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Prevalence and Transmission of Severe Acute Respiratory Syndrome Coronavirus Type 2 in Childcare Facilities: A Longitudinal Study

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Objective To evaluate the role of childcare facilities in the transmission of severe acute respiratory syndrome coronavirus type 2 (SARS-CoV-2) in a longitudinal study to gain further knowledge of SARS-CoV-2 prevalence, transmission, and spread among preschool children, their parents, and their caregivers.

Study design Children aged 1-6 years, their parents, and their caregivers in 14 childcare facilities in Dresden, Saxony/Germany were invited to participate in the KiTaCoviDD19-study between July 2020 and January 2021. Seroprevalence of SARS-CoV-2 antibodies was assessed up to 4 times during the study period in all participating adults, and demographic characteristics, as well as epidemiologic information on personal SARS-CoV-2 history were obtained. Samples for stool virus shedding of SARS-CoV-2 were analyzed by polymerase chain reaction every 2-4 weeks in all participating children.

Results In total, 318 children, 299 parents and 233 childcare workers were enrolled. By January 2021, 11% of the participating adults were found to be seropositive, whereas the percentage of children shedding SARS-CoV-2 was 6.8%. Overall, we detected 17 children with SARS-CoV-2 virus shedding in 8 different childcare facilities. In 4 facilities, there were a maximum of 3 connected cases in children. Approximately 50% of SARS-CoV-2 infections in the children could not be connected to a secondary case in our study population.

Conclusions This study does not provide evidence of relevant asymptomatic ("silent") spread of SARS-CoV-2 in childcare facilities in both low- and high-prevalence settings. Our findings add to the evidence that childcare and educational settings do not have a crucial role in driving the SARS-CoV-2 pandemic. *(J Pediatr 2021;237:136-42)*.

Since the beginning of the coronavirus disease 2019 (COVID-19) pandemic, school and childcare closures have been one of the main strategies to limit transmission.[1] These measures are based on the assumption that children play a similar role in transmitting severe acute respiratory syndrome coronavirus type 2 (SARS-CoV-2) as they do in transmitting influenza, and that school and childcare facility closures will effectively lower the overall transmission rate.[2]

However, most countries, including Germany, report a much lower proportion of cases in children compared with their proportion of the population.[3-5] In addition, COVID-19 most often leads to no or mild symptoms in children and carries a low risk of a serious course of disease in children. The disease's impact of childcare facility and school closures, as well as limited social interactions on children's mental health is becoming increasingly apparent, however.[6,7]

Although several studies in the United Kingdom and Norway identified only a very limited spread of COVID-19 in primary and secondary schools,[8,9] similar studies in preschools and childcare facilities are lacking. Given the difficulty of implementing relevant distancing and hygiene measures in this age group, SARS-CoV-2 transmission in childcare facilities is of particular interest. In this study, we investigated SARS-CoV-2 transmission in childcare facilities attended by children aged 1-6 years.

According to the prevalence of SARS-CoV-2 infections, mitigation strategies were implemented and tightened stepwise by the Federal State of Saxony during the study period. The study was continued even throughout a period of strict lockdown with limited childcare services.

COVID-19	Coronavirus disease 2019
ELISA	Enzyme-linked immunosorbent assay
IgG	Immunglobulin G
PCR	Polymerase chain reaction
SARS-CoV-2	Severe acute respiratory syndrome coronavirus type 2

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Methods

Children aged 1-6 years, their parents, and childcare workers in 14 childcare facilities in Dresden were invited to participate in the KiTaCoviDD19-study. A maximum of 1 corresponding parent per participating child was included in the study.

The study was approved by the Ethics Committee of Technische Universität Dresden (BO-EK-180052020), registered on July 30, 2020, and assigned clinical trial number DRKS00022729 (https://www.drks.de/drks_web/navigate.do?navigationId=trial.HTML&TRIAL_ID=DRKS00022729).

