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SHORT COMMUNICATION

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Real-life evaluation of a COVID-19 rapid antigen detection test in hospitalized children

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Abstract

Rapid antigen detection (RAD) tests for the detection of SARS-CoV-2 are simpler, faster, and less expensive than the reverse-transcription polymerase chain reaction (RT-PCR) that is currently considered the gold standard for the diagnosis of coronavirus disease 2019 (COVID-19). The objective of this study was to determine the performance of the PANBIO COVID-19 Ag RAD (Abbott) test, a lateral flow immunoassay that detects the nucleocapsid protein, using as a reference RT-PCR method the Cobas®8800 System (Roche Diagnostics). This prospective study was conducted in a tertiary Children's Hospital and included individuals aged ≤16 years with COVID-19-related symptoms or epidemiological criteria for COVID-19. Two nasopharyngeal samples were collected to perform the PANBIO RAD test and RT-PCR. Of 744 children included, 51 (6.86%) had a positive RT-PCR result. The RAD test detected 42 of 51 PCR-positive children while there were no false-positive results. The overall sensitivity and specificity were 82.35% (95% CI, 71.9%–92.8%) and 100%, respectively. Sensitivity was >95% in symptomatic children. The assay performed poorly in asymptomatically infected children. In agreement with previous studies in adults, the PANBIO RAD test can be useful in screening for COVID-19 in children admitted with symptoms suggestive of the disease, especially in the first days of the illness.

KEYWORDS
children, COVID-19, diagnosis, rapid antigen test, SARS-CoV-2

1 | INTRODUCTION

The ongoing severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) pandemic challenges public health systems worldwide. Rapid and accurate diagnosis of coronavirus disease 2019 (COVID-19) is essential to facilitate treatment, reduce the spread of disease, protect healthcare workers and ensure the optimal use of personal protective equipment. Reverse-transcription quantitative polymerase chain reaction (RT-qPCR) in nasopharyngeal samples is the currently recommended method to diagnose acute infection due to its favorable performance characteristics. However, molecular techniques require trained personnel and specialized equipment and the turnaround time may be considerably longer. In contrast, rapid antigen detection (RAD) tests are considered less sensitive, but they have the advantages of simple operation, short turnaround time, and low cost.[1,2] To date, several RAD tests have been evaluated but to our knowledge data on children are scarce.

The study was conducted in "P. and A. Kyriakou" Children's Hospital, Athens, Greece.

We evaluated the PANBIO COVID-19 Ag RAD test (Abbott-Chicago) in children ≤16 years of age admitted to a tertiary Children's hospital in Athens during the second and third pandemic waves using as a reference RT-PCR method, the Cobas®8800 System (Roche Diagnostics).

2 | MATERIALS AND METHODS

This prospective study was conducted between September 25th, 2020 to February 28th, 2021 in the second largest pediatric hospital of the country and a reference center for children with COVID-19. Children admitted to our hospital and tested for SARS-CoV-2 using both RAD test and RT-PCR were included. According to local protocol, children were tested either because they had symptoms suggestive of COVID-19 or they were considered at high risk of infection based on regional epidemiological data such as belonging to minority populations or originating from high prevalence regions. On admission, two nasopharyngeal samples were simultaneously collected, each one from a separate nostril. Oral informed consent was obtained from the patients' parents for double sampling. One of the samples was taken with the swab provided by the PANBIO RAD test and the other one with a swab that was then embedded in a universal transport medium for RT-PCR (Copan flocked swabs with UTM™, Universal Transport Medium). The PANBIO COVID-19 Ag RAD test is a lateral flow immunoassay that detects the nucleocapsid protein providing results in 15 min. The Cobas®8800 System detects ORF1 a/b, a nonstructural region unique to SARS-CoV-2, and the structural protein envelope E-gene that is found in all the members of Sarbecovirus subgenus. The turnaround time of the molecular assay performed at a National Reference Centre was 6–12 h. All RAD tests were performed immediately at the bedside, according to the manufacturer's instructions. Samples used for molecular testing were transferred to the National Reference Centre by three daily shipments. The lab personnel who handled these samples was blinded to the results of the RAD test. Both tests' results, the symptoms and number of days since their onset, Cycle threshold (C_t) values for RT-PCR, and demographic data were prospectively recorded for all participants. The study was approved by the Ethics Committee of the "P. and A. Kyriakou" Children's Hospital (EC 18155/02.11.2020).

