	26	35	688.779	5.5	58.496	25.121

Table 2 - Distribution of study participants by age group; SD = standard deviation.

Daycare	Approximated capacity	N°	Adults				Children			
			Male	Female	Average Age	SD Age	Male	Female	Average Age	SD Age
1	30	20	1	8	47	±10	6	5	3	±1
2	790	61	3	30	37	±6	15	13	3	±0.8
3	110	19	0	10	39	±11	7	2	2	±1.2
4	250	21	5	2	33	±4	6	8	2	±0.9
5	560	19	1	10	41	±11	2	6	2	±,1.4
6	200	39	-	-	-	-	20	19	3	±0.9
7	150	22	0	11	47	±11	7	4	2	±0.7
Total	-	201	10	71	40	±9,82	63	57	3	±1.08

real-time Reverse Transcription polymerase chain reaction (RT-qPCR) using Seegene’s Allplex™ 2019-nCoV Assay kit (Seegene, Songpa-gu, Seoul, Korea). The samples were tested in duplicated amplification parameters as follows: 1 cycle at 50 °C per 20 min; 1 cycle at 95 °C per 15 min; 45 cycles at 94 °C per 15 s, and 58 °C per 30 s. The samples were assessed as positive following the parameters established by the kit manufacturer. Furthermore, negative and positive controls were included in all test plates.

RESULTS

Seven daycare facilities located in the south of Sao Paulo were included in the study. These daycare centers serve a needy population that lives on the outskirts of the city. The sociodemographic data are shown in [Table 1](#).

Only daycare 1 serves children from 0 to 5 years old. Daycare centers 2 to 7 serve children from 0 to 3 years old. The adults that were invited to participate in the study were workers with a mean age of 40 years old (9.82) in close

contact with the children. We collected 201 samples and the participant profiles are listed in [Table 2](#).

Only one sample tested positive for SARS-CoV2 ([Table 3](#)). The sample was collected from a 33-year-old woman who is an employee at daycare center 3. She did not have any comorbidities or previous SARS-CoV2 infection. Although vaccination started in the country in January 2021, the participant was not in the eligible age group to receive a vaccine at the time of sampling. She lives in a poor area of Sao Paulo and earns around two Brazilian minimum wages, which represents about US\$ 432. Her household didn’t receive any federal government financial aid. She lives in a small house close to her working place. Although she remained in quarantine isolation in her room for 14 days, her husband and daughter were monitored and tested at the local health center, and both of them tested positive for SARS-CoV2. Although 20 children had symptoms such as runny nose and nasal congestion, none of them tested positive for SARS-CoV-2.

DISCUSSION

Brazil was among the countries to be most seriously affected by the pandemic. Presenting a high number of cases, hospitalizations, and deaths even with the rollout of vaccination¹³. In Sao Paulo, children aged from 0 to 9 years represent 2.9% of accumulated COVID-19 cases¹⁴. This study took place when they returned to school after a

Table 3 - SARS-CoV2 frequency in daycare centers.

	Participants		SARS-Cov2 frequency	
	N	%	N°	%
Adults	81	40.3%	1	1.2%
Children	120	59.7%	0	-
Total	201	100%	1	0.5%

long period without in-person activities. At the time of the collection, only 35% of the children were allowed to attend daycare centers daily and student attendance was optional.

It is important to note that poor world regions lack assistance, and therefore are more sensitive to viral transmission and more likely to present a higher death risk¹⁵. In the region where the study was conducted, most of the population is of socio-economic vulnerability, and 50% of its districts are in slums. Most families depend on government social services and have limited employment opportunities and a per capita income below the minimum wage. Nevertheless, local daycares are overcrowded, leading to long waiting lists for vacancies¹⁶. Due to the pandemic, daycares did not have in-person activities from March 2020 to February 2021. When the mayor of Sao Paulo city authorized the resume of in-person activities, there was only at 35% of student capacity and 100% of daycare workers' attendance. It is important to note that data from the Department of Health indicate that this period had been extremely sensitive for the country. During March and April 2021, the case and death averages were extremely high, mainly due to the Gamma variant (P.1)¹⁷ outbreak across the country.

Following the return to school, many measures were implemented to reduce the spread of SARS-CoV-2 in the school environment such as measuring the temperature at the entrance, and frequent hand washing or disinfection with 70% alcohol. Moreover, the frequent cleaning of surfaces and objects, the ventilation of environments, the physical distance, and the use of masks by adults and children as of 4 years of age. In addition, recreation activities and meals were conducted on a rotational basis to prevent mass gatherings. People suspected to have the virus were isolated, children with flu-like symptoms could not remain in daycare centers, and those with positive SARS-CoV-2 test would stay in quarantine, a measure that was applied to both workers and students. Parents were instructed not to take their children to daycare if they had flu-like symptoms or there was a case of SARS-CoV2 in a family member of the same household. Unfortunately, many of the infections are likely to be asymptomatic¹⁸, which highlights the need for mass testing. According to this finding, the positive case we found was a completely asymptomatic employee and, up to the time of collection, without any positive case in the family. No secondary cases were reported by the school.

Despite the high number of cases that have occurred in the country, this finding could be explained by several reasons. First, the non-pharmaceutical measures applied in daycare centers were effective and the transmission of SARS-CoV-2 in environments can indeed be lowered. Second, child attendance was significantly reduced and

the actual transmission rate among children should only be assessed under normal conditions. Third, it is known that the Variant of Concern (VOC), which was prevalent across the country, is more symptomatic than previous circulating SARS-CoV2 strains¹⁹. Therefore, it could lead the symptomatic individual to be away from the daycare either because of hospitalization or mild symptoms. Finally, an earlier study described that children infected with SARS-CoV-2 can be asymptomatic more often than adults. Kociolek *et al.*²⁰ found that asymptomatic children have reduced viral loads.

CONCLUSION

Despite the reasons that may have interfered with the rates of SARS-CoV2 transmission in daycare centers, our study was rolled out during the worst moment of the pandemic in the Brazilian population. Importantly, our findings show that the viral circulation tends to be low if testing and non-pharmacological preventive measures are considered part of the protocols. Today, immunization is accessible to the people, which has extensively reduced the cases and deaths of COVID-19. However, the transmission can still occur and non-pharmacological preventive measures should always be considered to avoid transmission in mass gathering sites such as daycares.

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