After informed consent was obtained, a 5-mL sample of peripheral venous blood was collected from adult participants at 3 or 4 defined time points during the study period, and serologic testing for immunglobulin G (IgG) antibodies against SARS-CoV-2 was conducted (**Figure 1**). No blood sampling or serologic testing was performed in the children. Parents were asked to collect stool samples from their child every 2 weeks. Excretion of SARS-CoV-2, as mostly noninfectious particles, is known to occur in infected people, especially children, for at least 2 weeks.[10] This served as an easy-to-obtain specimen in the childcare population. These stool samples were tested by polymerase chain reaction (PCR) to detect stool excretion of SARS-CoV-2 RNA. All blood and stool samples were collected between July 15, 2020, and January 31, 2021.

In addition, data on age, household size, comorbidities, regular medication, previously diagnosed SARS-CoV-2 infections in the participants and their household contacts, quarantine episodes, utilization of daycare during lockdown, number of contacts other than household contacts, and the occurrence of respiratory symptoms were collected.

Laboratory Analysis

Stool samples were frozen at −80 °C and then thawed to perform PCR testing after a minimum of 4 weeks of storage. According to the study protocol, stool samples (collected from each child) were initially examined at a 4-week interval. If at least 1 stool sample from a childcare facility tested positive, then all samples from this facility collected 2 weeks earlier and 2 weeks later were tested as well.

A liquid handling system (Nimbus; Seegene) was used for nucleic acid extraction using a modified protocol supplied by the manufacturer. A spatula point of stool in 1 mL of ASL buffer (Qiagen), mixed thoroughly and incubated for at least 10 minutes and then centrifuged at 16 060 × *g* for 2 minutes, was used for the extraction procedure as well as for the PCR setup. Extraction was followed by real-time reverse-transcriptase PCR (RT-PCR) using Seegene's Allplex 2019-nCoV assay. This assay detects 3 target regions within the genome of SARS-CoV-2: the *E* gene, *RdRP* gene, and *N* gene. The automatic calculation software supplied by Seegene was used for interpretation of the results. A specimen was considered positive if at least 1 target region was detected. In each test series a stool specimen from a patient with confirmed SARS-CoV-2 infection was run in parallel as a positive control for correct extraction and the PCR procedure. In the event of test inhibition (internal control), the assay was repeated once.

Blood samples were assessed for SARS-CoV-2 IgG antibodies using a commercially available chemiluminescence immunoassay technology for the quantitative determination of anti-S1– and anti-S2–specific IgG antibodies to SARS-CoV-2 (LIAISON SARS-CoV-2 S1/S2 IgG Assay; DiaSorin). Antibody levels >15.0 AU/mL were considered positive, and levels between 12.0 and 15.0 AU/mL were considered equivocal.

All samples with a positive or equivocal LIAISON test result, as well as all samples from participants with a

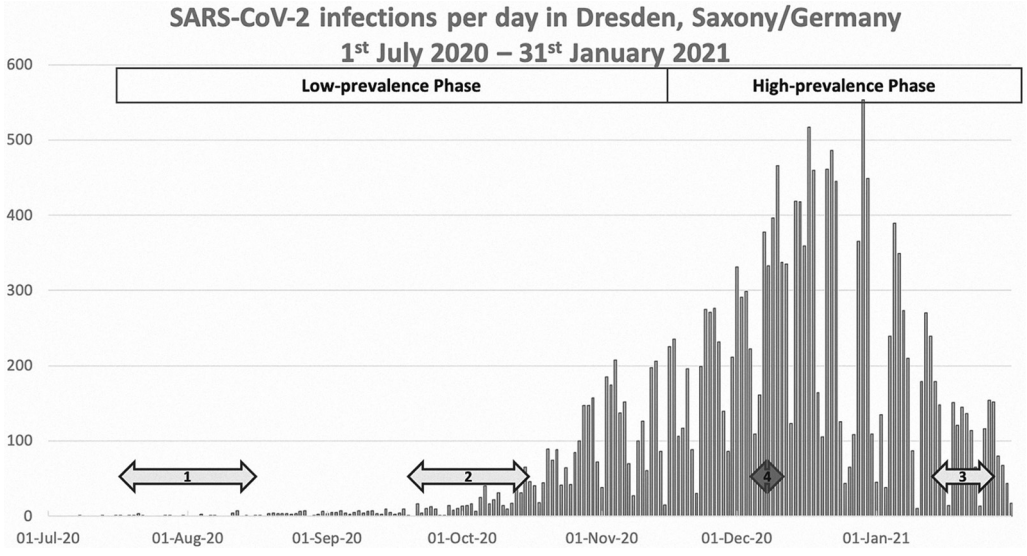

Figure 1. Timeline of serologic testing (1, baseline; 2, second serologic testing; 3, third serologic testing; 4, additional serologic testing in December 2020) and reported numbers of SARS-CoV-2 infections in Dresden, Saxony/Germany.[16]