The sensitivity and specificity of the PANBIO RAD test, with 95% confidence intervals (CI), were calculated using the RT-PCR results as the standard. Sensitivity was calculated for all patients and for specific groups of patients according to the presence of symptoms and RT-PCR C_t values. The level of agreement between the tests was estimated using Cohen's κ index. Statistical analyses were performed using the free, open-source software environment R.

3 | RESULTS

A total of 744 children were included; 57.9% (431 of 744) were male. Their median age was 7.7 years (1.4–13.2) and 65.1% (484 of 744) had symptoms suggestive of COVID-19 while 34.9% (260 of 744) were admitted with non-COVID-19 related symptoms. RT-PCR result was positive in 51 of 744 (6.86%) children; 82.4% (42 of 51) were symptomatic. Although hospitalized, 39 of 42 (92.9%) of the PCR-positive symptomatic children had mild disease while only two developed moderate disease with hypoxemia, and one was diagnosed with pericarditis. The median duration of symptoms was 2 days (1–3, range 0.5–8). The RAD test detected 42 of 51 (82.35%) of the PCR-positive children, 40 of 42 (95.24%) of symptomatic children with PCR confirmed COVID-19, and 2 of 9 (22.2%) children admitted with asymptomatic SARS-CoV-2 infection. In total, there were 17.6% (9 of 51) false-negative results whereas all PCR negative children had a negative RAD test ($n = 693$). The agreement between the two methods was 98.79% (κ score: 0.897; 95% confidence interval (CI) 0.83–0.96). Sensitivity was inversely related to the C_t values

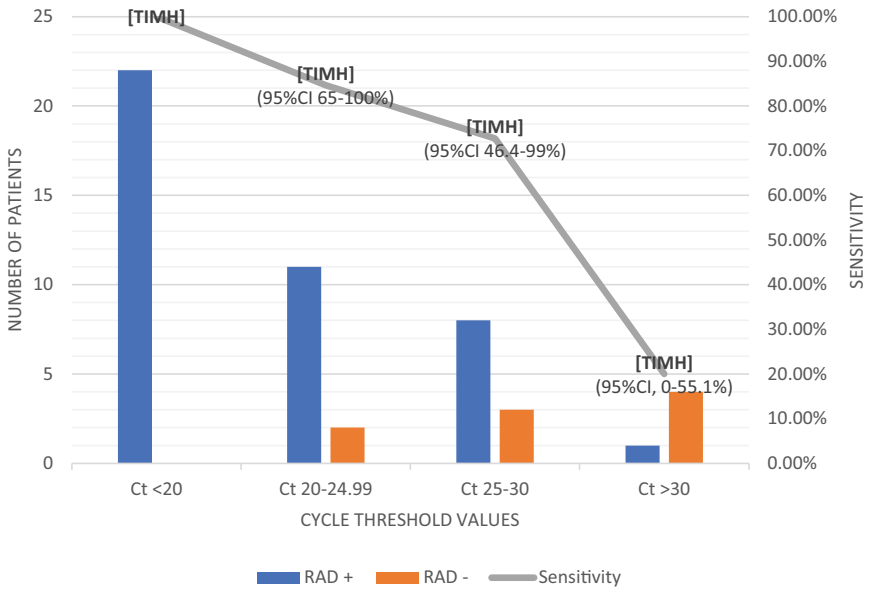

FIGURE 1 Association between cycle threshold (C_t) values, rapid antigen detection (RAD) test results, and sensitivity