PCR-confirmed SARS-CoV-2 infection via nasopharyngeal swab, were tested with 2 additional serologic tests; a chemiluminescent microparticle immunoassay intended for the qualitative detection of IgG antibodies to the nucleocapsid protein of SARS-CoV-2 (ARCHITECT SARS-CoV-2 IgG; Abbott Diagnostics) and an enzyme-linked immunosorbent assay (Anti-SARS-CoV-2 ELISA; EUROIMMUN) to detect IgG against the S1 domain of the SARS-CoV-2 spike protein.

Definitions

Participants with detectable antibodies in at least 2 assays were considered seropositive. Cases of SARS-CoV-2 infections within a facility were considered possibly linked if there was an epidemiologic and temporal association. An epidemiologic association was defined as at least 2 SARS-CoV-2 infections occurring in the same facility, and a temporal association was defined as cases occurring within the infectious period of each other, defined as 48 hours before onset of symptoms or a positive PCR test (on a stool sample or nasopharyngeal swab) until 14 days after the onset of symptoms. For stool samples, we extended this period to 14 days before and after collection of the positive sample. In cases of consecutive positive samples, only the date of the first positive stool sample was taken into consideration, given that previous studies have shown that SARS-CoV-2 RNA can be shed through stool for multiple weeks, whereas the infectious period is usually limited to the first 10 days.[11] In what follows, "PCR" generally refers to PCR testing via nasopharyngeal swab if not clearly stated otherwise.

Prevalence of Community SARS-CoV-2 and Mitigation Strategies in Childcare Facilities

According to the prevalence of SARS-CoV-2 infections in Dresden, we divided our study period (July 15, 2020, to January 31, 2021) into a low-prevalence phase and a high-prevalence phase (Figure 1). In the low-prevalence phase (July 15, 2020, to November 15, 2020), cumulative reported cases tripled in Dresden, whereas in the high-prevalence phase (November 16, 2020, to January 31, 2021), cumulative cases increased by more than 10-fold.

Only children with at least 2 donated stool samples in the respectively defined periods were included in this analysis; 232 of 318 children (73.0%) met these criteria in the low-prevalence phase, and 222 of 318 children (69.8%) did so in the high-prevalence phase (Table I). No positive stool samples were excluded by these criteria.

In the low-prevalence period, mitigation strategies implemented by the Federal State of Saxony initially included a mask mandate only for parents and not for childcare workers or children. At the end of this period, starting on November 2, 2020, fixed separation of different childcare groups was required; before then, contacts among the children had not been limited. Furthermore, access for parents was limited during drop-off and pick-up times.

In the high-prevalence period, mitigation strategies were tightened to a strict lockdown starting on December 14, 2020, during which childcare facilities were required to limit their services to emergency care for children of essential workers and to maintain the separation of different childcare groups and limited access for parents. In addition, childcare workers were required to wear masks when in contact with other adults (ie, parents/coworkers).

Furthermore, during the entire study period, children with a confirmed SARS-CoV-2 infection and those with close contact with an infected individual or individuals with respiratory symptoms or fever were not allowed to attend daycare facilities. The study continued throughout the period of limited childcare services. Parents whose children did not visit the facility during this time were invited to drop off their children's stool samples nonetheless.

Table I. Number of participants at each time point and number/cumulative prevalence of seropositive adult study participants and SARS-CoV-2–positive children during the low- and high-prevalence phases

Participants	Baseline serology, n/N	Low-prevalence phase, n/N (%)	High-prevalence phase, n/N (%)
All seropositive participants	0/361	2/350 (0.6)	48/424 (11.3)
Seropositive parents	0/206	1/196 (0.5)	25/236 (10.6)
Seropositive childcare workers	0/155	1/154 (0.6)	23/187 (12.3)
SARS-CoV-2 positive children	Not available	2/232 (0.9)	15/222 (6.8)

Statistical Analyses

Analyses were performed using SPSS 25.0 (IBM) and Excel 2010 (Microsoft). The Fisher exact test was used to determine categorical variables for the statistical analysis. A P value $\leq .05$ was considered to indicate statistical significance.

Results

Study Population/Demographics

In total, 318 children, 299 parents, and 233 childcare workers in 14 childcare facilities were enrolled between July 15, 2020, and January 31, 2021, representing 72% (50%-100%) of all staff members and 18% (12%-27%) of children attending these institutions (Table II). A corresponding parental serostatus was available for 314 of the 318 children (99%). Thirty-eight (12%) of the enrollees were siblings.

The median age of the children was 4 years (IQR, 2-5 years), that of their parents was 37 years (IQR, 34-40 years), and that of the childcare workers was 39 years (IQR, 32-49 years). The median household size was 4 (IQR, 3-4) for the children and parents and 3 (IQR, 2-3) for the childcare workers (Table II).

Low-Prevalence Period (July 15, 2020, to November 15, 2020)

At baseline, none of the participants was SARS-CoV-2 seropositive. In the low-prevalence phase until mid-November, 2 study participants—1 childcare worker (1 of 154; 0.6%) and

Table II. Baseline demographic data

Characteristics	Childcare workers	Parents	Children
Number of participants	233	299	318
Age, y, median (IQR)/(range)	39 (32-49)/(17-72)	37 (32-40)/(25-55)	4 (2-5)/(1-6)
Female sex, %	88.2	68.2	49.5
Household size, median (IQR)	3 (2-3)	4 (3-4)	

1 parent (1 of 196; 0.5%)—became seropositive. Both participants reported a known PCR-confirmed infection. They were not associated with the same childcare facility.

During the same period, we detected 2 positive stool samples in attendees, resulting in a cumulative prevalence of SARS-CoV-2 positivity of 0.9% (2 of 232 children) (Table I). These 2 children attended different childcare facilities. One case was detected only retrospectively by our study; there was no epidemiologically linked case to another child or to a childcare worker in the same facility. The other child showed symptoms of COVID-19, and SARS-CoV-2 was detected by PCR at the time of infection; one epidemiologically linked case occurred in another child in the same facility.

In both cases of positive stool samples, the parents did not have detectable antibodies and did not report positive PCR testing for SARS-CoV-2.

High-Prevalence Period (November 16, 2020, to January 31, 2021)

At the end of January 2021—after the second wave of the pandemic—seropositive rates were higher, but no statistically significant difference in the seropositivity of parents and childcare workers was detected (10.6% [25 of 236] vs 12.3% [23 of 187]; $P = .64$) (Table I).

During the same period, 15 of 222 children (6.8%) had at least 1 SARS-CoV-2–positive stool sample. In 6 of these cases, no connection to other children with SARS-CoV-2–positive stool samples could be found. The remaining 9 cases occurred in 4 different childcare facilities and were recorded as possibly epidemiologically linked according to our definitions.

Seropositivity: Detected/Undetected Cases

Twenty of 25 seropositive parents (80.0%) had previously tested positive for SARS-CoV-2, and 1 parent reported a confirmed SARS-CoV-2 infection in a household member. Similarly, 18 of 23 childcare workers (78.3%) had a personal history of SARS-CoV-2, and 1 childcare worker had a positive history in a household member. The ratio of undetected to detected SARS-CoV-2 infections did not differ significantly between parents and childcare workers (0.19 [4 of 21] vs 0.21 [4 of 19]).

Ten of 25 seropositive parents (40%) and 5 of 23 seropositive childcare workers (21.7%) identified a confirmed SARS-CoV-2 contact outside the childcare facility as a source of infection.

Seropositivity: Additional Results

Seroprevalence was significantly higher in parents whose children remained in emergency care during the strict lockdown compared with parents whose children did not attend the facility during this time (18.8% [12 of 64] vs 8.6% [13 of 152]; $P = .038$). Seroprevalence was also significantly higher in childcare workers assigned mainly administrative tasks compared with those with mainly childcare-related duties (20.8% [10 of 48] vs 8.1% [8 of 99]; $P = .034$).