A.	RADT+	RADT−	Total
Number of children	42	702	744
PCR positive	42	9	51
PCR negative	0	693	693
COVID-19	40	2	42
Asymptomatic SARS-CoV-2 infection	2	7	9
Duration of symptoms (days), median (range)	1 (0.5–4)	4.5 (1–8)	2 (0.5–8)
C_t median (Q1–Q3)	20.1 (16.9–24.9)	28.1 (24.8–33.6)	23.2 (19.6–26.4)

B.	Patients with symptoms suggestive of COVID-19	Asymptomatic children	Total
Prevalence of infection	8.7%	3.5%	6.86%
Sensitivity (95% CI)	95.2% (88.8–100)	22.2% (−4.9 to 49.4)	82.4% (71.9–92.8)
Specificity (95% CI)	100%	100%	100%
Positive predictive value (95% CI)	100%	100%	100%
Negative predictive value (95% CI)	99.5% (98.9–100)	97.3% (0.053–0.992)	98.7% (0.979–0.996)
κ score (95% CI)	0.973 (0.937–1)	0.355 (−0.005 to 0.716)	0.897 (0.83–0.96)

TABLE 1 (A) COVID-19 symptoms and duration, PCR positivity and median cycle threshold (C_t) values among SARS-CoV-2 RAD test positive and negative children. (B) Diagnostic performance of PANBIO RAD test in children who had symptoms suggestive of COVID-19 and in asymptomatic children tested for epidemiological reasons

(Figure 1). The overall sensitivity and specificity of the PANBIO RAD test were 82.35% (95% CI, 71.9%–92.8%) and 100%, respectively. The positive predictive value and the negative predictive value in this pediatric cohort with a prevalence of 6.86% were 100% and 98.72% (95% CI, 97.9%–99.6%), respectively. Of note, significant differences in the diagnostic performance of the assay were observed between symptomatic children and asymptomatically infected individuals (Table 1). Among the 9 of 51 (17.6%) falsely negative children, 7 were asymptomatic. Of the latter, 4 had C_t values ≥30, and 3 had C_t values between 25 and 30. The two false-negative children with SARS-CoV-2 had C_t values between 22 and 23 while one of them had symptoms for more than 7 days.

4 | DISCUSSION

The ongoing COVID-19 pandemic requires an ever-increasing number of tests for the early detection of SARS-CoV-2 infection. Rapid antigen detection tests could fill the gap between increased diagnostic needs and laboratory testing capacity. These tests are simple to perform by minimally trained healthcare personnel with no need for special equipment. They are also less expensive and can be performed at the point of care while the result is almost immediately available. However, in several early studies, RAD tests demonstrated poor performance for the COVID-19 diagnosis that precluded their extensive use.[3,4] Newer RAD tests show improved diagnostic characteristics that permit their implementation in several settings as part of a broader strategy for COVID-19 diagnosis and control.[5]

In this study, the PANBIO RAD test had a good clinical performance, with an overall sensitivity of 82.35% and a specificity of 100%. According to WHO guidelines, sensitivity ≥80% and specificity ≥of 97% are required for these tests compared to the RT-PCR assay.[1] The results of the current study agree with previously reported findings in adults.[6] However, previously conducted studies in children have demonstrated lower performance of antigen tests.[7] In a primary care setting, the PANBIO RAD test had a significantly higher sensitivity in adults (82.6%) compared to pediatric patients (62.5%).[8] Masiá et al.[9] found that younger age was independently associated with the poorest performance. Researchers claim that there are difficulties in collecting nasopharyngeal swabs in young children, dating of symptoms onset may be more inaccurate or C_t values may be lower in children compared to adults. However, the first of the above-mentioned studies included 85 children while the median age of participants in the second study was 40.6 years (23.0–55.6). In a retrospective, multicenter study that included more than 1600 symptomatic children attending the ED, the overall sensitivity of the PANBIO RAD test was 45.4%.[10] By contrast, in a prospective multicenter study conducted in ten university hospitals, the RAD assay was positive in all six out of 58 pediatric patients who had a positive RT-PCR result.[11] However, the small number of cases in this study does not permit any robust conclusions. Given that viral loads do not differ significantly between children and adults,[12] we

have shown that the PANBIO RAD test is a useful assay for the diagnosis of symptomatic children hospitalized with COVID-19. Nonetheless, appropriate collection of the nasopharyngeal swabs and accurate information about symptoms onset are necessary.