Eleven seropositive participants (2 childcare workers and 9 parents) had an additional serum sample taken in mid-December; 3 of these 11 (27.3%) were already seropositive at that time, and the other 8 (72.7%) seroconverted during the strict lockdown starting on December 14, 2020.

Sixty-eight of 187 childcare workers (36.4%) and 93 of 236 parents (39.4%) reported symptoms of an upper respiratory tract infection during the study period.

Children Who Were SARS-CoV-2 Positive

SARS-CoV-2 RNA was detected in 22 of the total 1168 stool samples analyzed, belonging to 17 different children, with 5 children having 2 consecutive positive samples.

Among 17 parents with a SARS-CoV-2–infected child, 6 (35.3%) were seropositive, 8 (47.1%) were negative, and 3 parents did not undergo serologic testing after their children's positive stool sample.

Two of 17 children with stool virus shedding had siblings who were also enrolled in this study. In both cases, the stool samples from the respective sibling yielded negative results.

Almost 30% of parents (64 of 219) reported that their children attended the childcare facility during the strict lockdown starting on December 14, 2020. The cumulative SARS-CoV-2 prevalence did not differ significantly between children attending emergency care and those who did not (6.3% [4 of 64] vs 8.6% [13 of 152]; $P = .78$).

Children Who Were SARS-CoV-2 Positive: Detected/Undetected Cases

Of 17 children with positive stool PCR test results, 8 (47%) had either themselves or a household contact nasopharyngeal specimen PCR-confirmed SARS-CoV-2 clinical infection, for a 1.125 (9:8) ratio of undetected to detected SARS-CoV-2 infections in the participating children. Seven of the remaining 9 children with a subclinical SARS-CoV-2 infection attended their childcare facilities during virus shedding. In 3 cases, 0 or 1 possibly linked cases were detected. Four cases were linked to outbreaks; however, given the retrospective design of our study, we cannot make assumptions about the index case of the outbreaks.

Children Who Were SARS-CoV-2 Positive: Transmission in Childcare Facilities

The 17 SARS-CoV-2–positive children attended 8 different childcare facilities. The maximum number of children with a SARS-CoV-2–positive stool sample per childcare facility was 4. Five of the 17 children (29.4%) did not attend the childcare facility before or during virus shedding, thereby

eliminating the possibility of an infection connected to their childcare facility. The remaining 12 children with a SARS-CoV-2–positive stool sample attended their childcare facility at the time of virus shedding, affecting 5 institutions. In 7 cases, at least 1 epidemiologically associated SARS-CoV-2 infection was detected. In the other 5 cases, there were no epidemiologically associated SARS-CoV-2 infections within the same facility.

SARS-CoV-2 Outbreaks: Overall Transmission in Childcare Facilities

We detected at least 1 SARS-CoV-2–positive participant (via serologic testing or stool PCR) in 13 of 14 childcare facilities (92.8%). In 8 of these 14 facilities (57.1%), none or only isolated cases without possible epidemiologic association occurred. In 3 of the 14 facilities (21.4%), there were a maximum of 2 SARS-CoV-2 infections with a possible epidemiologic link, and in 3 facilities (21.4%) there were outbreaks involving at least 3 cases (range, 3-10). In all outbreaks, positive stool samples in children could be detected (**Figure 2**).

Quarantine

Eleven of the 14 participating childcare facilities had at least 1 episode of quarantine mandated by the local health department because of a PCR-confirmed SARS-CoV-2 infection at their facility. At all 3 facilities with detected outbreaks, quarantine was ordered accordingly. In 2 facilities without quarantine episodes, we detected a total of 3 SARS-CoV-2–positive stool samples while some of the children attended their childcare facility during the time of stool virus shedding. In all 3 childcare facilities without quarantine episodes, we detected only isolated cases of SARS-CoV-2 with no epidemiologic links.

Discussion

The rate of seropositive participants during the low-prevalence phase following the first wave in the Federal State of Saxony/Germany was low, with only 2 seropositive adults detected. Both of these participants previously knew of their infections, quarantine warranties were issued accordingly, and we could not find any cases connected to either participant. This supports the assumption that the testing and quarantine methods in Germany during the low-prevalence phase were effective and successfully prevented an undetected spread of SARS-CoV-2. In accordance with this, we found evidence of only 1 clinically undetected infection in a child during this period. Even though this child attended the childcare facility and no special hygiene or distancing measures were taken by either the parents or the childcare workers, we did not detect a secondary case.

The increasing number of SARS-CoV-2 cases during the second wave in the general population was mirrored in our study population. Although the rate of seropositive participants rose considerably by January 2021, this increase was proportional to the increase within the general population in Dresden. The number of infections detected only by antibody testing in our studied adults continued to be lower than previously assumed by some authors.[12] We did not find a significant difference in the seropositivity rate between parents and childcare workers. The percentage of stool SARS-CoV-2 PCR-positivity was considerably lower in the children than in the adults during the high-prevalence study phase. This finding is supported by previous studies that also reported lower infection rates in children.[5,13]

Over the entire study period, we detected a maximum of 3 connected cases in children. There was a risk of underestimating secondary spread among children, given the participation rate of 20%. However, the fact that no outbreaks among staff were detected in childcare facilities without SARS-CoV-2 virus–shedding children supports the hypothesis that children are not a major source of uncontrolled clusters.[14] Also noteworthy is that in 2 cases, an infection was detected in children with a sibling also in childcare, and that in both cases, the other sibling did not test positive in the time-corresponding stool samples. Roughly 50% of the clinical SARS-CoV-2 infections in our study children could not be connected to a secondary case.

Although in this study the rate of participation in children was only approximately 20%, we included an average of 72% of childcare workers. Because of this high participation rate, we are confident that we analyzed an adequate number of samples to evaluate the SARS-CoV-2 seroprevalence rate in this group. We did not detect a significantly higher rate of seropositivity in childcare workers compared with the participating parents. Although the seropositive rate was significantly higher in parents whose child attended the childcare facility during the lockdown period, this also infers a working environment without the possibility of a home office. Therefore, this effect also could be related to the larger number of regular contacts within the work environment outside the household. This assumption is supported by the lower percentage of SARS-CoV-2–positive children in emergency care compared with those who stayed at home during the strict lockdown.

The percentage of seropositive childcare workers whose infection was possibly connected to their childcare facility was considerably higher than among seropositive parents. The significantly higher rate of seropositivity in childcare workers with mainly administrative duties compared with those without such duties, the higher infection rate in childcare workers compared with children, as well as the extremely limited spread linked to undetected SARS-CoV-2–positive children, suggest that transmission within childcare facilities occurs more frequently between adults than between children and adults. Although they cannot be directly attributable, hygiene and distancing measures themselves might be key measures in these facilities and more important between childcare workers than between children and these workers.

There are several limitations to our study. We enrolled a limited number of participants, and thus it was possible that some infections remained undetected. Moreover, the lower