Although in most pediatric cases COVID-19 is mild, timely testing is required to ensure accurate diagnosis and optimal management, as well as proper isolation, especially in the hospital setting. The vast majority of symptomatic patients with COVID-19 will be diagnosed with the PANBIO RAD test during the first days of illness. In our study, only 2 of 42 (4.8%) children with symptomatic SARS-CoV-2 infection had a false negative RAD test; in one of these cases with a moderate disease the test was performed 8 days after the onset of symptoms. According to WHO recommendations, RAD tests should be performed in the first 5–7 days of symptomatic COVID-19.

In agreement with previous studies, the PANBIO RAD test performed poorly in patients with asymptomatic SARS-CoV-2 infection. WHO recommends that RAD tests should not be used in asymptomatic individuals unless they are contacts of a confirmed case.[1] This recommendation highlights the uncertainties regarding their use for screening purposes that have been demonstrated by several studies.[13,14] However, many experts suggest that frequent testing with a simple, inexpensive and quick RAD test will help to limit the asymptomatic spread of SARS-CoV-2 and reopen educative, professional, and social activities.[15] Of note, Ct values do not differ significantly between symptomatic and asymptomatic individuals. The performance of RAD tests highly depends on the setting and the prevalence of the disease; further large studies are needed to evaluate the clinical utility of these tests in the community, before their extensive implementation in national or international prevention strategies.[13,14]

The strengths of this study are the rather large number of participants, its prospective nature, and the evaluation of the PANBIO RAD test under real-life conditions. A limitation may be that the study was conducted in one hospital that serves as a COVID-19 referral center for children in central and southern Greece, so symptomatic children may be over-represented. In addition, given that the study population included hospitalized children with COVID-19, although mostly mild cases, the results can't be generalized in the whole pediatric population that often experiences asymptomatic disease.

In conclusion, this study confirms the high specificity, positive and negative predictive value of the PANBIO COVID-19 RAD in hospitalized pediatric patients with COVID-19 as well as its low performance in asymptomatically infected children. The assay showed an overall sensitivity of 82.35% that comes to an agreement with findings in adults. When including only children with symptoms suggestive of COVID-19 the sensitivity improves to 95.35%. Although the PANBIO RAD test meets the criteria set by WHO, it can't replace PCR especially in hospital settings where optimal sensitivity is required for accurate diagnosis and for the prevention of SARS-CoV-2 spread.[14] However, the assay performs well in symptomatic children hospitalized with COVID-19 and it has a much shorter turnaround time compared to RT-PCR. Therefore, it can be useful in screening for COVID-19 in children admitted with symptoms suggestive of the disease, permitting their early diagnosis, isolation, and management, especially in the first days of the illness.

CONFLICT OF INTERESTS

The authors declare that there are no conflict of interests.

DATA AVAILABILITY STATEMENT

The data that support the findings of this study are available from the corresponding author upon reasonable request.

AUTHOR CONTRIBUTIONS

Conceptualization: Irini Eleftheriou, Maria Tsolia; *methodology*: Irini Eleftheriou, Foteini Dasoula, Dimitra Dimopoulou, Evangelia Lebessi, Eftihia Serafi, Nikos Spyridis, Maria Tsolia; *formal analysis and investigation*: Irini Eleftheriou, Foteini Dasoula, Dimitra Dimopoulou, Evangelia Lebessi, Eftihia Serafi, Nikos Spyridis, Maria Tsolia; *writing—original draft preparation*: Irini Eleftheriou; *writing—review and editing*: Foteini Dasoula, Dimitra Dimopoulou, Evangelia Lebessi, Eftihia Serafi, Nikos Spyridis, Maria Tsolia; *supervision*: Maria Tsolia.

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