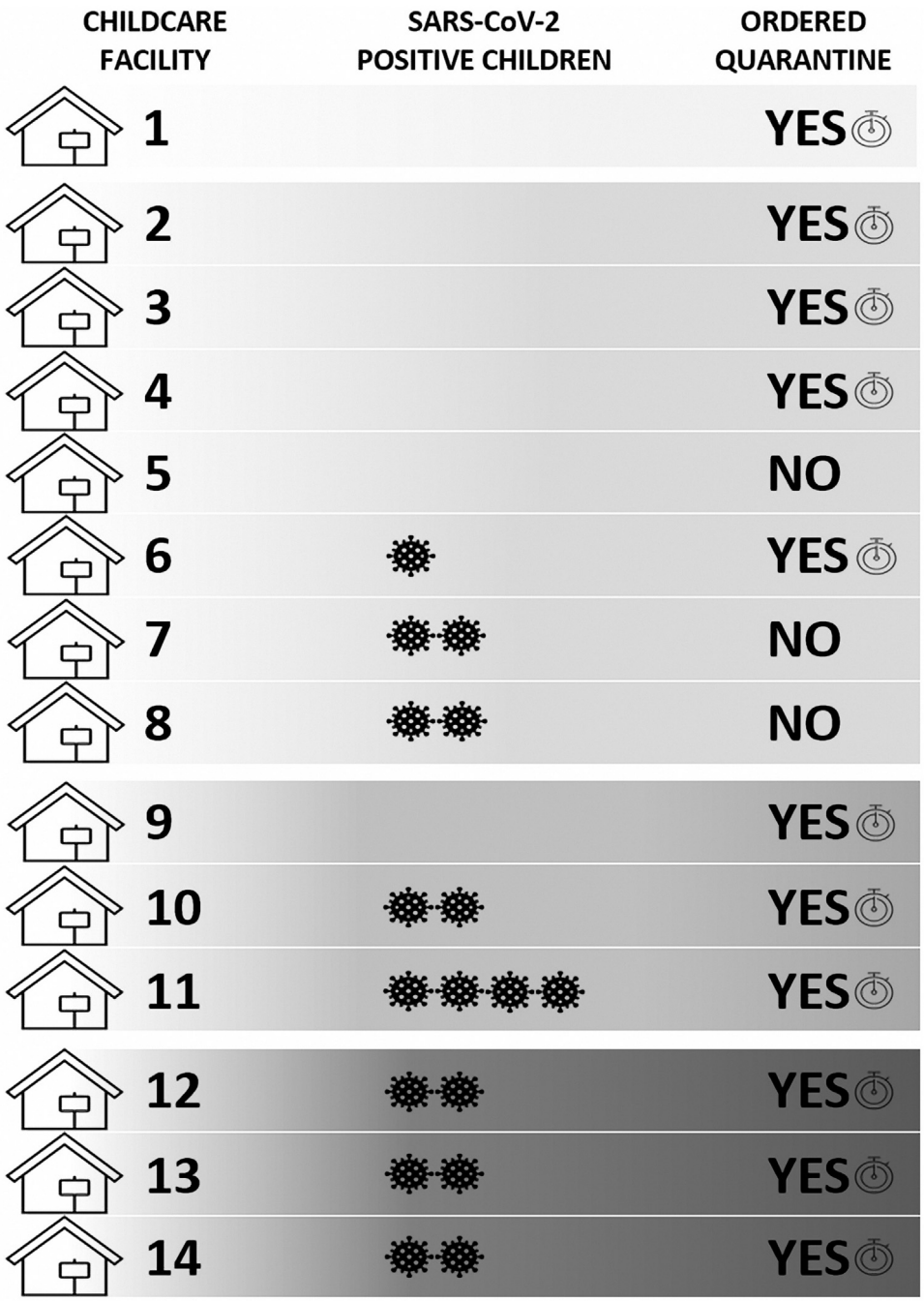

Figure 2. Positive SARS-CoV-2–infected children and the occurrence of epidemiologically linked transmissions within the single participating childcare facilities. Childcare facility 1, no epidemiologic link detected; childcare facilities 2-8, single cases without an epidemiologic link detected; childcare facilities 9-11, 2 epidemiologically linked cases detected; childcare facilities 12-14, outbreaks with >2 epidemiologically linked cases detected; stopwatch, officially mandated quarantine by local health department during the study period.

sensitivity of PCR testing for SARS-CoV-2 in stool samples than in oropharyngeal and nasopharyngeal swabs also might contribute to underestimating the true numbers of infections in the participating children.[15] Although we could detect only a limited spread of SARS-CoV-2 infections in childcare facilities, future developments, such as novel virus variants, may change this dynamic.

The inclusion of both low- and high-prevalence settings during the study period and the high participation rate of childcare workers in all facilities are strengths of our study. In addition, the use of antibody testing instead of PCR testing in parents and childcare workers reduced the possibility of missed undetected SARS-CoV-2 cases, at least in the adults.

This study does not provide evidence for a relevant asymptomatic ("silent") spread of SARS-CoV-2 in childcare facilities, despite the lack of hygiene or distancing measures in this age group. In addition, adults seem to transmit SARS-CoV-2 more frequently than children. These findings add to the evidence that childcare and educational settings do not have crucial roles in driving the SARS-CoV-2 pandemic. ■

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Data Statement

Data sharing statement available at http://www.jpeds.com.

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